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Short Communication

Prenatal diagnosis and molecular cytogenetic characterization of a derivative chromosome der(18;18)(q10;q10)del(18)(q11.1q12.1)del(18)(q22.1q22.3) presenting as apparent isochromosome 18q in a fetus with holoprosencephaly

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Abstract

Objective: To present prenatal diagnosis and molecular cytogenetic characterization of a derivative chromosome der(18;18)(q10;q10)del(18)(q11.1q12.1)del(18)(q22.1q22.3).

Materials, Methods, and Results: A 32-year-old woman was referred for genetic counseling of prenatally detected isochromosome 18q [i(18q)]. She had undergone amniocentesis at 19 gestational weeks because of a trisomy 18 risk of 1/39 derived from abnormally low levels of maternal serum unconjugated estriol, inhibin A, α-fetoprotein, and total β-human chorionic gonadotropin. Amniocentesis revealed a karyotype of 46,XX,i(18)(q10). Parental karyotypes were normal. Prenatal ultrasound showed alobar holoprosencephaly. Repeated amniocentesis was requested and performed at 21 gestational weeks. Array-comparative genomic hybridization analyses revealed a 14-Mb deletion of 18p11.32-p11.21, a 37.8-Mb duplication of 18q12.1-q22.1, and a 6.9-Mb duplication of 18q22.3-q23. Metaphase fluorescence *in situ* hybridization study showed the absence of an 18q12.1-specific probe signal in one arm and the absence of an 18q22.2-specific probe signal in the other arm of the derivative chromosome. Quantitative fluorescent polymerase chain reaction analysis determined a paternal origin of the derivative chromosome. The cytogenetic result was 46,XX,der(18;18)(q10;q10)del(18)(q11.1q12.1)del(18)(q22.1q22.3). The fetus postnatally manifested cebocephaly.

Conclusion: Concomitant monosomy 18p and trisomy 18q can be associated with holoprosencephaly and abnormal maternal serum screening results. Array-comparative genomic hybridization, fluorescence *in situ* hybridization, and quantitative fluorescent polymerase chain reaction are useful in genetic counseling of prenatally detected isochromosomes by providing information on the origin and genetic components of the isochromosome.

Keywords: 18q deletion; der(18;18)(q10;q10); Holoprosencephaly; Isochromosome 18q; Monosomy 18p; Prenatal diagnosis; Trisomy 18q

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Introduction

Isochromosome 18q [i(18q)] is a rare but distinctive syndrome and can be associated with clinical manifestations that closely resemble those of 18p deletion syndrome and trisomy 18. Concomitant occurrence of i(18q) and holoprosencephaly (HPE) is even rare. In a review of 23 patients with a pure isochromosome of the long arm of chromosome 18, Chen et al [1] found that only eight cases (34.8%) had HPE. To date, at least 11 cases of pure i(18q) with HPE have been reported [2–12]. In this article, we report our experience of prenatal diagnosis and molecular cytogenetic analyses of a derivative chromosome der(18;18)(q10;q10)del(18)(q11.1q12.1)del(18)(q22.1q22.3) presenting as an i(18q) in a fetus with alobar HPE and cebocephaly.

