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ARTICLE

Maternal UPD 20 in a hyperactive child with severe growth retardation

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Maternal uniparental disomy was observed in a 4-year-old boy with severe pre- and postnatal growth retardation (body height: 85 cm = 12 cm < third percentile, head circumference: 48 cm = 10 cm < third percentile), a few minor facial findings, and with apparent hyperactivity. His intelligence is within the normal range for his age. Karyotype analysis revealed two cell lines, one apparently normal with 46,XY, the other with a tiny marker (47,XY, + mar).

Microdissection and reverse chromosome painting using the marker DNA library as a probe, as well as PCR analysis revealed that the marker is from chromosome 20 and contains only the centromere and pericentromeric segments, but none of the pericentromeric loci for microsatellites. Microsatellite analysis of 25 chromosome 20 loci disclosed maternal uniparental disomy for all 16 informative markers. Maternal heterodisomy was evident for seven loci of the short arm segment 20p11.2-pter. Maternal isodisomy was found at five loci, three of them map to the proximal 20p11.2 segment and two to 20q. To our knowledge, this is the first case of maternal disomy 20 in humans.

Keywords: unimaternal disomy 20; microdissection; marker chromosome; reverse painting; microsatellite analysis

Introduction

Uniparental disomy (UPD) is a condition in which a chromosomally disomic individual inherited both copies of a chromosome from one parent only.[1] Depending on the identity of both chromosome homologues, UPD is defined as either heterodisomy (inheritance of both homologues from one parent) or isodisomy (presence of two identical copies of one homologue). UPD is associated with advanced maternal age, implying that it is frequently derived from meiotic nondisjunction followed by rescue of trisomic or monosomic conceptuses.[2] Abnormal phenotypes in UPD are explained by the detrimental effect of the lack of a normal homologue to an imprinted gene, homozygosity for recessive genes in case of isodisomy or partial isodisomy,[3] and confined placental mosaicism with trisomy and/or monosomy. UPD has already been reported for more than two-thirds of human chromosomes.[2,4]

Pure UPD for chromosome 20 has not been known previously. This is surprising as mosaic trisomy 20 is observed rather frequently in prenatal diagnosis and is found in about 6% of amniotic fluid cell cultures.[5] However, children born thereafter are almost always normal, and the trisomy 20 cell clone is thought to

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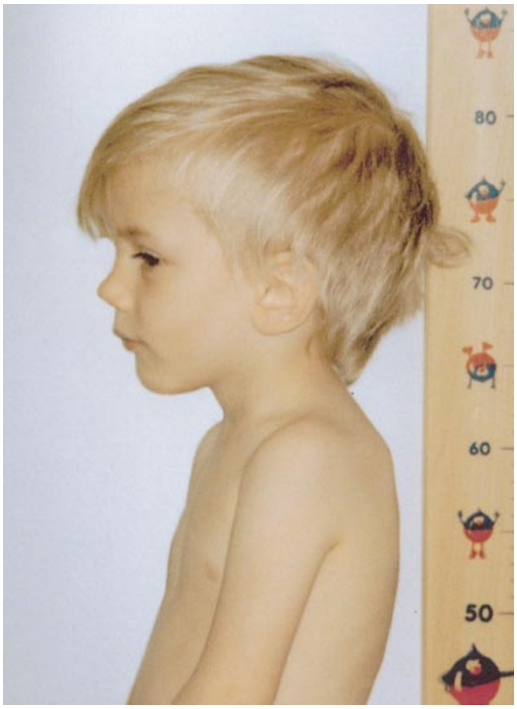

Figure 1 *The patient at the age of 3 10/12 years*

originate from epithelia of the urogenital tract. There is only one report on a phenotypically severely affected child with paternal UPD 20, which was found in addition to mosaicism for trisomy 20 in several tissues.[6] The second cell line had a dicentric chromosome due to an end-to-end short arm translocation of two paternal chromosome 20 homologues.

Here we report on the first case of maternal UPD 20 in a proband with severe pre- and postnatal growth retardation and hyperactivity.

Materials and Methods

Clinical information

The male patient (Figure 1) was referred for cytogenetic investigation because of growth retardation and minor dysmorphic facial features. The pregnancy ended by section at 32 weeks of gestation because of a cervix carcinoma in the mother. At time of conception the mother was 40-years-old. The pregnancy was normal until 30 weeks of gestation, when ultrasound showed that growth had decreased (birth weight: 1420 g and body length: 40 cm). It was the mother's sixth pregnancy.

