Case Report
J Genet Med 2023;20(1):25-29
https://doi.org/10.5734/JGM.2023.20.1.25
ISSN 1226-1769 (Print) 2383-8442 (Online)

JGM Journal of Genetic Medicine

Cholesterol side-chain cleavage enzyme deficiency caused by a novel homozygous variant in P450 side-chain cleavage enzyme gene (*CYP11A1*) in a 46,XX Korean girl

Ye Ji Kim[1], Sun Cho[1], Hwa Young Kim[2], Young Hwa Jung[2], Jung Min Ko[1,3], Chang Won Choi[2,3], and Jaehyun Kim[2,3,*]
[1]Department of Pediatrics, Seoul National University Children's Hospital, Seoul, Korea
[2]Department of Pediatrics, Seoul National University Bundang Hospital, Seongnam, Korea
[3]Department of Pediatrics, Seoul National University College of Medicine, Seoul, Korea

The *CYP11A1* gene encodes for the cholesterol side-chain cleavage enzyme (P450scc), which initiates steroid hormone biosynthesis. Defective P450scc activity results in severe glucocorticoid and mineralocorticoid deficiencies. We describe a case of P450scc deficiency due to a novel homozygous *CYP11A1* variant inherited from the mother with a possibility of uniparental disomy (UPD). The patient was a female, had no family history of endocrine disease, and showed adrenal insufficiency at 13 days of age. Hormonal analysis with an adrenocorticotropic hormone stimulation test showed both glucocorticoid and mineralocorticoid deficiencies, presumed to be a defect of the early stage of steroidogenesis. Exome sequencing reported a novel homozygous frameshift variant of *CYP11A1* (c.284_285del, p.Asn95Serfs*10), which was inherited from the mother. Additionally, homozygosity in 15q22.31q26.2, which included *CYP11A1*, was identified using a chromosomal microarray. It was suggested that the possibility of maternal UPD was involved as the cause of a P450scc deficiency by unmasking the maternally derived affected allele. To our understanding, P450scc deficiency associated with UPD encompassing *CYP11A1* had not been reported in Korea before. Genetic analysis can help diagnose rare causes of primary adrenal insufficiency, including P450scc deficiency.

Key words: Cholesterol side-chain cleavage enzyme (P450scc), CYP11A1, Adrenal insufficiency, Uniparental disomy.

Introduction

Primary adrenal insufficiency (PAI) is a disorder issued from a deficient production of glucocorticoids and/or mineralocorticoids that are normally synthesized in the adrenal cortex [1-3]. Cholesterol side-chain cleavage enzyme (P450scc) is an enzyme that initiates steroid hormone biosynthesis and is encoded by the *CYP11A1* gene. P450scc deficiency is a rare autosomal recessive disorder, with approximately 60 patients from more than 40 families reported to date [4-9]. Until now, around 40 pathogenic or likely pathogenic variants of *CYP11A1* have been reported [10]. Defective P450scc activity inhibits normal steroidogenesis

Received: 26 March 2023, Revised: 27 April 2023, Accepted: 28 May 2023, Published: 30 June 2023
*Corresponding author: Jaehyun Kim, M.D., Ph.D. https://orcid.org/0000-0002-0203-7443
Department of Pediatrics, Seoul National University Bundang Hospital, 82 Gumi-ro 173beongil, Bundang-gu, Seongnam 13620, Korea.
Tel: +82-31-787-7287, Fax: +82-31-787-4054, E-mail: pedendo@snubh.org
Conflict of interest: The authors declare that they do not have any conflicts of interest.

resulting in severe adrenal and gonadal deficiencies [4,11]. We report the first Korean case of P450scc deficiency caused by a novel *CYP11A1* variant with an association with uniparental disomy (UPD). This study was approved by the Institutional Review Board of Seoul National University Bundang Hospital (NO. B-2112-727-701). Informed consent was received from the patient's parents.

