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Influences of Genetic Variation of Endothelin-1 Gene on Effects of 16-Week Combined Exercise on Clinical and Physical Parameters in Middle-Aged Women

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Key Words: *combined exercise; endothelin-1; Korean population; physical fitness; women.*

Summary. *Background and Objective. In this study, we attempted to determine whether the C/T polymorphism of the ET-1 gene was significantly associated with changes in several clinical characteristics after the 16-week combined exercise program in overweight middle-aged women.*

Material and Methods. The C/T polymorphism of the ET-1 gene was assayed using polymerase chain reaction, i.e., the TaqI restriction fragment length polymorphism method.

Results. The genetic variation in the ET-1 gene showed a significant association with the serum LDL cholesterol level as well as several parameters of physical fitness, including muscular endurance and power of the participants ($P<0.05$). In addition, this genetic variation showed a significant association with changes in muscular strength of the participants after the 16-week combined exercise program ($P<0.05$).

Conclusions. Our data suggest that the C/T substitution on intron 4 of the ET-1 gene may be a useful genetic marker influencing muscular strength through a gene-exercise interaction and is associated with an interindividual difference of cardiovascular risk factors and parameters of physical fitness.

Introduction

Morbidity and mortality rates from cardiovascular diseases are high among the general population as those from cancer; it is also a serious threat to public health resulting in significant medical costs and lowering life expectancy of patients. Therefore, many studies on the causes, diagnosis, and cure for cardiovascular diseases are being conducted throughout the world (1).

Cardiovascular diseases including essential hypertension, coronary artery disease, angina pectoris, and myocardial infarction are multifactorial diseases resulting from a long-term interaction of various genetic components and environmental factors. Thus, the cause and the condition of the outbreak differ significantly among individuals, creating difficulties in the investigation of the cause and in the development of the treatment (2).

Dysfunction of the vascular wall is known to be one of the major factors related to the outbreak of cardiovascular diseases (3). Vasoactive substances are produced and secreted in the innermost layer of blood vessels in order to regulate contraction and expansion of blood vessels (4).

Endothelin-1 (ET-1), a vasoactive substance, is a peptide that represents the function of strong vasocontraction (5–7). The ET-1 gene encoding this peptide has been noted as a strong candidate gene related to the pathogenesis of various cardiovascular diseases, including essential hypertension and atherosclerosis, through experimental changes related to the structure and the function of the vascular endothelium (8).

The human ET-1 gene is located at 6p23-24 region of human chromosome 6, reported as having constructed from 5 exons and 4 introns (5). In addition, within the ET-1 gene, the existence of several polymorphisms has been confirmed, including the C/T substitution located on intron 4; the existence of such a polymorphism provides means for its use as a genetic marker for the examination whether the ET-1 gene is the causative gene of the outbreak of various clinical diseases (9). According to the results of various genetic epidemiological research studies conducted to date, the polymorphisms of the ET-1 gene have been reported to show a relationship with various clinical diseases, including essential hypertension; however, not all studies have presented congruent results (10–18).

In general, aerobic exercise is widely known for the fact that it positively influences the cardiovascular function by improving serum lipid profiles, insulin sensitivity, and body composition related to obesity. In the case of performing combined exercise

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by conducting aerobic exercise along with strength training, it is known that a complementary health-improving effect could be achieved by the improvement of the musculoskeletal system, which cannot be improved by aerobic exercise only (19).

However, this health improvement effect has been known to be interindividually different, and it is predicted that at least genetic predisposition has partial effects (20).

Thus, to our knowledge, we first investigated the distribution of the C/T substitution in the ET-1 gene in the general Korean population. Secondly, we tried to investigate the relationship between the C/T substitution of the ET-1 gene and various risk factors for cardiovascular diseases as well as physical fitness by selecting a homogeneous group of obese middle-aged women as the participants to engage in combined exercise for a period of 16 weeks.

Material and Methods

Subjects. A total of 420 unrelated Korean individuals, including 153 men (average age, 52.4 years [SD, 11.2]) and 267 women (average age, 54.8 years [SD, 13.6]), were recruited in order to investigate the distribution of the polymorphism due to the C/T substitution within the ET-1 gene in the general Korean population. Among them, 79 middle-aged women (average age, 46.6 years [SD, 12.0]) voluntarily agreed to participate in the combined exercise program for a period of 16 weeks.

