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Mucopolysaccharidosis type I due to maternal uniparental disomy of chromosome 4 with partial isodisomy of 4p16.3p15.2

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ABSTRACT

Mucopolysaccharidosis type I (MPS I) is a rare lysosomal storage disease caused by biallelic mutations in *IDUA*, the gene coding for the lysosomal enzyme alpha *L*-iduronidase. Clinically MPS I is a chronic progressive multisystem disease typically presenting with coarse facial features, skeletal deformities, joint contractures, and multi-organ involvement. Hurler syndrome (MPS IH) represents the severe end of the spectrum of mucopolysaccharidosis type I and is characterized by central nervous system involvement leading to childhood dementia.

Here we report on a severe affected MPS IH patient who is homozygous for a splice site mutation (c.158 + 1G > A) in the *IDUA* gene. Further analyses revealed maternal uniparental disomy of chromosome 4 with partial isodisomy of the telomeric end of chromosome 4 (4.p16.3p15.2), representing an extraordinary mode of inheritance with a much lower re-occurrence risk for MPS I in the family.

1. Introduction

Mucopolysaccharidoses are a group of lysosomal storage disorders caused by impaired enzymatic degradation of glycosaminoglycans (GAGs) [8]. Hurler syndrome (mucopolysaccharidosis type I, MPS I) represents the severe end of the phenotype-spectrum of mucopolysaccharidosis type I, whereas Morbus Scheie and Hurler-Scheie correspond to the mild and moderate forms of MPS I, respectively. The incidence is approximately 1 in 100.000 livebirths [2,12]. MPS I is caused by biallelic mutations in *IDUA*, the gene coding for alpha *L*-iduronidase [12] and subsequent accumulation of the GAGs dermatan and heparan sulfate within the lysosomes [26]. The impaired lysosomal function affects multiple organs over time. Loss of function (LOF) mutations in the *IDUA* gene are typically associated with the severe form of MPS I (MPS IH) due to complete deficiency of the encoded enzyme. First symptoms are usually observed between 6 and 8 months of age and include developmental delay and intellectual disability. Furthermore, dysmorphic facial features, hydrocephalus, cardiac and lung disease, organomegaly, corneal clouding and hearing impairment are frequently found. The characteristic phenotype also comprises the radiologically diagnosed skeletal dysostosis multiplex, with short stature, spine deformities and hip dysplasia. Joint contractures and spinal cord compression leading to spastic paralysis are also typical symptoms. MPS I follows an autosomal-recessive inheritance pattern. Usually, the parents of an affected individual are found to be asymptomatic heterozygous carriers and the re-occurrence risk for another affected child is 25%. After genetic analysis of the index patient, segregation analysis is recommended. For MPS I fewer de novo mutations than in MPS II have been reported [29]. Prenatal testing is available after the pathogenic molecular change is identified in the family [22].

Here, we report a 5 year old boy with severe MPS IH who is homozygous for a splice site mutation (c.158 + 1G > A) in the *IDUA* gene. Further analyses revealed maternal uniparental disomy of chromosome 4.

2. Patient & methods

2.1. Case report

The patient was born spontaneously at 38 weeks of gestation as the second child of healthy, non-consanguineous Caucasian parents. The boy first presented with neonatal pneumonia at 4 weeks of age, followed by recurrent upper and lower airway infections with multiple hospitalizations. At 6 months of age obstructive sleep apnea syndrome (OSAS) was diagnosed and he was treated with nocturnal non-invasive ventilation. During a hospital stay at 9 months of age coarse facial

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features (broad nose, frontal bossing, macroglossia) were noticed and led to biochemical workup with suspicion of MPS I. The results showed a significantly reduced activity of the alpha *L*-iduronidase (AI) and the diagnosis of MPS I was confirmed after genetic testing. Further clinical features included inner ear hearing impairment with hearing aids, mild joint stiffness, thoracolumbar kyphosis, hepatosplenomegaly and mild septic hypertrophy with a mild tricuspid valve insufficiency consistent with the severe type of MPS I, Morbus Hurler.