Materials, methods, and results

A 32-year-old, gravida 2, para 1 woman was referred for genetic counseling of prenatally detected i(18q) at 21 weeks of gestation. She and her husband were healthy and had an 11-year-old healthy son. During this pregnancy, she had undergone amniocentesis at 19 weeks of gestation because of a trisomy 18 risk of 1/39 derived from abnormal maternal serum screening results of unconjugated estriol = 0.59 multiples of median (MoM); inhibin A = 0.45 MoM; α-fetoprotein = 0.57 MoM; and total β-human chorionic gonadotropin = 0.25 MoM. Amniocentesis revealed a karyotype of 46,XX,i(18)(q10). Parental karyotypes were normal. Prenatal ultrasound at 21 weeks of gestation showed a female fetus with alobar HPE (Fig. 1). Repeated amniocentesis was requested and performed at 21 weeks of gestation. Oligonucleotide-based array-comparative genomic hybridization (aCGH) analyses using CytoChip Oligo Array (BlueGnome, Cambridge, UK) and Human CGH 12 × 135 K Whole-Genome Tiling Array v3.1 (Roche NimbleGen, Madison, WI, USA) were applied in uncultured amniocytes. The aCGH analyses revealed a 14-Mb deletion of 18p11.32-p11.21, a 37.8-Mb duplication of 18q12.1-q22.1, and a 6.9-Mb duplication of 18q22.3-q23 (Figs. 2 and 3). The aCGH result was arr cgh 18p11.32p11.21 (138,992–14,071,857) × 1, 18q12.1q22.1 (26,192,404–64,116,181) × 3, 18q22.3q23 (69,242,924–76,113,787) × 3 according to CytoChip Oligo Array (Fig. 2) [University of California Santa Cruz genome browser on human March 2006 (National Center for Biotechnology Information 36/human genome 18) assembly], and the aCGH result was arr cgh 18p11.32p11.21 (15,636–14,067,730) × 1, 18q12.1q22.1 (26,302,599–64,104,233) × 3, 18q22.3q23 (69,176,955–76,108,540) × 3 according to Human CGH 12 × 135 K Whole-Genome Tiling Array v3.1 (Fig. 3). The genomic imbalance was a deficiency of 18p and a duplication of 18q, including 18q12.1-q22.1 and 18q22.3-q23. Metaphase fluorescence *in situ* hybridization (FISH) study of the cultured amniocytes using an 18q12.1-specific bacterial artificial chromosome clone probe RP11-467D2 (25,363,767–25,551,469; spectrum green) and an 18q22.2-specific bacterial artificial chromosome clone probe RP11-184J20 (65,767,717–65,942,481; spectrum red) showed the absence of one green signal in one chromosome arm of the derivative chromosome and the absence of one red signal in the other chromosome arm of the derivative chromosome (Fig. 4). The result was consistent with different deletions in two chromosome arms on either side of the centromere in the derivative chromosome. Interphase FISH study of the uncultured amniocytes using an 18p subtelomeric probe RP11-324G2 (168,384–340,511; spectrum green) and an 18q subtelomeric probe RP11-154H12 (75,538,734–75,701,257; spectrum red) showed one green signal and three red signals (Fig. 5). The result was consistent

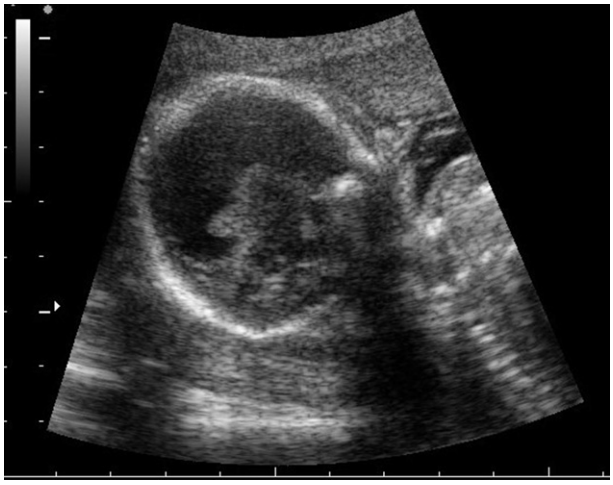

Fig. 1. Prenatal ultrasound at 21 weeks of gestation shows alobar holoprosencephaly.