Genotyping. A Miniban Automatic Blood DNA Isolation Kit (Bionex, Co. Ltd., Korea) was used for the isolation of total genomic DNA from the venous blood of the study subjects, and isolated total genomic DNA was used for the analysis of the polymorphism due to the C/T substitution of the ET-1 gene.

The analysis of the polymorphism of the ET-1 gene was performed using polymerase chain reaction (PCR) and *Taq*I, a restriction enzyme. In the case of PCR, 50 μL of amplicon, including approximately 150 ng of total genomic DNA isolated from the study participants, 10 pmol of 1 pair of primers, 200 μM of 4 dNTPs (dATP, dCTP, dTTP, and dGTP), and 5 μL of 10× buffer made by the manufacturer was placed in a Perkin-Elmer 9700 thermal cycler, and the reaction proceeded. The sequences of the primer set used to carry out this reaction were as follows: forward 5'-CAAACCGATGTCCTCTGTA-3' and reverse 5'-ACCAAACACATTTCCCTATT-3' (21).

PCR was first performed at 1 cycle, including 3 minutes at 94°C, 45 seconds at 94°C, 1 minute at 60°C, and 1 minute at 72°C. There were a total of 35 cycles, and finally, after carrying out an extension reaction for 10 minutes at 72°C, all the processes were ended.

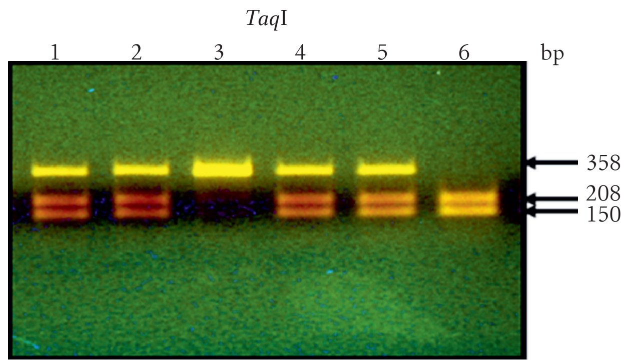

Fig. C/T substitution of the ET-1 gene
Lanes 1–2 and 4–5, CT genotypes; lane 3, CC genotype; lane 6, TT genotype.

The final PCR products were digested with 10 units of *Taq*I and then incubated for approximately for 6 hours at 65°C in a dry oven in order to end the digestion reaction.

The PCR products treated with the restriction enzyme were finally analyzed by using electrophoresis with the 2.5% agarose gel with the addition of ethidium bromide. The study subjects who showed a 358-bp band pattern by electrophoresis were considered as having the CC genotype and those who showed 280-bp and 150-bp band patterns were considered as having the TT genotype (Fig.). In addition, the study subjects with 358-bp, 208-bp, and 150-bp band patterns were considered as having the CT genotype (22).

Combined Exercise Program. As noted, a total of 79 middle-aged women participated in the combined exercise program; the subjects included obese people with more than 30% body fat (Table 1). The combined exercise program included aerobic and resistance exercises, where the aerobic exercise was treadmill walking exercise with an intensity of approximately 50%–60% of maximum oxygen consumption (VO_{2max}) performed 3 days per week for 30 minutes per day. The resistance exercise was dumbbell exercise performed 3 days per week for 30 minutes per day at the level of approximately 50%–60% of 1 repetition maximum. The number of dumbbell exercises was increased from initial 20 times, setting a repeating set as 2 sets for the first 2 weeks and as 3 sets later. A 3- to 5-minute break was given between sets.

Among the subjects participating in the combined exercise program, some individuals quited the program, and the sample size at the end of the exercise program was reduced in comparison with the initial stage.