He was first started on enzyme replacement therapy at the age of 9 months and underwent hematopoietic stem cell transplantation at the age of 12 months; the procedure was well tolerated. He learned to walk independently at 19 months, speaks complex sentences and is generally well adjusted. His cognitive development is slightly under the normal range (at a chronological age of 42 months the BSID-III revealed an AEqs of 34 months (developmental quotient (DQ) 90%). After an ENT-surgery at the age of 3 years nocturnal non-invasive ventilation was no longer indicated. At the age of 4 years he underwent reconstructive hip surgery due to progressive hip dysplasia. He currently visits a regular kindergarten. He presents with mild short stature, mild characteristic facies, mild corneal clouding, inner ear hearing impairment with hearing aids and mild musculoskeletal symptoms (shoulder joint contractures, mild genua valga and pes planovalgus) (see Image 1). Cardiac manifestations involves mitral und aortic valve insufficiencies grade II and high blood pressure (treated with an ACE-inhibitor). Due to suspected allergic asthma inhalative medication with steroids and ipratropium bromide is needed. His latest measurements at the age of 52/12 years were 16.3 kg (−1.48z), length 104 cm (−1.91z) and head circumference 51 cm (+0.60z). Donor cell chimerism was at 99.9% at that time; AI enzyme activity was within normal range (648 pmol/spot*20 h) and glycosaminoglycans (GAGs) in urine were moderately elevated.

2.2. Genetic testing

All biological samples were obtained following written informed consent from the studied individuals legal representatives. DNA samples from whole blood were isolated by standard procedures. PCR and Sanger sequencing of the *IDUA* gene (GenBank database accession number NM_000203.4) was performed in the index patient as described previously [23]. Consecutively, segregation analysis for the familial variant was performed on DNA samples of the healthy parents. This was followed by a microsatellite analysis confirming the paternity of the father and marker analysis [27]. Consecutively, SNP array analysis was performed using the Genome-Wide Human SNP Array 6.0 (Affymetrix) according to the manufacturer's instruction. Copy number and loss of heterozygosity (LOH) calculation was performed by the Genotyping Console 4.1 and CHAS 3.0 software (Affymetrix) Abnormalities were mapped according to Genome Build hg19.

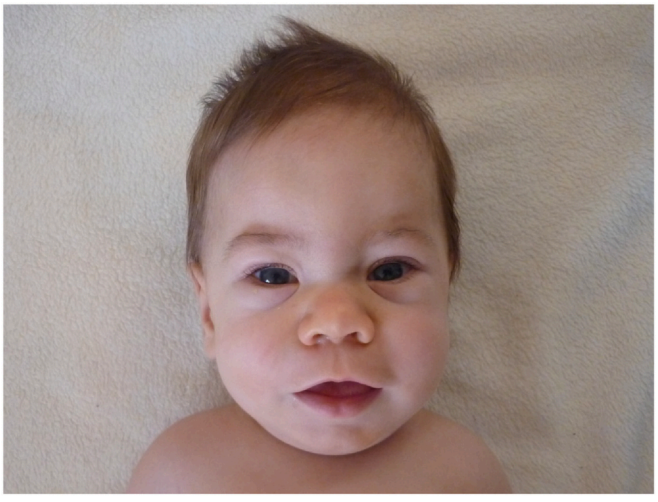

Image 1. Picture of the reported patient (frontal view, aged 7 months).

