Prenatal Diagnosis and Molecular Analysis of Triploidy in a Fetus With Intrauterine Growth Restriction, Relative Macrocephaly and Holoprosencephaly

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A 24-year-old, gravida 4, para 1, woman was referred to the hospital at 16 weeks of gestation because of fetal structural abnormalities. Level II ultrasound revealed a fetus with intrauterine growth restriction (IUGR), a normal amount of amniotic fluid, a small non-cystic placenta, a singleton fetus with a crown-rump length of 5.5 cm (12 weeks' equivalent), alobar holoprosencephaly (HPE), and relative macrocephaly with an anteroposterior diameter about half of the crown-rump length (Figure 1). The urinary tracts were normal. Amniocentesis revealed a karyotype of 69,XXX. The pregnancy was subsequently terminated. A 40-g female fetus was delivered with a small placenta (Figure 2). At birth, the proband displayed relative macrocephaly, premaxillary agenesis, median facial cleft, and a small thin trunk. The placenta was small and non-cystic. Postnatal cytogenetic analysis of the fetal skin revealed a karyotype of 69,XXX (Figure 3). DNA was obtained from the uncultured tissues of fetal skin and parental blood. Quantitative fluorescent polymerase chain reaction (QF-PCR) assays and polymorphic short tandem repeat markers for chromosomes 13, 18 and 21 were used for determination of the parental origin of the extra chromosomes (Table). The skin specimen showed a diallelic pattern with a dosage ratio of 1:2 (paternal allele to maternal allele ratio) for the chromosome 21-specific and chromosome 18-specific pericentromeric markers,

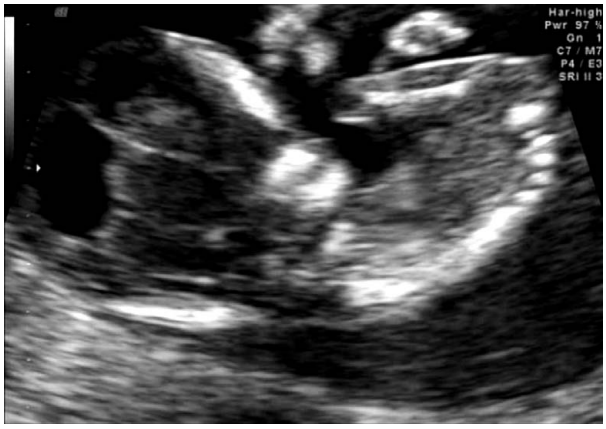

Figure 1. Prenatal ultrasound of the fetus at 16 weeks of gestation shows a relative large head and brain malformations.

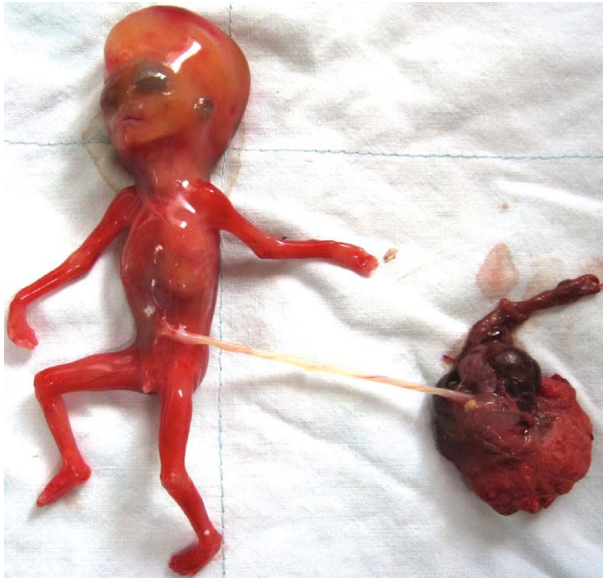

Figure 2. The fetus and the placenta at birth.

