LHCGR gene analysis in girls with Non - clasic Central Precocious Puberty

by

Hwal Rim Jeong

Major in Medicine

Department of Medical Sciences

The Graduate School, Ajou University

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Supervised by

Jin Soon Hwang, M.D., Ph.D.

Major in Medicine

Department of Medical Sciences

The Graduate School, Ajou University

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This certifies that the dissertation

of Hwal Rim Jeong is approved.

SUPERVISORY COMMITTEE

Jin Soon Hwang

Jun Eun Park

Jang Hoon Lee

Da Eun Jung

Min Ho Jung

The Graduate School, Ajou University

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- Abstract-

Background: Luteinizing hormone (LH) is a useful parameter in diagnosing precocious puberty. The pubertal response of serum LH to a GnRH stimulation test is varied, and clinical symptoms of precocious puberty are sometimes disproportionate with serum LH concentrations. Many patients present in a state of precocious puberty that advances rapidly, but the post-GnRH peak LH remains prepubertal. LH receptor mutations are suspected of involvement in the non-classic type of central precocious puberty (CPP).

Objective: To examine the association between LHCGR polymorphism and non-classic CPP in subjects exhibiting a peak LH < 5 IU/L on a GnRH stimulation test.

Methods: In total, 102 girls with non-classic CPP and 100 normal adult women were enrolled. All subjects underwent LHCGR gene analysis by the Sanger method, and patients and controls were compared. Auxological data and gonadotropin concentrations were analyzed in the 102 patients. Of these patients, 75 completed GnRH agonist treatment, and the treatment outcomes were analyzed.

Results: total of seven variants were identified, including two missense mutations (g.48698754 G/A and g.48688613 G/A) that were found in the patient group (no patients contained both mutations). In silico analysis of these missense mutations suggested the possibility of damaging the LHCGR. However, no significant association was found between the identified LHCGR variants and non-classic CPP. GnRH agonist treatment decreased bone age advancement and increased predicted adult height.

Conclusion: LHCGR gene polymorphisms do not appear to be a major causative factor for the relatively low concentration of LH in patients with non-classic CPP. GnRH agonist treatment improved clinical parameters in these patients.

Key words: LHCGR gene, precocious puberty, GnRH agonist

TABLE OF CONTESTS

LIST OF FIGURES

LIST OF TABLES

I. Introduction

Central precocious puberty (CPP) develops with early activation of the hypothalamic pituitary gonadotropin (HPA) axis, and is also known as gonadotropin-dependent precocious puberty or complete precocious puberty. CPP is diagnosed by early pubertal signs, advanced bone age, and pubertal gonadotropin concentrations. The gold standard is the GnRH stimulation test: a peak luteinizing hormone concentration (LH) above 5 IU/L suggests progressive CPP, but there is some overlap between prepubertal and early pubertal values of LH [1, 2].

Some patients present with breast development and advanced bone age, as well as a low LH peak following GnRH stimulation. These patients appear to have clinically progressing CPP, even though the HPA axis is not fully activated; the LH peak post-GnRH stimulation does not satisfy the current diagnostic criteria. This suggests that the current diagnostic criteria of CPP are not perfect and thus are debatable. It may also indicate that overreaction to the weaker LH stimulation plays a major role in this non-classic type of CPP. Non-classic CPP may be associated with luteinizing hormone receptor (LHCGR) mutations, and patients with mutations may present an increased gonadal response to relatively lower LH concentrations post-GnRH, preceding pubertal progression.

The LHCGR gene encodes the luteinizing hormone/choriogonadotropin receptor; this is a seven-transmembrane, G-protein-coupled receptor, and its activity is mediated by G proteins that activate adenylate cyclase. The LHCGR gene encodes a receptor that mediates the actions of both LH and human chorionic gonadotropin. It is found on human chromosome 2p21 and comprises 11 exons [3]. The receptor is found predominantly in the gonads (theca cells of the ovary and Leydig cells of the testicles), uterus, breast, and adipose tissue [4]. Loss-of-function mutations in the LHCGR gene can lead to infertility in females [5] and hypospadia or a micropenis in males [4]. Activating mutations in the LHCGR gene induce familial male-limited precocious puberty, also called testotoxicosis [6–9].

