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[Article]

Clinical spectrum of male patients with *OFD1* mutations

Running title: Phenotypes of male *OFD1* patients

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Conflict of Interest

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Abstract

Oral-facial-digital syndrome type 1 (OFD1) is a ciliopathy characterized by oral, facial, and digital malformations that are often accompanied by polycystic lesion of the kidney and central nervous involvement. OFD1 shows an X-linked recessive inheritance caused by mutation in the *OFD1* gene (Xp22.2). The disease is generally considered embryonic lethal for hemizygous males. However, males with *OFD1* mutations were recently reported. Here, we report four additional Japanese male patients with *OFD1* variants and describe the variable clinical manifestation and disease severity among the four patients. Patient 1 with pathogenic indels including a 19-bp deletion and 4-bp insertion (c.2600-18_2600delinsACCT) had end-stage renal disease (ESRD) with bilateral cystic kidneys and sensory hearing loss. He showed neither intellectual disability nor facial or digital dysmorphism. Patient 2 with a missense variant in exon 7 (c.539A>T, p.Asp180Val) presented head circumference enlargement, brachydactyly, high-arched palate, micropenis, severe global developmental delay, and ESRD. Patient 3 had a single base substitution at the splice donor site of intron 16 (c.2260+2T>G) causing a 513-bp deletion at the transcript level. The patient had chronic kidney disease and speech delay, but no oral, facial, or digital dysmorphism.

His uncle (patient 4) carried the same *OFD1* variant and showed ESRD with extra-renal malformations including obesity and micropenis, which was previously diagnosed as Bardet-Biedl syndrome. The *OFD1* mutations were not lethal in these four male patients, likely because the three mutations were in-frame or missense. This report provided insights into the onset mechanism and phenotype-genotype association in patients with *OFD1* mutations.

Keywords

Oral-facial-digital syndrome, *OFD1*, ciliopathy, male patients

Introduction

Oral-facial-digital syndrome type 1 (OFD1; MIM #311200) is a rare X-linked congenital disorder characterized by oral, facial, and digital abnormalities and is mostly seen in females. The incidence of OFD1 is approximately 1 in 50 000 live births [1]. This disorder is often accompanied with polycystic lesion of the kidney and a broad range of central nervous involvement. Cystic changes in the liver and pancreas have also been described in patients with OFD1. Central nervous involvement, which may include intellectual disability, hydrocephalus, cerebellar anomalies, porencephaly, and corpus callosum defects, is seen in 40% of patients with OFD1 [2]. Polycystic lesion of the kidney occurs in approximately 15–50% of female patients, most of whom progress to end-stage renal disease (ESRD) [1, 2]. In contrast, those without polycystic lesion of the kidney have normal renal function [3]. Some patients without external features are diagnosed by visceral features such as polycystic kidney in adulthood [4].

OFD1 is located on the short arm of the X chromosome (Xp22.2), and its mutations are responsible for OFD1 [5]. To date, at least 157 *OFD1* mutation sites have been cataloged in the Human Gene Mutation Database (HGMD) (http://www.hgmd.cf.ac.uk/). *OFD1* encodes a centrosome/basal

82 body protein necessary for the assembly of primary cilia and left-right axis
83 determination [6]. Mouse mutants of *Ofd1* have absent or malformed cilia in
84 various tissues [7]. Thus, OFD1 is considered a form of ciliopathy. Phenotypic
85 variability is often seen in affected females including those within the same
86 family, which may be partly due to the varying degree of somatic mosaicism
87 resulting from random X inactivation [8].
88 OFD1 is generally thought to cause lethality in males during the fetal
89 period, and almost all affected patients are females. However, recently, males
90 with *OFD1* mutation have been described. As in classical OFD1, there are four
91 phenotypes of *OFD1* mutation in males: X-linked Joubert syndrome with
92 polydactyly and retinal involvement (JBTS10), Simpson-Golabi-Behmel
93 syndrome type 2 (SGBS2), retinitis pigmentosa 23 (RP23), and unclassified
94 malformation syndromes [9]. However, male patients are so rare that very
95 little is currently known about the phenotype-genotype correlation for OFD1
96 in males. We now report four Japanese male patients from three families who
97 had novel *OFD1* mutations. They all had chronic kidney diseases including
98 ESRD, but with varying degrees of extra-renal malformations and central
99 nervous system involvement. Our findings may elucidate the phenotype-
100 genotype correlation in OFD1.

Materials and methods

Patients

Patient 1

Patient 1 was an 8-year-old boy who was born to non-consanguineous parents at 36 weeks and 5 days of gestation. His birth weight was 2881 g. During the neonatal period, he was diagnosed with an acute kidney injury due to a severe upper-pole primary ureteropelvic junction obstruction of the left kidney and non-functional right kidney (Fig. 1a). Ultrasound examination showed bilateral cystic kidneys with a multicystic dysplastic right kidney. After an emergency nephrostomy, his kidney function improved temporarily but then progressively declined. At the age of 8, he progressed to ESRD and underwent a kidney transplant. He also had bilateral sensory hearing loss, but no other abnormalities including facial, oral, or digital deformities. He had no failure of growth (height 116.9 cm, body weight 20.3 kg) or intellectual disability, and brain magnetic resonance imaging did not show abnormalities, including molar tooth sign. He is now 11 years old, and his transplanted renal function is preserved.