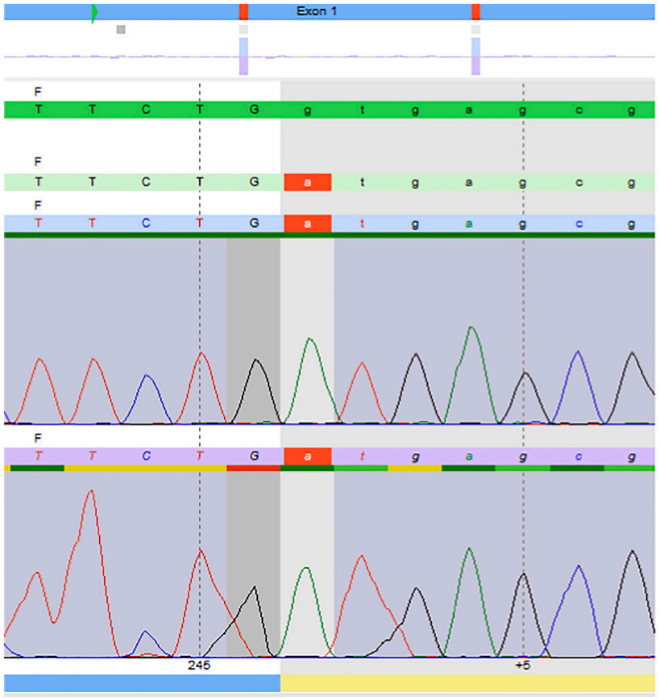

Fig. 1. Electropherograms for the familial *IDUA* mutation c.158 + 1G > A (Institute of Human Genetics, University Medical Center Hamburg-Eppendorf). (a) Electropherogram of the index patient (homozygous for the *IDUA* mutation). (b) Electropherogram of the mother (heterozygous for the *IDUA* mutation). (c) Electropherogram of the father (homozygous for the wildtype sequence).

3. Results

3.1. Genetic findings

Sanger sequencing of the *IDUA*-gene in the index patient identified a homozygous mutation c.158 + 1G > A. At the time of testing, the variant had not been previously described, but the alteration was predicted to disrupt the donor splice site at the end of exon 1. In 2017, the c.158 + 1G > A *IDUA* mutation was classified a pathogenic allele (HGMD accession number CS1718602) [10]. In 2019 it was described as a recurrent variant in a compound heterozygous state associated with a severe type of MPS I [6]. There is one heterozygous but no homozygous carrier of the mutation in gnomAD (databases accessed 28/05/19). Segregation analysis in our patient revealed that the *IDUA* mutation was also present in the DNA of the patient's mother, while DNA from the father only showed the wildtype allele (see Fig. 1(a)(c)). After paternity was confirmed via microsatellite analysis, marker analysis was conducted and revealed heterozygosity for 6 markers in the mother (D4S2366 (4p16.1), D4S2935 (4p16.1), D4S2639 (4p15.31), D4S391 (4p15.2), D4S1627 (4p13), D4S414 (4q22.1)), for 3 of which the index patient showed homozygosity (D4S2366 (4p16.1), D4S2935 (4p16.1) und D4S2639 (4p15.31)) (see Fig. 3). These findings confirmed maternal uniparental disomy with at least partial isodisomy for approx. 17.9 Mb on the short arm of the chromosome 4 in the index patient. The SNP array analysis in the index patient and his parents verified uniparental maternal disomy of the entire chromosome 4 (UPD(4)mat). Two long stretches of homozygosity also attested for the suspected partial interstitial isodisomy of chromosome 4 (ISCN 2016 karyotype: arr[hg19]_4p16.3p15.2(46691_25505932) x2hmz,4q32.2q34.3(163038241_179652804)x2hmz). As the *IDUA*