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Accepted: July 2, 2009

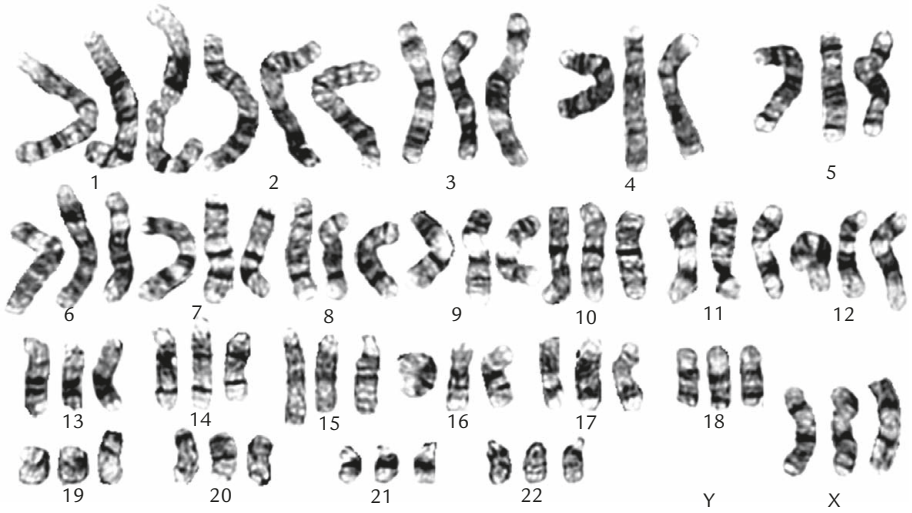

Figure 3. A karyotype of 69,XXX.

Table. Genotypic information of the fetus and the parents at short tandem repeat (STR) markers specific for chromosomes 13, 18 and 21 by quantitative fluorescent polymerase chain reaction assays*

STRs	Location	Father	Mother	Proband
D21S1434	21p11.2	182,194	190,198	182,190,190
D18S847	18q12.1	210,226	218	210,218,218
D18S1369	18q12.2	180	172	172,172,180
D13S1491	13q13.3	146,154	150,154	146,150,154
D13S1492	13q21.1	159	147,159	147,159,159
D13S766	13q33.1	129,133	121,125	121,125,129

**Alleles (basepair sizes) are listed below each individual.*

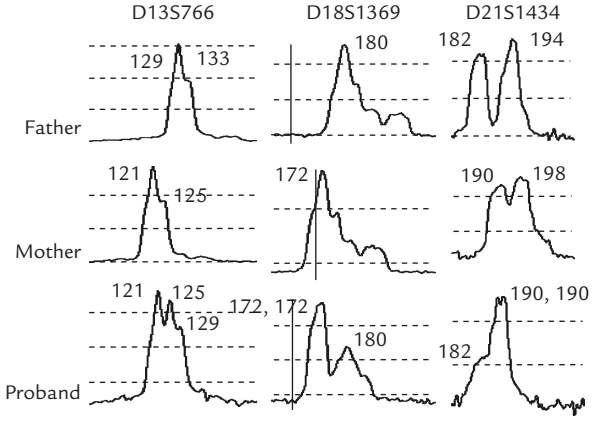

Figure 4. Representative electrophoretogram of quantitative fluorescent polymerase chain reaction assays at short tandem repeat markers for chromosomes 13, 18 and 21 using fetal tissues and parental blood. With the pericentromeric markers D21S1434 and D18S1369, the fetus showed a diallelic pattern with a dosage ratio of 1:2 (paternal allele to maternal allele ratio). With the non-pericentromeric and distal marker D13S766, the fetus showed a triallelic pattern with a dosage ratio of 1:1:1 (paternal allele to maternal allele to maternal allele ratio). Digynic triploidy in this fetus was most likely the result of meiosis II nondisjunction error of oogenesis.

and a triallelic pattern with a dosage ratio of 1:1:1 (paternal allele to maternal allele to maternal allele ratio) for the chromosome 13-specific non-pericentromeric and distal markers (Figure 4). The molecular data indicated a maternal origin of digynic triploidy. Triploidy in the fetal tissues in this case was most likely the result of meiosis II nondisjunction error of oogenesis. A recombination was observed. There were reductions to homozygosity at the pericentromeric markers and no reduction to homozygosity at the more distal loci.