Patient 2

Patient 2 was a 4-year-old boy. He was also born to non-consanguineous parents at 37 weeks and 1 day of gestation. His birth weight was 3160 g. A leptomeningeal cyst and hydrocephalus were detected during the neonatal period. He presented progressively worsening kidney dysfunction at the age of 1 and has been on peritoneal dialysis due to ESRD since the age of 2. Magnetic resonance imaging showed bilateral enlarged kidneys with numerous cysts (Fig. 1b), and kidney biopsy indicated nephronophthisis. He had congenital deformations including head circumference enlargement, brachydactyly, high-arched palate, micropenis, and optic nerve hypoplasia. He also displayed a severe intellectual disability and motor developmental delay. He is now 6 years old. He undergoes peritoneal dialysis and shows growth failure (height 82.0 cm, weight 12.3 kg).

Patient 3

Patient 3 was a 3-year-old boy. He was also born to non-consanguineous parents at 38 weeks and 4 days of gestation. His birth weight was 2638 g. At the age of 1, bilateral kidney enlargement with high intensity was detected by ultrasound (Fig. 1c and d). At that time, his estimated glomerular filtration rate was 62.2 ml/min/1.73 m^2, although his urinalysis findings were normal, and he was diagnosed with chronic kidney disease. He displayed a speech delay and

autistic features with hyperactivity, but no facial, oral, or digital abnormality. He exhibited short stature (85.6 cm) at the age of 3. He had a family history of renal disease, and his pedigree is shown in Figure 2. One of his uncles (II-1 in Fig. 2) also had ESRD and died at a young age. His mother (II-4) and grandmother (I-2) had normal urinalysis findings, with no oral and dental symptoms.

Patient 4

Patient 4 was a 29-year-old male and a maternal uncle of patient 3 (II-5 in Fig. 2). He had ESRD and underwent a renal transplantation at the age of 14. At the age of 13, he also had extra-renal malformations such as obesity (height 156 cm and body weight 91.7 kg), borderline intellectual disability (his intelligence quotient was 76 at the age of 12), and micropenis. His serum levels of follicle stimulating hormone and luteinizing hormone were normal (3.56 mIU/ml and 4.06 mIU/ml), and testosterone was low (0.91 ng/dl). He had no retinal disorder or hearing loss. He was diagnosed with Bardet-Biedl syndrome based on the clinical manifestations.

Analysis of *OFD1* mutations

Genetic analysis was performed after informed consent was received. This

158 study was approved by the institutional review board of Kobe University
159 School of Medicine. Genomic DNA was isolated from peripheral blood
160 leukocytes of the patients and their family using the QuickGene whole blood
161 kit S (Kurabo, Osaka, Japan). Additionally, total RNA was extracted from
162 blood leukocytes using the NucleoSpin RNA Blood (Macherey-Nagel, Hoerdt,
163 France) and an RNA stabilization agent (RNAlater; Thermo Fisher Scientific,
164 Waltham, MA, USA). Total RNA was reverse-transcribed to cDNA using
165 SuperScript III First-Standard Synthesis SuperMix (Thermo Fisher Scientific)
166 for cDNA analysis.
167 Targeted sequencing using next-generation sequencing (NGS) was
168 conducted for 91 genes in patient 1 (Supplementary Table 1) and 128 genes in
169 patient 2 (Supplementary Table 2) associated with congenital anomalies of the
170 kidney and urinary tract or nephronophthisis-related ciliopathies as cataloged
171 in the OMIM database (http://www.omim.org/) or PubMed
172 (http://www.ncbi.nlm.nih.gov/pubmed). NGS samples were prepared using the
173 HaloPlex target enrichment system kit (Agilent Technologies, Santa Clara,
174 CA, USA) according to the manufacturer's instructions. The captured DNA
175 samples were amplified by PCR and sequenced using the MiSeq platform
176 (Illumina, San Diego, CA, USA). Analysis of the NGS data was performed

using the SureCall software (Agilent Technologies). The mutations detected by NGS were confirmed by standard Sanger direct sequencing using the 3130 Genetic Analyzer (Thermo Fisher Scientific). For patient 3, the *OFD1* mutation was directly detected by standard Sanger sequencing without NGS because the patient's family tree indicated the mutation. The direct sequencing data were analyzed by CLC Main Workbench version 6.7.1 (CLC bio, Aarhus, Denmark). For variant descriptions, NM_003611.2 was used as a reference sequence of *OFD1*.

For prioritized candidate novel exonic variants, wANNOVAR (http://wannovar.wglab.org/) was used as an *in silico* evaluation tool to predict whether the mutations were pathogenic.

Results

We identified three different mutations by NGS and direct sequencing of *OFD1* in the four male patients. In patients 3 and 4, their pedigree indicated an X-linked dominant ciliopathy; therefore, their mutations were analyzed only by direct sequencing of *OFD1*.