At the age of 4 years and 2 months, the patient's height was 85 cm and his head circumference was 48 cm (both < third percentile). His face showed a prominent supra orbital region, a relatively short philtrum, a thin upper lip, a high palate and relatively large, slightly dysmorphic, backward rotated ears. Furthermore, he had hyperextensible wrist joints and fingers and partial soft tissue syndactyly of the fingers 2–5. He was reported to be a friendly and hyperactive child with a high-pitched voice. His IQ of 97 (Kramer-test) was within the normal range for his age. His neurologic and motor development were normal.

Cytogenetics

Cytogenetic examination was carried out on peripheral blood of the patient as well as on his parents and mentally retarded sister. Chromosome harvesting followed standard techniques and karyotyping was performed after GTG and CBG banding.

Microdissection

Microdissection was carried out as described by Senger *et al*.[7] Five marker chromosomes were excised under microscopical view using extended glass needles (inverted microscope IM 135, Zeiss, micromanipulator MR mot, Zeiss).

In order to gain information about the possible involvement of euchromatic material deriving either from the short or the long arm of chromosome 20, 20p and 20q specific microdissection libraries were generated. Five p-arms and five q-arms of chromosome 20 were collected in the same manner as the marker chromosome.

DOP-PCR

Amplification of the dissected chromosomal material was carried after protein digestion with Proteinase K (2 hours at 60°C) according to a procedure described in previous research.[8–10]

Fluorescence in situ *hybridisation (FISH)*

For FISH experiments, amplified DNA from the marker chromosome and the 20q library were labelled with Biotin-16-dUTP, the 20p library with Digoxigenin-11-dUTP.[11] Hybridisation was performed following standard procedures.[12] In reverse painting experiments on replication G-banded chromosomes, the biotinylated probe derived from the marker was detected with Avidin-Texas Red (Vector laboratories). Chromosome banding was obtained by incorporating BrdU into the late S-phase after synchronisation with Methotrexate.[13] A replication banding comparable to G-bands was obtained by incubation with FITC conjugated anti-BrdU. Slides were mounted in antifade solution (Vectashield) containing DAPI as a counterstain.

Slides were analysed using a Zeiss Axioskop microscope (Carl Zeiss Jena, Germany) equipped with a Pinkel #1 filter set (Chroma Technologies). Images were captured and processed using a standard integrating CCD video camera (Sony) and the *isis* software package (MetaSystems, Altlussheim, Germany).

Radioactive PCR

Genomic DNA was isolated from nucleated blood cells. The CA strand oligodesoxynucleotide primers were end-labelled for 30 min at 37°C in a 7 μl reaction containing 500 ng primer, 70 mM Tris (pH 7.6), 10 mM $MgCl_2$, 5 mM DTT, 7 units T4 polynucleotide kinase (New England Biolabs), and 1.25 μCi g^{32}P-dATP at 3000 Ci/mmol. Reactions were stopped by incubation at 95°C for 2 min. Polymerase chain reactions

were carried out in 25 µl volumes containing 100 ng of end-labelled primer, 100 ng of each unlabelled primer, 100 ng of genomic DNA template, 200 µM each dGTP, dCTP, dATP, and dTTP, 50 mM KCl, 10 mM Tris (pH 8.3), 1.5 mM $MgCl_2$ and 0.5 units *Taq* polymerase (Boehringer, Mannheim).

Amplification conditions were 1 min denaturation at 94°C, 1 min annealing at 55°C and 2 min extension at 72°C for 30 cycles. The last elongation step was lengthened to 7 min. Aliquots of the amplified DNA were mixed with 1 vol formamide sample buffer and were analysed on 8% denaturing polyacrylamide gels. Gels were then fixed and exposed to autoradiograph film for 1 d.

Loci used for PCR include D20S113, D20S199, D20S189, D20S98, D20S104, D20S114, D20S184, D20S103, D20S192, D20S188, D20S66, D20S112, and D20S195;[14] D20S59, D20S41, D20S50, D20S64;[15] D20S175, D20S186, D20S119, D20S102, and D20S100;[16] D20S17;[17] UT246;[18] D20S27;[19] 1H5-CA (Giannakudis, pers. com.); and F2B6-CA (Franke, unpublished).

Results

Cytogenetic analysis of 50 metaphases from blood lymphocytes revealed a karyotype of 46,XY (28)/47,XY, + mar(20). The small marker chromosome represents approx. twenty per cent of the length of chromosome 21q (Figure 2). According to CBG-banding it consists entirely of constitutive heterochromatin. Under the light microscope, there is no evidence of pericentromeric G-band negative material. Parental karyotypes and the karyotype of the sister were normal in 50 metaphases analysed from each.

