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BRIEF COMMUNICATION

Expanding the clinical and molecular spectrum of the *CWC27*-related spliceosomopathy

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Abstract

Cyclophilins are a type of peptidyl-prolyl *cis-trans* isomerases. CWC27, one of the known human cyclophilins, is recruited by the spliceosome for the pre-mRNA splicing process. Biallelic deleterious variants in *CWC27* lead to a spectrum of overlapping phenotypes including retinal degeneration, skeletal anomalies, short stature, and neurological defects. The present work reports a woman showing these clinical features, in addition to hypergonadotropic hypogonadism, hypoplastic/agenesic teeth, and cataracts, not previously associated with such phenotypic spectrum. Whole exome sequencing on this patient identified a novel *CWC27* homozygous variant predicted to originate a severely truncated protein and the consequent loss of functionality. The clinical and genetic characterization of such patient could provide further insight into the underlying causes of the spliceosomopathies.

Introduction

Cyclophilins are a type of peptidyl-prolyl *cis-trans* isomerases involved in binding to proline-containing peptides and protein folding. In humans, there are 8 cyclophilins associated with the spliceosome [1]. One of them, CWC27, shows an N-terminal PPIase domain containing a proline-binding pocket and a large C-terminal repetitive low complexity region of unknown function [2]. Although CWC27 does not have prolyl-isomerase activity, its conserved ability to bind to proline could explain its role in the spliceosome [1, 3].

Biallelic deleterious *CWC27* variants have been associated with a rare phenotypic spectrum (MIM#250410) characterized by retinal degeneration, short stature, skeletal anomalies, and neurological disorders [4]. Herein, we report a woman with retinitis pigmentosa (RP), several skeletal defects, and other relevant clinical features which have never been associated before with the abovementioned phenotypic spectrum. Whole exome sequencing on this patient identified a homozygous *CWC27* variant predicted to cause a severely truncated protein product.

Material and methods

Patient and clinical data

Written informed consent for participation was obtained and subsequently revised by the ethics review board of the Hospital Clínico (Santiago de Compostela, Spain). The patient, 18 years old at the time of this report, was referred to the Department of Pediatrics at Hospital Clínico because

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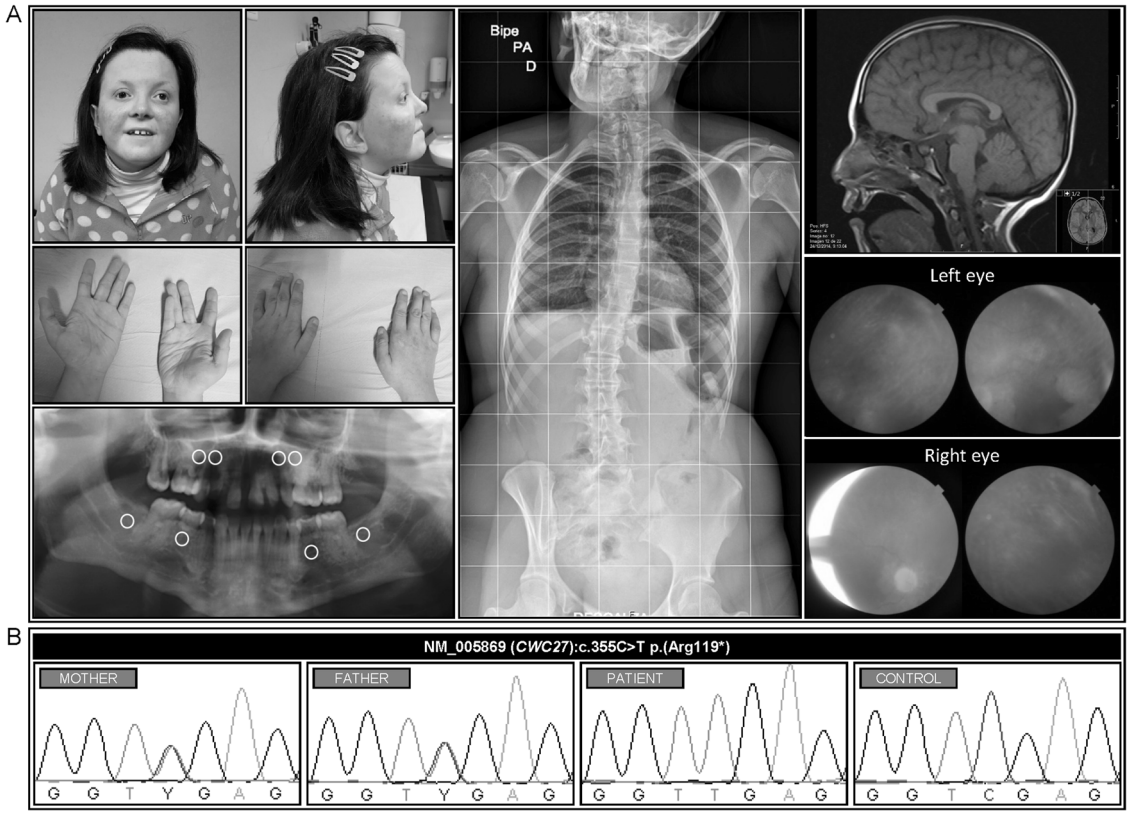