In patient 1, we identified a novel 19-bp deletion and 4-bp insertion (c.2600-18_2600delinsACCT) in exon 20 of *OFD1* and its splice acceptor site,

196 resulting in an in-frame mutation (p.Ser867_Asp869delinsAsn) (Fig. 3a and b,
197 left panels). His mother was heterozygous for this aberration.
198 In patient 2, we identified a novel missense variant in exon 7 of
199 *OFD1* (c. 539A>T, p.Asp180Val) (Fig. 3a, middle panel). His mother did not
200 carry this mutation; thus, the mutation was likely *de novo* in the patient. This
201 variant was estimated to be pathogenic by *in silico* analysis using
202 wANNOVAR. The analysis showed a CADD score of 27.7 and predicted the
203 mutation as deleterious by PROVEAN and SIFT algorithms, probably
204 damaging by PolyPhen2, and disease-causing by Mutation Taster.
205 In patient 3 and 4, a single base substitution at the splice donor site of
206 intron 16 (c.2260+2T>G) was detected. The result of the cDNA analysis
207 showed that this aberration caused a 513-bp in-frame deletion in exon 16 and
208 17 (Fig. 3a and b, right panels).
209
210 **Discussion**
211 We presented two familial and two sporadic male patients with novel
212 mutations in *OFD1*. All patients had severe renal dysfunction, but they
213 presented different extra-renal phenotypes.
214 Phenotype-genotype correlation in female OFD1 patients has been

shown to be dependent on the location of the mutation. Previous reports indicated that the location of mutations causing typical OFD1 in female patients extends only to exon 17 out of 23 coding exons in *OFD1* [1, 10]. Additionally, mutations in exon 3, 8, 9, 13, and 16 have been associated with intellectual disability [5], while those in exon 12 have been associated with renal cysts [1]. Phenotypic variability is often seen in affected females even within the same family. Thauvin-Robinet *et al.* suggested that skewed X inactivation is partially involved in the pathogenesis leading to the intrafamiliar clinical variability [2]. Bisschoff *et al.* reported that the direction of skewing is not correlated with disease severity [11]. Nevertheless, nearly all reports on the phenotype-genotype correlation in OFD1 were on female patients. Male OFD1 patients are exceedingly rare, and therefore, very little is currently known on the phenotype-genotype correlation of *OFD1* mutations in male patients.

The phenotypic spectrum associated with *OFD1* mutations has been recently extended to include JBTS10, SGBS2, RP23, and unclassified malformation syndromes as noted above. JBTS is characterized by a unique cerebellar and brain stem malformation, hypotonia, developmental delay, and unusual breathing pattern. Several male patients presenting symptoms of

234 classical JBTS who have mutations in *OFD1* have been reported [12, 13].
235 SGBS is characterized by overgrowth, coarse face, and other congenital
236 anomalies including heart defect. SGBS2 with an *OFD1* mutation of a male
237 patient has also been reported [14]. The patient had a frameshift mutation in
238 exon 16 and showed macrocephaly, severe intellectual disability, low-set ears,
239 digital malformations, and obesity. Except for the patient, other affected males
240 in his family died in their early postnatal period. In both JBTS10 and SGBS2,
241 females are clinically inconspicuous. Webb *et al.* reported a deep intronic
242 mutation in intron 9 of *OFD1* that produced an aberrant transcript and reduced
243 level of correctly spliced transcript, causing X-linked RP [15]. Sharma *et al.*
244 reported an atypical presentation of *OFD1* mutation in a male patient without
245 the classical OFD1 phenotype. This patient presented ESRD without any
246 evidence of polycystic kidney disease [16]. The spectrum of *OFD1* mutations
247 in males reported previously is summarized in Table 1. The findings
248 demonstrate phenotypic variability of patients with *OFD1* mutation. We were
249 the first to report a patient with *OFD1* mutation presenting a Bardet-Biedl
250 syndrome-like phenotype (patient 4).
251 Coene *et al.* reported a case of JBTS with *OFD1* mutation and
252 proposed that the inverse correlation between the length of OFD1 mutant

253 protein and phenotypic severity is due to differences in the binding of the
254 mutant protein to functionally interacting proteins and disruption of ciliary
255 localization [12]. They also suggested that variable degrees of RNA
256 degradation could contribute to the differences in phenotype. Thus, the
257 location and type of the mutation may be associated with the disease severity.
258 To the best of our knowledge, there have been three reports to date on
259 perinatal death of OFD1 male patients [9, 17, 20] with three different *OFD1*
260 mutations: a missense mutation in exon 2 (ref. 17), a splice site mutation in
261 intron 17 (ref. 20), and a frameshift mutation in exon 21 (ref. 9). Most *OFD1*
262 mutations in living male patients were located at the 3′ side of exon 7 or
263 downstream and were non-truncating mutations (Fig. 4). Male patients with
264 *OFD1* mutation in exon 21 are alive despite the mutation producing a
265 truncated OFD1 protein. Our patients survived past their perinatal period,
266 which may be due to them having a non-truncating *OFD1* mutation
267 downstream of exon 7. Three of the four patients reported here displayed
268 intellectual disability, and all had severe renal disease. Patient 1, who had an
269 in-frame *OFD1* mutation in exon 20, did not have intellectual disability. In
270 contrast, patient 2 with a missense mutation in exon 7 had severe intellectual
271 and psychomotor disabilities. Although there may be a strict genotype-

phenotype correlation in psychomotor development, this may not be the case with renal disease in male patients with *OFD1* mutation.