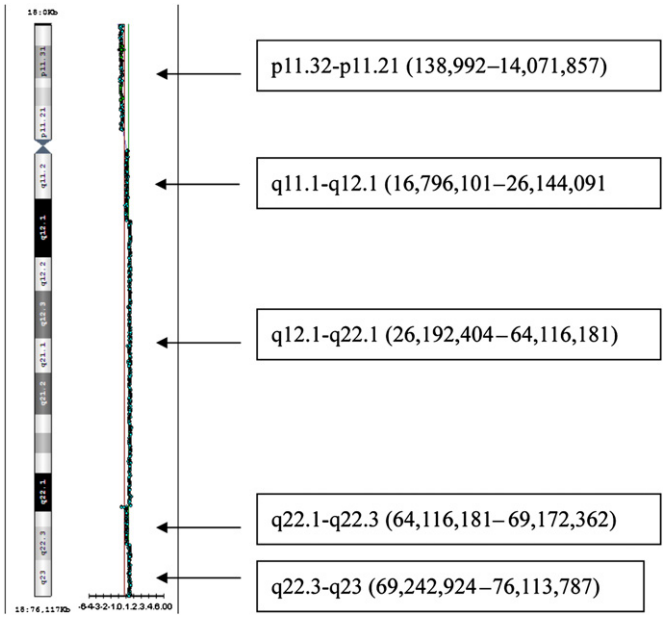

Fig. 2. Oligonucleotide-based array-comparative genomic hybridization analysis using CytoChip Oligo Array (BlueGnome, Cambridge, UK) shows a result of arr cgh 18p11.32p11.21 (138,992–14,071,857) × 1, 18q12.1q22.1 (26,192,404–64,116,181) × 3, 18q22.3q23 (69,242,924–76,113,787) × 3.

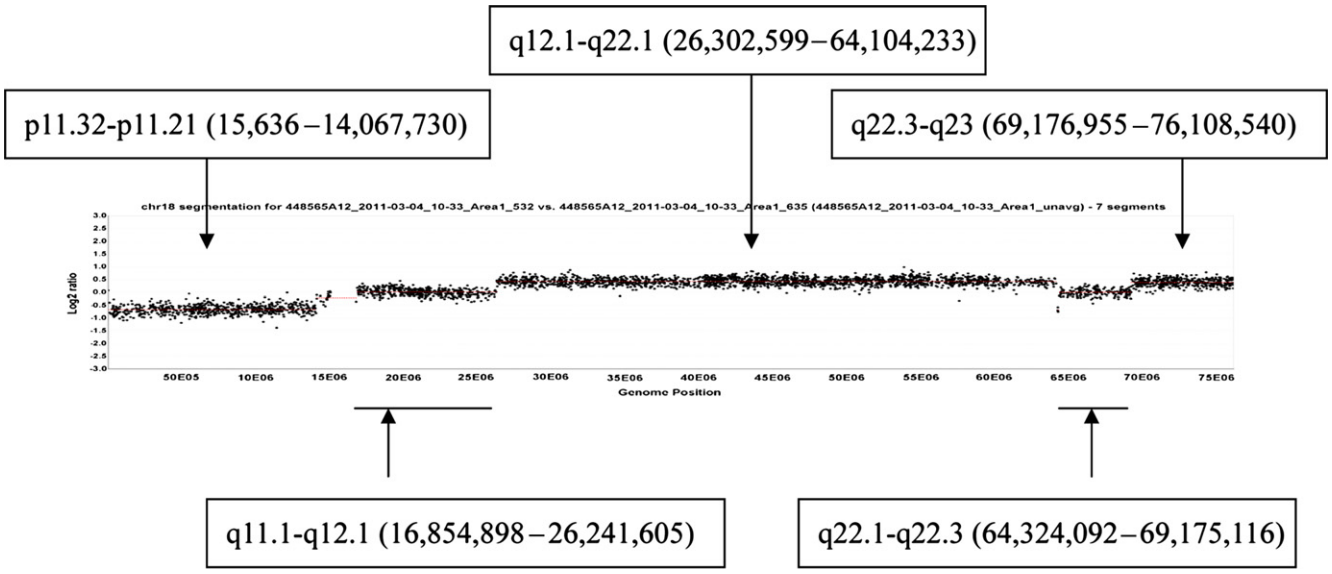

Fig. 3. Oligonucleotide-based array-comparative genomic hybridization aCGH analysis using Human CGH 12 × 135 K Whole-Genome Tiling Array v3.1 (Roche NimbleGen, Madison, WI, USA) shows a result of arr cgh 18p11.32p11.21 (15,636−14,067,730) × 1, 18q12.1q22.1 (26,302,599−64,104,233) × 3, 18q22.3q23 (69,176,955−76,108,540) × 3. The result is consistent with the diagnosis of a 14-Mb deletion of 18p11.32-p11.21, a 37.8-Mb duplication of 18q12.1-q22.1, and a 6.9-Mb duplication of 18q22.3-q23.

with monosomy 18p and trisomy 18q. The derivative chromosome was composed of two long arms of chromosome 18 with a deletion of 18q11.1q12.1 in one long arm and a deletion of 18q22.1q22.3 in the other long arm (Fig. 6). The cytogenetic result, thus, was 46,XX,der(18;18)(q10;q10)del(18)(q11.1q12.1)del(18)(q22.1q22.3). Quantitative fluorescent polymerase chain reaction (QF-PCR) analysis using polymorphic DNA markers revealed a paternal origin of the derivative chromosome (Fig. 7 and Table 1). The parents decided

