Chromosome 22q11 Deletion Syndrome : The First Three Cases Reported in Thailand

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Abstract

The DiGeorge, velocardiofacial, and conotruncal anomaly face syndromes were originally described as separate disorders due to different concerns regarding phenotypes. However, all these disorders have some common clinical manifestations, including congenital heart defect, facial anomaly, and developmental delay. It is now clear that most cases of these syndromes have a common cause resulting from microdeletion of chromosome 22q11. This study reports the first three cases of Thai children presented with developmental delays. All are females who were known cases of congenital heart diseases. Their minor facial anomalies were subtle and not previously recognized as of any syndromes. The chromosome study by fluorescent in situ hybridization technique yielded microdeletion of chromosome 22q11. Without known prevalence in Asian populations, except in Japanese children, further study for chromosome 22q11 deletion syndrome in Asian children with conotruncal heart defects, who also have minor facial anomalies or developmental delays, should be undertaken.

Key word : Chromosome 22q11 Deletion Syndrome, Velocardiofacial Syndrome, Conotruncal Heart Defect

DiGeorge syndrome (DGS) was first described by Angeo DiGeorge in 1965[1]. The significant clinical manifestations include absent thymus, abnormal immune system, and hypoparathyroidism. Lischner later correlated this syndrome with the abnormalities of the third and fourth pharyngeal pouches[2]. In 1979, Conley et al described that conotruncal heart defects were another major associated anomaly[3]. Shprintzen et al also described another group of patients, who had cleft palate, congenital heart disease, minor facial anomaly, and learning disability as velocardiofacial syndrome

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(VCFS)[4]. At about the same time, conotruncal anomaly face syndrome (CTAFS), which included conotruncal heart defect and facial anomaly, was described by Kinouchi et al[5].

All these syndromes have now a common cause resulting from a genetic disorder, microdeletion of chromosome 22q11[6-8]. By using fluorescent in situ hybridization technique (FISH), there are 80-90 per cent of DGS, 70 per cent of VCFS, and 15-30 per cent of CTAFS reported to be related to this genetic disorder[9-12]. Most of the cases are de novo; only 25 per cent are familial cases[13]. The prevalence in general population is estimated to be 1:5,000-10,000[14,15]. There was recently a collaborative European study of 558 cases. It was reported that 75 per cent had congenital heart defects, mainly conotruncal type; 68 per cent had mildly developmental delays; 46 per cent had abnormal palate, which mostly were velopharyngeal insufficiency (31%); and only 9 per cent had overt cleft palate[16].

We report 3 cases of known congenital heart defects with developmental delays diagnosed of chromosome 22q11 deletion by FISH technique in Thailand.

CASE REPORTS

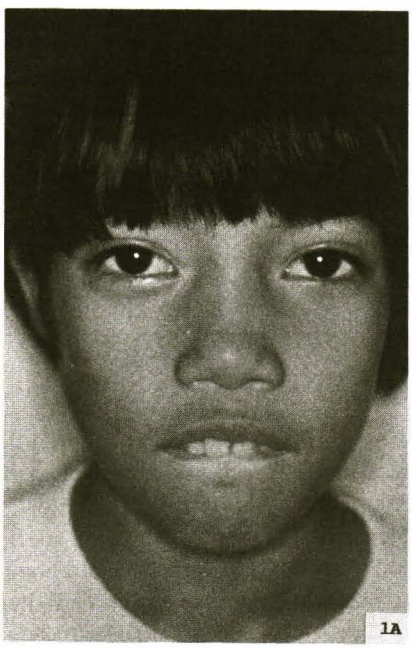
Case I

UM was a 7-year-old girl who was the 2.7 kg product of a term gestation to a 26-year-old mother. Pregnancy and delivery were uneventful. Her congenital heart defect was first recognized at 9 months of age. The subsequent diagnosis was atrial septal defect, secundum type, which was surgically corrected at 2 years. At 6 years old, she had bilateral herniotomy for her indirect inguinal hernia. When UM was 7 years old, she was referred for developmental evaluation due to her learning problem. Her height was 112 cm (10^{th} centile), weight was 18.5 kg (25^{th} centile). On examination (Fig. 1A), she had long face, lateral displacement of the inner canthi, a long prominent nose with bulbous tip, and a midsternal scar from previous cardiac surgery. Her speech was disarticulated. Her IQ, tested with Stanford-Binet, was 60.

UM had one younger brother who normally developed. The family history was unremarkable for any birth defects or developmental delays.