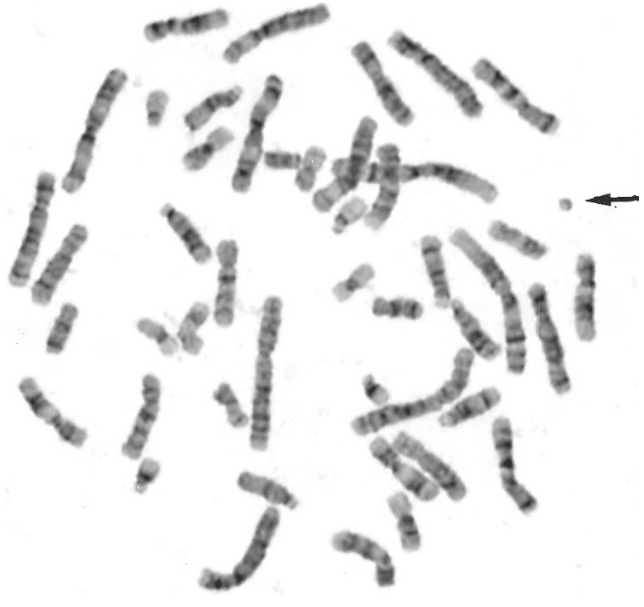

Figure 2 *Representative GTG-banded metaphase spread from the patient, containing the marker chromosome (←)*

Fluorescence *in situ* hybridisation (FISH) on normal metaphases from a control using the marker-specific microdissection library (reverse chromosome painting) revealed signals exclusively at the centromeres of both chromosomes 20, indicating its chromosome 20 origin. Reverse painting on metaphases from the proband revealed only two signals at the centromere of both normal chromosomes 20 in 46,XY cells but three signals (centromere of both chromosomes 20 and the marker) in 47,XY, + mar. The FISH signals on the marker appeared considerably brighter in comparison to the signals on both chromosomes 20 and covered the marker completely (Figure 3A and B).

The chromosome 20p- and 20q-specific microdissection probes were labelled differentially, hybridised to metaphase spreads of the proband and were analysed by two-colour FISH, with one colour for each arm, respectively. The two normal chromosomes 20 were clearly identified by differentially stained p- and q-arms, while no FISH signal were detected on the small marker (not shown).

Altogether, 25 highly polymorphic microsatellite markers for chromosome 20 were studied in this family by radioactive PCR. Results of genotyping are given in Table 1, where markers are listed according to the most likely order within the physical/genetic map (NCBI, genomes data base, release 3.8). An exclusively maternal inheritance was disclosed at 16 loci (see Table 1), the remaining nine loci are considered to be noninformative with respect to the segregation of alleles in the family.

Proband and mother share identical heterozygous alleles at seven loci (D20S113, D20S199, D20S189, 1H5, D20S41, UT246, D20S98). At each of these loci, the father is heterozygous for different alleles, thus revealing maternal heterodisomy. Maternal isodisomy was found at five loci (D20S104, D20S114, D20S184, D20S17, D20S119). Three of them (D20S104, D20S114, D20S184) map to the proximal third of the short arm. At four additional loci (D20S59, D20S195, D20S173, D20S64) mother and proband are homozygous for the same allele, while the father is heterozygous for two different alleles.

DNA of the microdissected and DOP-amplified marker was further used as template to narrow down the molecular content of the marker chromosome and, eventually, for identifying its parental origin. None of

three loci mapping closest to the centromere of chromosome 20 (D20S184, D20S195, D20S493) revealed any signal derived from that marker (data not shown).