Case

A 13-day-old female infant was hospitalized to Seoul National University Bundang Hospital with chief complaints of decreased oral intake and feeding cyanosis. She was delivered at 38 weeks of gestation, weighing 2.2 kg, without any perinatal complications. She was the second child of healthy nonconsanguineous parents. Newborn screening for inborn error of metabolism showed a normal level of 17-alpha-hydroxyprogesterone (17-OHP). At admission, the patient's height was 47 cm (–0.7 000 standard deviation score [SDS]), and weight was 2.0 kg (–2.6 SDS). On physical examination, her external genitalia appeared normal. Blood tests revealed hyperkalemia (9.5 mmol/L) and hyponatremia (125 mmol/L). She had elevated adrenocorticotropic hormone (ACTH) (531 pg/mL; reference range, 0-60), and increased plasma renin activity (84.5 ng/mL/h; reference range, 2.0-35.0). A standard-dose ACTH stimulation test (using 0.125 mg of synacthen [ACTH-(1-24)]) showed poor responses of cortisol and 17-OHP (Table 1). Adrenal imaging by ultrasonography revealed normal-sized adrenals. After the diagnosis of glucocorticoid and mineralocorticoid deficiency was made,

Table 1. Clinical and hormonal data

Items	Baseline	Latest visit	Normal age range (basal)
Age	13 days	5 yrs and 3 mos	NS
Height, cm (SDS)	47 (–0.7)	47 (–0.7)	NS
Weight, kg (SDS)	2.0 (–2.6)	47 (–0.7)	NS
External genitalia	Normal female	Normal female	NS
Blood glucose, mg/dL	71	87	70-110
Sodium, mmol/L	125	139	135-145
Potassium, mmol/L	9.5	4.3	3.5-5.5
Cortisol, μg/dL (basal/post ACTH)	3.5/8.2	11.0/ND	2-11
17OH-Progesterone, ng/mL (basal/post ACTH)	0.5/1.2	ND	0.1-0.8
ACTH, pg/mL	217	715	0-60
Plasma renin activity, ng/mL/h	84.5	0.9	2.0-35.0
Aldosterone, ng/dL	33	ND	5-175

SDS, standard deviation score; ACTH, adrenocorticotropic hormone; ND, Not done; NS, not significant.