Measurement of Body Composition and Physical Fitness. The anthropological parameters and the body composition of the test subjects participating in the combined exercise program were measured

Table 1. Clinical Characteristics of 79 Women Who Participated in a Combined Exercise Program

Variable	N	Mean (SD)
Age, years	79	46.6 (12.0)
Height, cm	78	158.1 (5.9)
Weight, kg	78	69.7 (13.0)
BMI, kg/m^2	78	31.5 (32.7)
Fat mass, kg	64	31.0 (37.3)
Percentage body fat, %	79	34.7 (6.2)
Muscular mass, kg	64	42.3 (7.5)
SBP, mm Hg	78	128.4 (17.4)
DBP, mm Hg	78	78.5 (12.4)
VO_{2max}, mL/kg/min	62	36.8 (10.3)
Muscular strength, kg	50	32.0 (6.4)
Muscular endurance, count/min	51	9.8 (5.6)
Power, cm	50	17.5 (10.7)
Flexibility, cm	66	10.0 (7.7)
Agility, ms	51	366.4 (116.0)
Balance, s	51	18.5 (21.9)
TC, mg/dL	79	205.2 (38.9)
TG, mg/dL	79	143.1 (88.3)
HDL cholesterol, mg/dL	64	61.1 (8.4)
LDL cholesterol, mg/dL	64	114.6 (31.7)
Glucose, mg/dL	79	98.9 (19.6)

Data are expressed as mean (standard deviation).
BMI, body mass index; SBP, systolic blood pressure; DBP, diastolic blood pressure; VO_{2max}, maximum oxygen consumption; TC, total cholesterol; TG, triglyceride; HDL, high-density lipoprotein; LDL, low-density lipoprotein.

using In Body 4.0 (Biospace, Co., Korea). Systolic blood pressure (SBP) and diastolic blood pressure (DBP) were measured using a mercury sphygmomanometer. In addition, 7 types of physical fitness factors, including cardiopulmonary endurance, muscular strength, muscular endurance, flexibility, instantaneous reactionary force, agility, and balance, were also measured. For cardiopulmonary endurance, the test subjects were attached with a 12-lead electrode on a treadmill, and the resting heart rate was measured, followed by the measurement of maximum oxygen consumption (VO_{2max}) using the Bruce protocol method wearing a Hans Rudolph mask. Muscle strength was measured with a hand gripper (Takei Co. Ltd., Japan). Muscular endurance within 1 minute was measured by performing a number of sit-ups; flexibility, by left anteversion; and instantaneous reactionary force, by the Sargent jump test. For agility, the whole body reaction time was measured, and for balance, the time standing on one foot with the eyes closed was measured.

Biochemical Assay. Approximately, 3–5 mL of blood was drawn from the antecubital vein of the subjects who had been in a fasting state for approximately 12–14 hours. Serum total cholesterol (TC) and triglyceride levels were measured using the cholesterol oxidase and glycerol phosphate oxidase enzymatic methods, respectively. The serum high-density lipoprotein (HDL) cholesterol level was measured using the dextran sulfate $MgCl_2$ sedimentation method. The serum low-density lipoprotein (LDL) cholesterol level was measured using the Friedewald equation (23). In addition, the serum glucose level was measured using the glucose oxidase method. All biochemical assays of the study participants were performed using a Hitachi 7180 automatic analyzer with enzyme treatment, except for the serum LDL cholesterol level.

Statistical Analysis. The analysis of the mean difference between the parameters of body composition, physical fitness, and serum biochemical parameters before and after the participation in the combined exercise program was performed using the paired *t* test. Deviation from the Hardy-Weinberg equilibrium and the difference in genotype and allele frequencies of the ET-1 gene polymorphism between the groups of the men and the women were analyzed by the χ^2 test. In addition, the differences in the effect of combined exercise on body composition, parameters of physical fitness, and serum biochemical parameters by each of the 3 genotypes composing the polymorphism in the ET-1 gene were tested by the unpaired *t* test for statistical significance by addition of the study participants with the CC genotype and the CT genotype, as the number of the study participants with the CC genotype was low. A multiple stepwise linear regression analysis was performed in order to evaluate the influence of the C/T polymorphism of the ET-1 gene on the change in muscle strength following the combined exercise after controlling for several independent variables. The following independent variables were considered: age, height, weight, weight change, body mass index (BMI), BMI change, percentage body fat, percentage body fat change, fat mass, fat mass change, muscle mass, and change in muscle mass. In addition, the polymorphism of the ET-1 gene was coded as follows: 0=CC+CT, 1 = TT. Statistical significance was determined by the α level of P=0.05, and all the statistical analyses were performed using the SPSSWIN version 17.0 software.