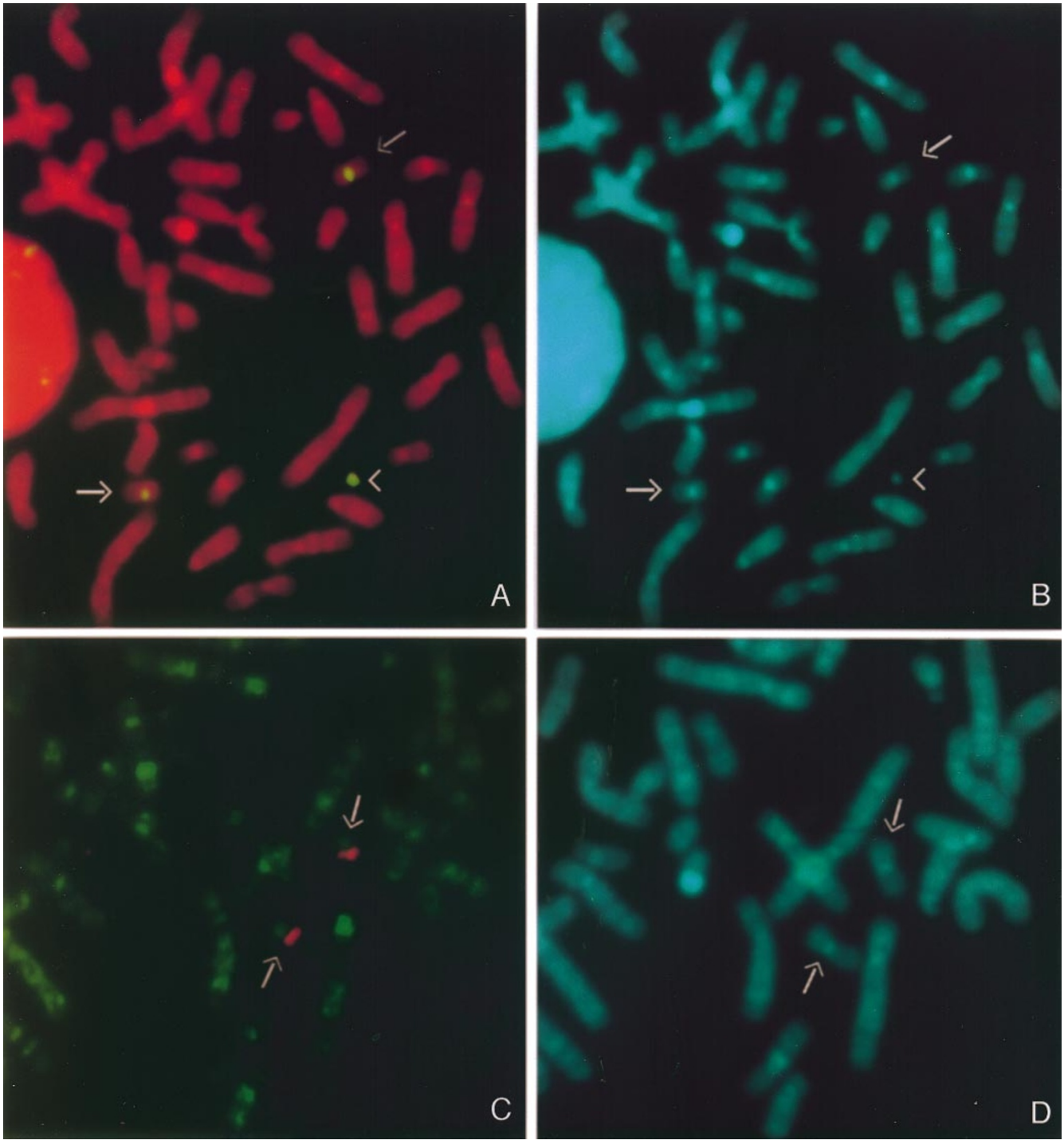

Figure 3 *A + B) FISH of the microdissection library to metaphase spreads of the patient containing the marker chromosome. Fluorescent signals are observed in the centromeric region of both chromosomes 20 (→) and on the marker chromosome (<). C + D) FISH of the microdissection library to normal metaphase spreads. A replication banding pattern corresponding to G-bands identifies the chromosomes indicated by the fluorescent signals as chromosome 20.*

Discussion

According to our FISH and microsatellite analysis, the marker of our proband comprises mainly the centromere of chromosome 20p and its pericentromeric heterochromatin. The presence of 20p and/or 20 specific DNA was examined using the respective arm-specific microdissection libraries. No FISH signal was revealed with any of them at the marker. The absence of FISH signals could have been due to technical limits, eg euchromatic DNA close to the centromere is not contained in the library or the amount of euchromatic pericentromeric DNA of the marker is too small to be detected by FISH. Nevertheless, these results suggest that pericentromeric euchromatin should only be a minor component of that marker, if present at all. This is confirmed by the analysis of pericentromeric microsatellites (eg D20S184, D20S195, D20S493). Trisomy was observed for none of these loci when genomic DNA was used as template. Furthermore, no allele was detected by PCR using microdissected and DOP-amplified marker DNA. From these data it is unlikely that the marker is causally related to the phenotype and hyperactivity of the proband, but they do not exclude the possibility that the marker contains one or a few genes that cause the proband's phenotype.

