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GENETICS

A novel homozygous mutation in *DNAJB13*—a gene associated with the sperm axoneme—leads to teratozoospermia

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Abstract

Purpose To evaluate the unknown genetic causes of teratozoospermia, and determine the pathogenicity of candidate variants.

Methods A primary infertile patient and his family members were recruited in the West China Second University Hospital of Sichuan University. Whole-exome sequencing was performed to identify causative genes in a man with teratozoospermia. Immunofluorescence staining and western blotting were applied to assess the pathogenicity of the identified variant. Intracytoplasmic sperm injection (ICSI) was used to assist fertilization for the patient with teratozoospermia.

Results We performed whole-exome sequencing (WES) and detected a novel homozygous frameshift mutation of c.335_336del [p.E112Vfs*3] in *DNAJB13* on a primary infertile male patient. Intriguingly, we identified abnormal sperm morphology in this patient, with recurrent respiratory infections and chronic cough. Furthermore, we confirmed that this mutation resulted in negative effects on DNAJB13 expression in the spermatozoa of the affected individual, causing ultrastructural defects in his sperm. Remarkably, our staining revealed that DNAJB13 was expressed in the cytoplasm of primary germ cells and in the flagella of spermatids during spermiogenesis in humans and mice. Finally, we are the first group to report a favorable prognosis using ICSI for a patient carrying this *DNAJB13* mutation.

Conclusion Our study revealed a novel homozygous frameshift mutation of c.335_336del [p.E112Vfs*3] in *DNAJB13* involved in teratozoospermia phenotype. Our study greatly expands the spectrum of limited *DNAJB13* mutations, and is expected to provide a better understanding of genetic counseling diagnoses and subsequent treatment of male infertility.

Keywords Teratozoospermia · *DNAJB13* · Frameshift mutation · Intracytoplasmic sperm injection

Introduction

Infertility has become one of three major factors affecting human health, involving 10–15% of global couples [1]. Teratozoospermia is one of the primary causes of male infertility and is characterized by decreased sperm motility and obvious morphological abnormalities [2]. Several gene mutations have thus far been detected in different types of teratozoospermia. For example, biallelic mutations in *SUN5* cause severely acephalic spermatozoa (AS) [3]; and dysfunction of DNAH1 [4] and CFAP family members [5–8] has been

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identified to be responsible for human multiple morphological abnormalities of the sperm flagella (MMAF). Additionally, mutations in *AURKC* [9] and *DPY19L2* [10] account for most cases of macrozoospermia and globozoospermia, respectively. These findings indicate that teratozoospermia exhibits high genetic heterogeneity and multiple phenotypes.

As chaperones of HSP70, the heat shock protein 40 (HSP40) family members participate in many biological processes that include protein folding and oligomeric protein complex assembly [11]. DNAJB13 is a member of the HSP40 co-chaperone family that is located on the radial spokes of the axoneme in sperm flagella and other flagellar structures [12]. In various germ cells of adult mice, DNAJB13 is localized in the cytoplasm of spermatids from steps 2 to 3 onwards, with the strongest expression at steps 9–10, and in the spermatid flagellum [12–14]. In mature spermatozoa, DNAJB13 is localized along the entire length of the sperm flagellum. This expression pattern indicates that DNAJB13 plays a key role in the formation of sperm flagella [15]. However, research studies on DNAJB13 in humans are limited; and only Khouri et al. identified two different homozygous loss-of-function mutations in *DNAJB13* in three unrelated infertility patients with primary ciliary dyskinesia (PCD) [16]. Thus, more clinical research is urgently needed.

In the present study, we identified a novel homozygous frameshift mutation in *DNAJB13* in a primary infertile male patient. Through Papanicolaou staining and clinical examination, we confirmed that this patient presents typical teratozoospermia and is characterized with PCD syndrome; and the detailed phenotype was demonstrated by electron microscopy. Furthermore, we uncovered no typical signs of DNAJB13 expression using western blotting analysis in vitro and immunofluorescence staining of the patient's sperm. We also correlated a favorable prognosis using intracytoplasmic sperm injection (ICSI) with the patient carrying the *DNAJB13* mutation. Our work, therefore, shows that the novel homozygous mutation in *DNAJB13* we observed led to inadequate morphological development and caused male primary infertility.

Materials and methods

Study participants

The primary infertile patient with teratozoospermia and his family members were enrolled at the West China Second University Hospital of Sichuan University, and a total of 200 men with normal fertility were recruited as the control group. This study was approved by the Ethical Review Board of West China Second University Hospital, Sichuan University. Informed consent was obtained from each study participant.

Genetic studies

We performed WES using patient DNA as previously described [17]. Targeted testing of the potentially pathogenic variants in the patient's parents and normal controls was performed by Sanger sequencing. The primers used in PCR analysis were as follows: F, 5′-GCTGGGTGTTACACA GGACA-3′; and R, 5′- AGTCCTCCCACCCAGGTAAG-3′.

