Available online at www.sciencedirect.com

SciVerse ScienceDirect

Taiwanese Journal of Obstetrics & Gynecology 50 (2011) 492–498

www.tjog-online.com

Short Communication

A *de novo* duplication of chromosome 21q22.11 → qter associated with Down syndrome: Prenatal diagnosis, molecular cytogenetic characterization and fetal ultrasound findings

Chih-Ping Chen [a,b,c,d,e,f,1,*], Hsu-Kuang Huang [g,1], Pei-Ying Ling [g], Yi-Ning Su [h], Ming Chen [i,j,k,l], Fuu-Jen Tsai [d,m,n], Pei-Chen Wu [a], Schu-Rern Chern [b], Yu-Ting Chen [b], Chen-Chi Lee [a], Wayseen Wang [b,o]

[a] *Department of Obstetrics and Gynecology, Mackay Memorial Hospital, Taipei, Taiwan*
[b] *Department of Medical Research, Mackay Memorial Hospital, Taipei, Taiwan*
[c] *Department of Biotechnology, Asia University, Taichung, Taiwan*
[d] *School of Chinese Medicine, College of Chinese Medicine, China Medical University, Taichung, Taiwan*
[e] *Institute of Clinical and Community Health Nursing, National Yang-Ming University, Taipei, Taiwan*
[f] *Department of Obstetrics and Gynecology, School of Medicine, National Yang-Ming University, Taipei, Taiwan*
[g] *Department of Obstetrics and Gynecology, Taiwan Adventist Hospital, Taipei, Taiwan*
[h] *Department of Medical Genetics, National Taiwan University Hospital, Taipei, Taiwan*
[i] *Department of Genomic Medicine, Changhua Christian Hospital, Changhua, Taiwan*
[j] *Department of Medical Research, Changhua Christian Hospital, Changhua, Taiwan*
[k] *Department of Obstetrics and Gynecology, Changhua Christian Hospital, Changhua, Taiwan*
[l] *Department of Obstetrics and Gynecology, College of Medicine, National Taiwan University, Taipei, Taiwan*
[m] *Department of Medical Research, China Medical University Hospital, Taichung, Taiwan*
[n] *Department of Medical Genetics, China Medical University Hospital, Taichung, Taiwan*
[o] *Department of Bioengineering, Tatung University, Taipei, Taiwan*

Accepted 30 November 2010

Abstract

Objectives: To present prenatal diagnosis and molecular cytogenetic characterization of *de novo* partial partial trisomy 21q (21q22.11 → qter) associated with clinodactyly and hypoplastic midphalanx of the fifth fingers, midface hypoplasia, and an intracardiac echogenic focus on prenatal ultrasound.
Materials, Methods, and Results: A 34-year-old gravida 2, para 1 woman underwent amniocentesis at 20 weeks of gestation because of fetal structural abnormalities on prenatal ultrasound. A level II ultrasound at 20 weeks of gestation showed polyhydramnios, clinodactyly and hypoplastic midphalanx of the fifth fingers, midface hypoplasia, and an intracardiac echogenic focus. Amniocentesis revealed an aberrant derivative chromosome 9, or der(9). Parental karyotypes were normal. Spectral karyotyping (SKY) and fluorescence *in situ* hybridization (FISH) analyses revealed that the der(9) contained a segment of chromosome 21 distal to chromosome 9q, and FISH analysis additionally showed that the distal subtelomeric region of 9q was not deleted. Array comparative genomic hybridization (aCGH) demonstrated a 14.8-Mb duplication of distal 21q encompassing the Down syndrome critical region (DSCR) but no genomic imbalance in the distal euchromatic region of chromosome 9. The karyotype was 46,XX,der(9)t(9;21) (q34.3;q22.11)dn. Polymorphic DNA marker analysis revealed the maternal origin of the aberrant chromosome. The pregnancy was subsequently terminated. A malformed female fetus was delivered with a characteristic phenotype of Down syndrome.
Conclusion: SKY, FISH and aCGH are useful in prenatal investigation of the nature of a *de novo* aberrant derivative chromosome. Partial trisomy 21q encompassing the DSCR may present characteristic Down syndrome features on prenatal ultrasound.

Keywords: 21q22; 21q duplication; Down syndrome; Partial trisomy 21q; Prenatal diagnosis; Ultrasound

* Corresponding author. Department of Obstetrics and Gynecology, Mackay Memorial Hospital, 92 Section 2, Chung-Shan North Road, Taipei, Taiwan.
E-mail address: cpc_mmh@yahoo.com (C.-P. Chen).
[1] Chih-Ping Chen and Hsu-Kuang Huang contributed equally to this work.

1028-4559/$ - see front matter
doi:10.1016/j.tjog.2011.10.016

Introduction

Prenatal diagnosis of a *de novo* unbalanced translocation may require molecular cytogenetic technologies, such as fluorescence *in situ* hybridization (FISH), spectral karyotyping (SKY) and array-based comparative genomic hybridization (aCGH) to identify the nature of the *de novo* derivative chromosome. We previously reported the usefulness of FISH, SKY and aCGH in the identification of a *de novo* derivative chromosome derived from an unbalanced translocation [1,2].