In conclusion, the phenotypic spectrum of *OFD1* mutations is very broad; thus, evaluation of *OFD1* mutation should not be restricted to patients presenting the classical OFD1 phenotype. A comprehensive genetic analysis using NGS is also useful to determine the presence of *OFD1* mutation in patients with no family history of OFD1. Further investigations are needed to clarify the mechanisms of such phenotypic diversity of *OFD1* mutations.

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291

292 **Conflict of Interest**

293 Kazumoto Iijima has received grant support from Daiichi Sankyo CO., Ltd.

294 and Zenyaku Kogyo Co., Ltd.

295

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Titles and Legends to Figures

Fig. 1 Renal imaging of patient 1 (a), patient 2 (b), and patient 3 (c and d). **a** Left renal hydronephrosis and right multicystic dysplastic kidney shown by abdominal computed tomography in patient 1. White arrow indicates left nephrostomy catheter. **b** Magnetic resonance image showing bilateral enlarged kidney and small cysts in patient 2. **c, d** Bilateral hyperechogenic kidney (c, left; d, right) in patient 3.

Fig. 2 Family tree of patient 3 and patient 4.

Fig. 3 Gene mutations identified in the patients. **a** Confirmation of the genetic analysis by Sanger sequencing. **b** cDNA analysis and schematic representation of the changes in *OFD1* mRNA.

Fig. 4 Known disease-related variants of male patients with *OFD1* mutation. JS, Joubert syndrome; RP, retinitis pigmentosa; SGBS2, Simpson-Golabi-Behmel syndrome type 2; †, perinatal death.

Supplementary Table 1: The gene list v.1 constructed using HaloPlex (91genes) for Patient 1.

ACE	*CITED1*	*GATA3*	*NPHP4*	*PAX2*	*SPRY1*
ACTN4	*CTDNEP1*	*GDNF*	*IQCB1*	*PAX8*	*TNXB*
AGT	*CXCL12*	*HOXA13*	*CEP290*	*PKD1*	*TP53*
AGTR1	*CXCR4*	*HNF1B*	*GLIS2*	*PKD2*	*TRAP1*
AHI1	*DSTYK*	*HOXD11*	*RPGLIP1L*	*PKHD1*	*TRPC6*
ANLN	*EP300*	*IFN2*	*NEK8*	*REN*	*UMOD*
APOL1	*EYA1*	*KAL1*	*SDCCAG8*	*RET*	*UPK3A*
BMP2	*FAT3*	*LMX1B*	*TMEM67*	*ROBO2*	*VANGL2*
BMP4	*FAT4*	*MDM2*	*TTC21B*	*ROR1*	*WNT4*
BMP7	*FOXD1*	*MI2B(CHD4)*	*WDR19*	*ROR2*	*WNT5A*
CC2D2A	*FGF2*	*MUC1*	*XNF423*	*SALL1*	*WNT7A*
CD2AP	*FGF9*	*MYO1E*	*CEP164*	*SIX1*	*WNT7B*
CDC5L	*FGF20*	*NPHP1*	*ANKS6*	*SIX2*	*WNT9B*
CHD1L	*FZD4*	*INVS*	*IFT172*	*SIX5*	*WNT11*
CHRM3	*FZD8*	*NPHP3*	*OFD1*	*SOX17*	*WT1*
					ZEB2

