A Novel Missense Mutation in the *SLC5A5* Gene in a Sudanese Family with Congenital Hypothyroidism

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Thyroid hormone synthesis requires the presence of iodide. The sodium–iodide symporter (NIS) is a glycoprotein that mediates the active uptake of iodide from the blood stream into the thyroid gland. NIS defects due to *SLC5A5* gene mutations are known to cause congenital hypothyroidism (CH). The probanda is a 28-year-old female whose origin is North Sudan where neonatal screening for CH is not available. She presented with severe constipation and a goiter at the age of 40 days. Laboratory testing confirmed CH, and she was started on levothyroxine. Presumably due to the delayed treatment, the patient developed mental retardation. Her younger sister presented with a goiter, tongue protrusion, and umbilical hernia, and the youngest brother was also diagnosed with CH based on a thyrotropin level $>100 \mu\text{IU/mL}$ at the age of 22 days and 8 days, respectively. The two siblings were treated with levothyroxine and had normal development. Their consanguineous parents had no history of thyroid disorders. Whole-exome sequencing was performed on the probanda. This identified a novel homozygous missense mutation in the *SLC5A5* gene—c.1042T>G, p.Y348D—which was subsequently confirmed by Sanger sequencing. All affected children were homozygous for the same mutation, and their unaffected mother was heterozygous. The NIS protein is composed of 13 transmembrane segments (TMS), an extracellular amino-terminus, and an intracellular carboxy-terminus. The mutation is located in the TMS IX, which has the most $\beta\text{-OH}$ group-containing amino acids (serine and threonine), which is implicated in Na^+ binding and translocation. In conclusion, a novel homozygous missense mutation in the *SLC5A5* gene was identified in this Sudanese family with CH. The mutation is located in the TMS IX of the NIS protein, which is essential for NIS function. Low iodine intake in Sudan is considered to affect the severity of hypothyroidism in patients.

Keywords: *SLC5A5*, NIS, sodium–iodide symporter, novel mutation, congenital hypothyroidism, goiter

Introduction

THE SODIUM–IODIDE SYMPORTER (NIS) is a glycoprotein that mediates the uptake of iodide from the bloodstream into the thyroid gland (1). In the thyroid, NIS encoded by the *SLC5A5* gene provides iodide crucial for thyroid hormone synthesis. To date, 15 *SLC5A5* mutations have been reported, resulting in congenital hypothyroidism (CH) (1). Herein, a Sudanese family with CH harboring a novel homozygous missense mutation in the *SLC5A5* gene is reported.

Patient

The probanda is a 28-year-old female from North Sudan. Since no neonatal screening exists in Sudan, she was found to have CH when she presented with severe constipation and a goiter at the age of 40 days. She had developed mental retardation, despite levothyroxine (LT4) treatment.

Her 25-year-old sister presented with tongue protrusion and umbilical hernia at the age of 22 days and was diagnosed with CH based on a thyrotropin (TSH) level of $>100 \mu\text{IU/mL}$. Her youngest brother was screened when he was eight days old because of the family history and was diagnosed with CH on the basis of a high TSH level ($>100 \mu\text{IU/mL}$). He was started on LT4 and had normal development. The other siblings and the consanguineous parents have no signs of thyroid disorders. Thyroid function tests (TFTs) were performed by Immulite 1000 system[®] (Siemens, Munich, Germany). At the time of TFT measurements, the three affected children were on LT4, and all of them had low TSH levels (Fig. 1A). The probanda and her brother had a slightly high free thyroxine level but normal total thyroxine and total triiodothyronine; only the probanda had a high serum thyroglobulin level (Fig. 1A). The mother had normal TFTs. The father and the unaffected siblings could not be analyzed.

