Detection of a de novo duplication of 1q32-qter by fluorescence in situ hybridisation in a boy with multiple malformations: further delineation of the trisomy 1q syndrome

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Abstract

We report a dysmorphic boy with a de novo partial trisomy 1q. The boy has microcephaly, bilateral cleft lip and palate, low set and dysmorphic ears, brain anomalies, pulmonary stenosis, duodenal obstruction, dysplastic kidneys, and bifid thumbs. The trisomic segment 1q32-qter is duplicated with an inverted insertion at 1p36.3. The aberration was initially detected at amniocentesis and confirmed and defined by GTG banding, chromosome microdissection, and FISH on post-natal blood samples. The parents had normal karyotypes. De novo partial duplications of chromosome 1q have rarely been reported. Comparison of our patient with other published pure trisomy 1q cases showed similarities which allowed the further delineation of the trisomy 1q syndrome.

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Trisomy 1q is a rare condition. In three recent reports clinical similarities and differences between patients with duplications of the long arm of chromosome 1 have been discussed and the cases published so far reviewed.^{1–3} The reported cases of partial trisomy 1q include trisomies of all segments of the q arm of chromosome 1 (1q21-qter), but most consist of trisomy 1q32-qter^{3–16} and 1q42-qter.^{1 2 17–24} Frequently, however, they are accompanied by other chromosome aberrations which have made the definition of a phenotype difficult.^{1–24} De novo duplications without another chromosome aberration appear to be rare since only three cases with pure trisomy 1q of the (1)(q32-qter) segment have been published.^{25–27} Here we describe a patient who has a duplication of the 1q32-qter segment, which is inserted at the terminal end of the short arm of the same chromosome in an inverted orientation. Our patient confirms the previously described features of trisomy 1q and allows further delineation of the associated syndrome. Diagnosis of the trisomic segment based on pure cytogenetic methods would have been difficult without the use of a chromosome paint, microdissection with reverse painting, a

telomere 1q probe, and a terminal 1p probe. Therefore this case illustrates the significant diagnostic value of FISH in cases where the parental chromosomes are normal or not available.

Materials and methods

CASE REPORT

The proband is the second child of a 30 year old mother and a 29 year old father. In the 30th week of gestation amniocentesis was performed because of hydramnios, a vitium cordis, and suspected renal agenesis and gastrointestinal obstruction on ultrasound. The boy was born at 37 weeks of gestation by caesarean section (weight 2600 g, length 45 cm, head circumference 36.5 cm, Apgar 4–8–8). The following dysmorphic features were observed on physical and clinical examination after birth (fig 1): hydrocephalus, an asymmetrical skull, wide sutures and large fontanelles, a prominent forehead, and dilatation of the lateral ventricles and the frontal and temporal subarachnoid space. There were also low set, posteriorly rotated, anteverted ears, abnormal helix and anthelix, hypoplastic lobes, tragus, and antitragus, and sensory hearing loss, as well as blepharophimosis, congenital ptosis, sparse eyelashes, downward slanting palpebral fissures, bilateral hypoplasia of the papilla of the optic nerve, and alternating divergent strabismus. He had a broad, beaked, pointed nose with a flat nasal bridge, bilateral cleft lip and palate, oesophageal stenosis, tracheal stenosis, tracheomalacia, pulmonary stenosis, atrial septum aneurysm, tricuspid insufficiency, duodenal obstruction and malrotation, widely spaced nipples, 13 pairs of ribs, multicystic dysplastic left kidney, dysplastic right kidney, a mal-descended testis, hypospadias and phimosis, bilateral preaxial polydactyly (bifid thumbs as a result of duplication of the terminal phalanx), and long and overlapping toes (IV over V).