The purpose of this study was to investigate whether an LHCGR gene mutation is associated with non-classic CPP, represented clinically by CPP characteristics but relatively low stimulated LH response, and to assess the efficacy of GnRHa treatment in patients with non-classic CPP.

II. Subjects and Methods

A. Subjects

The research protocol was approved by the ethics committee at Ajou University Hospital. All participants and/or their legal representatives received detailed information about the project and provided written informed consent.

In total, 102 patients with non-classic CPP and 100 healthy adult control women were included. The patient group was selected from the pediatric endocrinology outpatient clinic at Ajou University Hospital. They visited the clinic with breast engorgement and were evaluated for precocious puberty. All patients presented with breast engorgement before 8 years old and advanced bone age (1 year above chronological age, CA). They underwent a GnRH stimulation test and the peak LH response was < 5 IU/L. Their auxological data and endocrine parameters were evaluated. Patients who had been diagnosed with other endocrine diseases were excluded. The control group was recruited by advertising for volunteers. They were normal adult women, older than 20 years, and all reached menarche after 12 years old. All study subjects underwent genetic analyses for the LHCGR gene. All patients were treated with GnRH agonists; among them, 75 patients completed the treatment. The standard treatment regimen was the administration of subcutaneous leuprolide acetate, at a dose of 100 μg/kg, every four weeks.

Height was measured in the orthostatic position using a Harpenden stadiometer. Weight was measured with a calibrated digital scale. Body mass index (BMI) was calculated as weight/height2. The standard deviation scores (SDSs) for height, weight, and BMI were calculated based on the 2007 Korean National Growth Charts [10]. Pubertal development was evaluated according to Tanner's criteria [11]. Bone age was assessed by one endocrinologist using the Greulich and Pyle method [12]. Predicted adult height (PAH) was estimated using the Bayley-Pinneau method [13]. All patients underwent a GnRH stimulation test during the daytime using 100 μg GnRH (Relefact; Sanofi-Aventis, Frankfurt, Germany). Serum LH and FSH concentrations were determined at baseline and at 30, 45, 60, and 90 min after injection of GnRH. The basal estradiol (E2) concentration was measured before injection of GnRH.

Serum LH and FSH concentrations were measured by IRMA (BioSource, Nivelles, Belgium). The detection limits for LH and FSH were 0.1 IU/L and 0.2 IU/L, with an intra-assay coefficient of variation (CV) ranging from 1.4–3.9 % to 1.1–2.0 % and an inter-assay CV ranging from 3.4–8.0 % to 2.4–4.4 %, respectively. E2 concentrations were measured by radioimmunoassay with a detection limit of 5 pg/mL, an intra-assay CV ranging from 4.0–7.0 %, and an inter-assay CV ranging from 4.2–8.1 % (RIA; Coat-A-Count, Diagnostic Products Corp., Los Angeles, CA, USA).