Results

Endothelin-1 Gene Polymorphism. Table 2 shows the genotype and allele distributions for the C/T substitution of the ET-1 gene in our study subjects. After studying the frequency of the genotype in

Table 2. Genotype and Allele Frequencies of C/T Substitution in the Endothelin-1 Gene

	Genotype			Allele	
	CC	CT	TT	C	T
Male	6 (3.9)	70 (45.8)	77 (50.3)	82 (26.8)	224 (73.2)
Female	15 (5.6)	107 (40.1)	145 (54.3)	137 (25.7)	397 (74.3)
Total	21 (5.0)	177 (42.1)	222 (52.9)	219 (26.1)	621 (73.9)
χ^2		1.5951		0.0790	
df		2		1	
P		0.4504		0.7786	

Values are number (percentage).
Observed genotype distributions were in the Hardy-Weinberg equilibrium (χ^2=3.6520, *df*=1, *P*=0.0560).

this polymorphism, the frequencies of the CC, CT, and TT genotypes were 5.0%, 42.1%, and 52.9%, respectively, suggesting that the TT genotype was the most prevalent, while the CC genotype was the least prevalent. The allele frequencies of this polymorphism were 26.1% for the C allele and 73.9% for the T allele, suggesting a higher frequency of the T allele than the C allele. No significant differences in the genotype and the allele distribution of this polymorphism in the ET-1 gene were observed between men and women (for the genotype distribution, χ^2=1.5951, *df*=2, *P*=0.4504; for the allele distribution, χ^2=0.0790, *df*=1, *P*=0.7786). In addition, the observed genotype distribution was in the Hardy-Weinberg equilibrium (χ^2=3.6520, *df*=1, *P*=0.0560).

Effects of Combined Exercise Program. The analysis of changes in body composition and clinical parameters of obese middle-aged women who participated after the 16-week combined exercise program showed a significant decrease in body weight, body mass index, fat mass, percentage body fat, systolic blood pressure, diastolic blood pressure, and serum glucose level (*P*<0.05). In addition, a significant decrease was observed in muscular endurance, flexibility, and serum HDL cholesterol level (*P*<0.05) (Table 3).

Influences of ET-1 Gene Polymorphism on Effects of Combined Exercise Program. The comparison of baseline values and how they changed after the 16-week combined exercise program between the different genotype groups showed that there were no significant differences in any body composition or hemodynamic parameters (Table 4). However, there were significant differences in some parameters of physical fitness, namely muscular strength, muscular endurance, and power (Table 5). The study participants with the CC+CT genotype showed a significantly higher improvement in muscular strength after the 16-week combined exercise program than those with the TT genotype.

After entering the C/T polymorphism of the ET-1 gene, age, height, weight, weight change, body mass index, BMI change, percentage body fat, percentage body fat change, fat mass, fat mass change, muscle mass, and change in muscle mass as independent variables in the multiple stepwise regression analysis, the C/T polymorphism of the ET-1 gene was found to be an independent predictor of a change in muscular strength (adjusted R^2=0.157, *F*=4.733, *P*=0.042).

The comparison of baseline values of serum biochemical parameters and how they changed after the 16-week combined exercise program between the different genotype groups showed that there were no significant differences in any parameter except for the baseline serum LDL cholesterol level (Table 6).

Discussion

This study was carried out in order to investigate the distribution of the C/T substitution within the ET-1 gene in the general Korean population and to compare the clinical difference after the participation in the 16-week combined exercise program according to the C/T substitution in the ET-1 gene.