Fig 2 shows the boy at the age of 18 months. Surgery for the duodenal atresia, cleft lip, phimosis, and hypospadias was done within the first seven months. A tracheostomy was performed after two months. Whereas at birth the somatic data were in the normal range his development was delayed at the age of 18 months (weight 6380 g = <3rd centile, length 69 cm = <3rd centile, head circumference 42.5 cm = <3rd centile). At the last follow up at

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Figure 1 The proband with partial trisomy 1q32-qter.

Figure 2 The proband at the age of 18 months.

the age of 46 months examination showed a microcephalic boy with extreme developmental delay with all measurements far below the 3rd centile (weight 8440 g = <3rd centile, length 78 cm = <3rd centile, head circumference 46.5 cm = <3rd centile). From birth the boy has suffered from functional vomiting, recurrent aspirations, and recurrent infections of the respiratory, digestive, and urogenital tracts. Because of an uncorrected cleft palate and oesophageal stenosis the boy suffers from dysphagia and needs tube feeding. His vision and hearing are impaired and an examination at the department of development and movement disorders showed extreme psychomotor and mental retardation.

CYTOGENETICS

The first cytogenetic examination was performed on cultured amniocytes obtained by amniocentesis in the 30th week of gestation. The cells were harvested by the flask method²⁸ and GTG and QFQ banded according to Benn and Pearle.²⁹ After birth chromosomes of the proband and his parents were prepared from peripheral lymphocyte cultures using a modification of the synchronisation method of Yunis.³⁰ High resolution banding was performed by treatment with trypsin followed by staining with Giemsa to obtain a GTG band pattern (fig 3). No cell line is available.

FISH ANALYSIS AND MICRODISSECTION

Metaphase spreads were prepared from PHA stimulated lymphocytes using the standard 3:1 (v/v) methanol:acetic acid fixation. Slide treatment and hybridisation was performed according to the protocol of Lichter and Cremer³¹ with minor modifications. A paint specific for chromosome 1 (AGS, Germany), a telomere

1q probe (D1S555, Oncor, USA), and a terminal chromosome 1 midisatellite probe (D1Z2, Oncor, USA) were applied to the metaphase spreads according to the manufacturer's specifications. Five p arm fragments of the aberrant chromosome 1 were microdissected, amplified by DOP-PCR, and hybridised to normal control metaphases according to the protocol described by Müller-Navia *et al.*³²

Results

Analysis of chromosomes from amniocytes showed additional unknown material on the short arm of one chromosome 1. The karyotype was 46,XY,add(1)(p36.3) (fig 3). Hybridisation with a chromosome 1 specific paint also hybridised to the additional segment on chromosome 1 (fig 4). Hybridisation with the telomere 1q probe showed a signal on the q arm of the normal chromosome 1 and a signal on both ends of the aberrant chromosome 1 (fig 5). Hybridisation with the terminal chromosome 1 midisatellite probe showed a signal on the p arm of the normal and of the aberrant chromosome 1 (fig 6). Reverse painting with the amplified p arm fragment of the aberrant chromosome 1 showed a hybridisation signal on the p arm and on the end of the q arm of chromosome 1 (fig 7). In combination with analysis of GTG banded high resolution prometaphase chromosomes, the trisomic area was defined as 1q32-qter. This region is duplicated and inverted and inserted at 1p36.3. The final karyotype therefore is 46,XY,inv ins dup(1)(p36.3qter-q32). The parental karyotypes were normal.

Discussion

For most chromosomal derivatives, GTG banded high resolution chromosomes produce a banding pattern sufficient for chromosome

Figure 3 G banded partial karyotype of proband showing normal (a, c) and duplicated (b, d) chromosome and ideogram of chromosome 1 with arrows indicating the breakpoints at p36.3 and q32 and the duplicated segment q32-qter.

Figure 4 Identification of 1p+ material as derived from chromosome 1 by FISH with a chromosome 1 paint.