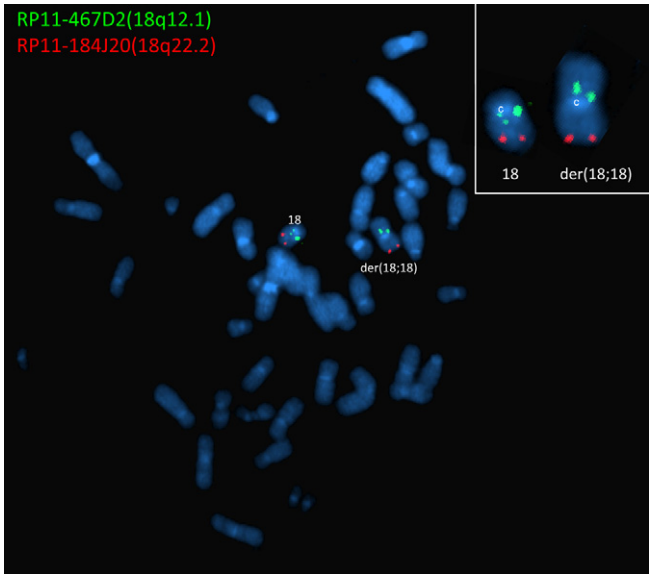

Fig. 4. Metaphase fluorescence *in situ* hybridization study of cultured amniocyte using an 18q12.1-specific bacterial artificial chromosome (BAC) clone probe RP11-467D2 (25,363,767−25,551,469; spectrum green) and an 18q22.2-specific BAC clone probe RP11-184J20 (65,767,717−65,942,481; spectrum red) shows the presence of one green signal and one red signal in the long arm of a chromosome 18, and the absence of one green signal in one chromosome arm of the derivative chromosome and the absence of one red signal in the other chromosome arm of the derivative chromosome. The result is consistent with the diagnosis of different deletions in two chromosome arms on either side of the centromere in the derivative chromosome. The inset shows amplified chromosome 18 and derivative chromosome 18, der(18;18). c = centromere.

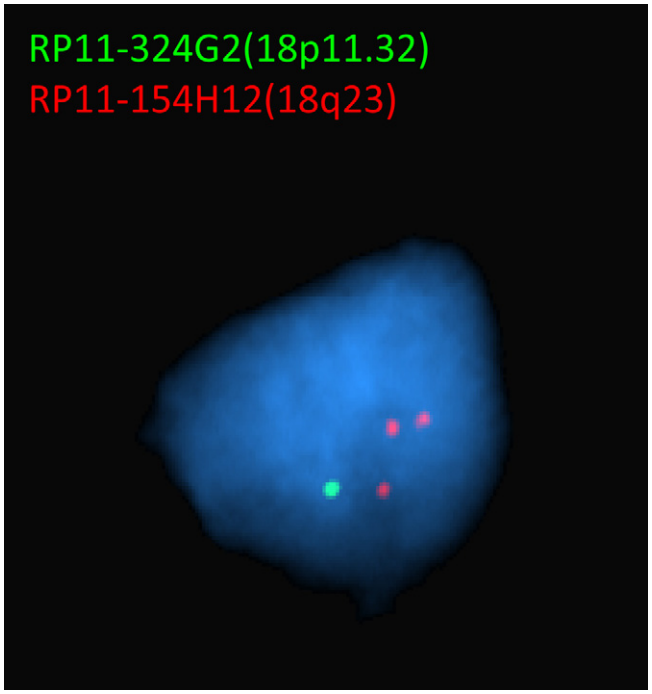

Fig. 5. Interphase fluorescence *in situ* hybridization study of the uncultured amniocyte using an 18p subtelomeric probe RP11-324G2 (168,384−340,511; spectrum green) and an 18q subtelomeric probe RP11-154H12 (75,538,734−75,701,257; spectrum red) shows one green signal and three red signals. The result is consistent with the diagnosis of monosomy 18p and trisomy 18q.