The study of the frequencies of the C and T alleles in the ET-1 gene polymorphism in 420 unrelated Korean subjects demonstrated that the frequency of the C allele was 26.1%, while the frequency of the T allele was 73.9%. This result suggests that within the Korean population, there is a higher frequency of the T allele in comparison with the C allele; these results were similar to those of other results

Table 3. Changes in Body Composition and Clinical Parameters Following 16-Week Combined Training

Variable	N	Pre	Post	*t* value	*P*
Height, cm	29	158.6 (5.6)	158.7 (5.6)	–1.000	0.326
Weight, kg	44	69.2 (14.9)	68.0 (13.9)	3.306	0.002
BMI, kg/m^2	44	28.1 (5.6)	27.4 (5.4)	3.779	<0.000
Fat mass, kg	30	28.5 (9.9)	27.2 (9.7)	2.803	0.009
Percentage body fat, %	45	34.4 (7.1)	32.5 (8.2)	5.623	<0.001
Muscular mass, kg	30	42.5 (8.6)	43.0 (6.3)	–0.456	0.652
SBP, mm Hg	44	131.3 (15.9)	125.6 (14.0)	2.781	0.008
DBP, mm Hg	44	80.1 (11.9)	76.4 (8.2)	2.238	0.030
VO_{2max}, mL/kg/min	32	33.5 (11.8)	34.7 (9.4)	–1.162	0.254
Muscular strength, kg	22	32.8 (6.0)	32.1 (5.3)	0.764	0.454
Muscular endurance, count/min	23	8.3 (6.0)	10.4 (6.8)	–3.436	0.002
Power, cm	22	15.3 (6.8)	16.5 (6.5)	–1.212	0.239
Flexibility, cm	38	10.1 (7.5)	15.8 (9.5)	–4.655	<0.001
Agility, ms	23	414.6 (139.2)	368.8 (96.8)	1.577	0.129
Balance, s	23	19.2 (24.1)	16.8 (15.8)	0.591	0.561
TC, mg/dL	45	201.2 (44.4)	193.0 (36.4)	1.398	0.169
TG, mg/dL	45	142.2 (60.7)	113.7 (43.3)	3.739	0.001
HDL cholesterol, mg/dL	30	59.0 (7.0)	52.8 (7.5)	5.284	<0.001
LDL cholesterol, mg/dL	30	109.9 (30.6)	115.6 (31.2)	–1.002	0.325
Glucose, mg/dL	45	101.2 (22.1)	95.8 (18.1)	2.274	0.028

Values are expressed as mean (standard deviation).
BMI, body mass index; SBP, systolic blood pressure; DBP, diastolic blood pressure; VO_{2max}, maximum oxygen consumption; TC, total cholesterol; TG, triglyceride; HDL, high-density lipoprotein; LDL, low-density lipoprotein.

Table 4. Body Composition and Hemodynamic Parameters at Baseline and Their Changes Following a 16-Week Combined Training Program According to Genotypes of C/T Substitution in the Endothelin-1 Gene

Variable	CC + CT		TT		*t* value	*P*
	N	Mean (SD)	N	Mean (SD)		
Height, cm						
Baseline	34	157.1 (5.1)	44	158.9 (6.4)	–1.370	0.175
Change	14	0.0 (0.0)	15	0.2 (0.8)	–1.000	0.334
Weight, kg						
Baseline	34	70.0 (14.7)	44	69.5 (11.7)	0.193	0.848
Change	22	–2.2 (2.8)	22	–1.0 (3.5)	–1.153	0.225
BMI, kg/m^2						
Baseline	34	28.4 (5.7)	44	33.9 (43.3)	–0.732	0.466
Change	22	–0.9 (1.1)	22	–0.4 (1.2)	–1.241	0.221
Fat mass, kg						
Baseline	26	28.0 (10.5)	38	33.0 (47.8)	–0.522	0.603
Change	14	–1.7 (2.3)	16	–1.0 (2.9)	–0.661	0.514
Percentage body fat, %						
Baseline	34	35.9 (6.3)	45	33.8 (6.0)	1.531	0.130
Change	22	–2.2 (2.4)	23	–1.6 (2.4)	–0.942	0.351
Muscular mass, kg						
Baseline	26	40.4 (7.4)	38	43.6 (7.3)	–1.752	0.085
Change	14	1.4 (8.3)	16	–0.3 (2.1)	0.804	0.428
SBP, mm Hg						
Baseline	33	129.0 (14.7)	45	128.0 (19.2)	0.257	0.798
Change	21	–6.1 (12.3)	23	–5.3 (15.0)	–0.180	0.858
DBP, mm Hg						
Baseline	33	78.5 (11.0)	45	78.5 (13.5)	–0.006	0.995
Change	21	–4.6 (9.2)	23	3.0 (12.8)	–0.491	0.626

BMI, indicates body mass index; SBP, systolic blood pressure; DBP, diastolic blood pressure.