Figure 5 (A) Identification of duplicated material on chromosome 1 by FISH with telomere 1q probe showing normal chromosome 1 with one FITC signal at qter (single white arrow) and duplicated chromosome 1 with two FITC signals (two white arrows) at qter and duplicated 1q segment at p arm. (B) Grey scaled DAPI image of the same metaphase showing qter of normal chromosome 1 (single black arrow) and terminal ends of duplicated chromosome 1 (two black arrows).

identification. In our case it was difficult to make a definitive assignment of the trisomic area to chromosome 1. Only the combined use of (1) a whole chromosome paint, (2) a telomere probe, (3) a midisatellite probe, and (4) microdissection with reverse painting was sufficient for unequivocal identification of the extra chromosomal material and for diagnosing the aberration as *inv ins dup(1)(p36.3qter-q32)*. The case presented here underlines the importance of combining different FISH methods for the identification of unknown chromosome segments, especially if the parents' chromosomes are normal or not available. By chromosome painting with a whole chromosome paint, the origin of the additional material could be determined and, subsequently, the exact assignment of the unknown chromosomal material to a defined chromosomal region was done by reverse painting with

Figure 6 Hybridisation with terminal chromosome 1 midisatellite probe (D1Z2, Oncor, USA), showing a signal on both chromosomes 1.

Table 1 Clinical findings in patients with pure trisomy 1q32-qter

	Our case	Clark et al ²⁷	Steffenson et al ²⁵	Flatz and Fonatsch ²⁶	Cases with trisomy 1q42-qter ^{12-17,24}
Low birth weight	-	+	+	+	+
Wide sutures and fontanelles	+	+	+	+	+
Prominent, wide forehead	+	-	+	+	+
Downward slanting palpebral fissures	+	-	+	-	+
Small palpebral fissures	+	+	-	+	+
Broad, flat nasal bridge	+	-	+	+	+
Pointed, beaked nose	+	-	-	+	+
Low set, posteriorly rotated ears	+	+	+	+	+
Malformed ears	+	+	+	-	+
Cardiopathies	+	+	+	+	+
Urogenital abnormalities	+	+	+	+	+
Abnormal fingers or toes or both	+	+	+	+	+
Retarded psychomotor development	+	-	+	-	+

+ = presence of a symptom, - = absence of a symptom or not mentioned in the clinical description.

Figure 7 Reverse painting on a normal control metaphase with microdissected and amplified fragments of the p arm of the aberrant chromosome 1, showing a signal on the whole p arm and the 1q32-qter region (white arrows).

microdissected fragments. Probes from telomeric and terminal regions proved the integrity of the terminal arms of the chromosome investigated.

Most previously reported cases with trisomy 1q show trisomy 1(q32-qter) or trisomy 1(q42-qter), frequently with additional chromosomal aberrations.¹⁻²⁹ Abnormalities which are regularly found in trisomy 1q cases include low birth weight, wide sutures and fontanelles, prominent, wide forehead, downward slanting and small palpebral fissures, broad, flat nasal bridge, pointed and beaked nose, low set, posteriorly rotated, and malformed ears, cardiopathies, urogenital abnormalities, abnormal fingers and toes, and delayed development. Cases of trisomy 1q spanning 1q32-qter without involvement of another chromosome have been described by Steffenson *et al*²⁵ and Flatz and Fonatsch.²⁶ They show a tandem duplication of 1q32-qter. The patient of Clark *et al*²⁷ and our case also have a pure duplication of the 1q32-qter region, though with a different position within the chromosome. Table 1 lists the relevant clinical characteristics of trisomy 1q32-qter cases and their presence/absence in the four cases with pure trisomy 1q32-qter and cases with trisomy 1q42-qter. These features are also found in patients with trisomy