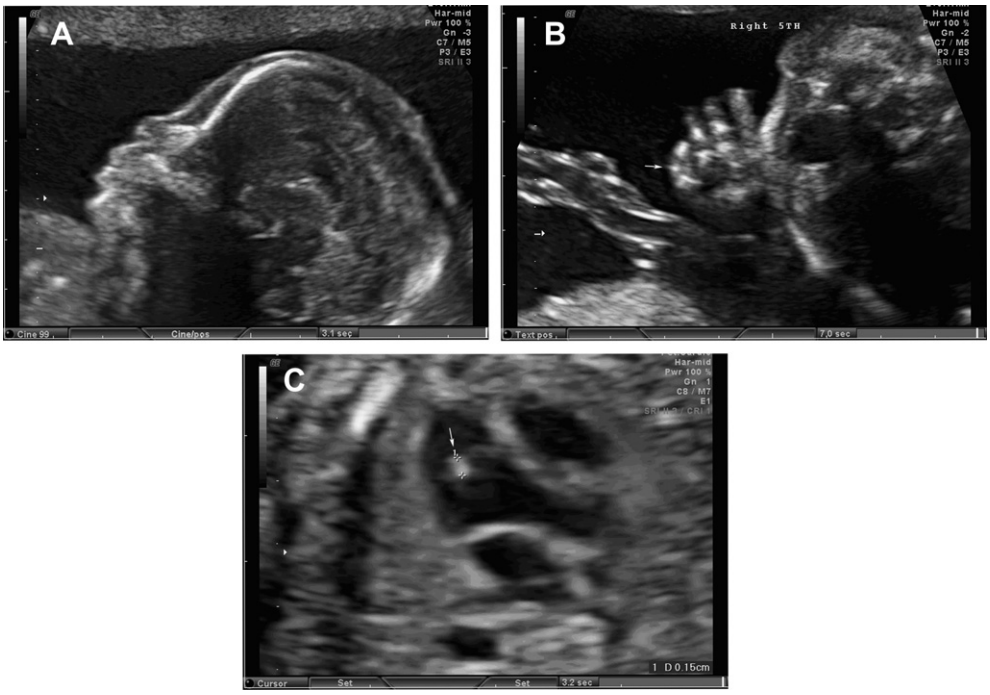

Fig. 1. Prenatal ultrasound at 20 weeks of gestation shows (A) midface hypoplasia, (B) clinodactyly of the fifth finger with hypoplasia of midphalanx (arrow) and (C) an intracardiac echogenic focus (arrow).

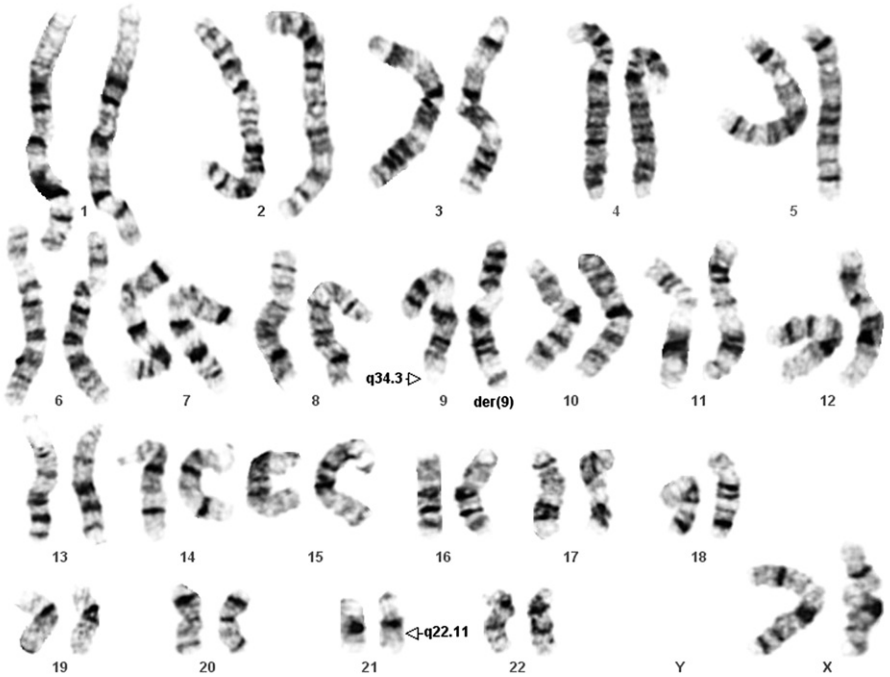

Fig. 2. G-banded karyotype shows a derivative chromosome 9, or der(9). The proband's karyotype is 46,XX,der(9)t(9;21)(q34.3;q22.11)dn.