Table 1 Results of microsatellite analysis for chromosome 20 in the proband and his parents (ni: noninformative; mat-iso: maternal isodisomy; mat-hetero: maternal heterodisomy; mat: maternal alleles only)

Marker	*Father*	*Proband*	*Mother*	*origin of alleles*
20p:				
D20S103	aa	ab	ab	ni
D20S192	ab	aa	aa	ni
D20S113	cc	ab	ab	mat-hetero
D20S199	ad	bc	bc	mat-hetero
D20S59	ac	bb	bb	mat
D20S175	ab	ab	ab	ni
D20S188	ab	ac	ac	ni
D20S189	bd	ac	ac	mat-hetero
D20S186	bc	ab	ab	ni
D20S27	aa	ab	ab	ni
1H5	ad	bc	bc	mat-hetero
F2B6	aa	ab	ab	ni
D20S41	ab	cd	cd	mat-hetero
UT246	bc	ad	ad	mat-hetero
D20S98	ad	bc	bc	mat-hetero
D20S104	ac	cc	bc	mat-iso
D20S114	bd	aa	ab	mat-iso
D20S184	bb	aa	ab	mat-iso
D20S493	aa	aa	aa	ni
centromere				
20q:				
D20S195	cd	aa	aa	mat
D20S17	ab	cc	ac	mat-iso
D20S119	bc	dd	ad	mat-iso
D20S173	aa	bb	bb	mat
D20S102	bb	bb	bb	ni
D20S64	bb	aa	aa	mat

Microsatellite analysis revealed maternal UPD 20 at all 16 informative loci. To our knowledge this proband represents the first case of maternal UPD for chromosome 20. Maternal heterodisomy was only observed for loci of the distal segment of 20p, including the proximal locus for D20S98, which has been mapped to 20p11.2 (unpublished observations). For three proximal 20p loci and two loci on 20q, genotyping revealed maternal isodisomy. The combination of two segments with heterodisomy and isodisomy can be explained by one recombination event between D20S98 and D20S104. Because that recombination creates partial isodisomy it cannot be decided whether maternal UPD of chromosome 20 is due to a meiosis I or meiosis II error. A meiosis II error, however, seems to be more likely as the patient has isodisomy at D20S184 in 20p and D20S17 at 20q. Both markers are close to the centromere, where recombinations are exceedingly rare. The karyotype of our proband is 47,XY, + mar.rev ish der(20)(:p11.2Ûq11.2:)/46,XY.

The question now is whether the severe prenatal and postnatal growth retardation of our proband is due to maternal UPD and imprinting, homozygosity for recessive genes within the isodisomic segment, or whether it is associated with placenta failure because of confined placental trisomy 20. A partial trisomy effect of the small marker is rather unlikely, due to the results obtained by FISH and microsatellites described above. A phenotypic effect of maternal UPD 20 on prenatal and postnatal growth is not unlikely, taking into account data from the literature where several cases with maternal UPD and growth retardation have been reported, such as for chromosome 7[21–24] and chromosome 16.[25,26] Maternal UPD for chromosome 16 was also detected in cases in which a trisomy 16 placental karyotype was identified in chorionic villi during prenatal diagnosis. In these cases one cannot clearly distinguish between a maternal UPD effect and placental insufficency due to confined placental mosaicism for trisomy 16. In none of these cases was a postnatal follow up possible. Two pregnancies ended as spontaneous abortion[25] and one infant died 140 days post partum.[26] Intrauterine growth retardation in our case

was detected at 30 weeks of gestation. At the age of four years our proband is still extremely small for his age and so far he has not caught up on his growth retardation. It is not unlikely, therefore, that his prenatal and postnatal growth retardation is due to maternal UPD and imprinting or due to homozygosity of one or several recessive gene(s).

Genetic studies in mice have led to the definition of 15 imprinting effects including retarded growth and behavioural abnormalities.[27] Two of them have been associated with a distal region of mouse chromosome 2, which shows homology with human chromosome 20.[28,29] Here, paternal UPD of this region led to hyperactivity and a short and broad body shape. Maternal UPD instead resulted in a counter type, totally inactive mice with a long and flat body. In contrast to the effects of UPD in mice, our case of maternal UPD 20 is associated with hyperactivity and not with inactivity as could have been expected from observations with the respective maternal UPD 2 mice. Even though the paternal origin of UPD in the hyperactive mice does not match our case, it is likely that the hyperactive behaviour of the boy is due to a maternal imprinting effect. Hyperactivity is not known in other members of the family.