Western immunoblotting analysis

Proteins were extracted from cultured cells using a universal protein-extraction lysis buffer (Bioteke) containing a protease inhibitor cocktail (Roche). Denatured proteins were separated on 10% SDS-polyacrylamide gels and transferred onto a polyvinylidene difluoride (PVDF) membrane (Millipore) for immunoblot analysis. The primary antibodies used were anti-DNAJB13 (1:500, Proteintech) and anti-GAPDH (1:1000, Abcam).

Immunofluorescence staining

Sperm and testicular tissues of the patient, normal control, and mouse sperm cells were fixed in 4% paraformaldehyde and then subjected to immunofluorescence staining as described in our previous study [17]. The primary antibodies used were anti-DNAJB13 (1:50, Proteintech) and α-tubulin (1:100, Abcam).

Electron microscopy and concentrated Papanicolaou staining

The appearance of spermatozoa was obtained by concentrated Papanicolaou staining and scanning electron microscopy (SEM), and ultrastructural assessments of flagellar cross-sections were performed with transmission electron microscopy (TEM). We executed Papanicolaou staining, SEM, and TEM as we described previously.

Minigene assay

A functional splicing reporter minigene assay was used to assess the impact of variants on splicing. A genomic segment encompassing intron 3, exon 4, intron 4, exon 5, and intron 5 of the *DNAJB13* gene was PCR-amplified from patient genomic DNA as well as the normal control and was cloned into the minigene vector pSPL3. After transient transfection into cultured cells, the splicing patterns of the transcripts generated from the wild-type and variant constructs were compared by RT-PCR analysis

and sequencing. The primers for genomic amplification were as follows: F, 5′ TTATGGGGTACGGGATCACCA GAATTCGCATGTCTGGGTCTCTGGAT3′; and R, 5′ CGGGATCACCAGATATCTGGGATCCATCCCATC TCACCCACTCTG3′. The primers for RT-PCR were as follows: SD6 (F), 5′ TCTGAGTCACCTGGACAACC 3′; and SA (R), 5′ ATCTCAGTGGTATTTGTGAGC 3′.

Results

Presentation of a primary infertile patient with teratozoospermia phenotype

The affected patient had been diagnosed with primary infertility for 2 years. Semen analysis was performed in triplicate and suggested that this patient had a normal sperm concentration and total sperm number, but diminished sperm motility and defective sperm morphology (Table 1). Papanicolaou staining revealed a typical teratozoospermia phenotype that was characterized by coiled, short, and/or irregular flagella and an abnormal head (Fig. 1a). Using SEM, we further observed the aberrant sperm morphology in detail (Fig. 1b). When we assessed sperm ultrastructure, TEM revealed that sperm from the patient possessed aberrant heads—including round-heads, tapered-heads, and pyriform heads—as well as anomalies of the flagella. For example, the central microtubules (CPs) were missing in the midpiece; the outer dense fibers (ODFs) and double-microtubule doublets (DMTs) were irregularly arranged in the principal piece, and the normal "9+2" structure was disorganized in the end piece (Fig. 1c). Furthermore, the patient's spermatozoa showed an abnormal ratio of head length to width and featured irregular shapes (Fig. 1d). In summary, these data suggested this patient was diagnosed with teratozoospermia.

A homozygous frameshift mutation in DNAJB13 identified in the teratozoospermia patient

We detected in our patient by WES a homozygous frameshift mutation of c.335_336del (p. E112Vfs*3) in *DNAJB13* that was not yet recorded in the public databases that included 1000 Genomes, ExAC database, and dbSNP. To confirm the inherited pattern of this variant in this patient, we further performed Sanger sequencing in his family (Fig. 2a). The unaffected father and mother both carried the heterozygous mutation of c.335_336del (Fig. 2a), suggesting that this homozygous mutation of *DNAJB13* was inherited as an autosomal recessive pattern, which is consistent with previous studies. Moreover, we did not detect this mutation in 200 normal controls, supporting the pathogenicity of this mutation. Given that dysfunction of DNAJB13 is related to PCD, we inquired our patient and he complained of having symptoms of chronic coughing and recurrent respiratory infections, without situs inversus (Supplementary Fig. S1). Based on all evidence, we hypothesized that the novel homozygous mutation of *DNAJB13* was responsible for the primary infertile phenotype in this patient.