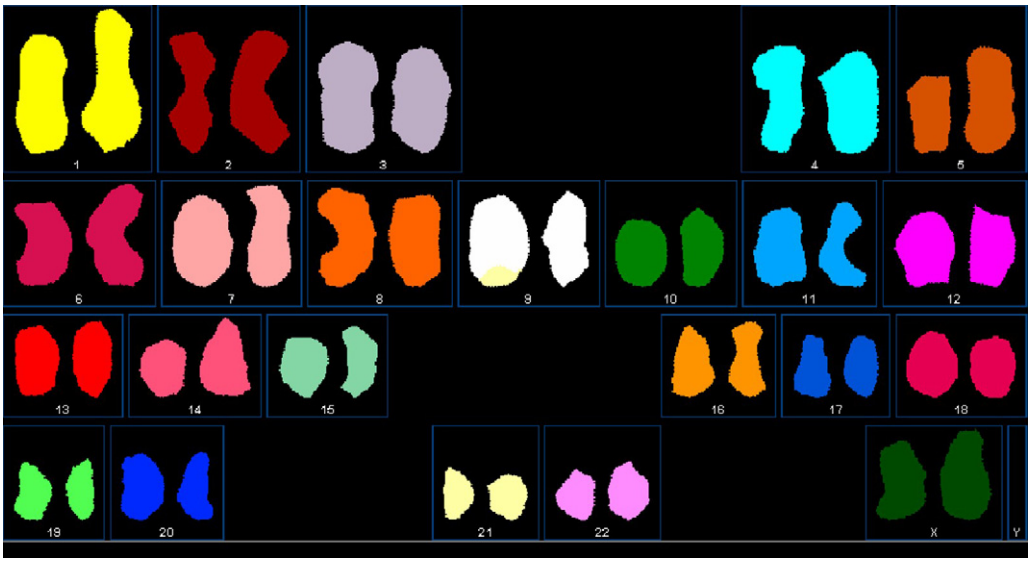

Fig. 3. Spectral karyotyping (SKY) using 24-color SKY probes shows a der(9) derived from a translocation between chromosomes 9 and 21.

Here, we additionally report prenatal diagnosis and molecular cytogenetic characterization of *de novo* partial trisomy 21q (21q22.11 → qter) derived from an unbalanced translocation associated with some of features of Down syndrome, such as clinodactyly and hypoplastic midphalanx of the fifth fingers, midface hypoplasia and an intracardiac echogenic focus on prenatal ultrasound.

Materials, methods and results

A 34-year-old gravida 2, para 1 woman was suggested to undergo amniocentesis at 18 weeks of gestation because of advanced maternal age, but she declined. Her husband was aged 34 years. She had a 2½-year-old healthy daughter. A level II ultrasound at 20 weeks of gestation showed polyhydramnios and some of the features of Down syndrome, such as clinodactyly and hypoplastic midphalanx of the fifth fingers, midface hypoplasia, and an intracardiac echogenic focus (Fig. 1). The woman decided to undergo amniocentesis. Amniocentesis revealed an aberrant derivative chromosome 9, or der(9) (Fig. 2). Chromosome preparations of the blood lymphocytes from the parents revealed normal karyotypes. The derivative chromosome was characterized by SKY using 24-color SKY probes (Applied Spectral Imaging, Carlsbad, CA, USA) and by FISH using DNA probe mixtures containing the 9q and 21q subtelomeric probes. The SKY analysis revealed that the der(9) contained a segment of chromosome 21 in the distal end of the long arm of a chromosome 9 (Fig. 3). The FISH analysis showed that the chromosome 21 segment in the distal end of the long arm of der(9) was of 21q in origin, and the distal subtelomeric region of 9q was not deleted (Fig. 4). Bacterial artificial chromosome (BAC)-based aCGH [CMDX BAC-based aCGH CA3000 chips (CMDX, Irvine, CA, USA)] demonstrated partial trisomy 21q [arr cgh 21q22.11q22.3 (RP11-367F15 → RP11-100I21) × 3] (Fig. 5). Oligonucleotide-based aCGH [Oligo HD Scan (CMDX, Irvine, CA, USA)] further demonstrated a 14.8-Mb duplication of distal 21q [arr cgh 21q22.11q22.3 (32,110,552–46,944,323)×3] (NCBI build 36, March 2006) (Fig. 6). Whole genome analysis by aCGH revealed no genomic imbalance in the distal euchromatic region of chromosome 9 (Fig. 6). The karyotype was 46,XX,der(9)t(9;21)(q34.3;q22.11)dn (Fig. 2). Polymorphic DNA marker analysis revealed the maternal origin of the aberrant chromosome. The parents opted to terminate the