Fig. 6. Partial karyotype of the fetus shows a chromosome 18 and a derivative chromosome der(18;18)(q10;q10)del(18)(q11.1q12.1)del(18)(q22.1q22.3).

to terminate the pregnancy, and a 432-g fetus was delivered. The fetus postnatally manifested hypotelorism, cebocephaly, a single nostril, a low nasal bridge, a round face, epicanthic folds, low-set ears, micrognathia, and a short neck (Fig. 8).

Discussion

The present case had two unidentical chromosome arms on either side of the centromere in the derivative chromosome. The present case most likely resulted from meiosis I recombination errors causing a deletion of 18q11.1q12.1 in one chromosome arm and a deletion of 18q22.1q23 in the other chromosome arm and a meiosis II centromere misdivision (Fig. 9). An isochromosome represents a mirror-image chromosome with two identical arms on either side of the centromere. It can be caused by a postzygotic centromere misdivision resulting in isochromosomes with complete isodisomy or by a meiosis II centromere misdivision resulting in isochromosomes with partial isodisomy [13] or by U-type reunion between sister chromatids [14]. In case of an isochromosome caused by a postzygotic centromere misdivision, the two chromosome arms are identical, and there is no heterozygosity for polymorphic DNA markers. However, in case of an isochromosome caused by a meiosis II centromere misdivision, recombination can be expected to result in heterozygosity for polymorphic DNA markers, although an isochromosome is defined as a derivative chromosome with cytogenetically identical chromosome arms [13,15–17].

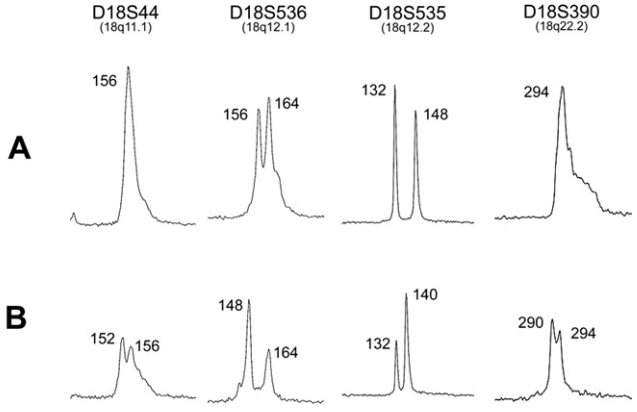

Fig. 7. Representative electrophoretograms of quantitative fluorescent polymerase chain reaction assays at short tandem repeat markers specific for chromosome arm 18q using fetal and maternal DNAs. With the markers D18S536 (18q12.1) and D18S535 (18q12.2), two alleles of 148 bp:164 bp and of 140 bp:132 bp, respectively, in the fetus, have a ratio of 2:1 (paternal:maternal), indicating paternal isodisomy in the derivative chromosome. With the markers D18S44 (18q11.1) and D18S390 (18q22.2), two alleles of 152 bp:156 bp and of 290 bp:294 bp, respectively, in the fetus, have a ratio of 1:1 (paternal:maternal), indicating heterozygous deletions in the different chromosome arms of the derivative chromosome. (A) Mother and (B) fetus.

Table 1
Genotypic information of the fetus and mother at short tandem repeat markers specific for chromosome arm 18q obtained by quantitative fluorescent polymerase chain reaction assays[a]

Markers	Locus	Position	Mother (bp)	Fetus (bp)
D18S44	18q11.1	16,999,595–16,999,740	156,156	152,156
D18S536	18q12.1	28,447,678–28,447,822	156,164	148,148,164
D18S1369	18q12.1	29,993,750–29,993,920	168,168	168,176,176
D18S535	18q12.2	35,011,115–35,011,256	132,148	132,140,140
D18S390	18q22.2	66,095,200–66,095,490	294,294	290,294
D18S380	18q22.3	69,862,961–69,863,107	150,154	150,158,158

[a] Alleles (basepair sizes) are listed below each individual.
bp = basepair.