B. Genetic analysis

Genomic DNA was isolated from peripheral blood leukocytes of all subjects using a DNA isolation kit (Qiagen GmbH, Hilden, Germany). The mutations were identified by conventional Sanger sequencing. The LHCGR gene contains one large exon of 1523 base pairs; this was polymerase chain reaction (PCR)-amplified using a cascade strategy. We obtained 11 overlapping fragments using the following 10 primer pairs (exons 5 and 6 were analyzed together because of their small size): F: 5′-CAG AAG GTG TGT GGA-3′ and R: 5′-CAA AGC GTT TTT CTC CAA GC-3′ (exon 1), F: 5′-TCT GAA GTC TCT GTC TCT CAC ACA-3′ and R: 5′-TCC TAA ACA CAA TTT CTT TCC TAG A-3′ (exon 2), F: 5′-CCA GTT GTT GGG TCA CAC AC-3′ and R: 5′-GGA TTC CTA GCA GTG GAT GC-3′ (exon 3), F: 5′-TCC TCC CAA GCA TAT GAA GG-3′ and R: 5′-ACA TGC AAA TAG TGC CAT GTA T-3′ (exon 4), F: 5′-AGT CCT GAT AAC ACC AAA CTC AA-3′ and R: 5′-CCA GTG AGT GAG GAA TGT GG-3′ (exons 5 and 6), F: 5′-GGT GGT GAT GAT GGA GAA CT-3′ and R: 5′-GCC CTG AGT TAG TTG CTG AA-3′ (exon 7), F: 5′-TTA CCC TTT TTC CCC CTT TT-3′ and R: 5′-CAG CAG CCT TTT CAG GTG TA-3′ (exon 8), F: 5′-CTT GCA GTG AGC CGA GGT-3′ and R: 5′-AAG TGA CCC CAT GTC TAC GG-3′ (exon 9), F: 5′- AAG AAT TTG CAT GAA CAA AGC A-3′ and R: 5′-GCA ACA GCT CCG TAA CCA AG-3′ (exon 10), and F: 5′-GGC TGA GGC TAT TAT GGC TTT-3′ and R: 5′-TTT CCT AAA TCC AAC CCT TTA TG-3′ (exon 11). PCR conditions were as follows: initial denaturation at 94 ℃ for 5 min, followed by 34 cycles of 94 ℃ for 1 min, annealing for 1 min, elongation for 1 min, and a final extension at 72 ℃ for 10 min. The PCR mixtures were subjected to electrophoresis on 1.5 % (w/v) agarose gels containing ethidium bromide to confirm that the sizes were

appropriate and that the amplicons were pure. DNA sequencing was performed using the same primer pairs and the BigDye Terminator V3.1 Cycle Sequencing Kit (Applied Biosystems, Foster City, CA, USA) according to the manufacturer's protocol. Sequencing reaction mixtures were electrophoresed and analyzed on an ABI3130xl Genetic Analyzer (Applied Biosystems) running the Sequencing Analysis software (ver. 5.2). PolyPhen-2 (http://genetics.bwh.harvard.edu/pph2/) was used to identify the possible effects of an amino acid substitution on the structural and functional assets of protein [14].

C. Statistical analysis

Statistical analyses were performed using SPSS software (ver. 20.0; SPSS Inc., Chicago, IL, USA). Allele frequencies were compared between the patient and control groups. Deviations from Hardy-Weinberg equilibrium were also evaluated by comparing the observed and expected genotype frequencies. Data are expressed as means ± SD or SDSs. Fisher's exact test and paired t-tests were used for data analysis. A p value < 0.05 was considered to indicate statistical significance.

III. Results

A. Clinical characteristics and results of the GnRH stimulation test in patients

Table 1 summarizes the pretreatment clinical characteristics and laboratory parameters in the patient group. In the patient group, the mean age at diagnosis was 8.37 ± 0.56 years, and breast engorgement was first observed at a mean age of 7.64 ± 1.18 years. The bone age at diagnosis was 10.72 ± 0.42 years, and the bone age advancement was 2.63 ± 0.63 years. The mean Tanner stage for breast development at diagnosis was 2.43 ± 0.68. Height SD and BMI SD at diagnosis were 1.16 ± 0.88 and 0.80 ± 1.04, respectively. The mid-parental height (MPH) SD was − 0.25 ± 0.75. The basal LH value and the post-GnRH peak LH were 1.04 ± 0.42 IU/L and 3.31 ± 0.99 IU/L, respectively. The basal LH/FSH ratio was 0.79 ± 0.50.

B. Identified polymorphisms in the LHCGR gene analysis

Direct sequencing of the LHCGR gene revealed seven polymorphisms (Fig. 1, Table 2). Among the seven polymorphisms, six had been previously reported, whereas the other was apparently novel (g.48687721C/A). The results met Hardy-Weinberg equilibrium expectations. One missense mutation, g.48694236 A/G, and one synonymous substitution, g.48688732 T/C, were found in both groups. These two mutations were found in all subjects in this study; no subjects had the wild-type sequences of g.48694236 A/A and g.48688732 T/T.