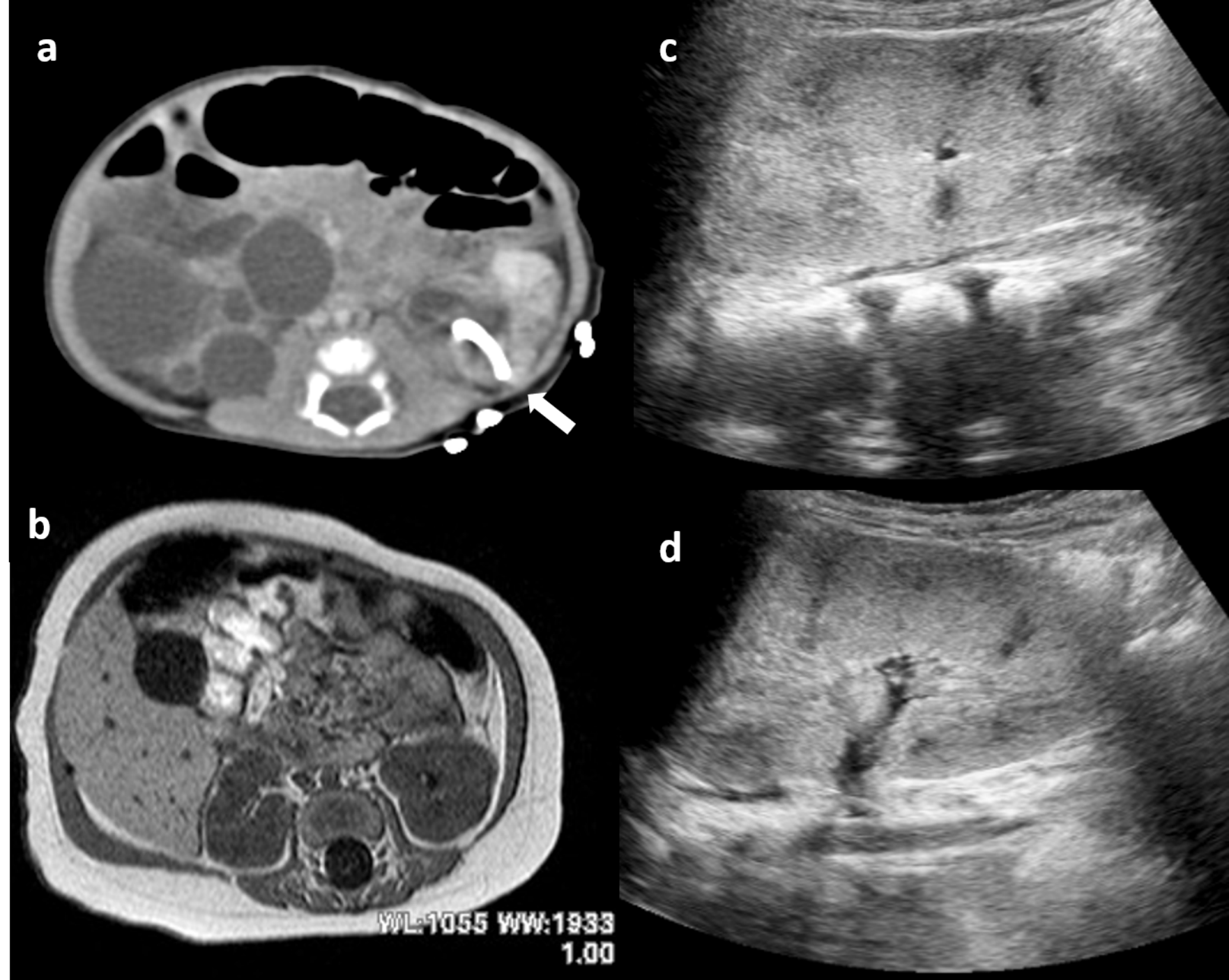
Supplementary Table 2: The gene list constructed using HaloPlex (128genes) for Patient 2.

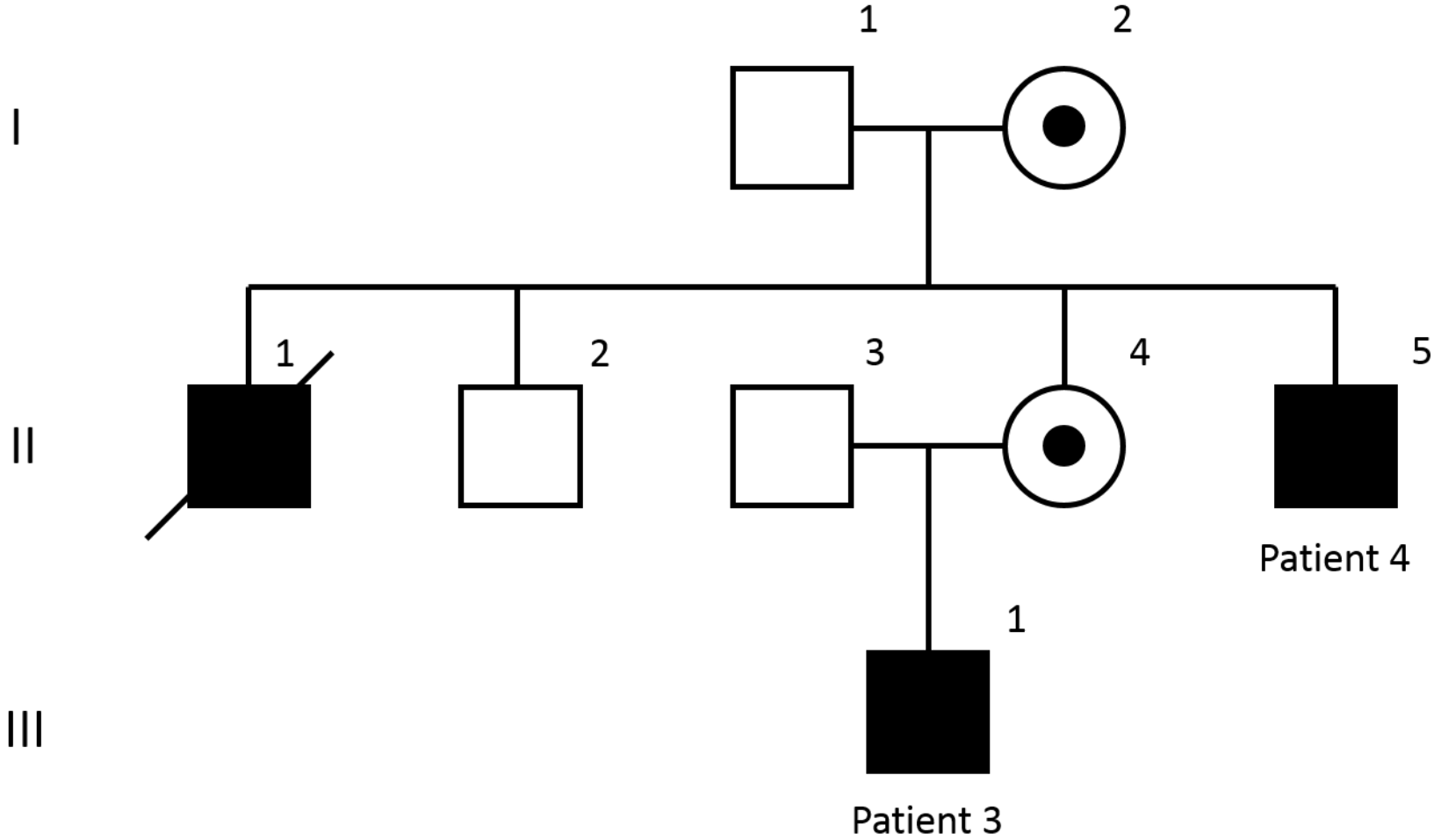
ACE	*BMP7*	*CXCL12*	*FZD4*	*IQCB1*	*NPHS2*	*RET*	*TBX18*	*WNT4*
ACTN4	*CC2D2A*	*CXCR4*	*FZD8*	*ITGA8*	*NUP107*	*ROBO2*	*TMEM138*	*WNT5A*
ADCK4	*CD2AP*	*DCDC2*	*GATA3*	*KAL1*	*NUP133*	*ROR1*	*TMEM231*	*WNT7A*
AGT	*CDC5L*	*DSTYK*	*GDNF*	*KIAA0586*	*NXF5*	*ROR2*	*TMEM237*	*WNT7B*
AGTR1	*CEP164*	*EP300*	*GLIS2*	*LAMB2*	*OFD1*	*RPGLIP1L*	*TMEM67*	*WNT9B*
AGTR2	*CEP290*	*EYA1*	*GREM1*	*LMNA*	*OSR1*	*SALL1*	*TNXB*	*WT1*
AHI1	*CEP41*	*FAT3*	*GRIP1*	*LMX1B*	*PAX2*	*SDCCAG8*	*TP53*	*ZEB2*
ALG13	*CEP83*	*FAT4*	*HNF1B*	*MDM2*	*PAX8*	*SIX1*	*TRAP1*	*ZNF423*
ANKS3	*CHD1L*	*FGF2*	*HOXA13*	*MKKS*	*PDE6D*	*SIX2*	*TRPC6*	
ANKS6	*CHD4*	*FGF20*	*HOXD11*	*MUC1*	*PKD1*	*SIX5*	*TTC21B*	
ANLN	*CHRM3*	*FGF9*	*IFN2*	*MYO1E*	*PKD2*	*SLIT2*	*UMOD*	
APOL1	*CITED1*	*FOXD1*	*IFT172*	*NEK8*	*PKHD1*	*SOX17*	*UPK3A*	
ARL13B	*COQ6*	*FRAS1*	*IFT81*	*NPHP1*	*PLCE1*	*SPRY1*	*VANGL2*	
BMP2	*CSPP1*	*FREM1*	*INPP5E*	*NPHP3*	*PODXL*	*SRGAP1*	*WDR19*	
BMP4	*CTDNEP1*	*FREM2*	*INVS*	*NPHP4*	*REN*	*TBX1*	*WNT11*	

