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Case Report

A novel deletion with two pathogenic variants of *UGT1A1* causing Crigler-Najjar syndrome in two unrelated Chinese

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ABSTRACT

Two Chinese female infants from two unrelated families were diagnosed with Crigler-Najjar syndromes-I (CNS-I) and CNS-II respectively. The CNS-I patient had Serum Total Bilirubin Concentration (STBC) peaked at 26.1 mg/dL. She was not responsive to Phenobarbital and received liver transplantation at 2-year-old. The CNS-II patient's STBC fluctuated between 10.2 mg/dL and 17.4 mg/dL and had a milder phenotype. Sequencing of *Uridine Diphosphate Glucuronosyl Transferase 1A1* (*UGT1A1*) revealed the CNS-I patient carried a heterozygous pathogenic variant in c.392 T > C (p.Leu131Pro) and the CNS-II patient carried a heterozygous pathogenic variant in c.1456 T > G (p.Tyr486Asp). Furthermore, a novel deletion spanning exons 2–4 of *UGT1A1* were detected in both patients. We studied two family members' genotyping results of *UGT1A1* to clarify the inheritance of this microdeletion. To our knowledge, this is probably the first time showing 2 CNS cases both carrying compound heterozygous variations of a known pathogenic variant and a novel microdeletion.

1. Case reports

We studied 2 Chinese female infants diagnosed with CNS-I and CNS-II from two unrelated families respectively. A novel deletion spanning exons 2–4 of *Uridine Diphosphate Glucuronosyl Transferase 1A1* (*UGT1A1*) were detected in both patients.

Patient 1 was recruited to our hospital at 49-day-old. Jaundice occurred on the 5th day of life, and peak Serum Total Bilirubin Concentration (STBC) reached 24.0 mg/dL. She was treated with phototherapy for 7 courses intermittently since jaundice always recurred once phototherapy was discontinued. She was not sensitive to Phenobarbital. A convulsion occurred at 1-year-old with a severe persisted jaundice reaching 26.1 mg/dL. The patient underwent liver transplantation at 2-year-old at another hospital. Patient 2 had a milder phenotype. Jaundice was observed on the 2nd day after birth. She first came to our hospital at 3-month-old. Her STBC fluctuated between 10.2 mg/dL and 17.4 mg/dL. Follow up at 14-month-old showed normal growth and development while jaundice persists. Pre-test counseling was performed and appropriate informed consent was signed by the patients' parents. The criteria of genetic testing received approval from the ethics committees of Children's Hospital of Fudan University (2016–235).

Genetic analysis of *UGT1A1* revealed that patient 1 had a maternally inherited variant: NM_000463.2: c.392T > C (NP_000454.1: p.Leu131Pro) on exon1. Patient 2 had a paternally inherited variant: NM_000463.2: c.1456T > G (NP_000454.1: p. Tyr486Asp) on exon5. We further analyzed and detected a novel heterozygous deletion spanning *UGT1A1*'s exons 2–4 in both patients inherited from her father or mother through CANOES (CNVs with an Arbitrary Number of Exome Samples) which was used for calling CNVs from Next generation sequencing (NGS) data at gene-level and region-level (Fig. 1F,G) [1]. Both two patients were first screened on the Illumina Hiseq 2000/2500 platform. The missense variants and microdeletions of the patients were confirmed by Sanger sequencing and real-time quantitative polymerase chain reaction (PCR), respectively. Family members were also analyzed by Sanger sequencing and real-time quantitative PCR (Fig. 1C, D, H, I). Combined with persistent bilirubinemia, responsiveness to phenobarbital treatment and neurodevelopment of follow-ups, patient 1 was