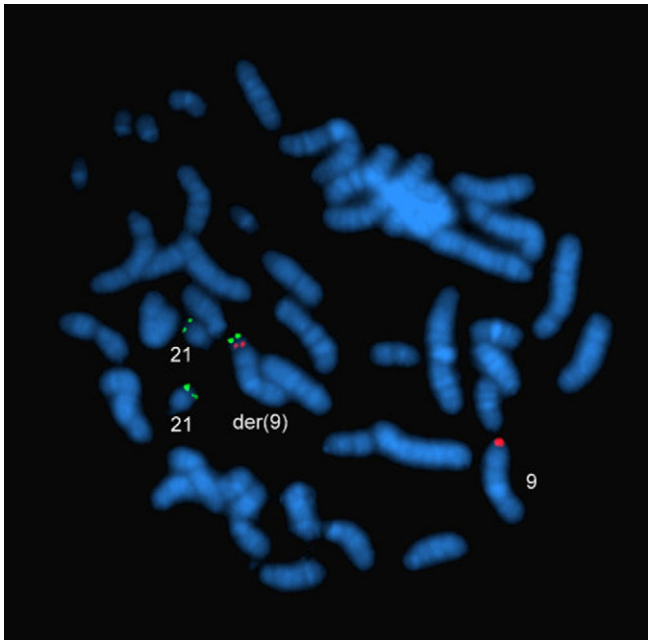

Fig. 4. Fluorescence *in situ* hybridization analysis using the 9q subtelomeric bacterial artificial chromosome (BAC) clone probe RP11-417A4 (TxR-labeled; red signal) (139,523,141–139,716,008 bp) (Build 36) and the 21q subtelomeric BAC clone probe RP11-345F15 (FITC-labeled; green signal) (45,593,807–45,808,641 bp) shows that the der(9) contains one red signal and one green signal, and the green signal is distal to the red signal.

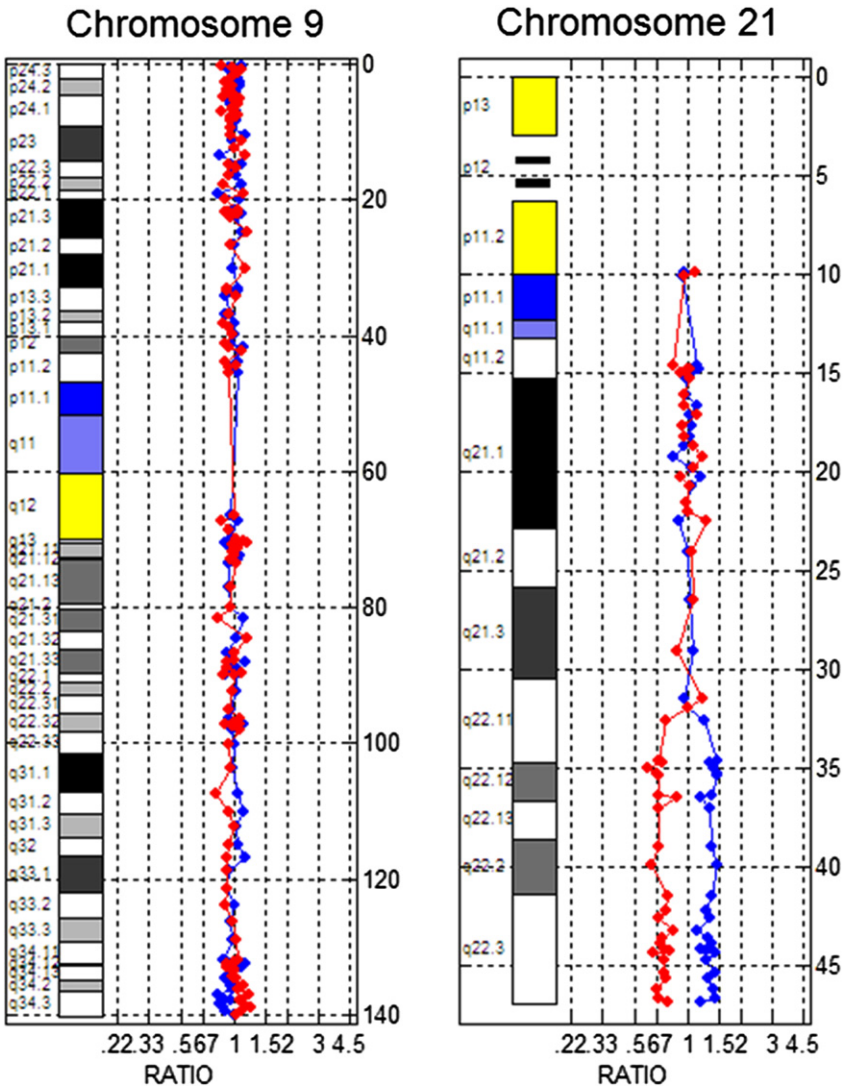

Fig. 5. BAC based-array comparative genomic hybridization (aCGH) shows a distal 21q duplication [arr cgh 21q22.11q22.3 (RP11-367F15 → RP11-1000I21)×3], but no genomic imbalance in the distal euchromatic region of chromosome 9q.

pregnancy. A malformed female fetus was delivered with a flat facial profile, hypertelorism, low-set ears, a depressed nasal bridge, clinodactyly, hypoplastic midphalanx of the fifth fingers, brachycephaly and epicanthic folds (Fig. 7).