The present case was associated with monosomy 18p, partial trisomy 18q, and HPE. The 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 [18]. Deletions involving chromosome 18p are among the most common aneuploidies detected in patients with HPE [19]. An HPE critical region on 18p11.3 has been defined as *HPE4* [Online Mendelian Inheritance in Man (OMIM) 142946] [19], and the responsible gene is *TGIF* (OMIM 602630) [20]. The 18p deletion syndrome (OMIM 146390) includes clinical features of growth and mental retardation, hypotonia,

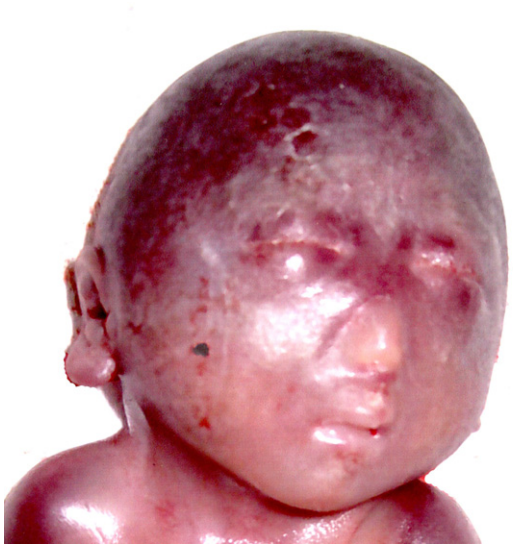

Fig. 8. Cebocephaly and facial dysmorphism of the fetus at birth.

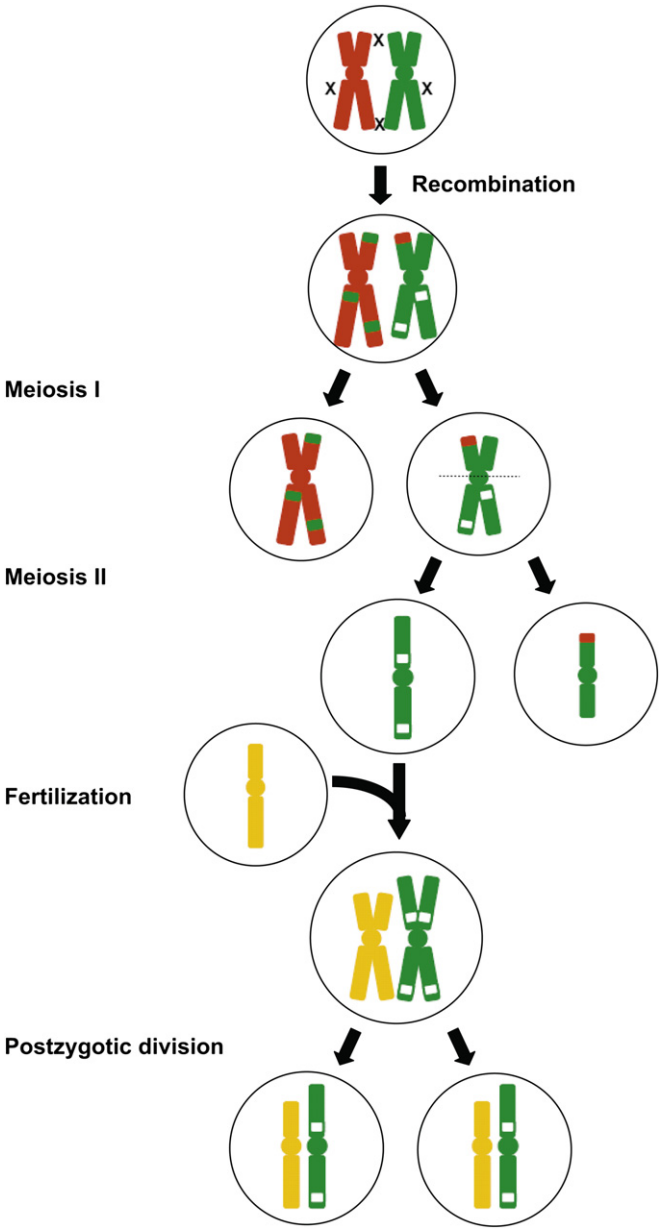

Fig. 9. Schematic representation of the most probable mechanism of formation of a cell line with der(18;18)(q10;q10)del(18)(q11.1q12.1)del(18)(q22.1q22.3).

epicanthic folds, ptosis, a low nasal bridge, a round face, micrognathia, a short neck, abnormal ears, small hands and feet, clinodactyly of the fifth finger, cardiac defects, abnormal genitalia, and cerebral malformations [21]. Only about 10% of cases with 18p deletion have HPE [22]. The low incidence of HPE in cases with *TGIF* haploinsufficiency and 18p deletion is because of autosomal dominant inheritance pattern with low penetrance [20]. HPE can also be associated with trisomy 18. Snijders et al [23] found HPE in 3% of fetuses with trisomy 18 (n =;137), and Chen [24] found HPE in 5.62% of fetuses with trisomy 18 ($n = 89$).