The negative effect of the homozygous frameshift mutation of c.335_336del on DNAJB13 expression

To further understand the harmful influence of this mutation in *DNAJB13* on its expression, we used an immunofluorescence assay on the patient's sperm as well as on the normal control. In the normal control group, DNAJB13 was primarily distributed in the head and flagellum of the sperm; however, bare signs of DNAJB13 were detected in the patient (Fig. 2b). Given that this mutation was likely to be located in a splice site, we carried out a minigene splicing assay. Our results showed this mutation did not disrupt the splicing process (Supplementary Fig. S2). In order to confirm the potential deleterious influence of the mutation in *DNAJB13*, we constructed expression

Table 1 Semen analysis of teratozoospermia patient

Semen parameters	Patient	Normal control	Normospermic parameters
Sperm volume (mL)	2.97±0.93	5.96±0.49	≥1.5
Sperm concentration (million/mL)	50.83±11.29	98.00±4.72	≥15
Motility sperm (%)	5.33±1.76	67.33±6.69	≥40
Vitality (%)	24.26±4.42	71.33±4.05	≥58
Absent flagella (%)	25.66±3.05	3.33±2.52	-
Short flagella (%)	26.33±5.51	5.67±2.08	-
Coiled flagella (%)	19.00±4.00	5.33±3.51	-
Bent flagella (%)	16.67±5.35	7.00±1.00	-
Flagella of irregular caliber	11.33±1.53	6.33±1.53	-

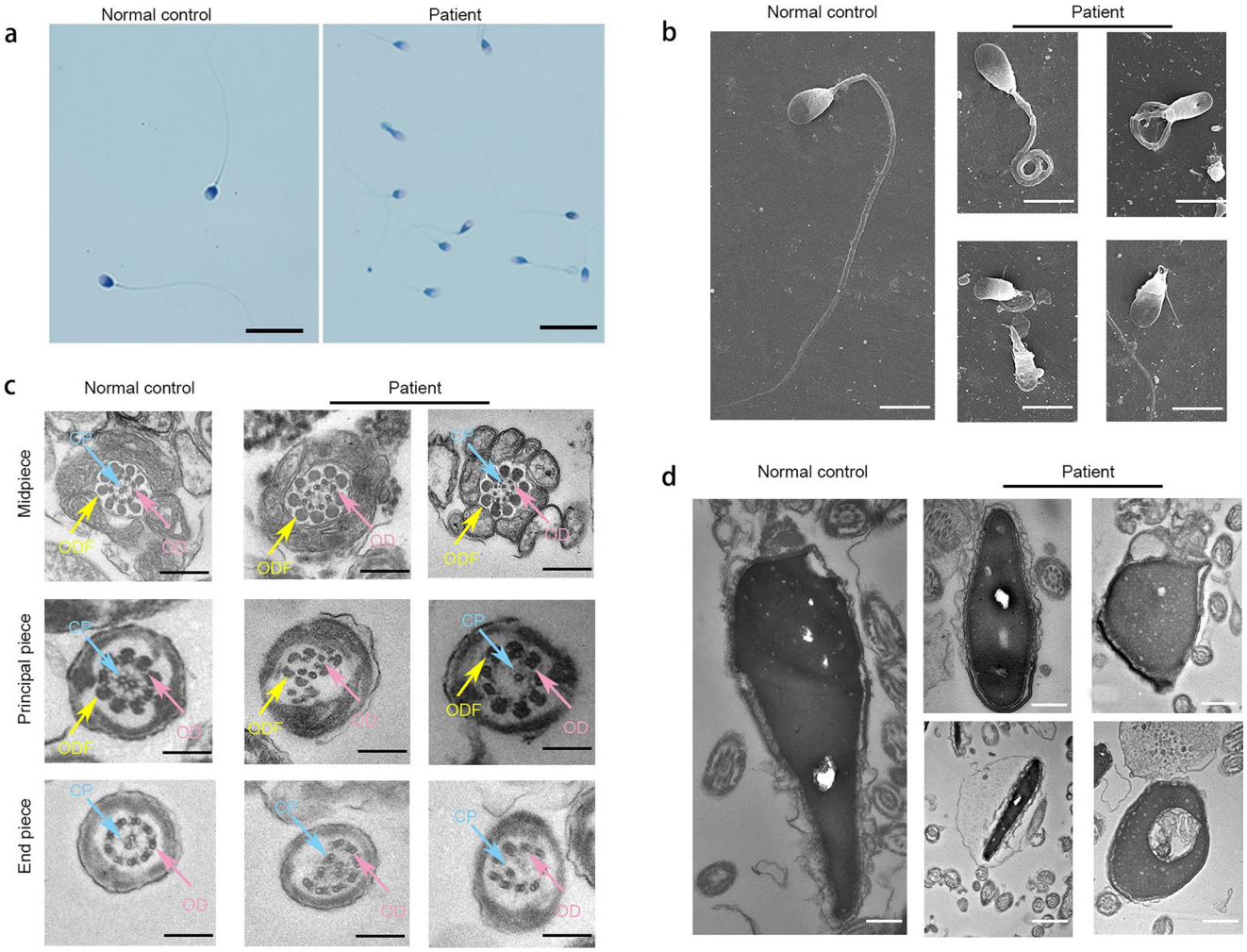

Fig. 1 The teratozoospermia phenotype of the patient. **a** Concentrated Papanicolaou staining of normal sperm and sperm from the patient. The majority of the sperm from the patient manifested anomalous morphology. **b** Detailed defects in sperm flagella were observed in the patient by SEM (scale bars, 5 μm). **c**, **d** TEM analyses of sperm from the patient and normal control. Ultrastructural abnormalities of the head and flagellum observed in the patient sperm compared to the normal control using TEM (scale bars, 100 nm)

