

한국인 다낭성 난소증후군 환자에서 CYP11 α 유전자 (tttta)_n 다형성 양상 및 역할

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CYP11 α (tttta)_n Microsatellite Polymorphism in Korean Patients with Polycystic Ovary Syndrome

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Objectives: To investigate the distribution and functional significance of CYP11 α (tttta)_n microsatellite polymorphism in Korean patients with polycystic ovary syndrome

Materials and Methods: Analysis of CYP11 α (tttta)_n microsatellite polymorphism was carried out on DNA samples from 97 patients with polycystic ovary syndrome and 70 normal controls. Comparison were done between PCOS patients and controls concerning CYP11 α (tttta)_n microsatellite polymorphism genotype or allele frequencies.

Results: The most frequent allele observed in the controls was an allele with six repeats (60.7%). Significant difference in the frequency of genotype (4R (-) genotype) having no copy of four-repeat-allele were observed between PCOS patients and controls (66.0% vs 34.0%, p=0.038, OR=1.939). But no significant difference was observed in the serum levels of total testosterone or free testosterone between 4R (+) genotype and 4R (-) genotype among PCOS patients. However, hyperandrogenic PCOS patients with 4R (+) genotype showed a higher serum testosterone levels compared to controls (mean \pm S.D: 0.49 \pm 0.21 ng/ml vs 0.37 \pm 0.18 ng/ml, p=0.037).

Conclusion: The allelic distribution of CYP11 α (tttta)_n microsatellite polymorphism in Korean subjects were different from those reported in Caucasians. CYP11 α (tttta)_n microsatellite polymorphism was associated with polycystic ovary syndrome in the Korean population, and may play a role in the synthesis of androgens in patients with polycystic ovary syndrome.

Key Words: CYP11 α gene, Hirsutism, Hyperandrogenism, Polycystic ovary syndrome, Microsatellite polymorphism, P450scc

가 4~7%

가

2

가

1.

2004 3 2004 10

97

2003 ARSM/ESHRE consensus meeting guideline (>35)

(

2~9 mm 가 12

가 10 cm³) 2

⁴ 2002 3 2004 10

(21 ~35) 가

70

P450sc (P450 side chain cleavage enzyme) coding CYP11 α

2.

1)

¹⁻³ CYP11 α

cholesterol side chain cleavage enzyme steroid rate limiting step , cholesterol pregnenolone

hirsutism score (modified Ferriman-Gallway) 8

BMI waist-to-hip ratio 3

528 bp (upstream) (ttta)_n microsatellite (polymorphism) LH, FSH, estradiol, TSH, PRL

total testosterone free testosterone 0.2~0.8 ng/ml, free testosterone 2.0 pg/ml

가

CYP11 α

가

2

nonclassic congenital adrenal hyperplasia 17- α hydroxy progesterone (OHP)

CYP11 α

CYP11 α

Table 1. Clinical characteristics of patients with PCOS and controls

	PCOS (n=97)	Control (n=70)	p value
Age (years)	26.0±5.4	32.6±4.8	<0.01
BMI (kg/m ²)	22.4±5.3	21.5±3.1	n.s.
Waist-to-hip ratio	0.77±0.07	0.77±0.08	n.s.
Total testosterone (ng/ml)	0.38±0.18	-	
Free testosterone (pg/ml)	1.73±1.13	-	

All values are means ± SD. BMI: Body mass index. n.s.: Not significant

2) (PCR)
 3 ml Wizard genomic DNA extraction kit (Promega)
 PCR 50 µl genomic DNA 0.1 µg, 10 mM Tris-HCl, pH 8.3, 50 mM KCl, 1.5 mM MgCl₂, 200 µM dNTPs, upstream primer downstream primer 500 nM, 1 U Taq polymerase
 PCR oligonucleotide primer upstream 6FAM-5'GGT GAA ACT GTG CCA TTG C3', downstream 5'CTG TAC CTG CTC CAC TTC AGC3' PCR 94
 1 denaturation 61 1 anealing 72 1 extension 35 (fluorescent PCR), gene scan analysis

homozygous sequencing (tttta)_n repeat , repeat 가 6 221 bp 4 repeat

3) CYP11α (tttta)_n (alleles) CYP11α (tttta)_n

CYP11α (tttta)_n testosterone , SPSS 11.5 package Student T test, chi square Fisher's exact test , p<0.05 (odds ratio, OR) Woolf , 가 0 Haldane's modification

