SHORT REPORT

Inv dup(22), del(22)(q11) and r(22) in the father of a child with DiGeorge syndrome

Anne Bergman and Elisabeth Blennow

Department of Clinical Genetics, Karolinska Hospital, Stockholm, Sweden

We here report a unique inherited case of DiGeorge syndrome. The asymptomatic father had a mosaic karyotype with a 21q11 deletion in three different cell lines. In two of the cell lines there was an additional supernumerary inv dup(22) or an r(22), respectively. In the third cell line the del(22) was the sole anomaly. FISH analysis showed that both the inv dup(22) and the r(22) included the DGS region. We hypothesize that an inter-chromosomal recombination between inverted repeats, together with a recombination between sister chromatids during meiosis I, gave rise to a deletion of 22q11 as well as an inv dup(22) containing the DGS region. The inv dup(22) was later rearranged into a ring chromosome during mitosis which was subsequently lost during cell division, thereby resulting in three different cell lines. This is the first case reported with an inv dup(22) and a del(22)(q11) in the same cell line. Our findings support a related mechanism in the formation of these two rearrangements mediated by low-copy repeats. *European Journal of Human Genetics* (2000) **8**, 801–804.

Introduction

DiGeorge syndrome (DGS) is caused by a defect in the development of the pharyngeal arches and pouches. The major features include outflow tract malformations of the heart, a dysmorphic facial appearance, thymic hypoplasia and hypocalcaemia.[1] Microdeletion of 22q11 is found in about 90% of patients with DGS. The deletion is inherited in approximately 20% of the cases and it is therefore recommended to screen parents for carrier status. Carrier parents frequently appear to be more mildly affected than their children. The fact that most DGS cases occur *de novo* indicates that the q11 region on chromosome 22 is especially prone to rearrangements.

Cat eye syndrome (CES) is another, less frequent 22q11 disorder with a variable phenotype including ocular coloboma, anal atresia, heart defects and dysmorphic features.[2] CES is associated with a duplication 22pter → 22q11, most often in the form of a supernumerary marker chromosome, an inv dup(22). The marker chromosome is reported to vary in size with some of the CES chromosome breakpoints corresponding to common breakpoints of the DGS deletion.

This suggests that the mechanism of formation of the inv dup(22) and the del(22)(q11) may be related.[3,4]

We here describe a child with DGS due to an inherited 22q11 deletion. The father has a complex mosaic karyotype that includes a 22q11 deletion as well as a 22q11 duplication in the form of an inv dup(22) or a r(22).

Materials and methods

Case history

The proband, the first son of healthy and unrelated parents, was born at term after an unremarkable pregnancy, with a birth weight of 3275 g and a birth length of 51 cm. Due to breathing difficulties at 2 days old, cardiac investigation with echography and angiography was performed and showed an interrupted aortic arch and a ventricular septal defect. He also had borderline hypocalcaemia and a dysmorphic facial appearance reminiscent of DGS.

Cytogenetic analysis

Peripheral blood samples were obtained from the proband, and subsequently from his parents and his paternal grandparents. Metaphase chromosome spreads were prepared from

Correspondence: Dr Elisabeth Blennow, Department of Clinical Genetics, Karolinska Hospital, S171 76 Stockholm, Sweden. Tel: + 46 8 517 75380; Fax: + 46 8 32 77 34; E-mail: ebl@gen.ks.se
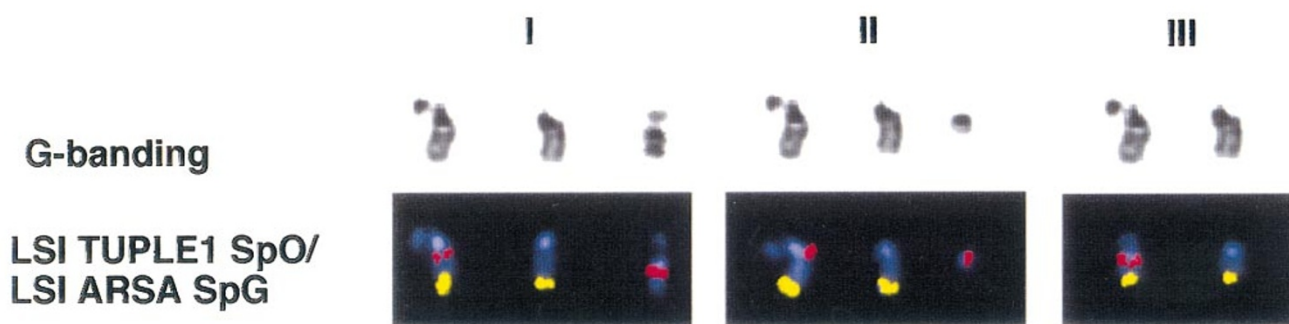
Figure 1 Mosaicism with three different cell lines. The G-banded chromosomes on top and the FISH results using LSI TUPLE1 SpO/LSI ARSA SpG below.

lymphocyte cultures and routine GTG-banding was performed according to standard procedures.