vectors for WT-*DNAJB13* (His-tagged wild-type human *DNAJB13*) and mutated *DNAJB13* (His-tagged human mutant *DNAJB13* with c.335_336del), and then transiently transfected them into 293T cells. Compared with WT-*DNAJB13* expression, the truncated protein was detected in mutated-*DNAJB13* expression vectors using western immunoblotting analysis; and this revealed that the mutation caused premature termination (p. E112Vfs*3), similar to the immunofluorescence results of the patient's sperm (Fig. 2c). Through three-dimensional protein-structure analysis, we predicted that this mutation may result in a complete deletion of the β-pleated sheet, which disrupts protein integrity and further loses function (Fig. 2d). These results strongly suggested that this mutation exerted a negative effect on DNAJB13 expression, and that it disrupted sperm flagellar development—ultimately leading to teratozoospermia.

Exploration of the expression pattern of DNAJB13 in the human and mouse testis

To further explore the roles of *DNAJB13* in male reproduction, we investigated the expression and location of DNAJB13 in human and mouse testis. Mouse testicular sections were used for immunofluorescence and showed that DNAJB13 was predominantly expressed in the nucleus and cytoplasm of the spermatogonia, round spermatids, and elongating spermatids, and in the flagella of spermatozoa (Fig. 3a). In addition, we evaluated the expression of DNAJB13 in human testis, showing that DNAJB13 was distributed in the nucleus and cytoplasm of spermatogonia and round spermatids (Fig. 3b). Moreover, staining results revealed that DNAJB13 was detectable in the head and flagellum of various germ cell types, especially those from steps Sc and Sd elongated spermatids (Fig. 3c). *DNAJB13*