97 25.9 ±5.4 (SD) , BMI 22.7±4.7 (SD) kg/m², waist-to-hip ratio 0.77±0.07 (SD) 70 32.4±4.7 , BMI 21.5 ±3.1 kg/m², waist-to-hip ratio 0.77±0.08 (Table 1). BMI waist-to-hip ratio 가 , (p<0.01). 5가 (alleles) 가 (Table 2). homozygous (genotypes) sequencing , 6 repeat (6R) PCR 221 bp 4R, 5R, 6R, 8R, 9R 22.5%, 0.3%, 65.0%, 11.7%, 0.6%

(Table 3). 4R 4R (+) 4 repeat 4R (-) 4 repeat 97 64 (66.0%) 4R (-) , 70 35 (50.0%) 4R (-) , 4R (-) 가 (p=0.038, OR=1.939, 95% CI 1.034~3.639) (Table 4).

Table 2. CYP11 α (ttta)_n microsatellite polymorphism genotypes in patients with PCOS and controls

Genotype	PCOS (n=97) (%)	Control (n=70) (%)	OR (95% confidence interval)
4/4	4 (4.1%)	3 (4.3%)	0.961 (0.208~4.434)
4/6	24 (24.7%)	25 (35.7%)	0.592 (0.302~1.159)
4/8	5 (5.2%)	7 (10.0%)	0.489 (0.149~1.610)
4/9	0 (0%)	0 (0%)	-
5/6	1 (1.0%)	0 (0%)	2.192 (0.226~21.290)
6/6	46 (47.4%)	27 (38.6%)	1.436 (0.769~2.638)
6/8	13 (13.4%)	6 (8.6%)	1.651 (0.595~4.580)
6/9	2 (2.1%)	0 (0%)	3.615 (0.418~31.285)
8/8	2 (2.1%)	2 (2.9%)	0.716 (0.098~5.208)
8/9	0 (0%)	0 (0%)	-

OR: Odds ratio

Table 3. Allelic frequencies of CYP11 α (ttta)_n microsatellite polymorphism of in patients with PCOS and controls

Repeat number	Total (n=334) (%)	PCOS (n=194) (%)	Control (n=140) (%)	OR (95% confidence interval)
4	75 (22.5%)	37 (19.1%)	38 (27.1%)	0.633 (0.377~1.061)
5	1 (0.3%)	1 (0.5%)	0 (0%)	2.178 (0.225~21.05)*
6	217 (65.0%)	132 (68.0%)	85 (60.7%)	1.378 (0.875~2.169)
8	39 (11.7%)	22 (11.3%)	17 (12.1%)	0.925 (0.472~1.816)
9	2 (0.6%)	2 (1.0%)	0 (0%)	3.649 (0.424~31.41)*

*Haldane's modification method

Table 4. Genotype distribution of CYP11 α (ttta)_n microsatellite polymorphism by the presence or absence of allele (s) with 4 repeats in patients with PCOS and controls

Study group	Genotype		OR (95% confidence interval)	p value*
	4R (-)	4R (+)		
PCOS (n=97)	64 (66.0%)	33 (34.0%)	1.939 (1.034~3.639)	0.038
Hyperandrogenic PCOS (n=51)	33 (64.7%)	18 (35.3%)	1.833 (0.874~3.847)	0.107
Control (n=70)	35 (50.0%)	35 (50.0%)		

*Compared to control

4R (+) , BMI, waist-to-hip ratio, testosterone, free testosterone, 37 (modified Ferriman-Gallwey score >8), 4R (-), 4R (+), rone >0.8 ng/ml, free testosterone >2.0 pg/ml