Our case represents a rare occurrence of triploidy with HPE presenting at 16 weeks of gestation. Triploidy occurs in about 1% of all conceptions [1]. The prevalence of triploidy has been estimated to be 1 per 50 at 6 gestational weeks, 1 per 350 at 8 gestational weeks, 1 per 1,000 at 10 gestational weeks, 1 per 3,500 at 12 gestational weeks, 1 per 10,000 at 14 gestational weeks, 1 per 30,000 at 16 gestational weeks, 1 per 100,000 at 18 gestational weeks, and 1 per 250,000 at 20 gestational weeks [2]. Jauniaux et al [3] reported HPE in 2.9% (2/70) of cases with triploidy at 13–29 weeks of gestation, and Mittal et al [4] reported HPE in 5% (1/20) of cases with triploidy at 14–25 weeks of

gestation. Chen et al [5] reported that triploidy represented 6.8% (5/73) of cases with fetal HPE detected in the second and third trimesters. However, Jauniaux et al [6] reported HPE in 22.2% (4/18) of cases with triploidy at 10–14 weeks of gestation, and Philipp et al [7] reported HPE in 11.1% (2/18) of cases with triploidy at 10–15 weeks of gestation. The frequency of HPE in prenatally diagnosed triploidy varies with gestational age at diagnosis. When the diagnosis of triploidy is made in early pregnancy, the frequency of HPE increases. The low frequency of triploidy in fetal HPE detected in the second trimester and beyond is because most of the conceptuses with triploidy abort in the first trimester and only a few survive to the second trimester [5].

The prevalence of HPE in registered births is about 1.2 per 10,000 [8,9]. The prevalence of HPE in the second trimester is about 1 per 8,000 [9]. Variable types of HPE range from severe alobar HPE with cyclopia, ethmocephaly, cebocephaly or premaxillary agenesis, to microforms with microcephaly, corpus callosum agenesis/dysgenesis, mental retardation, ocular hypertelorism only, or a single maxillary central incisor [10]. Chromosomal aberrations, Mendelian mutations, X-linked inheritance and teratogens are well known causes of HPE. Cytogenetic abnormalities have been reported in 24–25% of live births born with HPE, trisomy 13 being the most common [11]. Reported chromosomal abnormalities associated with HPE include trisomy 13, trisomy 18, triploidy, del(2p), dup(3p), del(7q), del(13q), del(18p), del(21q), and interstitial deletion of 14q13 [10,12–15]. Blaas et al [16] found chromosomal abnormalities in 36.7% (11/30) of fetuses with prenatally detected HPE including trisomy 13 ($n=5$), r(13) ($n=1$), del(13q) ($n=1$), triploidy ($n=1$), partial monosomy 14q ($n=1$), partial monosomy 11p ($n=1$), and t(8;14)(q21.1;q24.1) ($n=1$).

In the present case, digynic triploidy occurred in the fetus of a relatively young woman. Molecular analysis showed that the digynic triploidy was caused by an error in the second meiotic division of oogenesis. Digynic triploidy most often arises from fertilization by a normal haploid sperm with a primary oocyte or with a diploid ovum. The diploid ovum results from an error during meiosis I or II, or from retention of the second polar bodies. Digynic triploidy is characterized by marked asymmetric IUGR, a small placenta without partial mole, and a variety of congenital malformations that may affect almost every organ system. Diandric triploidy is characterized by normal fetal growth or a mild degree of symmetric growth restriction, normal or partial molar placenta, and a range of malformations similar to those of digynic triploidy. In the fetal period, cases with digynic triploidy are far more commonly observed than those with diandric triploidy [17,18]. Ultrasonography is the first choice of prenatal investigation of triploid fetuses, and prenatal magnetic resonance imaging may provide excellent anatomic details and a large field of view, especially under the circumstance of oligohydramnios [19]. This presentation highlights the usefulness of QF-PCR in the rapid diagnosis of digynic triploidy following prenatal sonographic diagnosis of IUGR, relative macrocephaly, and HPE.

Acknowledgments

This work was supported by research grants NSC-96-2314-B-195-008-MY3 and NSC-97-2314-B-195-006-MY3 from the National Science Council, and MMH-E-98004 from Mackay Memorial Hospital, Taipei, Taiwan.

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