Case II

NV was a 7-year-old girl who was born *via*



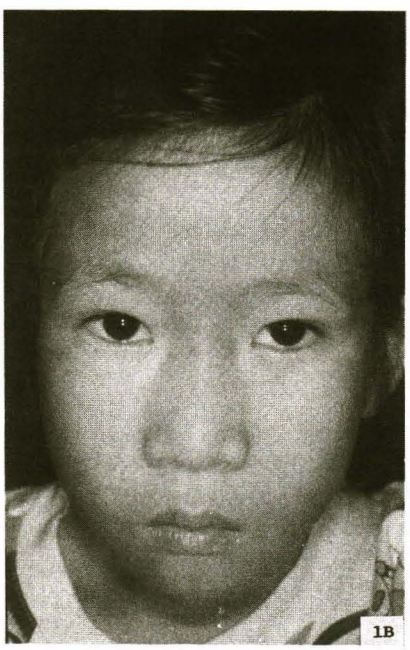


Fig. 1. The facial features of case I (A) and case II (B) are similar on long face, lateral displcement of the inner canthi, a long prominent nose with bulbous tip, and micrognathia.

Table 1. The clinical summary of the patients.

Clinical features	Case I	Case II	Case III
Congenital heart defect	ASD	PA with VSD	PA with VSD
Dysmorphic facial features			
- long face	+	+	+
- lateral displacement of inner canthi	+	+	+
- laterally built up nose (square nose)	+	+	+
- micrognathia	-	+	-
- velopharyngeal insufficiency (nasal voice)	-	+	+
Mental retardation	Mild	Mild	Moderate
Family history	-	Positive for congenital heart defect	-

ASD = atrial septal defect; PA = pulmonary atresia; VSD = ventricular septal defect

cesarean section with birth weight of 2.9 kg. A cyanotic heart disease was diagnosed shortly after birth. It was later confirmed as pulmonary atresia and ventricular septal defect. She had been on medical treatment and planned to have a surgical correction. At 7 years old, she was admitted for subacute bacterial endocarditis. Shortly before admission, NV had an IQ test due to her learning difficulty. Her IQ was 64, so the consultation for educational plan was made during the admission. She weighed 16.4 kg (3rd centile), had a height of 108 cm (3rd centile). Physical examination (Fig. 1B) showed a long face, lateral displacement of the inner canthi, a laterally built up nose, micrognathia, prominent cardiac murmur with central cyanosis, and long tapered fingers. NV also had nasal voice.

The family history was remarkable for her younger sister, who also was diagnosed with a congenital heart defect at birth. She died at one year of age at another medical center. Her detailed medical history was not obtained.

Case III

PP was a 9-year-old girl who was the 2.6 kg product of an uneventful term pregnancy. Pulmonary atresia with ventricular septal defect and patent ductus arteriosus were first diagnosed at 2 months of age. She later had a modified Blalock-Taussig shunt, and had been on medical treatment for congestive heart failure. Academically, she had learning difficulty since starting kindergarten. At the age of 9 years and 6 months, she was in the first grade. Her IQ score was 41, which was in the moderate range of mental retardation. Her weight was 18.3 kg, and height was 112 cm; both were below the 3rd centiles. Physical examination showed a long face, lateral displacement of the inner canthi, a laterally built up nose with flat nasal bridge, increased heart sound S2 with continuous murmur at left upper sternal border, central cyanosis, and clubbing fingers. Her speech was well-articulated, but with nasal voice. The family history was negative for congenital heart defects and for mental retardation in other family members.

The clinical summaries of patients are shown in Table 1.

LABORATORY METHODS

DNA probes

The DNA probes used in this study were located at 22q11.2. The clone 48F8 was localized in the DGS minimal critical region. 100C10 was distal to the critical region (Fig. 2). Both of them were isolated by Desmaze[17]. The cosmid clone PI 90-22, located at centromere of chromosome 22 was used as control probe. All of these probes were kindly provided by Dr. A Jauch, Institute of Human Genetics, University of Heidelberg, Germany.

Probe labelling and FISH analysis

Probe labelling was performed *via* nick translation[18]. Cosmid clones 48F8 and 100C10 were conjugated with biotin-14-dCTP(BRL) and centromere 22 specific probes were labelled with digoxigenin-11-dUTP(Boehringer). Aliquots of 200 ng or each cosmid were precipitated in the presence of 5 μg human Cot-I DNA(BRL) and 10 μg salmon sperm DNA(Sigma). DNA was dissolved in 10 μl of hybridization solution (50% formamide, 10% dextran sulfate, 2xSSC), denatured at 75°C for 5

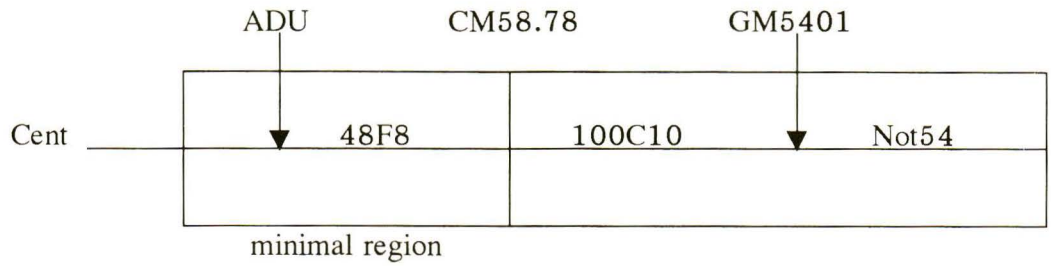


Fig. 2. 48F8 is the probe region detecting microdeletion on chromosome 22.