1q42-qter,^{1 2 17-24} which can be expected to share features with trisomy 1q32-qter. However, not all patients with trisomy 1q42-qter have these signs and no cases with pure trisomy 1q42-qter have been published. Therefore it is unclear whether and which clinical features are caused by the trisomy 1q42-qter in these patients. Clinical features which are shared by at least three of the patients with pure trisomy 1q32-qter are low birth weight, wide sutures and fontanelles, prominent, wide forehead, small palpebral fissures, broad, flat nasal bridge, low set, posteriorly rotated, and malformed ears, cardiopathies, urogenital abnormalities, and abnormal fingers or toes or both. Since the three previously described cases together with the case presented here share a single duplication of 1q32-qter, the phenotype cannot be attributed to another chromosome aberration, for example to a monosomic segment of another chromosome. Although the extent of the clinical descriptions is different in the reported cases, which makes comparison difficult, we conclude that the characteristics shared by at least three of these four patients represent abnormalities which define a specific trisomy 1q32-qter syndrome. A gene which is associated with the 1q32 segment is the gene for Van Der Woude syndrome (VWS) (OMIM 119300³³). Patients with VWS show cleft lip and palate. This suggests that the VWS gene may contribute to the phenotype found in our patient.

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1 Rasmussen SA, Frias JL, Lafer CZ, Eunpu DL, Zackai EH. Partial duplication 1q: report of four patients and review of the literature. *Am J Med Genet* 1990;36:137-43.
2 Johnson VP. Duplication of the distal part of the long arm of chromosome 1. *Am J Med Genet* 1991;39:258-69.
3 DuPont BR, Huff RW, Ridgway LE, Stratton RF, Moore CM. Prenatal diagnosis of partial trisomy 1q using fluorescent in situ hybridisation. *Am J Med Genet* 1994;50:21-7.
4 Sanger R, Alfi O, Donnell G. Partial trisomy 1q in 3 patients. *Am J Hum Genet* 1974;26:75A.
5 Schinzel A. Duplication-deletion with partial trisomy 1q and partial monosomy 3p resulting from a maternal reciprocal translocation rcp(1;3)(q32;p25). *J Med Genet* 1981;18:64-8.
6 Yunis E, Egel H, Zuniga R, Ramirez E, Torres de Caballero OM, Leibovici M. "De novo" trisomy 1q32-qter and monosomy 3p25-3pter. *Hum Genet* 1977;36:113-16.
7 Bourrouillou G, Colombies P, Blanc P. Trisomie 1q secondaire à une translocation réciproque maternelle. *C R Soc Biol (Paris)* 1978;172:359-62.