Several mechanisms have been reported resulting in UPD. They are regarded as mechanisms for aneuploidy correction and include gamete complementation, monosomy duplication and 'trisomy rescue'.[2,4,30] 'Trisomy rescue' is well documented for cases with confined placental trisomy 16 and uniparental disomy in the foetus[31] as well as for cases with Prader Willi Syndrome

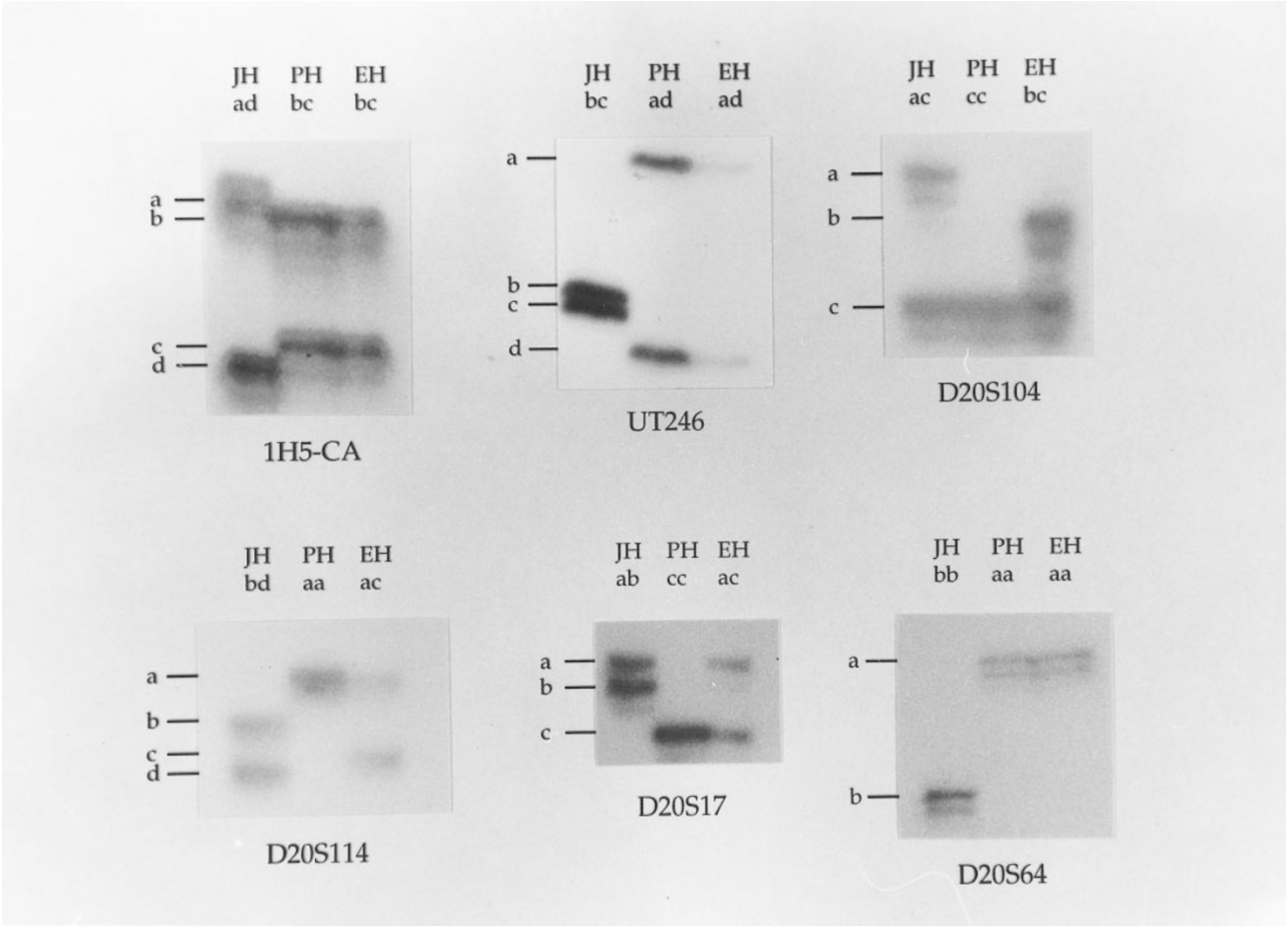

Figure 4 *Results of microsatellite analysis for 1H5, UT246, D20S104, D20S114, D20S17 and D20S64 in the proband (PH), father (JH) and mother (EH). Maternal heterodisomy is shown for 1H5, UT246 and maternal isodisomy for D20S104, D20S114 and D20S17. At D20S64, both mother and proband are homozygous for aa with absence of the paternal allele in the proband (noninformative maternal UPD)*