Discussion

We have presented prenatal sonographic and molecular cytogenetic characterization of a second-trimester fetus with some clinical features of Down syndrome resulting from a pure partial trisomy 21q (21q22.11 → qter) due to a *de novo* unbalanced translocation. Down syndrome with an unbalanced translocation involving a duplication of the Down syndrome critical region (DSCR) is very rare and accounts for less than 1% of the cases with Down syndrome [3]. DSCR is the critical region on chromosome 21 of which the duplication will be sufficient to cause the Down syndrome phenotype. DSCR has been narrowed down to 21q22 with an approximate length of 5.4 Mb within 21q22.1-q22.3 [4,5]. Ronan et al. [6] additionally reported a 4.3-Mb region within 21q22.13-q22.2 with typical facial features of Down syndrome but no congenital heart defects. Recently, Eggermann et al. [7] narrowed down the DSCR in 21q22 with a 0.46-Mb duplication in 21q22 affecting the *KCNE1* and *DSCR1/RCAN1* genes.

To date, at least 38 cases of Down syndrome with pure partial trisomy 21 have been reported [3,6–23]. Our case manifested some of the clinical features of Down syndrome similar to the cases reported by Nadal et al. [20] and Vaglio et al. [23]. Nadal et al. [20] reported a 2-year-11-month-old boy with pure partial trisomy 21q (21q22.1 → qter) due to an intrachromosomal duplication, short stature, brachycephaly, a flat face, oblique palpebral fissures, epicanthic folds, a flat nasal bridge, a high palate, malformed and low-set ears, short and broad hands with clinodactyly of the fifth fingers, moderate mental retardation and delayed psychomotor development. Vaglio et al. [23] reported a 4-year-10-month-old girl with pure partial trisomy 21q (21q22.11 → qter) due to a *de novo* intrachromosomal duplication, a flat broad face,