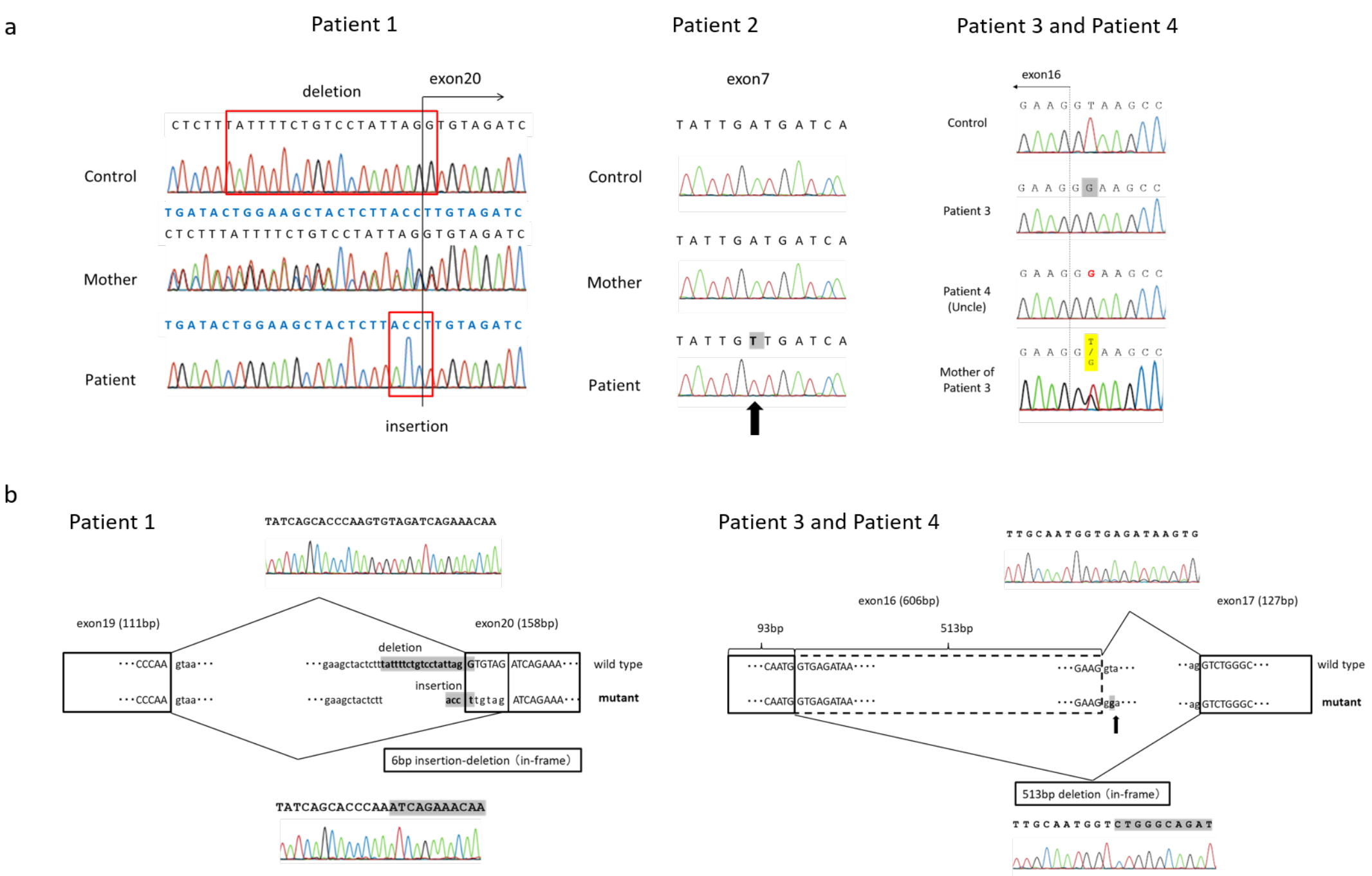
Table 1. Clinical manifestations in male patients with *OFD1* mutations