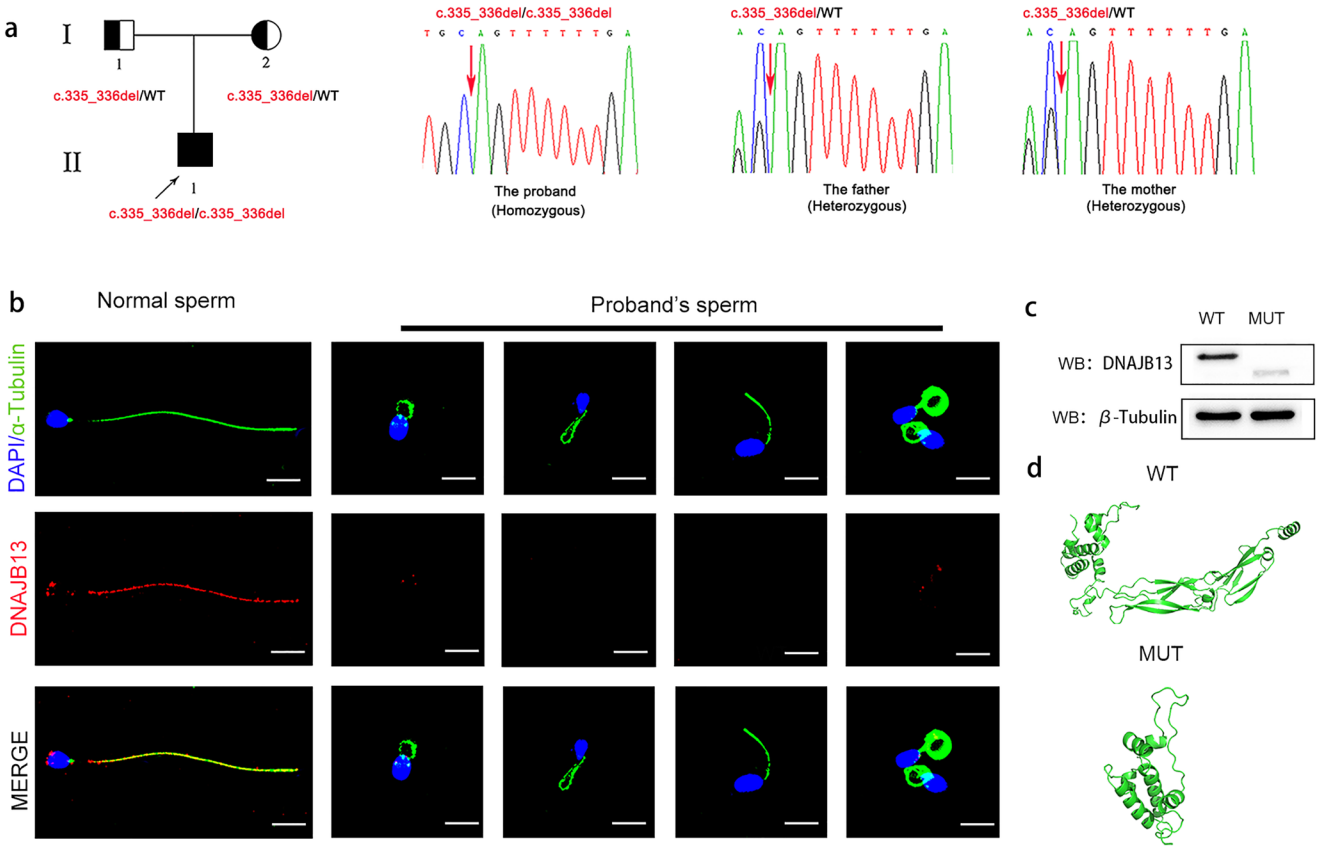

Fig. 2 Genetic findings in the patient with teratozoospermia. **a** A DNAJB13 frameshift mutation was identified in a consanguineous family. Pedigree structure of the family, with the proband indicated by a black arrow. The mutation in c.335_336del was confirmed by Sanger sequencing of the family. The mutation position is indicated by arrow. **b** Results of immunofluorescence staining obtained from the sperm of the patient showed barely perceptible expression of DNAJB13 relative to the normal control (scale bars, 5 μm; red, α-tubulin; green, DNAJB13; blue, DAPI). **c** The *DNAJB13* mutant plasmid or WT plasmid was transfected into 293T cells. Western blotting results showed the truncated protein of DNAJB13 was detectable in the mutant plasmid. **d** Structural illustration of the frameshift mutation in DNAJB13

is therefore a gene that contributes a crucial role for sperm development and is thus involved in male fertility.

Favorable prognosis with ICSI in a patient carrying a mutation in DNAJB13

An ICSI cycle was attempted for this couple, and we received signed informed consent from them to implement the procedure. The basal hormonal data of the patient's wife were normal (Table 2), and consequently, 11 oocytes were aspirated using follicular puncture; seven of these were in metaphase II. The ejaculated spermatozoa from the patient were then injected into the six oocytes for an ICSI cycle, and achieved 80% fertilization (Table 2); six of the seven embryos generated reached cleavage stages. All of the embryos developed to available D3 embryos, including one at 7 II, one at 8 II, two at 4BB, and two at 4BC. One 4BB embryo was then chosen for transplantation which resulted in the patient's wife becoming pregnant (Table 2). With this study, we demonstrated that male sterility associated with a *DNAJB13* mutation could be treated using ICSI, and that dysfunction in DNAJB13 did not hamper embryonic development, in accordance with a previous report.

Discussion

Abnormal sperm morphology is an important risk factor that contributes to male infertility; however, its pathogenesis remains unclear. In our study, we analyzed a primary infertile male affected by teratozoospermia, and our genetic studies revealed a novel homozygous frameshift mutation in the *DNAJB13* of the proband. This loss-of-function mutation disrupted DNAJB13 expression and resulted in defective spermiogenesis. In addition, we could reverse male infertility associated with the *DNAJB13* mutation using ICSI. Thus, our findings strongly supported our hypothesis that this novel homozygous frameshift