FISH
The probe LSI TUPLE1(22q11, DGS region) Spectrum Orange/LSI ARSA(22q11, control probe) Spectrum Green (Vysis, Downers Grove, IL, USA) was used according to the manufacturer's instructions. The probe alpha XT680 (centromere 14/22 specific probe) was labelled with FITC-

12-dUTP (Vector Lab, Burlingame, LA, USA) by nick translation and used as previously described.[5]

Results
FISH analysis using the probe LSI TUPLE1/LSI ARSA showed a microdeletion of 22q11 in the child, thus confirming the DGS diagnosis. Chromosomal and FISH analyses of the mother and the paternal grandparents were all normal. Cytogenetic analysis of the apparently asymptomatic and
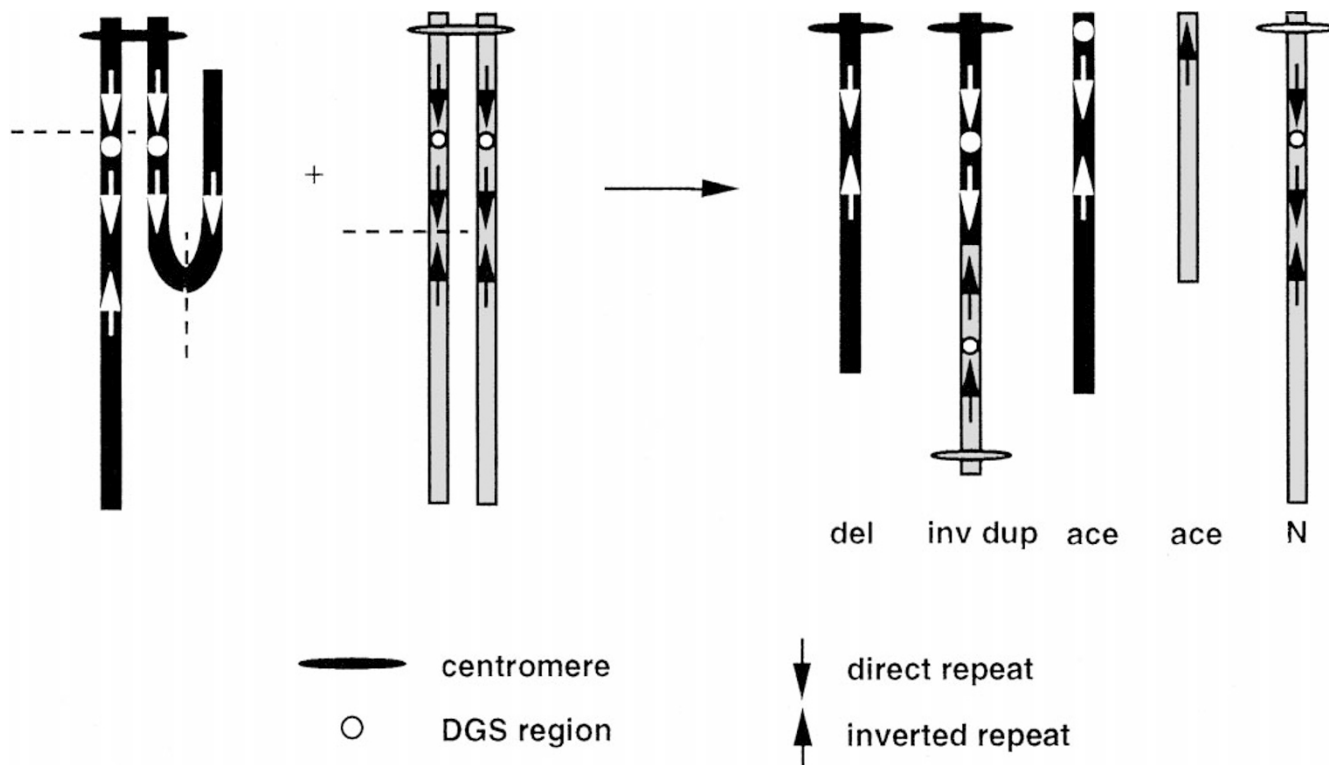

Figure 2 Inter-chromosomal recombination between inverted repeats combined with a recombination between sister chromatids resulted in a del(22)(q11) and an inv dup(22).

non-dysmorphic father revealed mosaicism with three different cell lines (Figure 1). FISH analysis using LSI TUPLE1/LSI ARSA showed a deletion of TUPLE1 in one of the cytogenetically normal chromosomes 22 in all three cell lines. In the first cell line (12% of the cells according to FISH analysis), there was also a supernumerary marker chromosome. Since the marker chromosome showed labelling with TUPLE1 but not with ARSA and double labelling with the 14/22 centromere specific probe, it was identified as an inv dup(22). The second cell line (23% of the cells) had a supernumerary ring chromosome in addition to the deleted chromosome 22. The ring chromosome showed labelling with TUPLE1 but not with ARSA and single labelling with the 14/22 centromere specific probe. The third cell line (65% of the cells) had deletion of 22q11 as the sole anomaly. In conclusion, the father's karyotype was:

46,XY,del(22)(q11q11)/47,idem,+r(22)(p?p?)/47,idem,
+inv dup(22)(pter → q11::q11 → pter)

Discussion

Mosaicism for structural aberrations is rare and the presence of different rearrangements involving the same chromosomal region is intriguing. We have found the simultaneous occurrence of a del(22)(q11) and an extra chromosome containing the 22q11 region, suggestive of a common

Figure 3 The evolution of the three different cell lines of the father.

mechanism of formation of these rearrangements. The fact that low-copy repeats on 22q11 may mediate rearrangements both for CES chromosomes and 22q11 deletions is consistent with a related origin.[6]

Microdeletions are considered to arise through unequal crossing over as the result of misalignment in homologous regions.[7] Low-copy-number repeat sequences flank the DGS region at 22q11,[8] and recent studies by Edelman et al have shown that the breakpoints of the common DGS deletion on 22q11 are located in those highly homologous low-copy repeats.[9] The formation of a deletion may also result in a reciprocal duplication product. This is thought to be the case in Charcot-Marie Tooth disease 1A and HNPP (hereditary neuropathy with pressure palsies), where the structural rearrangement is a duplication or a deletion of the same region on 17p.[10] Edelman et al described a family with an interstitial duplication of the 3 Mb region that is deleted in DGS patients.[9] To the best of our knowledge, a 22q11 duplication together with a deletion in the same patient has hitherto never been reported.

Inverted duplications have been suggested to arise as a U-type exchange between homologous chromosomes[11,12] or inter-chromosomal recombination between inverted sequences.[6,13] Another alternative is repair of a broken chromosome through replication and end to end fusion.[11]

We hypothesize that an inter-chromosomal recombination between inverted repeats combined with a recombination between sister chromatids during meiosis I gave rise to both a deleted 22q11 and an inv dup(22) containing the DGS region (Figure 2). The acentric fragments were lost. After fertilization the cell line included one normal chromosome 22, one deleted chromosome 22 and the inv dup(22). The inv dup(22) was later rearranged into a ring chromosome which, due to its unstable nature, was subsequently lost during mitotic cell division, thereby giving rise to three different cell lines (Figure 3).

A few cases with inv dup chromosomes and a deletion or a duplication of the homologous regions have previously been reported. Spinner et al described a patient with Angelman syndrome due to a deletion of one of the cytogenetically normal chromosomes 15 and an inv dup(15) that, in contrast to our case, did not include the deleted region.[14] Therefore, the inv dup(15) was not believed to contribute to the patient's phenotype. It was speculated that patients with inv dup(15)s may be at increased risk for other abnormalities involving 15q11–q13 and it was suggested that when one cytogenetic abnormality occurred, it could predispose to a second rearrangement during meiosis. Leana-Cox et al described a patient with an inv dup(15) and a dup(15),[15] and a similar finding was made by Abeliovich et al.[16] Mears et al reported the presence of an interstitial duplication of 22q in combination with an inv dup(22).[17] However, the present case is the first to be described with an inverted duplication and a deletion involving the same region on 22q.
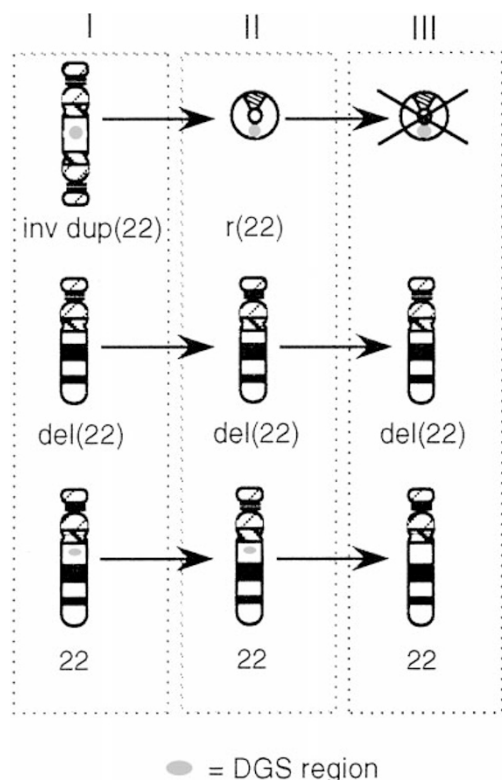
The risk of recurrence of DGS in our case is difficult to estimate due to the presence of mosaicism and many possible modes of segregation. The risk of DGS could be as high as 50%. In addition, there is also a theoretical risk of offspring with cat eye syndrome if the inv dup(22) or the r(22) is inherited together with a normal chromosome 22. Prenatal diagnosis may be offered to the couple, including chromosome analysis and FISH after chorionic villus sampling or amniocentesis.

The cytogenetic findings in this case provide a possible link between DGS and CES. Future similar cases will hopefully further elucidate the mechanism of formation of these structural rearrangements.

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