Fig. 1 **a** Clinical features of patient. White circles in the orthopantomography indicate the location of dental agenesis at 14 years of age. Authors have obtained consent to publication of the images. **b** Sanger validation of *CWC27* variant. Both consanguineous parents were heterozygous for the c.355C>T p.(Arg119*) variant, suggesting autosomal recessive inheritance

of intellectual disability, delayed physical growth, skeletal defects, and visual impairment. She is the first child of healthy consanguineous Spanish parents and was born at 37.5 weeks by spontaneous vaginal delivery with a birth weight of 2210 g (<3rd centile) and APGAR score of 8/9/9. She achieved to sit up around 10 months of age and independent walking at 19 months.

Electroencephalogram, serum/urinary amino acid analysis, karyotyping, and array CGH yielded normal results. Brain magnetic resonance revealed ventricular system asymmetry, hypoplasia of the corpus callosum and minimum cerebellar atrophy. She was found to have bilateral cataracts, retinal dystrophy, bilateral 10 dB hearing loss, hyperostosis frontalis interna, 11 pairs of ribs, osteopenia in hands, hypothyroidism (TSH: 13.8 [0.35–5.5mIU/l]; free T4: 1.26 [0.89–1.8 ng/dl]) and hypergonadotropic hypogonadism (FSH: 69 IU/l, LH: 61 IU/l, and 17β-estradiol: 22.4 pg/l). Pelvic ultrasound identified normal uterine structure and nonvisualized ovaries. Spontaneous puberty was absent and pubertal induction with estrogen replacement therapy was required.

Nowadays, she also shows divergent strabismus, malar telangiectasias, thinning of eyebrows, long philtrum, thin lips, hypoplastic teeth, dental agenesis, low weight (43.4 kg; −1.51 SDS; <3rd centile), short stature (139.7 cm; −3.64 SDS; <3rd centile), kyphoscoliosis, bilateral elbow, and left achilles contractures, lower limb dysmetria, clubbing of fingers, and ungueal dystrophy in feet (Fig. 1a). Furthermore, she presents echolalia, inattention, hyperactivity, cheerful character, and good social integration.

Whole exome sequencing (WES)

DNA was isolated from peripheral blood using the chemagic™ MSM I instrument (PerkinElmer chemagen Technologie GmbH, Baesweiler, Germany), following the manufacturer's instructions. WES was performed on the captured DNA library (xGen Exome Research Panel; Integrated DNA Technologies, Coralville, IA, USA) using Illumina NextSeq 500 (Illumina, San Diego, CA, USA); Average sequencing depth: 101×; Average coverage of target regions with sequencing depth >30×: 95.36%. Variants of interest were confirmed by Sanger sequencing on the family trio and submitted to Leiden Open Variation Database (LOVD; https://databases.lovd.nl/shared/genes/CWC27).