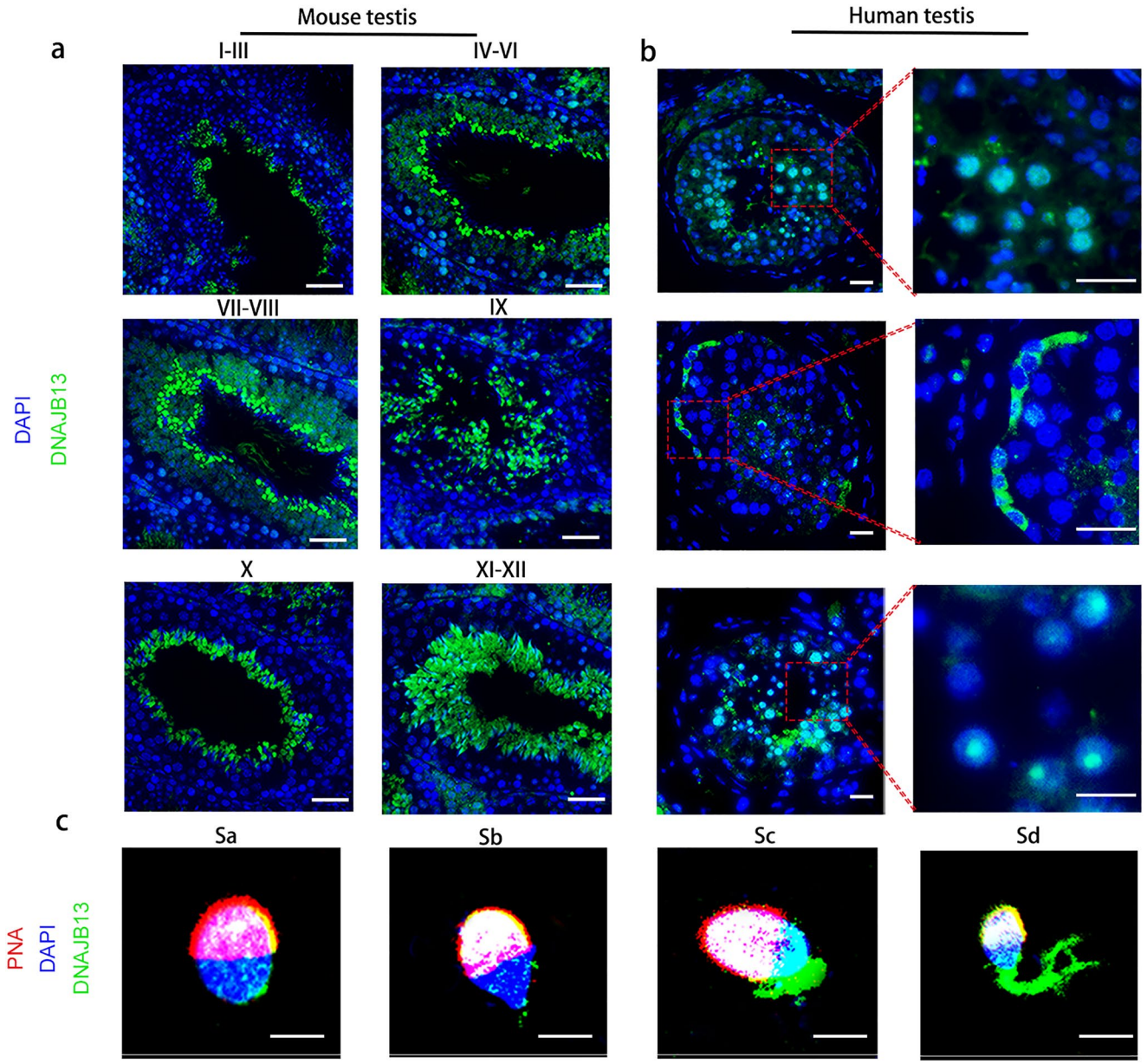

Fig. 3 The expression of DNAJB13 in human and mouse tissues. **a** Representative images of testicular tubules in a mouse showing that DNAJB13 is principally localized to the cytoplasm of round sperm cells and the flagella of different stages of spermatids (scale bar, 5 μm; green, DNAJB13; blue, DAPI). **b** DNAJB13 was detected in the cytoplasm of spermatogonia at different stages and in round spermatids (scale bar, 5 μm; green, DNAJB13; blue, DAPI). **c** Immunofluorescence staining indicated that DNAJB13 was primarily expressed in the cytoplasm of spermatogonia

mutation in *DNAJB13* was the direct genetic cause of the patient's infertility. Idiopathic infertility is an exciting research area in reproductive medicine. DNAJB13, a spermatogenesis-related gene, has attracted the increased attention of researchers, and its role in cilia and flagella has been widely confirmed [12–16]. In 2004, researchers first identified the novel gene *Dnajb13* of the HSP40 family [18] as highly expressed in mouse testicular tissues, and that it manifests high sequence identity between human and mouse. Several studies have revealed that DNAJB13 is localized to the cytoplasm of sperm cells and the tail of mature sperm using western immunoblotting and immunohistochemical analyses, and immunoelectron microscopy was used to further localize DNAJB13 as positioned in the middle piece of the fibrous sheath of the flagellum of mature sperm [13, 15]. Asami et al. knocked out *Dnajb13* in mice through CRISPR/Cas9 technology, and as a result, all of the homozygous mice died of hydrocephalus 4 weeks

Table 2 Clinical features of the patient with ICSI treatment

Age (years)		28
Length of primary infertility history (years)		2
BMI		19.2
Basal hormones	FSH (IU/L)	5.0
	LH (IU/L)	6.0
	E2 (pg/mL)	78
	PRL (ng/ml)	8.8
	Prog (ng/ml)	0.57
	Testo (ng/ml)	0.34
Cycle 1	Protocol	Long
	E2 level on the trigger day (pg/mL)	2688.1
	No. of follicles ≥ 14 mm on the trigger day	8
	No. of follicles ≥ 18 mm on the trigger day	3
	No. of oocytes retrieved	12
ICSI progress	Oocytes injected	11
	Fertilization rate (%)	80% (6/7)
	Cleavage rate (%)	100% (6/6)
	Available D3 embryos	6

after birth [19]. Thus, *Dnajb13* is considered to be a lethal gene in mice. Intriguingly, spermatozoa collected from the chimeric mice harboring homozygous *Dnajb13* KO embryonic stem cells (ESCs) showed tailless, short-tailed, and immotile spermatozoa [19]. Moreover, a *DNAJB13* missense mutation and a splice mutation were screened in three PCD patients who presented with an abnormal percentage of cilia lacking central microtubules. All findings indicated that DNAJB13 was critical for maintaining the integrity of the central complex in motile cilia and flagella [16].