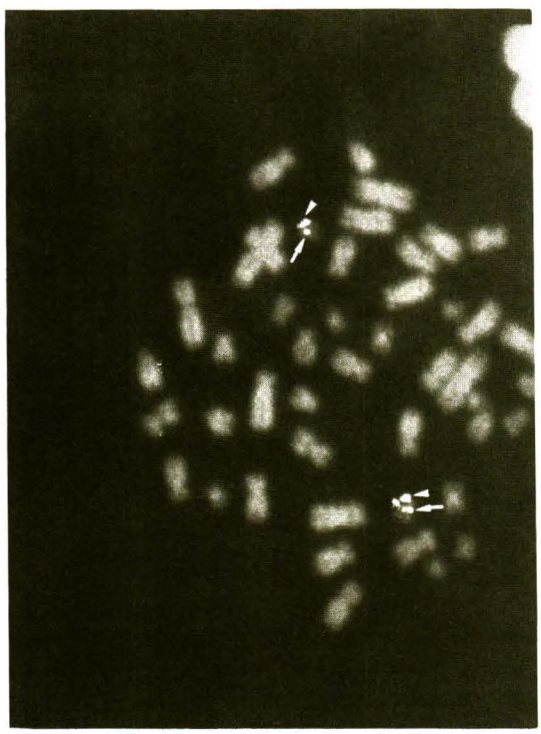


Fig. 3. When the metaphase chromosome of patients were hybridized with cosmid clone 100C10 (arrow) and specific probe of centromere of chromosome 22 (arrowhead), both probes were detected on both chromosome 22.

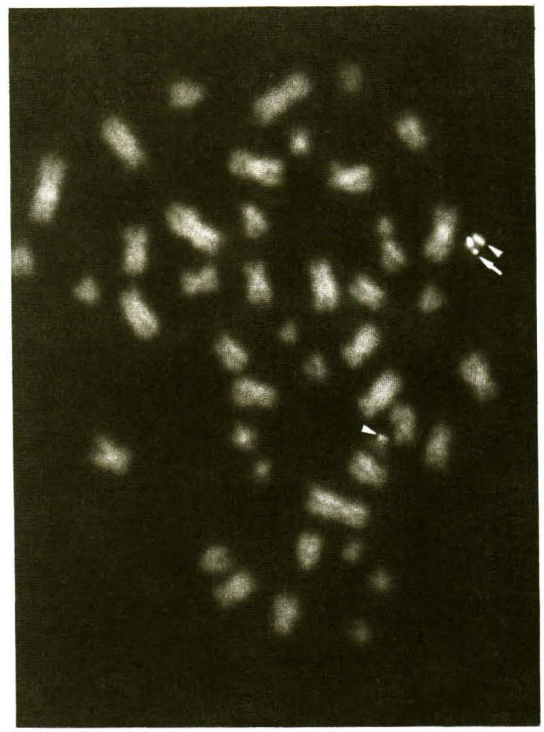


Fig. 4. Metaphase spreads obtained from a patient were hybridized with bio-labeled 48F8 and dig-labeled centromere 22. Signals of centromere of chromosome 22 (arrowhead) were detected on both chromosomes whereas only one of the chromosome 22 was labeled with cosmid clone 48F8 (arrow).

min, allowed to preanneal at 37°C for 20 min and applied to previously, denatured metaphase chromosome (at 75°C for 2 min in 70% formamide, 2xSSC pH 7.0). Hybridization took place overnight at 37°C. The DGS biotinylated probes were detected with Streptavidin Cy4(Sigma) and the Dig-probe was detected with mouse antidigoxiginin and anti-mouse FITC.

About 25 metaphases were analysed under fluorescence microscope (Nikon, Labophot). Photographs were taken from digital image printing (Metasystem).

All 3 girls and both parents of the first and second ones had chromosome studied by FISH technique. They all, except their parents, had microdeletion in the region of probe 48F8 DiGeorge minimal critical region but no deletion was found in the distal probe (100C10). The results are shown in Fig. 3 and 4.