- 8 Taysi K, Sekhon GS. Partial trisomy of chromosome No 1 in two adult brothers due to a maternal translocation (1q-;6p+). *Hum Genet* 1978;**44**:277-85.
- 9 Schinzel A. Cyclopia and cebocephaly in two newborn infants with unbalanced segregation of a familial translocation rcp(1;7)(q32;q34). *Am J Med Genet* 1984;**18**:153-61.
- 10 Schinzel A. A further case of cyclopia due to unbalanced segregation of a previously reported rcp(1;7)(q32;q34) familial translocation. *Am J Med Genet* 1986;**24**:205-6.
- 11 Boué J, Daketsé MJ, Deluchat C, Ravisé N, Yvert F, Boué A. Q and G banding techniques in the identification of chromosome anomalies in spontaneous abortions. *Ann Genet (Paris)* 1976;**19**:233-9.
- 12 Bonfante A, Stella M, Rossi G. Partial trisomy of the long arm of chromosome 1 due to a familial translocation t(1;10)(q32;q26). *Hum Genet* 1978;**45**:339-43.
- 13 Rehder H, Friedrich U. Partial trisomy 1q syndrome. *Clin Genet* 1979;**15**:534-40.
- 14 Fryns JP, de Muelenaere A, Pedersen J, van de Berghe H. Partial distal 1q trisomy. A distinct clinical dysmorphic syndrome in adulthood. *Ann Genet (Paris)* 1980;**23**:181-2.
- 15 Seabright M. The use of proteolytic enzymes for the mapping of structural rearrangements in the chromosomes of man. *Chromosoma* 1972;**36**:204-10.
- 16 Michels VV, Berseth CL, O'Brian JF, Dewald G. Duplication of part of chromosome 1q: clinical report and review of literature. *Am J Med Genet* 1984;**18**:125-34.
- 17 Leisti J, Aula P. Partial trisomy 1 (q42-qter). *Clin Genet* 1980;**18**:371-8.
- 18 Wade J, Morgan T, Allanson J. Child with deletion of 4q and duplication of 1q. *Am J Hum Genet* 1989;**33**:553-4.
- 19 Duillo MT, de Toni T, Cavaliere G, et al. Studio clinico e citogenetico di un caso di trisomia 1q per traslocazione familiare t(1;5)(q42;p15.3). *Pathologica* 1979;**71**:223-34.
- 20 Pierquin G, Van Regemorter N, Hayez-Delatte, et al. Two unrelated children with partial trisomy 1q and monosomy 6p, presenting with the phenotype of the Larsen syndrome. *Hum Genet* 1991;**87**:587-91.
- 21 Kiss P, Imrei J. Trisomy 1q. *Clin Genet* 1989;**36**:75-6.
- 22 Liberfarb RM, Breg WR, Atkins L, Holmes LB. Multiple congenital anomalies/mental retardation (MCA/MR) syndrome due to partial 1q duplication and possible 18p deletion: a study of four individuals in two families. *Am J Med Genet* 1979;**4**:27-37.
- 23 Chia NL, Bousfield LR, Poon CCS, Dtrudinger BJ. Trisomy (1q)(q42-qter): confirmation of a syndrome. *Clin Genet* 1988;**34**:224-9.
- 24 Kennerknecht I, Barbi G, Rodens K. Dup(1q)(q42-qter) syndrome: case report and review of the literature. *Am J Med Genet* 1993;**8**:1157-60.
- 25 Steffensen DM, Chu EHY, Speert DP, Wall PM, Meilinger K, Kelch RP. Partial trisomy of the long arm of human chromosome 1 as demonstrated by in situ hybridisation with 5S ribosomal RNA. *Hum Genet* 1977;**36**:25-33.
- 26 Flatz S, Fonatsch C. Partial trisomy 1q due to tandem duplication. *Clin Genet* 1979;**15**:541-2.
- 27 Clark BJ, Lowther GW, Lee WR. Congenital ocular defects associated with an abnormality of the human chromosome 1: trisomy 1q32-qter. *J Pediatr Ophthalmol Strabismus* 1994;**31**:41-5.
- 28 Rooney DE, Czepulkowski BH. Prenatal diagnosis and tissue culture. In: Rooney DE, Czepulkowski BH, eds. *Human cytogenetics I*. Oxford: Oxford University Press, 1992:55-89.
- 29 Benn PA, Perle MA. Chromosome staining and banding techniques. In: Rooney DE, Czepulkowski BH, eds. *Human cytogenetics I*. Oxford: Oxford University Press, 1992:91-118.
- 30 Yunis JJ. New chromosome techniques in the study of human neoplasia. *Hum Pathol* 1981;**12**:540-9.
- 31 Lichter P, Cremer T. Chromosome analysis by non-isotopic in situ hybridization. In: Rooney DE, Czepulkowski BH, eds. *Human cytogenetics I*. Oxford: Oxford University Press, 1992:157-92.
- 32 Müller-Navia J, Nebel A, Schleiermacher E. Complete and precise characterization of marker chromosomes by application of microdissection in prenatal diagnosis. *Hum Genet* 1995;**96**:661-7.
- 33 Online Mendelian Inheritance in Man, OMIM(TM). Johns Hopkins University, Baltimore, MD. MIM Number: 119300: 6/25/1994: World Wide Web URL: <http://www3.ncbi.nlm.nih.gov/omim>.