The other five polymorphisms were found in five subjects. The g.48698754 G/A (Glu→Lys) and g.48688613 G/A (Arg→His) mutations were found in two patients each. One missense mutation (g.48708972 G/A) was found in one control subject, one insertion (g.48755617ins TGCAGC) was found in another control subject, and a novel mutation (g.48687721 C/A) was detected in one control subject. Regardless of the phenotype group, the most frequent polymorphisms were g.48694236 A/G and g.48688732 T/C (100 %): these two mutations presented with g.48688732 T/C in almost all the same subjects.

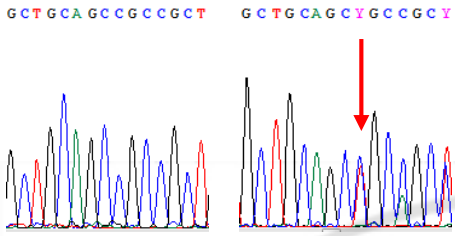
Table 3 lists the allele counts and frequencies in the two groups. Using Fisher's exact test, we evaluated the associations between the polymorphisms and the two phenotypes. No differences in the frequencies of the seven polymorphisms were observed between the two groups. In silico analysis predicted that the g.48698754 G/A (Glu→Lys) and g.48688613 G/A (Arg→His) might be damaging.

C. Clinical response to GnRH agonist treatment in the patient group

All patients received treatment with GnRH agonist, 75 of whom completed the treatment. Their auxologic parameters before and after treatment are shown in Table 4. The pre-treatment mean stimulated peak LH and peak FSH concentrations were 3.33 ± 1.02 and 10.24 ± 4.22 U/L, respectively. The mean treatment period was 2.81 ± 0.74 years, and the mean MPH was 159.53 ± 3.78 cm. After treatment, their mean height SDS decreased from 1.24 ± 0.85 and 0.75 ± 0.87 ($p < 0.001$), their Tanner stage increased from 2.42 ± 0.70 to 2.77 ± 0.78 ($p <$

0.001), and their bone age advancement decreased from 2.61 ± 0.69 to 0.69 ± 0.67 years ($p < 0.001$). The PAH increased from 154.52 ± 5.78 to 163.48 ± 5.19 cm ($p < 0.001$); the mean increase in PAH was 8.96 ± 5.04 cm.

(a)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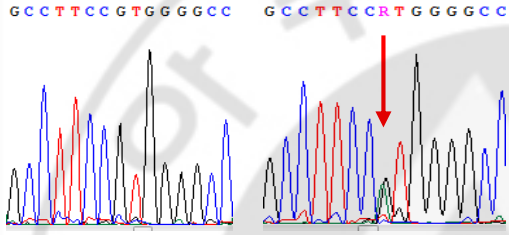
GCTGCAGCYGCCGCY

(b)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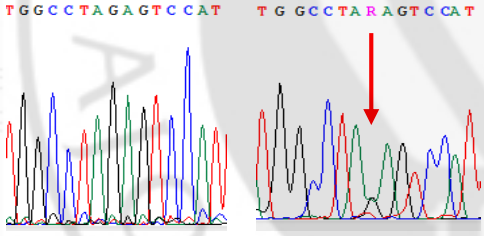
GCCTTCCRTGGGGCC