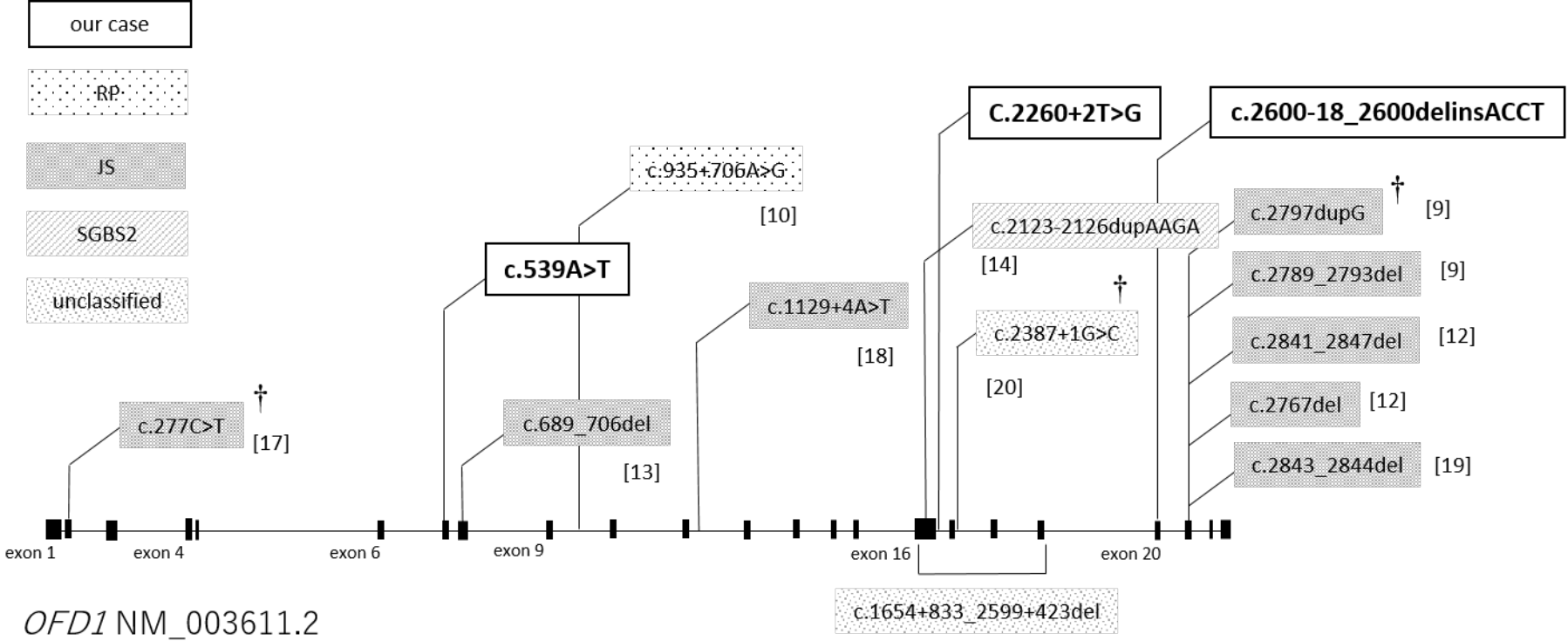
Patient or reference no.	Patient 1	Patient 2	Patient 3	Patient 4	9, 12, 13, 17, 18, 19	14	15	16, 20
No. of patients (families)					11 (8)	3 (1)	2 (1)	4 (2)
Clinical diagnosis	unclassified	unclassified	unclassified	BBS	JBTS	SGBS2	RP	unclassified
Facial								
Macrocephaly	-	+	-	-	5/9	1/3	N/A	3/3
Low-set ears	-	±	-	-	2/8	1/3	N/A	2/3
Microphthalmia	-	-	-	-	0/8	0/3	N/A	2/3
Hypertelorism	-	-	-	-	0/8	0/3	N/A	3/3
Short palpebral fissure	-	-	-	-	0/8	0/3	N/A	2/3
Oral								
High-arched palate	-	+	-	-	0/8	1/3	N/A	0/3
Cleft soft palate/lip	-	-	-	-	2/11	0/3	N/A	3/3
Tooth abnormality	-	+	-	-	1/8	0/3	N/A	N/A
Accessory frenulum	-	-	-	-	N/A	N/A	N/A	N/A
Digital								
Brachydactyly	-	+	-	-	4/11	1/3	N/A	0/2
Polydactyly	-	-	-	-	7/11	1/3	N/A	2/2
Neurological & Brain								
Intellectual disability	-	+	+	+	8/8	3/3	N/A	N/A
Ventricular dilatation	-	+	N/A	N/A	2/5	0/3	0/1	2/4
Molar tooth sign	-	-	N/A	N/A	11/11	0/3	0/1	0/1
Hypoplastic vermis	-	-	N/A	N/A	5/6	N/A	0/1	0/1
Hypoplastic gyri	-	-	N/A	N/A	0/4	N/A	0/1	1/2
Epilepsy	-	+	-	-	2/5	N/A	N/A	N/A
Dandy-Walker malformation	-	-	N/A	N/A	1/4	N/A	0/1	1/2
Renal								
Cystic kidney	+	+	-	-	1/11	0/3	N/A	0/4
Renal insufficiency	+	+	+	+	2/7	0/3	N/A	1/4
Others								
Obesity	-	+	-	+	2/8	1/3	N/A	1/1
Retinitis pigmentosa	-	+	-	-	1/5	1/3	2/1	N/A
Hearing loss	+	-	-	-	2/7	0/3	N/A	N/A
Micropenis	-	+	+	+	0/7	0/3	N/A	1/3
Cardiac malformation	-	-	-	-	0/7	0/3	N/A	3/3
Situs inversus	-	-	-	-	2/9	0/3	N/A	0/4
Liver fibrosis	-	-	-	-	2/2	N/A	N/A	N/A