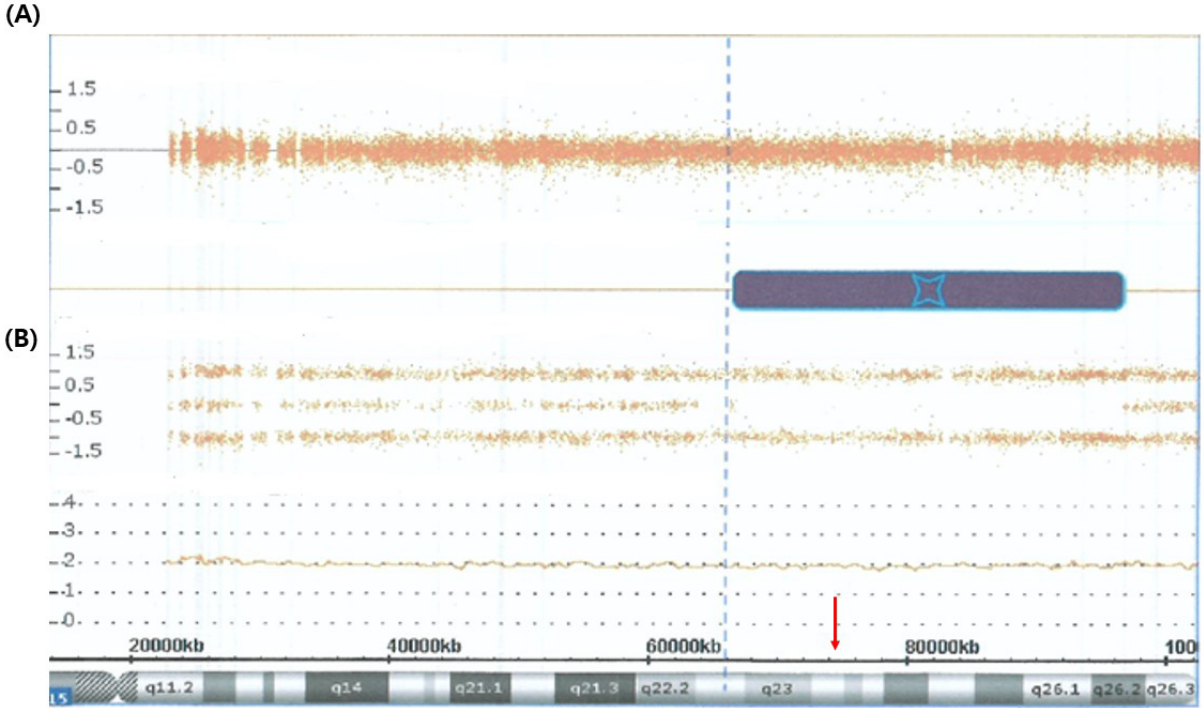

Fig. 1. A chromosomal microarray showed a 30-Mb region of homozygosity (black box with star). The homozygosity on chromosome 15 (human genome assembly GRCh37 [hg19], chr15: 66481835-96607321) demonstrating a copy number state of 2 (A) and no heterozygous single nucleotide polymorphisms (B). The red arrow denotes the location of the *CYP11A1* gene.

hydrocortisone with 0.8 mg three times per day (15.0 mg/m^2/d), and 9α- fludrocortisone with 0.1 mg once daily were prescribed to the patient.

The patient's karyotype was 46,XX. Whole exome sequencing was done on a NextSeq 500 system (Illumina Inc.,) with 2×150 paired-end reads. Reads were aligned to the human genome build 37 (Hg19). A novel homozygous frameshift variant of *CYP11A1* (c.284_285del, p.Asn95Serfs*10) was identified and confirmed using Sanger sequencing. The mother of the patient was a heterozygous carrier of the variant, while the father had none. As a UPD in the 15q23q26.1 region was suspected on exome sequencing, a single-nucleotide polymorphism (SNP) microarray using the Affymetrix Cytoscan 750K array (Affymetrix) was additionally conducted. A SNP microarray revealed a 30 Mb homozygosity of 15q22.31q26.2 region that includes *CYP11A1* (Hg19, chr15: 66481835-96607321), consistent with segmental UPD without any genomic copy number variations (Fig. 1). This finding, in conjunction with the patient's previous biochemical testing for PAI, provided confirmation that the etiology of P450scc deficiency was due to maternal UPD encompassing the *CYP11A1* variant.

While taking hydrocortisone and 9α-fludrocortisone, she showed catch-up growth and a stable clinical course without adrenal crisis. On her latest visit (5 years and 3 months of age), she was prescribed hydrocortisone, 3.3 mg three times per day (12.0 mg/m^2/d), and 9α-fludrocortisone (0.1 mg daily). Her height was 104.0 cm (–1.4 SDS), and her weight was 23.3 kg (–1.5 SDS). She had normal plasma renin activity (0.9 ng/mL/h; reference range, 1.0–6.5) but had high ACTH levels (715 pg/mL; reference range, 0–60) without hypoglycemia or electrolyte imbalance (blood glucose 87 mg/dL, sodium 139 mmol/L, and potassium 4.3 mmol/L, respectively) (Table 1).

Discussion

P450scc deficiency is an extremely rare cause of PAI and results from defects in the *CYP11A1* gene on chromosome 15q23-q24. P450scc deficiency is similar to congenital lipoid adrenal hyperplasia (CLAH) in clinical characteristics and hormonal profile but is not accompanied by adrenal enlargement, a typical finding in CLAH [4,11,12]. Most reported patients had highly elevated ACTH levels with low adrenal and gonadal steroid levels. Abdominal imaging revealed normal or small adrenal glands. In contrast to 46,XX patients, 46,XY patients show a disorder of sexual development.