Table 5. Anthropometric characteristics and serum testosterone levels of patients with PCOS and controls according to their genotypes

Study group	Genotype	Age (years)	BMI (kg/m ²)	Waist-to-hip ratio	Total testosterone (ng/ml)	Free testosterone (pg/ml)
PCOS	4R (-) (n=64)	26.3±5.6	22.1±5.8	0.77±0.07	0.35±0.16	1.70±1.13
	4R (+) (n=33)	25.2±4.9	23.0±4.3	0.77±0.07	0.42±0.20	1.81±1.14
Hyperandrogenic PCOS	4R (-) (n=33)	25.1±5.7	23.0±5.3	0.78±0.07	0.37±0.18*	2.21±1.20
	4R (+) (n=18)	24.7±4.6	23.2±4.2	0.77±0.06	0.49±0.21*	2.22±1.30
Control	4R (-) (n=35)	32.5±5.1	21.3±3.3	0.79±0.10	-	-
	4R (+) (n=35)	32.7±4.6	21.8±2.9	0.76±0.05	-	-

All values are means ± SD. *p=0.037

, 4R (-) 64.7% 1997 Gharani 69
(33/51) 50.0%
(p=0.107) (Table 4). 28 , 42
4R (-) 4R 27
(+) , BMI, waist-to-hip ratio cholesterol side chain cleavage enzyme (cytochrome
free testosterone , P450scc) aromatase (cytochrome P450arom) co-
4R (+) testosterone 가 0.49± ding
0.21 (mean±S.D.) ng/ml 4R (-) 0.37±0.18 ,
ng/ml (p=0.037) (Table 5). , cytochrome P450scc coding CYP11α
(ttta)_n
testosterone
CYP11α 5' regulatory region CYP11α (ttta)_n
(-528 bp) (ttta)_n
CYP11α promoter 4 repeat (-)
CYP11α 가 가 CYP11α
camp-regulated elements 가 CYP11α
5 528 bp (ttta)_n microsate-
tellite , 1997 Franks , 2000 Kandar-
akis 4 repeat (-) ,
(susceptibility locus) ,
1-3 2001 Millan
insulin gene VNTR CYP11α (ttta)_n ,
가 (functional polymorphism) , 7 2004 Gaasenbeek
6 230 , 331 ,

527 , 1062

가 , 3가

testosterone 2가

⁸ 2002 Baek 가

30 26

4 repeat (-)

14 (53.8%) 7 (23.3%) 4 repeat (-)

4 repeat (-)

가 (p= (p=0.107), 가

0.019).⁹

가 4 repeat (+)

가 testosterone 가 0.49±0.21 (mean

4 repeat ±S.D.) ng/ml 4 repeat (-) 0.37±0.18

5 ng/ml (p=0.037). 4

4 repeat repeat (-) 4 repeat (+)

52%~59%

testosterone

4 repeat ¹⁻³ 2001 Millan 92

가 27.1% (38/140), 6 repeat 가 33

60.7% (85/140) 6 repeat 가 가 , , 4

repeat (+) testosterone 가

Baek (2002)

6 repeat 가 42.3% ⁷

(22/52) 가 ,⁹ Zheng

(2004) CYP11α 26.0±5.4 , 32.6±4.8

(ttta)_n (p<0.001).

6 repeat가

가 (65.9%)

¹⁰ CYP11α

(ttta)_n , , 가

4 repeat , , ,

(-) 가 4 repeat

CYP11α (ttta)_n (p=0.038). 가

4 repeat

testosterone free testosterone (ttta)_n CYP11α

6 repeat가 가

4 repeat가 가

가
 CYP11 α (tttta)_n
 CYP11 α (tttta)_n

repressed by 12-O-tetradecanoylphorbol-13-acetate and A23817 through independent cis elements. *Mol Cell Biol* 1990; 10: 6013-23.

- 4 repeat (-)
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