(c)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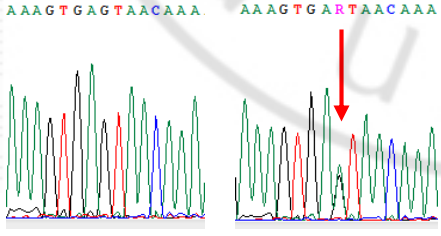
(d)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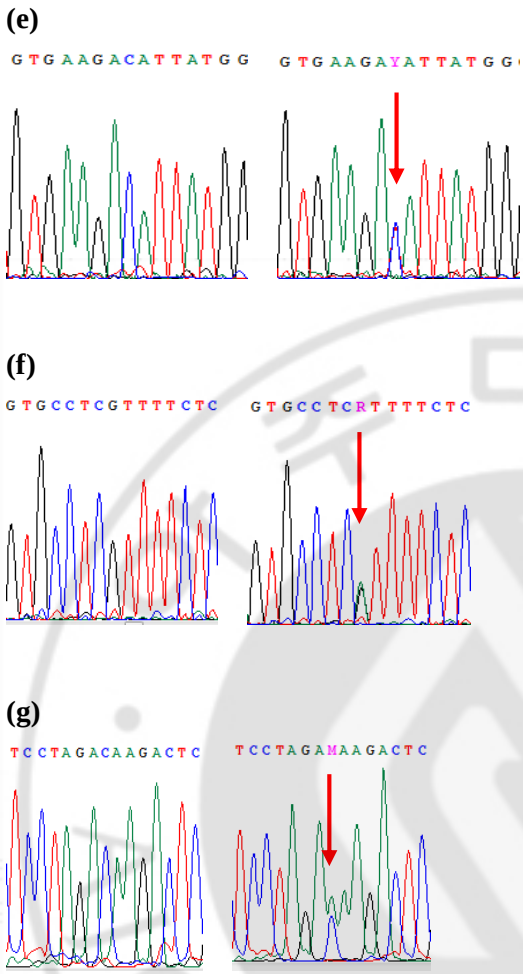

Figure 1. Partial sequences of the LHCGR gene show the polymorphisms detected in the present study. Lt: wild-type, Rt polymorphism (a) a heterozygote of g.48755617_48755618 insTGCAGC, (b) a heterozygote of g.48708972G>A (Lt), (c) a heterozygote of g.48698754G>A. (d) a homozygote (Lt) and a heterozygote (Rt) of g.48694236A>G, (e) a homozygote (Lt) and a heterozygote (Rt) of g.48694236T>C (Rt), (f) a heterozygote of g.48688613G>A, (g) a heterozygote of g.48687721 C>A. The positions of the polymorphisms are defined according to contig NM_022184.16 in this figure. *; Wild-types of g.48694236 and g.48688732 are AA and TT, respectively.

Table 1. Baseline characteristics of patients with non-classic CPP

	Patient group (*n* = 102)
Age of onset (years)	7.64 ± 1.18
Age of diagnosis (years)	8.37 ± 0.56
Tanner stage (breast)	2.43 ± 0.68
Bone age (years)	10.72 ± 0.42
BA–CA (years)	2.63 ± 0.63
MPH SDS	-0.25 ±0.75
Height SDS	1.16 ± 0.88
BMI SDS	0.80 ± 1.04
Basal LH (IU/L)	1.04 ± 0.42
Basal FSH (IU/L)	1.71 ± 1.23
Basal LH/FSH ratio	0.79 ± 0.50
Peak LH (IU/L)	3.31 ± 0.99
Peak FSH (IU/L)	10.71 ± 4.49

Abbreviations: BA–CA: bone age advancement, MPH: mid-parental height, SDS: standard deviation scores, LH: luteinizing hormone, FSH: follicle-stimulating hormone

Table 2. LHCGR gene polymorphisms identified by sequencing (*n* = 202)

No	Location	dbSNP ID	Polymorphism	AA position	Function	Group
1	Exon 1	rs142537840	g.48755617_48755618ins TGCAGC		insertion	Control
2	Exon 8	rs182707541	g. 48708972G>A	p.R219H	missense	Control
3	Exon 9	rs768846378	g.48698754G>A	p.E243K *	missense	Patient
4	Eon 10	rs2293275	g. 48694236A>G	p.N312S	missense	Both
5	Exon 11	rs11125179	g.48688732T>C	p.D355D	synonymous	Both
6	Exon11	rs754040582	g.48688613G>A	p.R395H*	missense	Patient
7	Exon 11	-	g.48687721C>A	p.D692E novel	missense	Control

Abbreviation: AA: amino acid, *: in silico analysis predicted possible damage to LHCGR
The positions of the polymorphisms are defined according to contig NT_022184.16 for genomic DNA and NM_000233 for cDNA.