Abbreviations: BBS, Bardet-Biedl syndrome; JBTS, Joubert syndrome; N/A, not available; RP, retinitis pigmentosa; SGBS2, Simpson-Golabi-Behmel syndrome type 2


a
b
c
d
WL:1055 WW:1933
1.00


I
II
III
1
2
1
2
3
4
5
Patient 4
1
Patient 3


a
Patient 1
deletion
exon20
CTCTTTATTTTCTGTCCTATTAGGTGTAGATC
Control
TGATACTGGAAGCTACTCTTACCTTGTAGATC
CTCTTTATTTTCTGTCCTATTAGGTGTAGATC
Mother
TGATACTGGAAGCTACTCTTACCTTGTAGATC
Patient
insertion
Patient 2
exon7
TATTGATGATCA
Control
TATTGATGATCA
Mother
TATTGTTGATCA
Patient
Patient 3 and Patient 4
exon16
GAAGGTAAGCC
Control
GAAGGGAAGCC
Patient 3
GAAGGGAAGCC
Patient 4
(Uncle)
GAAGG T/G AAGCC
Mother of
Patient 3
b
Patient 1
TATCAGCACCCAAGTGTAGATCAGAAACAA
exon19 (111bp)
exon20 (158bp)
deletion
···CCCAA gtaa···
···gaagctactcttttattttctgtcctattag GTGTAG ATCAGAAA···
wild type
insertion
···CCCAA gtaa···
···gaagctactctt acc ttgtag ATCAGAAA···
mutant
6bp insertion-deletion（in-frame）
TATCAGCACCCAAATCAGAAACAA
Patient 3 and Patient 4
TTGCAATGGTGAGATAAGTG
exon16 (606bp)
exon17 (127bp)
93bp
513bp
···CAATG GTGAGATAA····
···GAAG gta···
···ag GTCTGGGC···
wild type
···CAATG GTGAGATAA····
···GAAG gga···
···ag GTCTGGGC···
mutant
513bp deletion（in-frame）
TTGCAATGGTCTGGGCAGAT


our case
RP
JS
SGBS2
unclassified
c.277C>T †
[17]
c.539A>T
c.689_706del
[13]
c.935+706A>G
[10]
c.1129+4A>T
[18]
C.2260+2T>G
c.2123-2126dupAAGA
[14]
c.2387+1G>C †
[20]
c.2600-18_2600delinsACCT
c.2797dupG †
[9]
c.2789_2793del
[9]
c.2841_2847del
[12]
c.2767del
[12]
c.2843_2844del
[19]
exon 1
exon 4
exon 6
exon 9
exon 16
exon 20
c.1654+833_2599+423del
OFD1 NM_003611.2