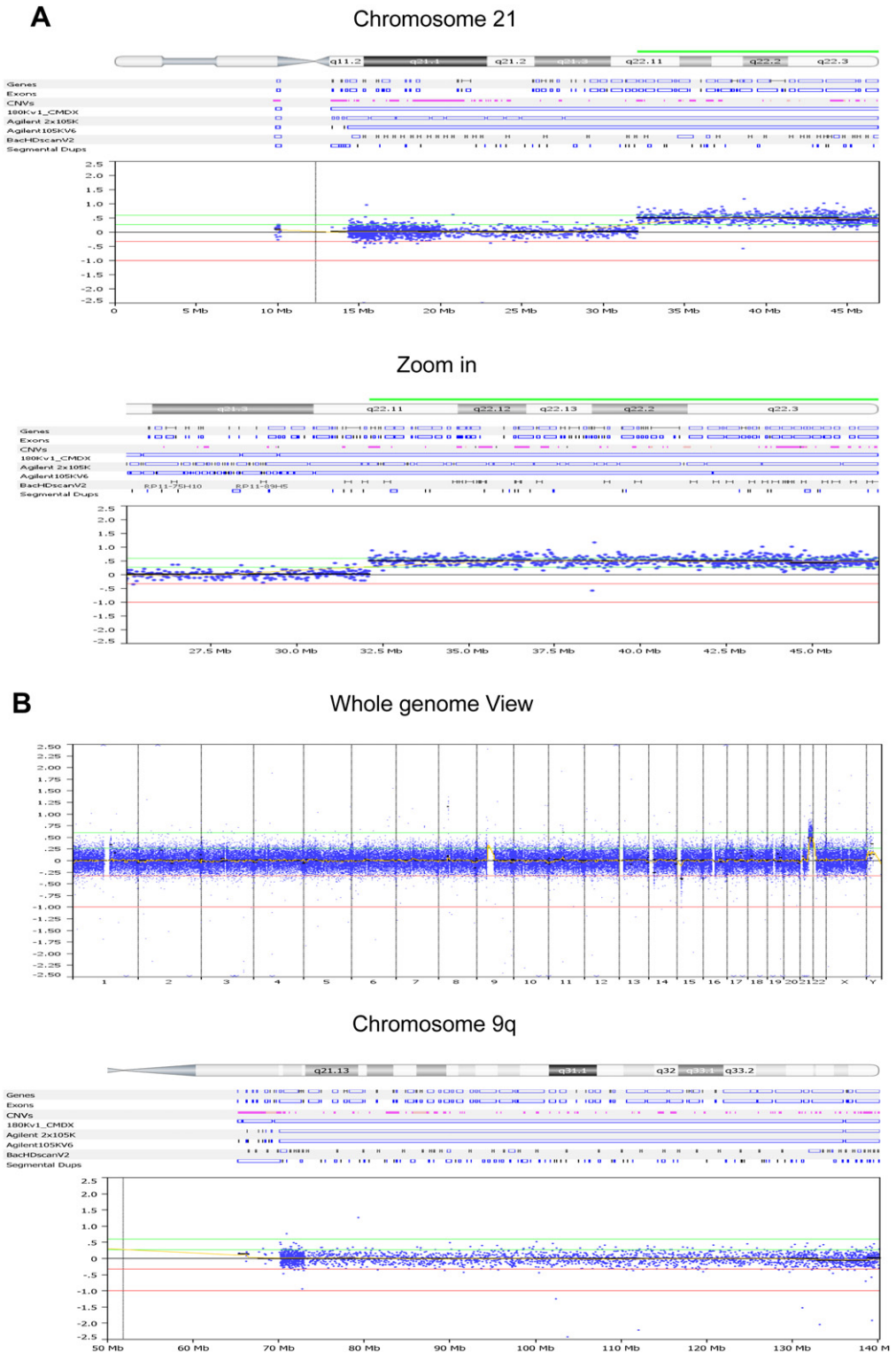

Fig. 6. Oligonucleotide-based aCGH shows (A) a 14.8-Mb distal 21q duplication [arr cgh 21q22.11q22.3 (32,110,552–46,944,323)×3], and (B) no genomic imbalance in the distal euchromatic region of chromosome 9q.

brachycephaly, a flat nasal bridge, upwardly slanted palpebral fissures, epicanthic folds, blepharitis, brushfield spots, strabismus, a wide mouth with downturned corners, a prominent low lip, a narrow tongue and a short palate, but the diagnostic criteria for mental retardation were not fulfilled.

The present case manifested clinodactyly, midface hypoplasia, an intracardiac echogenic focus and hypoplastic midphalanx of the fifth fingers on prenatal ultrasound. Prenatal diagnosis of pure partial trisomy 21q associated with Down syndrome is very rare. Lee et al. [22] reported prenatal diagnosis

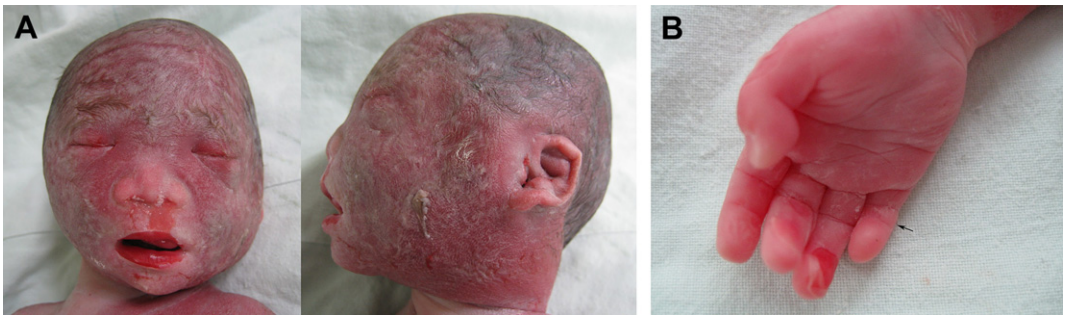

Fig. 7. (A) Craniofacial profile and (B) the abnormal fifth finger with hypoplastic midphalanx (arrow) at birth.

of pure trisomy 21q (21q13 → q22.2) due to an unbalanced cryptic insertion (4;21)(q21;q22.1q22.3) inherited from the carrier father. The mother was aged 32 years. The fetus had a karyotype of 46,XX,der(4)ins(4;21)(q21;q22.13q22.2)pat. The abnormal prenatal findings included a maternal serum screening Down syndrome risk of 1:17 and a thick nuchal fold. The fetus was delivered with clinical features of Down syndrome. Ronan et al. [6] reported prenatal diagnosis of bilateral septated cystic hygroma and hydrops in a fetus with pure trisomy 21q (21q22.13 → q22.2) due to a 4.3-Mb intrachromosomal duplication of 21q22.13-q22.2 inherited from the mother who had the facial gestalt of Down syndrome. Chorionic villus sampling (CVS) at 12 weeks of gestation failed to identify the subtle chromosome abnormalities. Postnatal reexamination of the CVS samples using FISH showed a microduplication of 21q22.13-q22.2 on one chromosome 21. The maternal DNA study using aCGH revealed a 4.3-Mb gain of one copy of the same region.