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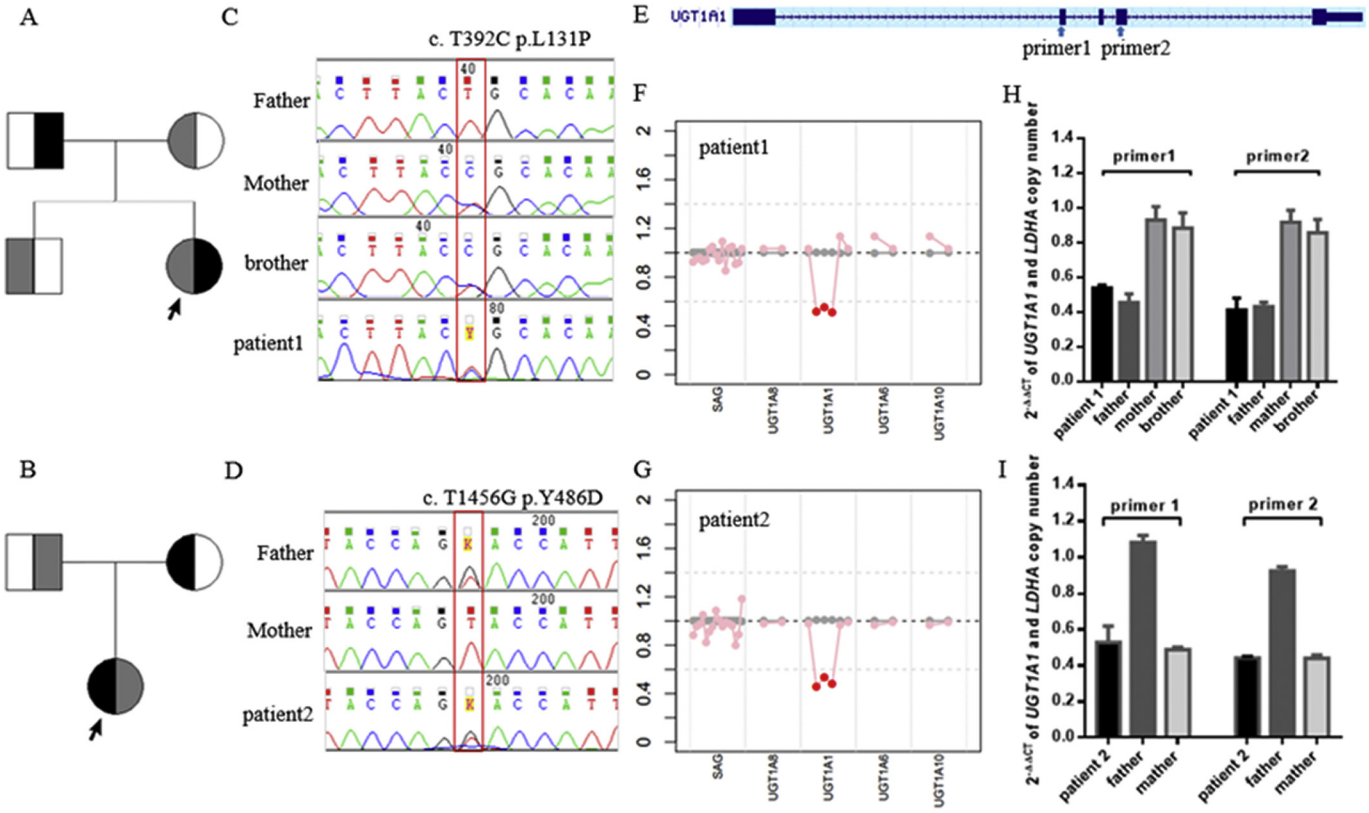

Fig. 1. Genetic analysis of *UGT1A1* of patient 1 and patient 2: The pedigrees of patient 1 (A) and patient 2 (B) (single nucleotide variant in grey, CNV in black); The Sanger sequencing of Leu131Pro and Tyr486Asp (C,D); A novel deletion of exons 2–4 in patient 1(F) and patient 2(G) was detected using CANOES; Real-time PCR confirmation result of patient 1 (H) and patient 2(I), primer 1 and 2 were designed to covering most part of exon 2 and exon 4 (E). The 2^(−ΔΔC_T)value of the normal control was considered to be 1.

diagnosed with CNS-I and patient 2 was diagnosed with CNS-II eventually (Fig. 1A, B).

2. Discussion

To our knowledge, compound heterozygous variations of a pathogenic variant and a microdeletion identified in CNS patient have rarely been reported. There is only one similar case: a 4.5 months old male who was a compound heterozygote for a novel frameshift mutation c.1253delT (p.Met418ArgfsX5) in one allele and an entire *UGT1A1* deletion on the other [2]. His STBC reached 29.0 mg/dL and he died of kernicterus at 13 months of age.

In addition, the glucuronidation activity of Tyr486Asp was reduced by > 95% but not all activity, but Leu131Pro loses total glucuronidation activity of UGT1A1 in enzyme activity study [3,4]. Therefore, we postulate that Leu131Pro is more deleterious than Tyr486Asp which explain that why our two patients developed different outcomes although they had similar heterozygous microdeletions of *UGT1A1*.

In summary, we present two cases of CNS with compound heterozygous variations of a known pathogenic variant and a novel microdeletion of *UGT1A1*, which suggest that when clinical performance can't be explained by heterozygous pathogenic variant, copy number variation might be a potential reason. Therefore, either partial or complete copy deletion of *UGT1A1* should be excluded during genetic analysis.

Author contributions

All 4 authors contributed to the work and meet the criteria for authorship:

Study concept and design: W Li, W Zhou and Y Zhou. Acquisition of data: W Li. and Lin Yang. Analysis and interpretation of data: W Li and Y Zhou. Drafting of the manuscript: W Li. Critical revision of the manuscript for important intellectual content: W Zhou and Y Zhou. Obtained funding: W Zhou and Y Zhou. Study supervision: W Zhou and Y Zhou.

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