To our knowledge, approximately 60 cases of P450scc deficiency caused by *CYP11A1* variants have been reported so far (Supplementary Table 1). Our 46,XX female case presented with decreased oral intake and feeding cyanosis in the neonatal period, with normal 17-OHP levels, giving the impression of defects of early stages in steroid biosynthesis. Among 13 previously reported female cases with proven 46,XX karyotype, four were followed until puberty; one developed hypergonadotropic hypogonadism at 12 years of age, and one female patient was prescribed medication for precocious puberty with normal menstruation cycle in adulthood [10,13]. Although our 5-year-old female patient had normal external genitalia without any signs of gonadal dysfunction, careful monitoring of pubertal progression will be needed.

The *CYP11A1* variant identified in our case was previously not reported and considered pathogenic according to the American College of Medical Genetics and Genomics (ACMG) criteria (PVS1+PM2+PP4) [14]. Chromosomal microarray analysis suggested a 30 Mb homozygosity region in 15q22.31q26.2, which included *CYP11A1*. UPD is a genetic condition characterized by the inheritance of both copies of a chromosome pair from one parent alone [15]. This phenomenon can result in disturbances of imprinted genes at certain imprinted loci in humans, leading to clinically recognizable imprinted disorders such as Prader-Willi syndrome (PWS) and Angelman syndrome (AS), which arise from maternal UPD15 and paternal UPD15, respectively [16]. In addition, UPD can rarely result in clinical conditions by allowing two copies of a recessive variant to be transmitted from a heterozygous carrier parent [17]. Notably, this unusual non-Mendelian form of inheritance related to UPD has been linked to several cases of recessive disorders, including cystic fibrosis [18]. Furthermore, UPD can result in an unusual association between two rare genetic disorders [19,20]. For instance, a case involving the co-occurrence of AS and P450scc deficiency has been reported, which originated from a large segmental UPD (15q11.1 to 15q26.2) unmasking a novel recessive variant in *CYP11A1* [20]. In our case, as only the mother carried the variant as a heterozygote, it was suggested that maternal UPD was involved as the cause of P450scc deficiency in the proband by unmasking the maternally derived affected allele. The homozygous region identified in the patient did not encompass the 15q11-q13 chromosomal region that is typically associated with PWS/AS. Furthermore, no clinical manifestations were observed in the patient that were indicative of PWS/AS. Moreover, a comprehensive analysis of the region of segmental UPD, which encompassed a total of 43 recessive disease-causing genes, was conducted using whole-exome sequencing. However, this analysis did not

reveal any additional novel nonsynonymous homozygous variants within this region.

The diagnosis of PAI in pediatric patients is often delayed as many symptoms and signs of PAI are nonspecific. In cases of normal 17-OHP levels, genetic analysis can be helpful in diagnosing rare causes of PAI, including CLAH, X-inked adrenoleukodystrophy, and P450scc deficiency as in this case who developed symptoms early in their infancy [3,5,12,13]. In conclusion, the diagnosis of P450scc deficiency in our case was established through a comprehensive genetic analysis that combined exome sequencing and a SNP array. This underscores the critical role of genetic analysis in uncovering the rare etiologies underlying PAI. Furthermore, our case serves as an illustrative example of the potential for UPD to reveal recessive disorders such as *CYP11A1* defects in patients who present with adrenal insufficiency.

Acknowledgements

The authors thank the patient and her parents, who consigned to participate in this report.

Authors' Comtributions

Conception and design: CWC, JK. Acquisition of data: YJK, SC, HYK, YHJ, JMK, CWC, JK. Analysis and interpretation of data: HWK, JMK. Drafting the article: YJK, SC. Critical revision of the article: YJK, HWK, JK. Final approval of the version to be published: YJK, SC, HYK, YHJ, JMK, CWC, JK.