Prenatal diagnosis of i(18q) associated with HPE is very rare. To our knowledge, only seven cases have been reported [3,4,6,9–12]. Shyu et al [3] first reported prenatal diagnosis of 46,XY,i(18q) at 29 weeks of gestation in a 30-year-old woman with abnormal ultrasound findings of polyhydramnios and HPE. The fetus postnatally presented with cebocephaly, aplasia of thumbs, and syndactyly of fingers. Nyberg et al [4] reported prenatal diagnosis of HPE at 29 weeks of gestation in a fetus with cyclopia, radial aplasia, polyhydramnios, esophageal atresia, tracheoesophageal fistula, and a karyotype of 46,XY,i(18q). Wurster-Hill et al [6] reported prenatal diagnosis of 46,XY,i(18q) at 19 weeks of gestation in a 31-year-old woman with abnormal ultrasound findings of HPE, flexion contractures of the wrists, right pleural effusion, absent stomach bubble, and mild polyhydramnios. Levy-Mozziconacci et al [9] reported prenatal diagnosis of 46,XX,i(18q) at 22 weeks of gestation in a 27-year-old woman with abnormal ultrasound findings of intrauterine growth restriction, brain anomalies, and omphalocele. The fetus postnatally manifested cyclopia. De Pater et al [10] reported prenatal diagnosis of i(18q) at 14 weeks of gestation in a 27-year-old woman with sonographic findings of HPE and contiguous orbits. Leonard et al [11] reported prenatal diagnosis of 46,XX,rec(18) dup(18q)inv(18)(p11.31q11.2)mat at 18 weeks of gestation in a 29-year-old woman with sonographic findings of HPE. The fetus postnatally manifested a proboscis. Sepulveda [12] reported first-trimester diagnosis of alobar HPE associated with 46,XX,i(18q) in a fetus with an increased nuchal translucency, a central facial cleft, and a single umbilical artery in a 39-year-old woman. The present case shows that abnormal maternal serum screening results can be a prenatal finding in addition to alobar HPE on prenatal ultrasound in fetuses with concomitant monosomy 18p and trisomy 18q.

With the advent of aCGH, FISH, and QF-PCR, the nature of an aberrant chromosome presenting as an isochromosome can be well characterized to be a derivative chromosome with multiple deletions and duplications. We conclude that aCGH, FISH, and QF-PCR are helpful in genetic counseling of prenatally detected isochromosomes by providing information on the origin and genetic components of the isochromosome.

Acknowledgments

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References

[1] Chen C-P, Chern S-R, Lee C-C, Town D-D. Isochromosome 18q in a fetus with congenital megacystis, intrauterine growth retardation and cloacal dysgenesis sequence. Prenat Diagn 1998;18:1068–74.
[2] Froster-Iskenius U, Coerdt W, Rehder H, Schwinger E. Isochromosome 18q with karyotype 46, XX, i(18q). Cytogenetics and pathology. Clin Genet 1984;26:549–54.
[3] Shyu S-K, Yang M-L, Ng H-T, Hsu I, Wang J-S, Ho D-M. Prenatal diagnosis of a congenital anomaly of isochromosome 18q—a case report. Clin Med J 1988;41:383–8.