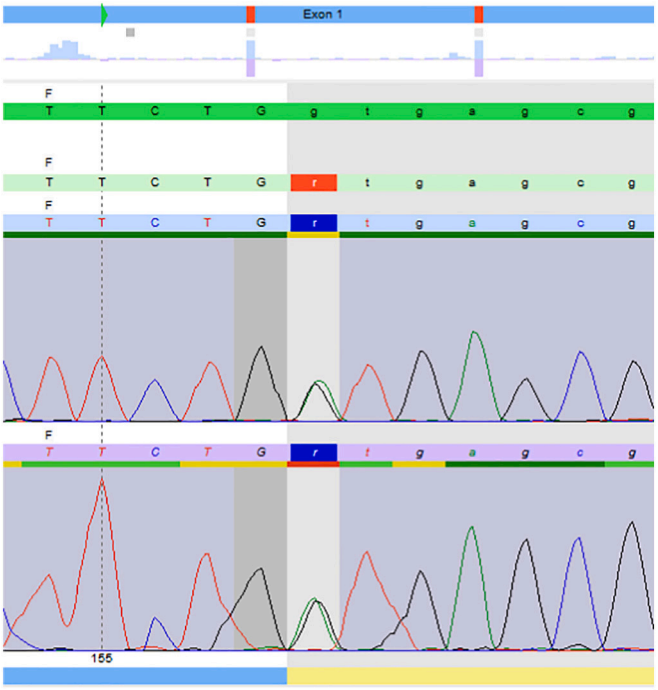

Fig. 1. (*continued*)

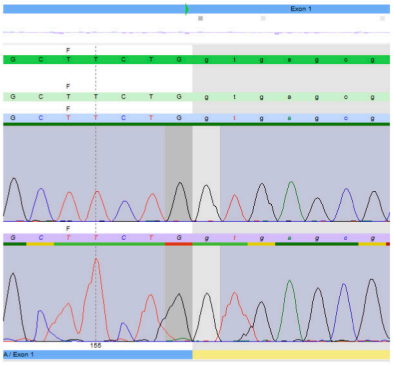

Fig. 1. (*continued*)

gene is located at 4p16.3 [25], this result explains the homozygosity of the detected splice site mutation in the index patient (see Fig. 2).

4. Discussion

This report describes for the first time how partial uniparental isodisomy can cause the autosomal recessive lysosomal disorder MPS I. As previously described, MPS I is usually inherited in an autosomal recessive manner with one heterozygous mutation being passed on by each healthy parent. Although it is not typically associated with a chromosomal aberration, few cases of UPD as a cause of MPS have been mentioned in the literature [5,13]. Uniparental disomy (UPD) describes a situation in which both copies of a chromosome are inherited from the same parent and the other parent's chromosome is not present. If chromosomes with genomic imprinted gene regions are affected this is associated with distinct syndromes e.g. Prader-Willi syndrome caused by maternal uniparental disomy of chromosome 15 [4,15,21] It is also a common finding in spontaneous abortions [14]. UPDs have been described for almost all chromosomes or chromosomal regions (partial or segmental UPD), with some incidental findings in healthy individuals [5,11]. The clinical consequences include imprinting disorders, autosomal recessive disorders due to the resulting homozygosity of the region as well as pathogenic effects of trisomy or mosaic trisomy on the placenta [9,15].

Typically, a constitutional UPD is the result of a nondisjunction event during meiosis. This aneuploidy is most commonly described for chromosomes X, Y, 15, 16, 21 and 22 [9]. The most common syndromes associated with UPD are described for chromosome 7, 11 and 15 (for a list of the most common described syndromes see Table 1) [3].

Advanced maternal age and/or the presence of structural abnormalities in the parents' karyotype may facilitate the occurrence of these events [1,15]. A higher risk for UPD (up to 15-fold) has also repeatedly been reported in children born after applying assisted reproductive techniques [7,18]; though recently, *Liehr* et al. have postulated this might not be the case [16].

The resulting trisomy in one of the gametes will in some cases undergo a “trisomy rescue” as a salvage procedure, meaning the ejection of one of the surplus chromosomes. In some cases, an unremarkable gamete with 23 maternal and paternal chromosomes will emerge. Occasionally, a gamete will retain the two homologous maternal or paternal chromosomes, resulting in uniparental disomy in the cell, called heterodisomy (hetUPD). It has also been postulated that - by reason of complementarity – two abnormal gametes' fusion (one nullisomic and one disomic) can lead to uniparental isodisomy (isoUPD) of