References

1. Arlt W, Allolio B. Adrenal insufficiency. Lancet 2003;361:1881-93.
2. Charmandari E, Nicolaides NC, Chrousos GP. Adrenal insufficiency. Lancet 2014;383:2152-67.
3. Bornstein SR, Allolio B, Arlt W, Barthel A, Don-Wauchope A, Hammer GD, et al. Diagnosis and treatment of primary adrenal insufficiency: an Endocrine Society clinical practice guideline. J Clin Endocrinol Metab 2016;101:364-89.
4. Tee MK, Abramsohn M, Loewenthal N, Harris M, Siwach S, Kaplinsky A, et al. Varied clinical presentations of seven patients with mutations in *CYP11A1* encoding the cholesterol side-chain cleavage enzyme, P450scc. J Clin Endocrinol Metab 2013;98:713-20. Erratum in: J Clin Endocrinol Metab 2013;98:4213.
5. National Center for Biotechnology Information. ClinVar. [https://www.ncbi.nlm.nih.gov/clinvar].
6. Buonocore F, Achermann JC. Primary adrenal insufficiency: New genetic causes and their long-term consequences. Clin Endocrinol (Oxf) 2020;92:11-20.
7. Kolli V, Kim H, Torky A, Lao Q, Tatsi C, Mallappa A, et al. Characterization of the *CYP11A1* nonsynonymous variant p.E314K in children presenting with adrenal insufficiency. J Clin Endocrinol Metab 2019;104:269-76.
8. Lara-Velazquez M, Perdomo-Pantoja A, Blackburn PR, Gass JM, Caulfield TR, Atwal PS. A novel splice site variant in CYP11A1 in trans with the p.E314K variant in a male patient with congenital adrenal insufficiency. Mol Genet Genomic Med 2017;5:781-7.
9. Matusik P, Gach A, Zajdel-Cwynar O, Pinkier I, Kudela G, Gawlik A. A novel intronic splice-site mutation of the *CYP11A1* gene linked to adrenal insufficiency with 46,XY disorder of sex development. Int J Environ Res Public Health 2021;18:7186.
10. Pomahačová R, Sýkora J, Zamboryová J, Paterová P, Varvařovská J, Šubrt I, et al. First case report of rare congenital adrenal insufficiency caused by mutations in the *CYP11A1* gene in the Czech Republic. J Pediatr Endocrinol Metab 2016;29:749-52.
11. Hauffa B, Hiort O. P450 side-chain cleavage deficiency--a rare cause of congenital adrenal hyperplasia. Endocr Dev 2011;20:54-62.
12. Yoo HW. Diverse etiologies, diagnostic approach, and management of primary adrenal insufficiency in pediatric age. Ann Pediatr Endocrinol Metab 2021;26:149-57.
13. Goursaud C, Mallet D, Janin A, Menassa R, Tardy-Guidollet V, Russo G, et al. Aberrant splicing is the pathogenicity mechanism of the p.Glu314Lys variant in *CYP11A1* gene. Front Endocrinol (Lausanne) 2018;9:491.
14. Richards S, Aziz N, Bale S, Bick D, Das S, Gastier-Foster J, et al.; ACMG Laboratory Quality Assurance Committee. Standards and guidelines for the interpretation of sequence variants: a joint consensus recommendation of the American College of Medical Genetics and Genomics and the Association for Molecular Pathology. Genet Med 2015;17:405-24.
15. Robinson WP. Mechanisms leading to uniparental disomy and their clinical consequences. Bioessays 2000;22:452-9.
16. Eggermann T, Perez de Nanclares G, Maher ER, Temple IK, Tümer Z, Monk D, et al. Imprinting disorders: a group of congenital disorders with overlapping patterns of molecular changes affecting imprinted loci. Clin Epigenetics 2015;7:123. Erratum in: Clin Epigenetics 2016;8:27.
17. Kotzot D, Utermann G. Uniparental disomy (UPD) other than 15: phenotypes and bibliography updated. Am J Med Genet A 2005;136:287-305.
18. Engel E. Uniparental disomies in unselected populations. Am J Hum Genet 1998;63:962-6.
19. Schinzel AA. Uniparental disomy and gene localization. Am J Hum

Genet 1991;48:424-5.
20. Kim A, Fujimoto M, Hwa V, Backeljauw P, Dauber A. Adrenal insufficiency, sex reversal, and Angelman syndrome due to uniparental disomy unmasking a mutation in *CYP11A1*. Horm Res Paediatr 2018;89:205-10.