(PWS) and maternal disomy.[32] Another probable example of incorrect 'trisomy rescue' comes from a case with PWS and a supernumerary marker chromosome 15 and maternal heterodisomy for that chromosome.[33] A similar mechanism can be assumed for the present case. UPD resulting from nondisjunction and related to advanced maternal age has been reported in the literature.[2] In our case it is also very likely that the zygote was trisomic for chromosome 20, with two chromosomes derived from the mother who was 40 years at conception of the proband and that breakage of the paternal chromosome subsequently. The DNA of the proband may help to identify imprinted genes on chromosome 20.

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References

1 Engel E: A new genetic concept: uniparental disomy and its potential effect, isodisomy. *Am J Med Genet* 1980; **6**: 137–143.
2 Ledbetter DH, Engel E: Uniparental disomy in humans: development of an imprinting map and its implications for prenatal diagnosis. *Hum Mol Genet* 1995; **4**: 1757–1764.
3 Pulkkinen L, Bullrich F, Czarnecki P, Weiss L, Uitto J: Maternal uniparental disomy of chromosome 1 with reduction to homozygosity of the LAMB3 locus in a patient with Herlitz junctional epidermolysis bullosa. *Am J Hum Genet* 1997; **61**: 611–619.
4 Engel E: Uniparental disomy revisited: the first twelve Years. *Am J Med Genet* 1993; **46**: 670–674.
5 Hsu L, Kaffe S, Perlis TE: A revisit of trisomy 20 mosaicism in prenatal diagnosis – an overview of 103 cases. *Prenat-Diagn* 1991; **11**: 7–15.
6 Spinner NB, Rand E, Bucan M, Jirik R, Gogolin-Ewens C, Riethman HC, McDonald-McGinn DM, Zackai EH: Paternal uniparental isodisomy for human chromosome 20 and absence of external ears. *Am J Hum Genet* 1994; **55**: A118 (Abstract).
7 Senger G, Lüdecke HJ, Horsthemke B, Claussen U: Microdissection of banded human chromosomes. *Hum Genet* 1990; **84**: 507–511.
8 Telenius H, Carter NP, Bebb CE, Nordenskjold M, Ponder BA, Tunnacliffe A: Degenerate oligonucleotide-primed PCR: general amplification of target DNA by a single degenerate primer. *Genomics* 1992; **13**: 718–725.
9 Rubtsov N, Senger G, Kuzcera H, Neumann A, Kelbova C, Junker K, Beensen V, Claussen U: Interstitial deletion of chromosome 6q: report of a case and precise definition of the breakpoints by microdissection and reverse painting. *Hum Genet* 1996; **97**: 705–709.
10 Chudoba I, Rubtsov N, Senger G, Junker K, Bleck C, Claussen U: Improved detection of chromosome 16 rearrangements in acute myeloid leukemias using 16p and 16q specific microdissection DNA libraries. *Oncology Report* 1996; **3**: 829–832.
11 Pinkel D, Staume T, Gray JW: Cytogenetic analysis using quantative, high-resolution fluorescence hybridization. *Proc Natl Acad Sci USA* 1986; **83**: 2934.
12 Kallionemi O-P, Kallionemi A, Kurisu W, Thor A, Chen L-C, Smith HS, Waldman FM, Pinkel D, Gray JW: *ERBB2* amplification in breast cancer analyzed by fluorescence in situ hybridization. *Proc Natl Acad Sci USA* 1992; **89**: 5321.
13 Yunis JJ: Chromosomal rearrangements, gene, and fragile sites in cancer: clinical and biological implications. In: Devita VT, Hellman S, Rosenberg SA (eds). *Important advances in oncology*. JB Lippincott: Philadelphia, 1986,pp 93–123.
14 Gyapay G, Morissette J, Vignal A, Dib C, Fizames C, Millasseau P, Marc S, Bernadi G, Lathrop M, Weissenbach J: The 1993–1994 Genethon human linkage map. *Nat Genet* 1994; **7**: 246–339.
15 Hazan J, Dubay C, Pankowiak MP, Becuwe N, Weissenbach J: A genetic linkage map of human chromosome 20 composed entirely of microsatellite markers. *Genomics* 1992; **12**: 183–189.
16 Weissenbach J, Gyapay G, Dib C, Vignal A, Morissette J, Millasseau P, Vaysseiz G, Lathrop M: A second generation linkage map of the human genome. *Nature* 1992; **359**: 794–801.
17 Rothschild CB, Akots G, Hayworth R, Pettenati MJ, Rao PN, Wood P, Stolz FM, Hansmann I, Serino K, Keith TP, Fajans SS, Bowden DW: A genetic map of chromosome 20q12-q13.1: Multiple highly polymorphic microsatellite and RFLP markers linked to the maturity-onset diabetes of the young (MODY) locus. *Am J Hum Genet* 1993; **52**: 110–123.
18 Melis R, Bradley P, Elsner T, Robertson M, Lawrence E, Gerken S, Albertsen H, White R: Polymorphic SSR (simple-sequence-repeat) markers for chromosome 20. *Genomics* 1993; **16**: 56–62.
19 Weber JL, May PE: Dinucleotide repeat polymorphism at the D20S27 locus. *Nucl Acids Res* 1990; **18**: 2202.
20 Callen DF, Ringenbergs ML, Fowler JCS, Freemantel CJ, Haan EA: Small marker chromosomes in man: origin from pericentric heterochromatin of chromosomes 1, 9 and 16. *J Med Genet* 1990; **27**: 155–159.
21 Spence JE, Perciaccante RG, Greig GM, Willard HF, Ledbetter DH, Heitmancik JF, Pollak MS, O'Brien WE, Beaudet AL: Uniparental disomy as a mechanism for human genetic disease. *Am J Hum Genet* 1988; **42**: 217–226.
22 Voss R, Ben-Simon E, Avital A, Godfrey S, Zlotogora J, Dagan J, Tikochinsky Y, Hillel J: Isodisomy for chromosome 7 in a patient with cystic fibrosis: could uniparental disomy be common in humans? *Am J Hum Genet* 1989; **45**: 373–380.