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FIG. 1. (A) Pedigree of the family and results of thyroid function tests. Each generation corresponds to a roman number. Arabic numbers above each symbol identify the subjects. Laboratory data are aligned below each symbol. Abnormal values are in bold type. L-T₄, levothyroxine; TT₄, total thyroxine; TT₃, total triiodothyronine; FT₄, free thyroxine; TSH, thyrotropin; TBG, thyroxine binding globulin; TG, thyroglobulin; TPOAb, anti-TPO antibody; TG Ab, antithyroglobulin antibody; N/A, not available. The International System of Units: TT₄, µg/dL = 12.87 nmol/L; TT₃, ng/dL = 0.0154 nmol/L; fT₄, ng/dL = 12.87 pmol/L; TBG, µg/mL = 0.0185 µmol/L. (B) Chromatograms of direct sequencing results of a heterozygote (wild-type/mutant) and a homozygote (mutant/mutant) individual covering the region of the mutation in exon 8 of the SLC5A5 gene (c.1042T>G, p.Tyr348Asp). Color images available online at www.liebertpub.com/thy

The molecular genetic study was approved by The Human Subject Research Office of the University of Miami. Written informed consent was obtained from all study participants, as well as the mother of the subject <18 years of age. Genomic DNA was extracted from whole blood by standard extraction procedures. Whole-exome sequencing (WES) was performed using an Illumina HiSeq 2500 System[®] with a sequencing depth of 50× (Illumina, San Diego, CA) with the DNA of the probanda and the mother, and the data were analyzed assuming an autosomal recessive inheritance pattern.

WES identified a novel homozygous missense mutation in exon 8 of the SLC5A5 gene: c.1042T>G, p.Y348D (RefSeq transcript NM_000453) in the probanda. Sanger sequencing confirmed that all affected siblings were homozygous for the same mutation, while the mother was heterozygous (Fig. 1B). Without blood from the father, it can only be assumed that the father was also heterozygous for the mutations or, less likely, that maternal uniparental disomy occurred. This variant was not present in the Genome Aggregation Database (gnomAD), dbSNP, and Exome Variant Server. The variant was pre-

dicted to be deleterious by *in silico* bioinformatics tools: SIFT; 0.001, Damaging, Polyphen2; 0.992, Probably damaging, Mutation Taster; 1.000, Disease causing.

Discussion

A novel homozygous missense mutation in the SLC5A5 gene causing CH was identified. The NIS protein is composed of 13 transmembrane segments (TMS), an extracellular amino-terminus extremity, and an intracellular carboxy-terminus (1). The variant in the present case is located in TMS IX. The TMS IX contains the highest number of β-OH group-containing amino acids (serine and threonine) and appears to be involved in Na⁺ binding and translocation (2). Interestingly, the residues in the segment are highly conserved in other SLC5A family members (2), indirectly supporting that TMS IX plays a key role for NIS function. The p.Y348D mutation does not substitute one of the serines or threonines, but it is possible that a structural change affects the function of TMS IX. Previous studies have shown that investigating the functional consequences of amino-acid

substitutions in NIS detected in patients with CH yield important mechanistic information (3). Therefore, further functional analysis is important in the present case.

The proposita had hypothyroidism, resulting in mental retardation. Iodine intake in the Sudanese population was found to be much lower compared to the recommended intake (4), suggesting that the affected patients living in North Sudan also had low iodine intake. The degree of nutritional iodine intake has been shown to influence thyroid function in patients with NIS defects (5). Most patients with hypothyroidism due to iodide transport defect respond well to high-dose iodine treatment, with rapid restoration of the euthyroid state because high iodine exposure can lead to nonspecific iodide uptake that allows for some or sufficient thyroid hormone synthesis (5,6). Currently, the affected patients in this study are treated with LT4. However, administration of potassium iodide could be considered, either alone or together with LT4.

In conclusion, a novel homozygous mutation in the *SLC5A5* gene was identified in the affected individuals of this Sudanese family with CH. The mutation is located in TMS IX of the NIS protein, which is essential for Na⁺ binding and translocation. Low iodine intake in Sudan is expected to contribute to the severity of hypothyroidism in these patients.

Acknowledgments

This research was supported by funds from the Esformes Thyroid Research Fund and National Institutes of Health grant MD010722 to R.W.

Author Disclosure Statement

No competing financial interests exist.

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