The present case had a duplication of 21q22.11-qter encompassing some genes associated with the pathophysiology of Down syndrome such as *DSCR1* (OMIM 602917), *DSCR2* (OMIM 605296), *DSCR3* (OMIM 605298), *DSCR4* (OMIM 604829), *DSCR6* (OMIM 609892), *DSCR8* or *MMA1* (OMIM 613396), *DSCR10*, *SIM2* (OMIM 600892) and *DYRK1A* (OMIM 600855). DSCR1 or RCAN1 (regulator of calcineurin 1) (gene map at 21q22.11-q22.2) protein is overexpressed in the brain of the fetuses with Down syndrome. DSCR1 protein interacts with calcineurin A, and overexpression of DSCR1 will inhibit calcineurin-dependent gene transcription through the inhibition of NFAT (OMIM 600489) signaling pathway [24]. Ermak et al. [25] suggested that overexpression of DSCR1 will inhibit calcineurin activity and causes accumulation of hyperphosphorylated tau protein and production of neurofibrillary tangles causing Alzheimer's disease. DYRK1A (dual-specificity tyrosine phosphorylation-regulated kinase 1A) (gene map at 21q22.1) has been suggested to be involved in the abnormal neurogenesis found in Down syndrome [26]. Arron et al. [5] suggested that in Down syndrome, with a 1.5-fold increase in the gene dosage within DSCR, *DSCR1* and *DYRK1A* will cooperatively destabilize the NFAT genetic regulatory circuit, leading to NFAT dysregulation and abnormal phenotypes in Down syndrome. *SIM2* (homolog of Drosophila single-minded 2) gene plays an important role in Drosophila development and encodes a basic helix-loop-helix protein that is a transcription factor in the midline development of CNS. Chen et al. [27] suggested that human *SIM* gene is a candidate for facial and skull dysmorphology, brain abnormalities and mental retardation of Down syndrome. In the mouse model, overexpression of *Sim2* gene contributes to the pathogenesis of Down syndrome [28,29].

In conclusion, aCGH is useful for rapid identification of the genomic imbalance associated with a *de novo* derivative chromosome, and SKY, FISH and aCGH are useful in prenatal investigation of the nature of a *de novo* aberrant derivative chromosome. Partial trisomy 21q encompassing the DSCR may present characteristic Down syndrome features on prenatal ultrasound.

Acknowledgments

This work was supported by research grants NSC-96-2314-B-195-008-MY3, NSC-97-2314-B-195-006-MY3 and NSC-99-2628-B-195-001-MY3 from the National Science Council, and MMH-E-99004 from Mackay Memorial Hospital, Taipei, Taiwan.

References

[1] Chen C-P, Chen M, Su Y-N, Tsai F-J, Chern S-R, Hsu C-Y, et al. Prenatal diagnosis and molecular cytogenetic characterization of *de novo* partial trisomy 7p (7p15.3 → pter) and partial monosomy 13q (13q33.3 → qter) associated with Dandy-Walker malformation, abnormal skull development and microcephaly. Taiwan J Obstet Gynecol 2010;49:320–6.

[2] Chen C-P, Chen M, Su Y-N, Hsu C-Y, Tsai F-J, Chern S-R, et al. Chromosome 1p36 deletion syndrome: prenatal diagnosis, molecular cytogenetic characterization and fetal ultrasound findings. Taiwan J Obstet Gynecol 2010;49:473–80.

[3] Gardner RJM, Sutherland GR. Down syndrome, other full aneuploidies, and polyploidy. In: Gardner RJM, Sutherland GR, editors. Chromosome abnormalities and genetic counseling. 3rd ed. New York: Oxford University Press; 2004. p. 249–63.

[4] Delabar J, Theophile D, Rahmani Z, Blouin J, Prieur M, Noel B, et al. Molecular mapping of twenty-four features of Downs syndrome on chromosome 21. Eur J Hum Genet 1993;1:114–24.

[5] Arron JR, Winslow MM, Polleri A, Chang CP, Wu H, Gao X, et al. *NFAT* dysregulation by increased dosages of *DSCR1* and *DYRK1A* on chromosome 21. Nature 2006;441:595–600.

[6] Ronan A, Fagan K, Christie L, Conroy J, Nowak NJ, Turner G. Familial 4.3-Mb duplication of 21q22 sheds new light on the Down syndrome critical region. J Med Genet 2007;44:445–8.

[7] Eggermann T, Schönherr N, Spengler S, Jäger S, Denecke B, Binder G, et al. Identification of a 21q22 duplication in a Silver-Russell syndrome