23 Spotila LD, Sereda L, Prockop DJ: Partial isodisomy for maternal chromosome 7 and short stature in an individual with mutation COLIA2 locus. *Am J Hum Genet* 1992; **51**: 1396–1405.
24 Kotzot D, Schmitt S, Bernasconi F, Robinson WP, Lurie IW, Ilyina H, Méhes K, Hamel BCJ, Otten BJ, Hegersberg M, Werder E, Schoenle E, Schinzel A: Uniparental disomy in Silver Russel syndrome and primordial growth retardation. *Hum Mol Genet* 1995; **4**: 583–587.
25 Vaughan J, Zehra A, Bower S, Bennett P, Chard T, Moore G: Human maternal uniparental disomy for chromosome 16 and fetal development. *Prenat Diagn* 1994; **14**: 751–756.
26 Whiteford ML, Coutts J, Al-Roomi L, Mather A, Lowther G, Cooke A, Vaughan JI, Moore GE, Tolmie JL: Uniparental isodisomy for chromosome 16 in a growth-retarded infant with congenital heart disease. *Prenat Diagn* 1995; **15**: 579–584.
27 Cattenach BM, Jones J: Genetic imprinting in the mouse: implication for gene regulation. *J Inher Met Disease* 1994; **17**: 403–420.
28 Cattenach BM, Kirk M: Differential activity of maternally and paternally derived chromosome regions in mice. *Nature* 1985; **315**: 496–498.
29 DeBry RW, Seldin MF: Human/Mouse homology relationships. *Genomics* 1996; **33**: 337–351.
30 James RS, Temple IK, Dennis NR, Crolla JA: A search for uniparental disomy in carriers of supernumerary marker chromosomes. *Eur J Hum Genet* 1995; **3**: 21–26.
31 Kalousek DK, Langlois S, Barett I, Yam I, Wilson DR, Howard-Peebles PN, Johnson MP, Georgiutti E: Uniparental disomy for chromosome 16 in humans. *Am J Hum Genet* 1993; **52**: 8–16.
32 Cassidy SB, Lai L-W, Erickson RP, Magnuson L, Thomas E, Gendron R, Herrmann J: Trisomy 15 with loss of the paternal 15 as a cause of Prader-Willi syndrome due to maternal disomy. *Am J Hum Genet* 1992; **51**: 701–708.
33 Robinson WP, Wagstaff J, Bernasconi F, Baccichetti C, Artifoni L, Franzoni E, Suslak L, Shih L-Y, Aviv H, Schinzel A: Uniparental disomy explains the occurrence of Angelman or Prader-Willi syndrome in patients with an additional small inv dup(15) chromosome. *J Med Genet* 1993; **30**: 756–760.