Table 5. Physical Fitness Parameters at Baseline and Their Changes Following a 16-Week Combined Training Program According to Genotypes of C/T Substitution in the Endothelin-1 Gene

Variable	CC + CT		TT		*t* value	*P*
	N	Mean (SD)	N	Mean (SD)		
VO_{2max}, mL/kg/min						
Baseline	26	37.3 (11.4)	36	36.4 (9.7)	0.326	0.745
Change	15	2.5 (5.0)	17	0.2 (7.1)	1.048	0.303
Muscular strength, kg						
Baseline	19	30.6 (5.0)	31	32.9 (7.1)	–1.199	0.237
Change	12	1.1 (3.9)	10	–2.9 (4.2)	2.293	0.033
Muscular endurance, count/min						
Baseline	20	7.2 (4.9)	31	11.5 (5.4)	–2.851	0.006
Change	13	1.4 (2.2)	10	2.9 (3.5)	1.281	0.214
Power, cm						
Baseline	20	13.3 (10.5)	30	20.3 (10.1)	–2.356	0.023
Change	12	0.8 (4.1)	10	1.7 (5.3)	–0.476	0.639
Flexibility, cm						
Baseline	28	9.2 (7.6)	38	10.5 (7.8)	–0.673	0.503
Change	21	7.1 (9.4)	17	4.0 (4.2)	1.262	0.215
Agility, m						
Baseline	20	374.4 (127.3)	31	361.2 (110.0)	0.394	0.695
Change	13	–16.8 (150.6)	10	–83.4 (119.5)	–1.147	0.264
Balance, s						
Baseline	20	15.8 (19.9)	31	20.3 (23.3)	–0.710	0.481
Change	13	–2.8 (21.9)	10	–1.9 (17.7)	–0.111	0.912

VO_{2max}, maximum oxygen consumption.

obtained not only in the Korean population but also in a Caucasian population (9, 21–22).

After the analysis of the effect of the 16-week combined exercise program in 79 postmenopausal obese women, no improvement was observed in weight, body mass index, body fat, and percentage body fat as well as in SBP and DBP. In addition, among 7 parameters of physical fitness, muscular endurance and flexibility showed an improvement, and in the case of biochemical parameters, although serum HDL cholesterol decreased, the serum TG and glucose levels also decreased, suggesting that our study participants showed actual health-promoting effects through the improvement on several

Table 6. Biochemical Parameters at Baseline and Their Changes Following a 16-Week Combined Training Program According to Genotypes of C/T Substitution in the Endothelin-1 Gene

Variable	CC + CT		TT		*t* value	*P*
	N	Mean (SD)	N	Mean (SD)		
TC, mg/dL						
Baseline	34	211.8 (43.6)	45	200.2 (34.5)	1.319	0.191
Change	22	–3.0 (48.5)	23	10.2 (29.5)	–1.110	0.273
TG, mg/dL						
Baseline	34	148.3 (61.0)	45	139.1 (104.9)	0.454	0.651
Change	22	13.5 (65.2)	23	6.6 (50.8)	0.399	0.692
LDL-cholesterol, mg/dL						
Baseline	26	125.4 (27.9)	38	107.2 (32.4)	2.329	0.023
Change	14	–2.8 (34.9)	16	13.2 (26.4)	–1.429	0.164
HDL-cholesterol, mg/dL						
Baseline	26	61.1 (7.8)	38	61.0 (8.9)	0.041	0.967
Change	14	–6.4 (7.5)	16	–6.2 (5.7)	–0.070	0.945
Glucose, mg/dL						
Baseline	34	96.8 (18.3)	45	100.4 (20.7)	–0.799	0.427
Change	22	3.3 (12.9)	23	5.3 (18.9)	–0.409	0.684

TC, total cholesterol; TG, triglyceride; HDL, high-density lipoprotein; LDL, low-density lipoprotein.

parameters related to cardiovascular disease.

