Diagnosis of a complex chromosomal rearrangement using fluorescent in situ hybridisation

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Abstract

We report the use of fluorescent in situ hybridisation (FISH) to clarify a complex chromosomal rearrangement (CCR) carried by a woman presenting with recurrent miscarriages. CCRs are rare cytogenetic rearrangements involving three or more chromosomes, which can be difficult to interpret using routine cytogenetic studies with GTG banding. FISH was used to establish a correct interpretation of the maternal karyotype before amniocentesis in a present pregnancy.

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Key words: complex chromosomal rearrangements (CCR); fluorescent in situ hybridisation (FISH); prenatal diagnosis; recurrent spontaneous abortion.

Complex chromosomal rearrangements (CCR) involve rearrangement of chromosomal material between three or more chromosomes with three or more breakpoints, which present difficulty in interpretation.^{1,2} The rearrangements can include deletions, insertions, or inversions that distort GTG banding patterns.³ Fluorescent in situ hybridisation (FISH) has been used to identify the origin of rearranged chromosomal material.³⁻⁵ We report a case of a CCR carrier identified through recurrent spontaneous abortion in whom FISH studies were vital for correct interpretation of an unbalanced fetal chromosome karyotype in a continuing pregnancy.

Case report

The proband's obstetric history included three spontaneous abortions at 12, 14, and 20 weeks, as well as delivery of a healthy son in her third pregnancy. Tissue from her 20 week loss had shown an abnormal fetal karyotype interpreted by GTG banding as 46,XX,-5,-7,+der(5)t(5;7)(p15;q31),+der(7)t(7;11)(q31;q13). When she presented for prenatal diagnosis at 16 weeks' gestation in her fifth pregnancy, she was identified as a carrier of a CCR involving chromosomes 5, 7, and 11 (fig 1). Her parents and her husband had normal karyotypes.

In order to perform prenatal diagnosis, FISH was performed using a maternal blood sample in separate hybridisations using two probes from Oncor, Inc, Gaithersburg, MD: cri du chat cosmid probe with assignment at 5p15.3 (fig 2A) and chromosome 7 specific painting probe (fig 2B). The study showed a small chromosome segment containing 5p15.3 inserted into the q arm of a submetacentric chromosome identified on GTG banding as a derivative 7. Inversion of the inserted segment is possible, because its orientation could not be determined. FISH using the 7 painting probe showed chromosome 7 derived material on a large submetacentric chromosome identified as a derivative 5. The 7 paint probe did not hybridise evenly, but was satisfactory for identification of the rearranged material on the derivative chromosomes. A chromosome 11 paint probe was not necessary as the 11 material was clearly identifiable on routine banding. Combining the information from GTG band-

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Figure 1 Partial ideogram and GTG banded karyotype of the patient showing normal chromosomes 5, 7, 11, and their derivatives. Ideogram shows rearrangements as interpreted after FISH studies.

Figure 2 Partial view of metaphase from lymphocyte culture showing FISH study: (A) cri du chat cosmid probe indicating 5p15.3 inserted into derivative chromosome 7 and normal homologue, (B) painting probe for chromosome 7 showing translocated 7 material on derivative 5, derivative 7, and normal homologue.

ing and FISH analysis, the breakpoints of the three way translocation were established as 5p15.1, 5p15.3, 7q31.2, and 11q13.3. The final interpretation of the balanced maternal karyotype was 46,XX,t(5;7;11)(5qter→5p15.1::7q31.2→7qter;7pter→7q31.2::5p15.3 5p15.1::11q13.3 → 11qter;11pter → 11q13.3::5p15.3→5pter).

Ultrasound studies at 17 weeks, when amniocentesis was performed, did not detect any fetal abnormalities. Cytogenetic analysis of fetal cells showed an unbalanced karyotype trisomic for 7q31.2 to 7qter and monosomic for 5p15.1 to 5pter: 46,XY,-5,+der(5)t(5;7;11)mat. Since the derivative chromosome 5 was identified through FISH studies of the maternal karyotype, GTG studies were sufficient for prenatal diagnosis.

Duplication of 7q in the region of 7q3 has resulted in pre- and postnatal growth retardation, severe to profound mental retardation, hypotonia, seizures, hydrocephalus, and neonatal death.⁶ Deletion of 5p results in the cri du chat syndrome, associated with severe mental retardation, microcephaly, growth retardation, epicanthic folds, and a monochromatic cry similar to the mewing of a cat.⁷ Because of the severity of the prognosis, the patient decided to terminate the affected pregnancy.

Figure 3 Theoretical multivalent chromosome pairing in pachytene of meiosis I showing the intricacy involved in chromosome pairing and assortment with the 5, 7, 11 translocation.

Discussion and conclusions

Most carriers of CCRs are ascertained through recurrent miscarriages or the birth of an abnormal child.⁸ Recurrence risks for pregnancy loss are empirically estimated to be as high as 48%, and the risk for birth of an abnormal child may be as high as 18%.⁹ Fig 3 shows the theoretical meiotic pairing of the rearranged chromosomes to illustrate the intricate segregation needed to produce a balanced gamete in this case.

In previously reported CCR cases, when FISH was used to supplement GTG studies, the rearrangements were correctly identified to be more complex.^{3 10 11} In the reported case, FISH studies allowed accurate prenatal diagnosis of complex cytogenetic findings and more specific prediction of potential fetal abnormalities.

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