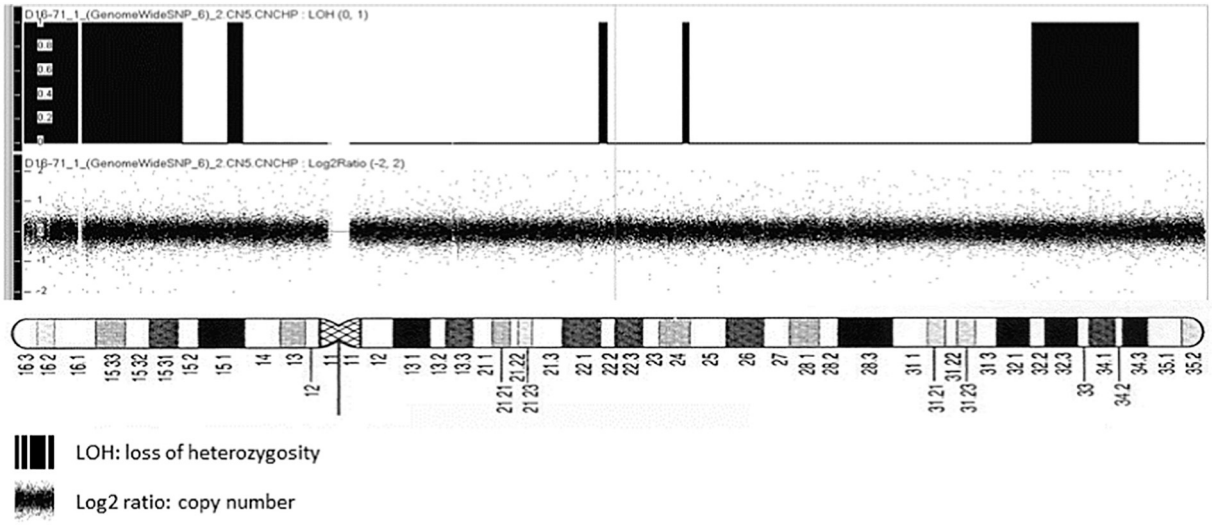

Fig. 2. SNP array analysis, stretches of homozygosity and copy number profile [image courtesy of the Institute of Human Genetics, UKSH Campus Kiel].

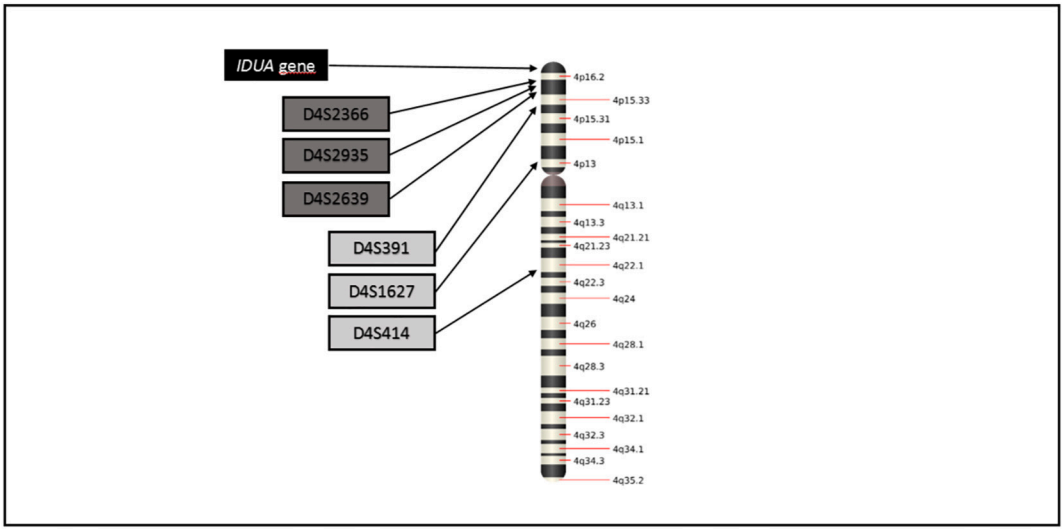

Fig. 3. Chromosome 4: areas of heterodisomy (heterozygosity) and isodisomy (homozygosity). Results from the marker analysis (*dark grey marker*: homozygous; *light grey marker*: heterozygous) [image adapted from www.lookfordiagnosis.com].

Table 1
Disorders associated with the most common forms of UPD [image adapted from [3]].