Table 3. Allele frequencies of the LHCGR polymorphisms from 102 patients and 100 controls

Polymorphism	Group	Allele frequency		*P*-value*
		1	2	
g. 48755617_48755618ins TGCAGC Wild = 1; Ins TGCAGC = 2	Patient Control	204(100%) 199(99.5%)	0(0.0%) 1(0.05%)	0.495
g. 48708972G>A G=1; A=2	Patient Control	204(100%) 199(99.5%)	0(0.0%) 1(0.05%)	0.495
g.48698754G>A G=1; A=2	Patient Control	203(99.5%) 200(100%)	1(0.4%) 0(0.0%)	1.00
g. 48694236A>G A=1; G=2	Patient Control	10(4.9%) 12(6%)	194(95.1%) 188(94%)	0.657
g.48688732T>C T=1; C=2	Patient Control	10(4.9%) 11(5.5%)	194(95.1%) 189(94.5%)	0.821
g.48688613G>A G=1; A=2	Patient Control	203(99.5%) 200(100%)	1(0.5%) 0(0.0%)	1.00
g.48687721C>A C=1; A=2	Patient Control	204(100%) 199(99.5%)	0(0.0%) 1(0.05%)	0.311

* Comparison of the allele frequencies between the patient and control groups.

Table 4. Response to gonadotropin releasing hormone agonist treatment in 75 patients

	Pre-treatment (n = 75)	Post – treatment (n =75)	P-value
Age (years)	8.35 ± 0.60	11.17 ± 0.62	0.000
Bone age (years)	10.73 ± 0.45	11.87 ± 0.23	0.000
Height (cm)	134.23 ± 4.70	150.62 ± 5.01	0.000
Height SDS	1.24 ± 0.85	0.75 ± 0.87	0.000
BMI SDS	0.79 ± 0.92	0.86 ± 1.04	0.468
Tanner stage	2.42 ± 0.70	2.77 ± 0.78	0.000
BA–CA (years)	2.61 ± 0.69	0.69 ± 0.67	0.000
PAH (cm)	154.52 ± 5.78	163.48 ± 5.19	0.000

Abbreviations: BA–CA: bone age advancement, PAH: predicted adult height

IV. Discussion

To our knowledge, this is the first report on LHCGR gene polymorphisms in patients with non-classic CPP. A total of seven LHCGR gene polymorphisms were identified in this study. Although the identified mutations were not significantly associated with nonclassic CPP, two missense mutations (g.48698754G > A and g.48688613G > A) might be damaging to the LHCGR. These mutations might have a causative role in patients exhibiting CPP characteristics but with a low LH response. Further studies are required o investigate the functional role of these mutations with respect to the clinical history of the patients.

The patient with the LHCGR mutation g.48698754G > A exhibited breast engorgement at 2 years old and had advanced bone age. She underwent the GnRH stimulation test, but the peak LH result was < 5 U/L. Since then, we have observed her clinical course every 3–6 months in the outpatient clinic. Her Tanner stage was 2 for breasts and has not changed; however, her bone age has advanced steadily. She underwent the GnRH stimulation test more than five times, and her peak LH concentration each time was less than < 5 U/L. Moreover, she had normal brain MRI images and pelvis USG. Finally, her parents wanted her to be treated with a GnRH agonist at 8 years old. At that time, she was Tanner stage 2 for breasts and her bone age was advanced by more than 3 years. Her height and its SD were 131.40 cm and 1.17 SD. Her MPH was 153.0 cm and -1.58 SD. Her BMI was 18.30 kg/m2 and 0.92 SD. Interestingly, her pre-treatment basal LH and FSH concentrations were 1.30 and 2.00 U/L, respectively. However, her stimulated peak LH and FSH concentrations were 2.10 and 7.00 U/L, respectively. She was treated with a GnRH agonist for 3 years; her Tanner stage did not change, and her BMI was 19.93 kg/m2 and 0.75 SD at the last injection. Her PAH was 153.26 cm pre-treatment and 161.92 cm at the end of treatment. At the age of 13 years, her final height was 161.80 cm, and her bone age was 15 years old.