[4] Nyberg DA, Fitzsimmons J, Mack LA, Hughes M, Pretorius DH, Hickok D, et al. Chromosomal abnormalities in fetuses with omphalocele: significance of omphalocele contents. J Ultrasound Med 1989;8:299−308.
[5] Spinner NB, Eunpu DL, Austria JR, Mamunes P. Holoprosencephaly in a newborn girl with 46, XX, i(18q). Holoprosencephaly in a newborn girl with 46, XX, i(18q). Am J Med Genet 1991;39:11−2.
[6] Wurster-Hill DH, Marin-Padilla JM, Moeschler JB, Park JP, McDermet M. Trisomy 18 and 18p- features in a case of isochromosome 18q [46, XY, i(18q)]: prenatal diagnosis and autopsy report. Clin Genet 1991;39:142−8.
[7] van Essen AJ, Schoots CJF, van Lingen RA, Mourits MJE, Tuerlings JHAM, Leegte B. Isochromosome 18q in a girl with holoprosencephaly, DiGeorge anomaly, and streak ovaries. Am J Med Genet 1993;47:85−8.
[8] Chow BHC, Loh SF, Yan YL, Ang HK, Yeo GSH. Holoprosencephaly and chromosomal anomalies. Singapore Med J 1996;37:394−7.
[9] Levy-Mozziconacci A, Piquet C, Scheiner C, Adrai J, Potier A, Pelissier MC, et al. i(18q) in amniotic and fetal cells with a normal karyotype in direct chorionic villus sampling: cytogenetics and pathology. Prenat Diagn 1996;16:1156−9.
[10] de Pater JM, Scheres JM, Brons J. Abnormal chromosome 18 in prenatal diagnosis with holoprosencephaly. Prenat Diagn 1997;17:887−8.
[11] Leonard NJ, Tomkins DJ, Demianczuk N. Prenatal diagnosis of holoprosencephaly (HPE) in a fetus with a recombinant (18)dup(18q) inv(18)(p11.31q11.2)mat. Prenat Diagn 2000;20:947−9.
[12] Sepulveda W. Monosomy 18p presenting with holoprosencephaly and increased nuchal translucency in the first trimester: report of 2 cases. J Ultrasound Med 2009;28:1077−80.
[13] van den Berg C, Pijpers L, Halley DJ, Opstal DV, Los FJ. Prenatal detection of trisomy 18 caused by isochromosome 18p and 18q formation. Am J Med Genet 1999;86:151−5.
[14] de la Chapelle A. How do human isochromosomes arise? Cancer Genet Cytogenet 1982;5:173−9.
[15] Bugge M, Blennow E, Friedrich U, Petersen MB, Pedutour F, Tsezou A, et al. Tetrasomy 18p *de novo*: parental origin and different mechanisms of formation. Eur J Hum Genet 1996;4:160−7.
[16] Eggermann T, Engels H, Apacik C, Moskalonek B, Müller-Navia J, Schwanitz G, et al. Tetrasomy 18p caused by paternal meiotic nondisjunction. Eur J Hum Genet 1997;5:175−7.
[17] Kotzot D, Bundscherer G, Bernasconi F, Brecevic L, Lurie IW, Basaran S, et al. Isochromosome 18p results from maternal meiosis II nondisjunction. Eur J Hum Genet 1996;4:168−74.
[18] Chen C-P. Prenatal sonographic features of fetuses in trisomy 13 pregnancies (II). Taiwan J Obstet Gynecol 2009;48:218−24.
[19] Overhauser J, Mitchell HF, Zackai EH, Tick DB, Rojas K, Muenke M. Physical mapping of the holoprosencephaly critical region in 18p11.3. Am J Hum Genet 1995;57:1080−5.
[20] Gripp KW, Edwards MC, Mowat D, Meinecke P, Richieri-Costa A, Zackai EH, et al. Mutations in the transcription factor TGIF in holoprosencephaly. Am J Hum Genet 1998;63:A32.
[21] Chen C-P, Kuo Y-T, Lin S-P, Su Y-N, Chen Y-J, Hsueh R-Y, et al. Mosaic ring chromosome 18, ring chromosome 18 duplication/deletion and disomy 18: perinatal findings and molecular cytogenetic characterization by fluorescence in situ hybridization and array comparative genomic hybridization. Taiwan J Obstet Gynecol 2010;49:327−32.
[22] Schinzel A. In: Schinzel A, editor. Catalogue of unbalanced chromosome aberrations in man. 2nd ed. Berlin, Germany: Water de Gruter, GmbH & Co; 2001. p. 717−22.
[23] Snijders RJM, Farrias M, von Kaisenberg C, Nicolaides KH. Fetal abnormalities. In: Snijders RJM, Nicolaides KH, editors. Ultrasound markers for fetal chromosomal defects. New York: Parthenon Publishing Group; 1996. p. 1−62.
[24] Chen C-P. Central nervous system anomalies associated with fetal trisomy 18. Prenat Diagn 2005;25:419−21.