Chromosome	Parental origin	Disorder	OMIM
UPD 6	mat	Transient neonatal diabetes mellitus (TNDM)	#601410
UPD 7	mat	Silver Russell syndrome	#180860
UPD 11	mat	Silver Russell syndrome	#180860
UPD 11	pat	Beckwith-Wiedemann syndrome	#130650
UPD 14	mat	Temple syndrome	#616222
UPD 14	pat	Paternal UPD(14) syndrome Kagati-Ogama syndrome	#608149
UPD 15	mat	Prader Willi syndrome	#176270
UPD 15	mat	Angelman syndrome	#105830
UPD 20	mat	Mulchandani-Bhoj-Conlin syndrome	#617352

the entire chromosome concerned [9,15,16,28].

Herein, the proband presented maternal disomy (hetUPD) of the entire chromosome 4, likely resulting from a non-disjunction event during the first meiotic division and an attempt of "trisomy rescue". We hypothesize, that after a successful trisomy rescue 2/3 of the cells would carry an "unremarkable biparental" gene set.

The two long segments of isodisomy in our patient would most likely be caused by an earlier event, such as recombination during maternal meiosis prior to the first division (meiotic crossing over) and thus result in partial isodisomy (partial isoUPD) [28]. The phenotype of the affected individual (if live-born) would depend greatly "on the timing of the maldistribution (first or second meiosis)", the rearrangement of the chromatids after crossing over and the amount of crossovers per pair as well as the hereby affected genes [9,28].

It should be taken into consideration that long segments of homozygosity – as found in our patient – could hint at potential other autosomal recessive disorders that may complicate or obliterate a patient's clinical presentation. Also, if consanguinity is reported, diagnosis of uniparental disomy should not exclude the hypothesis of homozygous pathogenic changes in the patient due to shared genetic ancestry even of the two homologous (not isologous, i.e. identical) chromosomal pairs.

Furthermore, the counselling of the affected individual's family has to be adjusted accordingly: in contrast to usual autosomal recessive inheritance in MPS I with a typical reoccurrence risk for further affected children of 25%, the reoccurrence risk for a uniparental isodisomy affecting the telomeric end of chromosome 4 is diminishingly small. In general, the estimated incidence for UPD – at least with a clinical manifestation – used to be low; the occurrence of longer stretches of isodisomy is even less common [16,20]. Naturally, risk factors like age of the mother, the possibility of more complex chromosomal arrangements in the parents and the genetic base line risk of 2–5% have to be taken into consideration [19]. Recently, larger cross-sectional studies revealed that UPD in the general "healthy" population might be much higher than estimated, often without the prevalence of symptoms; i.e. "hidden" partial / segmental isodisomy (isoUPD) with a frequency of 0.578% in healthy individuals (currently 3.650 cases documented [16,20,24]). Interestingly, it was also shown that mosaic partial UPD (i.e. of 4q and 14q) can be acquired with age [17]. We hypothesize that UPD might in fact sometimes be well tolerated by the cell but might potentially also be (partly) causative for other more common disorders in the population, not just distinct syndromes. More research is needed to look into the mechanisms and risk factors that lead to aneuploidy in gametes, the consecutive salvation pathways and their potential clinical outcomes.

5. Conclusion

This is the first detailed description of severe MPS IH caused by uniparental disomy (hetUPD) of chromosome 4 with partial isodisomy of the telomeric end (4p16.3, partial isoUPD) leading to homozygosity for a pathogenic donor splice site mutation c.158 + 1G > A in the *IDUA* gene. This rare mode of inheritance is of great importance for the counselling of the parents because of the greatly reduced risk of re-occurrence. This case also formally adds MPS I to the list of diseases potentially associated with uniparental disomies (UPDs). Looking forward, the incidence of UPD in the general population seems to be much higher than expected; giving rise to the assumption that UPD is often well tolerated but might also represent a pathomechanism responsible for much more common disorders in the population.

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Declaration of Competing Interest

The authors declare no conflict of interest.

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