The patient with the g.48688613G > A mutation exhibited breast engorgement at 7 years old and her bone age was 10.5 years old. Her height and SD were 131.7 cm and 1.32 SD. Her BMI was 18.16 kg/m2 and 0.89 SD. Her MPH was 159.50 cm and -0.25 SD, and her PAH was 153.66 cm pre-treatment. Interestingly, her basal LH and FSH concentrations were 1.30 and 1.20 U/L, respectively, and her GnRH-stimulated peak LH and FSH concentrations were 2.90

and 9.10 U/L, respectively. She is currently under GnRH agonist treatment.

In our study, g.48694236A > G and g.48688732 T > C were the most frequently identified polymorphisms. Wild-type g.48694236 AA and g.48688732 TT were not found in any study subject. All subjects had heterozygous or homozygous polymorphisms. According to the allele frequency per the 1000 Genomes Browser (http://www.ncbi.nlm.nih.gov/vatiation/tools/1000genomes/?assm = GCF_000001405.25), the allele frequencies of g.48694236 are consistent with those in the Chinese and Japanese populations.

The g.48694236A > G mutation is associated with polycystic ovarian syndrome (PCOS). Recently, Thathapudi et al. reported that this mutation was associated with an increased risk of PCOS in South Indian women: especially, 3.36-fold in the homozygous G/G genotype [15]. In contrast, Valkenburg et al. found no association between this mutation and the increased risk for PCOS in Caucasians [16]. All of our subjects had the LHCGR mutation g.48694236A > G, predominantly the homozygous GG genotype. The conflicting results of an association between PCOS and this mutation might be due to ethnic differences. Nevertheless, these results suggest a correlation between the pathogenesis of precocious puberty and PCOS; however, additional research is required.

Testotoxicosis is representative of peripheral precocious puberty and is caused by LHCGR gene-activating mutations [6, 7, 17, 18]. It is a gonadotropin-independent precocious puberty characterized by rapid virilization, growth acceleration, bone age advancement, and elevated testosterone concentrations despite prepubertal concentrations of LH [7, 19]. To date, 18 mutations in the LHCGR gene have been reported in boys with testotoxicosis, mostly located in exon 11, which differs from the results of our study [20]. The serum LH response to GnRH reflects the pubertal status: LH concentration 1 h post-GnRH agonist stimulation of ≥ 3.2 U/L is 90 % sensitive, and that of ≥ 5.5 U/L is 95 % specific for the onset of puberty in girls [21–23]. In addition, an early morning basal LH > 0.6 U/L has been reported to be 62–95 % sensitive and 92– 100 % specific for the diagnosis of CPP in girls [23–25]. In our study, patients exhibited the interesting laboratory findings that the basal LH concentration was 1.04 ± 0.42 U/L but the peak LH response to GnRH 3.31 ± 0.99 U/L. It is difficult to both diagnose and rule out CPP in these patients. From this perspective, how to classify and diagnose such patients is a matter of dispute.

Rosenfield et al. suggested that precocious puberty can be viewed as a spectrum, ranging

from benign premature thelarche through slowly progressive complete precocity to rapidly progressive complete precocity, and that GnRH agonist therapy is necessary in rapidly progressive precocious puberty [26]. However, the patients in our study did not fall within the standard disease spectrum of progressive CPP, because peak LH concentrations > 5 IU/L suggest progressive CPP(1).The clinical signs of our patients suggested that they were more representative of progressive CPP rather than the natural and physiological course of puberty. The elevated basal LH concentration suggests that our patients had CPP. However, the patients had a lower stimulated LH concentration than the current standard for progressive CPP. Furthermore, there was no evidence of peripheral precocious puberty in these patients. In our study, all patients were treated with a GnRH agonist, and their clinical parameters improved. Indeed, GnRH agonist treatment decreased bone age advancement and increased their PAH. In the 75 patients who completed GnRH agonist treatment, their midparental height was 159.53 ± 3.78 cm; their pre- and post-treatment PAH were 154.52 ± 5.78 and 163.48 ± 5.19 cm, respectively; and their PAH increase was 8.96 ± 5.04 cm. These findings are consistent with previous reports [27–30]. Unfortunately, data on the actual final height are not yet available, since the vast majority of patients have not yet reached their final height.