patient further narrows down the Down syndrome critical region. Am J Med Genet 2010;152A:356−9.
[8] Aula P, Leisti J, von Koskull H. Partial trisomy 21. Clin Genet 1973;4: 241−51.
[9] Hagemeijer A, Smit E. Partial trisomy 21. Further evidence that trisomy of band 21q22 is essential for Down's phenotype. Hum Genet 1977;38:15−23.
[10] Leonard C, Gautier M, Sinet P, Selva J, Huret J. Two Down syndrome patients with rec(21), dupq, inv(21)(p11;q2109) from a familial pericentric inversion. Ann Genet 1986;29:181−3.
[11] Delabar J, Sinet P, Chadefaux B, Nicole A, Gegonne A, Stehelin D, et al. Submicroscopic duplication of chromosome 21 and trisomy 21 phenotype (Down syndrome). Hum Genet 1987;76:225−9.
[12] Huret J, Delabar J, Marlhens F, Aurias A, Nicole A, Berthier M, et al. Down syndrome with duplication of a region of chromosome 21 containing the CuZn superoxide dismutase gene without detectable karyotypic abnormality. Hum Genet 1987;75:251−7.
[13] McCormick M, Schinzel A, Petersen M, Stetten G, Driscoll DJ, Cantu ES, et al. Molecular genetic approach to the characterization of the "Down syndrome region" of chromosome 21. Genomics 1989;5:325−31.
[14] Petersen MB, Tranebjaerg L, McCormick MK, Michelsen N, Mikkelsen M, Antonarakis SE. Clinical, cytogenetic, and molecular genetic characterization of two unrelated patients with different duplications of 21q. Am J Med Genet 1990;7(Suppl):104−9.
[15] Rahmani Z, Blouin J, Creau-Goldberg N, Watkins PC, Mattei JF, Poissonnier M, et al. Down syndrome critical region around D21S55 on proximal 21q22.3. Am J Med Genet 1990;7:98−103.
[16] Korenberg JR, Bradley C, Disteche CM. Down syndrome: molecular mapping of the congenital heart disease and duodenal stenosis. Am J Hum Genet 1992;50:294−302.
[17] Korenberg JR, Chen XN, Schipper R, Sun Z, Gonsky R, Gerwehr S, et al. Down syndrome phenotypes: the consequences of chromosomal imbalance. Proc Natl Acad Sci USA 1994;91:4997−5001.
[18] Barnicoat A, Bonneau J, Boyd E, Docherty Z, Fennell SJ, Huret JL, et al. Down syndrome with partial duplication and del(21) syndrome: study protocol and call for collaboration. Study 1: clinical assessment. Clin Genet 1996;49:20−7.
[19] Forster-Gibson CJ, Davies J, MacKenzie JJ, Harrison K. Cryptic duplication of 21q in an individual with a clinical diagnosis of Down syndrome. Clin Genet 2001;59:438−43.
[20] Nadal M, Vigo CG, Melaragno MI, Andrade JA, Alonso LG, Brunoni D, et al. Clinical and cytogenetic characterisation of a patient with Down syndrome resulting from a 21q22.1 → qter duplication. J Med Genet 2001;38:73−6.
[21] Kosaki R, Kosaki K, Matsushima K, Mitsui N, Matsumoto N, Ohashi H. Refining chromosomal region critical for Down syndrome-related heart defects with a case of cryptic 21q22.2 duplication. Congenit Anom (Kyoto) 2005;45:62−4.
[22] Lee J, Stanley JR, Vaz SA, Mulvihill JJ, Wilson P, Hopcus-Niccum D, Li S. Down syndrome with pure partial trisomy 21q22 due to a paternal insertion (4;21) uncovered by uncultured amniotic fluid interphase FISH. Am J Med Genet 2005;132A:206−8.
[23] Vaglio A, Milunsky A, Quadrelli A, Huang XL, Maher T, Mechoso B, et al. Clinical, cytogenetic, and molecular characterization of a girl with some clinical features of Down syndrome resulting from a pure partial trisomy 21q22.11-qter due to a *de novo* intrachromosomal duplication. Genet Test Mol Biomarkers 2010;14:57−65.
[24] Fuentes JJ, Genesca L, Kingsbury TJ, Cunningham KW, Perez-Riba M, Estivill X, et al. DSCR1, overexpressed in Down syndrome, is an inhibitor of calcineurin-mediated signaling pathways. Hum Mole Genet 2000;9:1681−90.
[25] Ermak G, Morgan TE, Davies KJA. Chronic overexpression of the calcineurin inhibitory gene *DSCR1* (Adapt78) is associated with Alzheimer's disease. J Biol Chem 2001;276:38787−94.
[26] Song W-J, Sternberg LR, Kasten-Sportes C, Keuren ML, Chung S-H, Slack AC, et al. Isolation of human and murine homologues of the Drosophila minibrain gene: human homologue maps to 21q22.2 in the Down syndrome 'critical region'. Genomics 1996;38:331−9.
[27] Chen H, Chrast R, Rossier C, Gos A, Antonarakis SE, Kudoh J, et al. Single-minded and Down syndrome? Nat Genet 1995;10:9−10.
[28] Ema M, Ikegami S, Hosoya T, Mimura J, Ohtani H, Nakao K, et al. Mild impairment of learning and memory in mice overexpressing the *mSim2* gene located on chromosome 16: an animal model of Down's syndrome. Hum Mole Genet 1999;8:1409−15.
[29] Chrast R, Scott HS, Madani R, Huber L, Wolfer DP, Prinz M, et al. Mice trisomic for a bacterial artificial chromosome with the single-minded 2 gene (*Sim2*) show phenotypes similar to some of those present in the partial trisomy 16 mouse models of Down syndrome. Hum Mole Genet 2000;9:1853−64.