DISCUSSION

Even though there have been no previous reported documented in Thai literature search, DiGeorge syndrome has been known as a clinical syndrome for a long time in Thailand. Velocardiofacial and conotruncal anomaly face syndromes are hardly mentioned among Thai physicians. All these 3 clinical syndromes are currently known to be caused by a genetic disorder, microdeletion of chromosome 22q11. Due to mild dysmorphic features and other medical concerns, congenital heart diseases are then commonly recognized as patients' health problem[19].

The 3 patients studied had been known cases of congenital heart diseases before being diagnosed with chromosome 22q11 microdeletion. Most types of cardiac anomaly in this syndrome are conotruncal defects[20]. Two patients had pulmonary atresia and ventricular septal defect, which are common findings in other reports. Atrial septal defect in the third one, even though is not a conotruncal type, was also previously reported. Developmental delay is another common finding, which varies from 60-90 per cent of cases[16-21]. Most have delayed speech or mild mental retardation[22, 23]. All patients in this report had definite mental deficit, which was in mild and moderate range of retardation.

With awareness, the faces of these girls had typical dysmorphic features as described in the literature. However, no cleft palate was found. Two patients had nasal voice, which was a suggestive sign of velopharyngeal insufficiency. Only 2 patients had serum calcium checked, and all were in the normal range (data is not shown).

Genetically, all 3 patients presumably have *de novo* deletions because parents of 2 had normal chromosome study by FISH technique, and the family history was unremarkable in the third one. However, in case II, whose parents did not have microdeletion of chromosome 22 shown in our study, her younger sister reportedly died because of a congenital heart defect at one year. There was a study hypothesized that gonadal mosaicism could be a mode of genetic transmission in a family of normal parents with 2 affected children. It is unfortunately impossible to prove this theory of transmission in this family because the second affected sibling died many years ago.

Since prenatal diagnosis is available for familial cases, early recognition in the first-born child can help families have an alternative choice about second child.

ACKNOWLEDGEMENT

We would like to thank Mrs. Pannee Prasertsan for her help in obtaining blood samples from subjects.

(Received for publication on October 15, 1999)

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กลุ่มอาการโครโมโซม 22q11 deletion : รายงานผู้ป่วย 3 รายแรกของไทย

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กลุ่มอาการ DiGeorge (DG), velocardiofacial (VCF) และ conotruncal anomaly face (CTAF) เคยถูกจัดเป็นกลุ่มโรคที่แยกจากกันชัดเจน แต่พบว่ามีลักษณะทางคลินิกบางอย่างพบร่วมกัน ได้แก่ ความผิดปกติของหัวใจแต่กำเนิด ลักษณะใบหน้าผิดปกติเล็กน้อย และพัฒนาการช้าหรือปัญญาอ่อน ปัจจุบันเป็นที่ทราบกันว่าผู้ป่วยเหล่านี้ส่วนมากมีการขาดหายไป (microdeletion) ของตำแหน่งที่ 11 บนแขนยาวของโครโมโซมคู่ที่ 22 ผู้วิจัยรายงานผู้ป่วยเด็ก 3 รายแรกของไทยที่มาด้วยปัญหาพัฒนาการช้า เด็กทั้ง 3 คนเป็นผู้ป่วยที่มีปัญหาหัวใจผิดปกติแต่กำเนิด มีลักษณะผิดปกติของใบหน้าเล็กน้อย ซึ่งได้แก่ ใบหน้ายาว ตาห่าง สันจมูกกว้างออกด้านข้าง เป็นต้น การตรวจโครโมโซมโดยเทคนิคพิเศษคือ fluorescent *in situ* hybridization (FISH) พบว่ามีการขาดหายไปของตำแหน่งดังกล่าวบนโครโมโซมคู่ที่ 22 นอกจากการศึกษาในกลุ่มประชากรญี่ปุ่นที่เป็นโรคหัวใจผิดปกติแต่กำเนิด ซึ่งพบว่ามีความผิดปกติของโครโมโซมนี้ใกล้เคียงกับประเทศตะวันตก ยังไม่เคยมีการศึกษาความชุกของกลุ่มอาการนี้ในประชากรเอเชียอื่นซึ่งน่าจะมีจำนวนมากพอสมควร จึงควรมีการศึกษาหาความผิดปกติทางพันธุกรรมชนิดนี้ในเด็กทุกคนที่มีข้อบ่งชี้ โดยเฉพาะที่มีความผิดปกติของหัวใจแต่กำเนิดชนิด conotruncal defect ร่วมกับมีความผิดปกติเล็กน้อยของใบหน้า หรือมีพัฒนาการช้าผิดปกติ

คำสำคัญ : Chromosome 22q11 Deletion Syndrome, Velocardiofacial Syndrome, Conotruncal Heart Defect

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