In girls with non-classic CPP, we examined the LHCGR gene and clinical responses to GnRHa treatment. Our findings must be understood in view of the following limitations. First, this study had a relatively small sample size and a heterogeneous patient group. Second, the use of GnRH agonist treatment in patients with nonclassic CPP is controversial, because a consensus has not yet been established. Third, FSHR (FSH receptor) gene analysis was not performed. FSH binds to the FSHR and induces the expression of LHCGR [31, 32], and the response of LH depends strictly on the prior FSH-dependent expression of LHCGR [33]. Thus, further study is required to investigate the FSHR gene in this patient group.

V. Conclusion

In this study, LHCGR gene mutations were identified in patients with non-classic CPP. We also found that, although some mutations are predicted to impair the function of LHCGR, this mutation does not seem to be the common cause of non-classic CPP. Furthermore, treatment

with GnRH agonist was effective in these patients. Our findings extend the spectrum of CPP by presenting a non-classic CPP phenotype; however, more research is needed.

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-국문요약-

성조숙증 여아에서 황체화호르몬 수용체 유전자 (LHCGR gene)의 다형성 연구

아주대학교 대학원의학과

정 활 림

(지도교수 : 황 진 순)

목적: 황체화호르몬은 성조숙증의 진단에 있어 유용한 지표이다. 성선자극호르몬방출호르몬 자극검사에서 혈중 황체화호르몬의 사춘기 반응은 다양하게 나타나고 때때로 성조숙증의 임상 증상은 황체화호르몬의 혈중 농도와 일치하지 않는다. 많은 수의 환자들이 빠르게 진행하는 성조숙증의 임상증상을 나타내면서도 성선자극호르몬방출호르몬 자극검사에서 황체화호르몬의 혈중 최고 농도는 사춘기 이전상태로 나타나기도 한다. 본 연구에서는 이러한 비전형적인 중추성 성조숙증 환자를 대상으로 황체화호르몬 수용체 유전자의 변이와 이의 임상적 의의를 확인하고자 하였다.

방법: 아주대 병원 소아청소년과에 내원하여 비전형적인 중추성 성조숙증으로 진단받은 102 여아들이 환자군으로 모집되었으며, 대조군은 100명의 정상 사춘기 발달 과정을 거친 건강한 한국인 성인 여자들을 대상으로 황체화호르몬 수용체 유전자 분석을 시행하였다. 비전형적인 성조숙증 환자는 성조숙증의 임상양상을 보이면서 성선자극호르몬방출호르몬 자극검사에서 황체화호르몬의 혈중 최고 농도가 5IU/L 미만인 환자로 정의하였다. 총 102명의 환자 중 75명은 성선자극호

르몬방출호르몬 작용제 치료를 완료하였고 이들에게서 치료 효과를 분석하였다.

결과: 102명의 성조숙증 환아와 100명의 정상 대조군의 황체화호르몬 수용체 유전자 분석을 시행하였다. 유전자 분석 결과 총 7개의 유전자 변이가 발견되었으나 이러한 황체화호르몬 유전자의 변이와 비전형적인 성조숙증 간에 임상적으로 유의한 관련성은 발견하지 못하였다. 그러나 이들 중 2명의 환자에서 각각 발견된 2개의 과오돌연변이는 인 실리코 분석에서 황체화호르몬 수용체에 손상을 줄 가능성이 있는 것으로 나타났다. 한편, 비전형적인 성조숙증 환자에서 성선자극호르몬방출호르몬 작용제는 골연령을 늦추고 성인 예측키를 증가 시키는 결과를 보였다.

결론: 본 연구에서 황체화호르몬 수용체 유전자의 변이가 비전형적인 성조숙증 환자의 주요한 원인은 아님을 알 수 있었다. 성선자극호르몬방출호르몬 작용제는 이들 환자에서 성조숙증의 임상 지표들을 향상시킨다는 것을 알 수 있었다.

핵심어: 성조숙증, 황체화호르몬 수용체 유전자, 성선자극호르몬방출호르몬 작용제