Regrettably, the current literature is principally focused on animal models and research on *DNAJB13* in humans is extremely limited. Therefore, it is of paramount importance to identify additional pathogenic mutations in infertile patients so as to assist in their clinical diagnosis and treatment. So far, only Khouri et al. detected two different homozygous loss-of-function mutations in *DNAJB13* in three unrelated infertile patients with PCD [16]. This study constructs the relationship between *DNAJB13* mutations and infertile phenotype in humans. More importantly, this report identified *DNAJB13* mutations involved in male infertility are linked to recessive inheritance. However, another study detected a heterozygous mutation c.106T>C (p.Ser36Pro) in *DNAJB13* in 9 of 92 idiopathic asthenozoospermia patients and ICSI can help these patients to get offspring successfully [20]. Considering the findings of Khouri et al. and our study, as well as the fertile heterozygous *Dnajb13* KO mice [19], we believed that the inherent pattern associated with *DNAJB13* mutations in male infertility is a recessive inheritance. Thus, it is indicated that the heterozygous *DNAJB13* mutations are not the causative mutations of the infertile patients.

It is worth mentioning that Kartagener syndrome is a specific subtype of PCD, accompanied by situs inversus [21]. Certain PCD-causing genes displayed phenotype heterogeneity, for example, *DNAH5* and *DNAI1* [22, 23]. Individuals carrying pathogenic mutations in *DNAH5* or *DNAI1* present Kartagener syndrome with randomization [22, 23]. However, our patient is characterized by typical PCD syndrome, but no situs inversus. Also, three PCD patients harboring *DNAJB13* mutations in Khouri's report did not present situs inversus as well [16]. Based on these considerations, no existing evidence could demonstrate that loss-of-function in DNAJB13 is associated with Kartagener syndrome, and more future studies might provide clues of the relationship between *DNAJB13* mutations and Kartagener syndrome.

In conclusion, our genetic and functional results provide powerful evidence that the homozygous mutation of c.335_336del (p. E112Vfs*3) in the *DNAJB13* gene is a novel genetic cause of teratozoospermia, and is responsible for defective development of the sperm head and motor disturbances of the flagellum. Our study greatly expands the spectrum of *DNAJB13* mutations in teratozoospermia and confirms a favorable clinical prognosis in male infertility caused by a *DNAJB13* mutation upon using ICSI. The information garnered from this study will provide a better understanding of genetic counseling diagnoses and the subsequent treatment of male infertility caused by teratozoospermia.

Supplementary Information The online version contains supplementary material available at https://doi.org/10.1007/s10815-022-02431-1.

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Author contribution Y.S. designed and supervised the study experiments. Y.Y. collected data and conducted the clinical evaluations. J.L. and C.J. performed TEM and SEM. M.L. wrote the first article draft; M.L., Y.Z., and Y.S. performed immunofluorescence staining and Minigene assay. All authors revised and approved the article.

Data availability Data are available on request from the corresponding author.

Declarations

Ethics approval The study had been approved by the Ethics Committee of West China Second University Hospital, Sichuan University.

Consent to participate Obtained

Conflict of interest The authors declare no competing interests.