However, such clinical changes caused by the combined exercise program indicated interindividual differences; such interindividual differences may be caused, at least in part, by genetic predisposition (20). Thus, in order to explore genetic factors affecting interindividual differences in clinical changes from the combined 16-week exercise program, this study used the C/T substitution in the ET-1 gene as a genetic marker and attempted to determine whether this polymorphism could affect changes in clinical parameters resulting from the participation in the combined exercise program of 16 weeks.

According to the analysis, this polymorphism of the ET-1 gene did not indicate a remarkable effect on changes in body composition and blood pressure values but showed a significant effect on changes in muscular strength after the participation in the combined exercise program as well as in baseline muscular endurance and power. In the case of muscular endurance and power, the study participants with the TT genotype had higher baseline values of physical endurance and power compared with those with the CC and CT genotypes. However, in the case of muscular strength, the study subjects with the CC and CT genotypes showed a noticeable increase in muscular strength after the 16-week combined exercise program, while those with the TT genotype actually showed a decrease in muscular strength, suggesting the usefulness of this polymorphism as a genetic marker that shows an interindividual difference in muscular strength after the 16-week combined exercise program. Also, in the multiple regression analysis including the C/T polymorphism of the ET-1 gene, age, height, weight, weight change, BMI, BMI change, percentage body fat, percentage body fat change, fat mass, fat mass change, muscle mass, muscle mass change as independent variables, only the C/T polymorphism of the ET-1 gene was considered as a unique predictor with statistical significance.

To date, a precise mechanism related to the influence of this polymorphism on baseline muscular endurance or power or changes in muscular strength after the 16-week combined exercise program is still unknown. As a probable mechanism, posttranscriptional processing such as RNA stability and RNA splicing may be considered. In addition, because this polymorphism is located within intron 4 site of the ET-1 gene, the possibility that such an effect is indicated through linkage disequilibrium with the causative allele that exerts the actual effect on the ET-1 gene expression or its function must not be excluded (9). Nevertheless, our data suggest that the statistical power for the C/T polymorphism of the ET-1 gene was modest. It is unlikely that a genetic marker such as the C/T polymorphism of the ET-1 gene may strongly contribute to the change in muscular strength of the participants following our combined exercise program because the change in muscular strength might be due to a complex interaction of various genetic and environmental factors. Therefore, further molecular biological studies under various experimental conditions will be conducted in order to test these possibilities.

Regarding serum biochemical parameters, this polymorphism showed a significant association only with the baseline serum LDL cholesterol level, and the study subjects with the TT genotypes showed a higher serum LDL cholesterol level than those with the CC and CT genotypes. These results suggest the possibility of this polymorphism in the ET-1 gene as a useful genetic marker related to the risk of cardiovascular disease because harmful changes in serum lipid profiles such as the serum LDL cholesterol level may be considered as risk factors of various cardiovascular diseases. To date, there has been no known report on the effect of this polymorphism on serum lipid profiles; however, the Lys198Asn polymorphism as another genetic variation in the ET-1 gene has shown a significant association with the serum HDL cholesterol level (16). Although no exact mechanism is known, ET-1 has been reported to increase insulin resistance and decrease the transport of glucose to skeletal muscles (24–25). Therefore, this polymorphism is assumed to be one of the components affecting the serum LDL cholesterol level and acts as a factor related to insulin resistance. However, regarding this issue, further investigations are needed.

Conclusions

In summary, the C/T substitution of the ET-1 gene showed a significant association with not only the baseline serum LDL cholesterol level and the several baseline parameters of physical fitness, such as muscular endurance and power, but also with the changes in muscular strength after the 16-week combined exercise program for obese middle-aged women. However, as there is no exact mechanism known for this polymorphism to have such associations, further studies to clarify our finding are believed to be necessary. Moreover, as the sample size in this study was not large enough to conduct a genetic epidemiological study, further investigations using a larger sample size will be required.

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Statement of Conflict of Interest

The authors state no conflict of interest.

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