Results

WES identified a NM_005869 (*CWC27*):c.355C>T p.(Arg119*) variant not present in Human Gene Mutation Database, Exome Sequencing Project, Exome Aggregation Consortium or Genome Aggregation Database (Table 1 Supplementary information). The c.355C>T transition is predicted to cause the replacement of a moderately conserved Arginine located within the PPIase domain by a stop codon, resulting in a truncated protein product (Alamut Visual version 2.11, Interactive Biosoftware, Rouen, France). Sanger sequencing confirmed the homozygous state of c.355C>T in the patient and the heterozygous in her consanguineous parents (Fig. 1b).

Discussion

Association of RP, short stature and brachydactyly has been previously reported in a few studies [4–6]. The largest one collected 10 patients from 7 families of different ethnicity, who showed biallelic *CWC27* variants and the abovementioned common phenotype in combination with other clinical features [4]. This study suggested that deleterious variants within the N-terminal region—especially the proline-binding pocket—would be responsible for the more severe phenotypes. In this regard, it is likely that the truncated protein originated by the p.(Arg119*) variant identified by WES in our patient, offspring of a consanguineous couple, would cause the loss of the protein structure stability and the subsequent impaired function of the spliceosome. The severe phenotype observed in our patient is characterized by several skeletal defects, hypoplastic/agenesic teeth, cataracts, and hypergonadotropic hypogonadism; in addition to the common phenotype shared with patients from previous studies (Table 1). Two of such patients showed elevated FSH levels, unspecific endocrinological dysfunction, normal genitalia, and secondary sexual development [6]. Our patient also shows elevated FSH and LH since early adulthood, but she did not present spontaneous pubertal development. These new findings strengthen the assumption about the possible effect of *CWC27* over the ectodermal and internal organs, apart from its critical roles over development and functional preservation of retina and skeletal tissue [4].

Despite defects in certain spliceosomal components (i.e., PRPF3, SNRNP200, or RP9) lead to RP, different phenotypes could be seen when deleterious variants affect to other genes encoding spliceosome-related proteins; i.e., altered *SNRPB* is associated with cerebrocostomandibular syndrome or *PUF60* with Verheij syndrome [7–9]. Some of the clinical features of these syndromes could resemble those

Table 1 Clinical features of patients with *CWC27* deleterious variants

Patient	DNA variant	Craniofacial defects	Neurological	Ocular	BD	SS	Other
1: II-3 [4]	c.943G>T p.(Glu315*)	+	SD; DW	RP	+	+	Cafe-au-lait spots
1:II-4 [4]	c.943G>T p.(Glu315*)	+	SD, DW	RP	+	+	Cafe-au-lait spots, hallux valgus both sides, flat feet
2:II-1 [4, 6]	c.495G>A p.Leu167Glyfs*3	+	ID, PR	RP	+	+	Unspecific endocrinological dysfunction
2:II-2 [4, 6]	c.495G>A p.Leu167Glyfs*3	+	ID	RP	+	+	Unspecific endocrinological dysfunction
3:II-1 [4]	c.1002dupA p.(Val335Serfs*13)	–	–	RP	–	–	–
4:II-3 [4]	c.599+1G>A p.[Val191Lysfs*3; Val166Lysfs*3]	+	ID; FD; PR	fERG; aVEP	+	+	Alopecia, absent eyebrows and eyelashes, ichthyosis, multiple KC
4:II-4 [4]	c.599+1G>A p.[Val191Lysfs*3; Val166Lysfs*3]	+	ID; FD; PR; CA	fERG; aVEP	+	+	Alopecia, absent eyebrows and eyelashes, ichthyosis, ectopic testis
5:II-1 [4]	c.19C>T p.(Gln7*) c.427C>T p.(Arg143*)	+	ID; SD; DW; FD; ACM	RP	+	+	Neonatal hypotonia, inguinal hernia, bladder cyst, heart murmur
6:II-1 [4]	c.19C>T p.(Gln7*) c.427C>T p.(Arg143*)	+	–	Normal	+	+	Bilateral SVC, VSD, horseshoe kidney
7:II-1 [4]	c.617C>A p.(Ser206*) c.1002dupA p.(Val335Serfs*13)	–	–	LCA	+	+	–
1 [this study]	c.355C>T p.(Arg119*)	+	ID	RP; bilateral cataracts	+	+	VBSA, hypoplasia of the corpus callosum, minimum cerebellar atrophy, bilateral hearing loss, hypoplastic teeth, dental agenesis, malar telangiectasias, thinning of eyebrows, hypergonadotropic hypogonadism.