References

1. Krausz C, Riera-Escamilla A. Genetics of male infertility. Nat Rev Urol. 2018;15(6):369–84. https://doi.org/10.1038/s41585-018-0003-3.
2. Coutton C, Escoffier J, Martinez G, Arnoult C, Ray PF. Teratozoospermia: spotlight on the main genetic actors in the human. Hum Reprod Update. 2015;21(4):455–85. https://doi.org/10.1093/humupd/dmv020.
3. Zhu F, Wang F, Yang X, Zhang J, Wu H, Zhang Z, et al. Biallelic SUN5 mutations cause autosomal-recessive acephalic spermatozoa syndrome. Am J Hum Genet. 2016;99(4):942–9. https://doi.org/10.1016/j.ajhg.2016.11.002.
4. Wang X, Jin H, Han F, Cui Y, Chen J, Yang C, et al. Homozygous DNAH1 frameshift mutation causes multiple morphological anomalies of the sperm flagella in Chinese. Clin Genet. 2017;91(2):313–21. https://doi.org/10.1111/cge.12857.
5. Tang S, Wang X, Li W, Yang X, Li Z, Liu W, et al. Biallelic mutations in CFAP43 and CFAP44 cause male infertility with multiple morphological abnormalities of the sperm flagella. Am J Hum Genet. 2017;100(6):854–64. https://doi.org/10.1016/j.ajhg.2017.04.012.
6. Liu C, Tu C, Wang L, Wu H, Houston BJ, Mastrorosa FK, et al. Deleterious variants in X-linked CFAP47 induce asthenoteratozoospermia and primary male infertility. Am J Hum Genet. 2021;108(2):309–23. https://doi.org/10.1016/j.ajhg.2021.01.002.
7. Li W, Wu H, Li F, Tian S, Kherraf ZE, Zhang J, et al. Biallelic mutations in CFAP65 cause male infertility with multiple morphological abnormalities of the sperm flagella in humans and mice. J Med Genet. 2020;57(2):89–95. https://doi.org/10.1136/jmedgenet-2019-106344.
8. He X, Liu C, Yang X, Lv M, Ni X, Li Q, et al. Bi-allelic loss-of-function variants in CFAP58 cause flagellar axoneme and mitochondrial sheath defects and asthenoteratozoospermia in humans and mice. Am J Hum Genet. 2020;107(3):514–26. https://doi.org/10.1016/j.ajhg.2020.07.010.
9. Ben Khelifa M, Zouari R, Harbuz R, Halouani L, Arnoult C, Lunardi J, et al. A new AURKC mutation causing macrozoospermia: implications for human spermatogenesis and clinical diagnosis. Mol Hum Reprod. 2011;17(12):762–8. https://doi.org/10.1093/molehr/gar050.
10. Elinati E, Kuentz P, Redin C, Jaber S, Vanden Meerschaut F, Makarian J, et al. Globozoospermia is mainly due to DPY19L2 deletion via non-allelic homologous recombination involving two recombination hotspots. Hum Mol Genet. 2012;21(16):3695–702. https://doi.org/10.1093/hmg/dds200.
11. Jha P, Laskar S, Dubey S, Bhattacharyya MK, Bhattacharyya S. Plasmodium Hsp40 and human Hsp70: a potential cochaperone-chaperone complex. Mol Biochem Parasitol. 2017;214:10–3. https://doi.org/10.1016/j.molbiopara.2017.03.003.
12. Guan J, Yuan L. A heat-shock protein 40, DNAJB13, is an axoneme-associated component in mouse spermatozoa. Mol Reprod Dev. 2008;75(9):1379–86. https://doi.org/10.1002/mrd.20874.
13. Guan J, Kinoshita M, Yuan L. Spatiotemporal association of DNAJB13 with the annulus during mouse sperm flagellum development. BMC Dev Biol. 2009;9:23. https://doi.org/10.1186/1471-213X-9-23.
14. Li W, Liu G. DNAJB13, a type II HSP40 family member, localizes to the spermatids and spermatozoa during mouse spermatogenesis. BMC Dev Biol. 2014;14:38. https://doi.org/10.1186/s12861-014-0038-5.
15. Guan J, Ekwurtzel E, Kvist U, Hultenby K, Yuan L. DNAJB13 is a radial spoke protein of mouse '9+2' axoneme. Reprod Domest Anim. 2010;45(6):992–6. https://doi.org/10.1111/j.1439-0531.2009.01473.x.
16. El Khouri E, Thomas L, Jeanson L, Bequignon E, Vallette B, Duquesnoy P, et al. Mutations in DNAJB13, encoding an HSP40 family member, cause primary ciliary dyskinesia and male infertility. Am J Hum Genet. 2016;99(2):489–500. https://doi.org/10.1016/j.ajhg.2016.06.022.
17. Shen Y, Zhang F, Li F, Jiang X, Yang Y, Li X, et al. Loss-of-function mutations in QRICH2 cause male infertility with multiple morphological abnormalities of the sperm flagella. Nat Commun. 2019;10(1):433 Published 2019 Jan 25. 10.1038/s41467-018-08182-x.
18. Liu G, Lu GX, Xing XW. Molecular cloning of TSARG6 gene related to apoptosis in human spermatogenic cells. Acta Biochim Biophys Sin (Shanghai). 2004;36(2):93–8. https://doi.org/10.1093/abbs/36.2.93.
19. Oji A, Noda T, Fujihara Y, Miyata H, Kim YJ, Muto M, et al. CRISPR/Cas9 mediated genome editing in ES cells and its application for chimeric analysis in mice. Sci Rep. 2016;6:31666. https://doi.org/10.1038/srep31666.
20. Li WN, Zhu L, Jia MM, Yin SL, Lu GX, Liu G. Missense mutation in DNAJB13 gene correlated with male fertility in asthenozoospermia. Andrology. 2020;8(2):299–306. https://doi.org/10.1111/andr.12685.
21. Omran H, Häffner K, Völkel A, Kuehr J, Ketelsen UP, Ross UH, et al. Homozygosity mapping of a gene locus for primary ciliary dyskinesia on chromosome 5p and identification of the heavy dynein chain DNAH5 as a candidate gene. Am J Respir Cell Mol Biol. 2000;23:696–702. https://doi.org/10.1165/ajrcmb.23.5.4257.
22. Olbrich H, Häffner K, Kispert A, Völkel A, Volz A, Sasmaz G, et al. Mutations in DNAH5 cause primary ciliary dyskinesia and randomization of left-right asymmetry. Nat Genet. 2002;30(2):143–4. https://doi.org/10.1038/ng817.
23. Guichard C, Harricane MC, Lafitte JJ, Godard P, Zaegel M, Tack V, et al. Loss-of-function mutations in a human gene related to Chlamydomonas reinhardtii dynein IC78 result in primary ciliary dyskinesia. Am J Hum Genet. 1999;65(6):1508–19. https://doi.org/10.1086/302683.