In brackets, article reference

BD brachydactyly, *SS* short stature, *SD* speech delay, *DW* delay of walking, *RP* retinitis pigmentosa, *ID* intellectual disability, *PR* psychomotor retardation, *FD* feeding difficulty, *KC* kidney cysts, *CA* cortical atrophy, *fERG* flat electroretinogram signals, *aVEP* altered visual evoked potential response, *ACM* Arnold–Chiari malformation, *SVC* superior vena cava, *VSD* ventricular septal defect, *LCA* leber congenital amaurosis, *VBSA* ventricular brain system asymmetry

shown by our patient, highlighting the phenotypic variability caused by deleterious variants in *CWC27* [4]. Furthermore, it is possible that CWC27 could exert other possible functions by interaction with other proteins/structures through additional domains/motifs [2]. Such assumption has already been proven in the multidomain cyclophilin PPIE which, apart from its function in mRNA splicing, participates in chromatin remodeling complexes and is also related to transcription-coupled DNA repair. Nevertheless, it is still unknown whether CWC27 may interact with other proteins or structures, just as happens with PPIE. In this context, it should be noted that only the PPIase domain of CWC27 was modeled and around 200 residues remain uncharacterized [2].

In summary, the novel homozygous NM_005869 (*CWC27*):c.355C>T p.(Arg119*) variant identified in our patient expands the clinical and molecular spectrum of the disease phenotype associated with this cyclophilin. These new findings could be helpful for early diagnosis, management, and genetic counseling of patients with spliceosomopathies.

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Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

References

1. Ulrich A, Wahl MC. Structure and evolution of the spliceosomal peptidyl-prolyl cis-trans isomerase Cwc27. Acta Crystallogr D Biol Crystallogr. 2014;D70:3110–23. https://doi.org/10.1107/S1399004714021695.
2. Rajiv C, Davis TL. Structural and functional insights into human nuclear cyclophilins. Biomolecules. 2018;8:E161. https://doi.org/10.3390/biom8040161.
3. Davis TL, Walker JR, Campagna-Slater V, Finerty PJ, Paramanathan R, Bernstein G, et al. Structural and biochemical characterization of the human cyclophilin family of peptidyl-prolyl isomerases. PLoS Biol. 2010;8:e1000439. https://doi.org/10.1371/journal.pbio.1000439.
4. Xu M, Xie YA, Abouzeid H, Gordon CT, Fiorentino A, Sun Z, et al. Mutations in the spliceosome component CWC27 cause retinal degeneration with or without additional developmental anomalies. Am J Hum Genet. 2017;100:592–604. https://doi.org/10.1016/j.ajhg.2017.02.008.
5. Phillips CI, Wynne-Davies R, Stokoe NL, Newton M. Retinitis pigmentosa, metaphyseal chondrodysplasia, and brachydactyly: an affected brother and sister. J Med Genet. 1981;18:46–9.
6. Lorda-Sanchez I, Trujillo MJ, Gimenez A, Garcia-Sandoval B, Franco A, Sanz R, et al. Retinitis pigmentosa, mental retardation, marked short stature, and brachydactyly in two sibs. Ophthalmic Genet. 1999;20:127–31.
7. Růžičková Š, Staněk D. Mutations in spliceosomal proteins and retina degeneration. RNA Biol. 2017;14:544–52. https://doi.org/10.1080/15476286.2016.1191735.
8. Tooley M, Lynch D, Bernier F, Parboosingh J, Bhoj E, Zackai E, et al. Cerebro-costo-mandibular syndrome: clinical, radiological, and genetic findings. Am J Med Genet A. 2016;170A:1115–26. https://doi.org/10.1002/ajmg.a.37587.
9. Low KJ, Ansari M, Abou Jamra R, Clarke A, El Chehadeh S, FitzPatrick DR, et al. PUF60 variants cause a syndrome of ID, short stature, microcephaly, coloboma, craniofacial, cardiac, renal and spinal features. Eur J Hum Genet. 2017;25:552–9. https://doi.org/10.1038/ejhg.2017.27.