Williams Syndrome and the Elastin Gene in Thai Patients

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Abstract

Williams syndrome (WS) has long been known as a complex disorder of dysmorphic facial features, described as elfin face, mental retardation or learning disability, loquacious personality, and supravalvular aortic stenosis. The etiology is now known to be due to deletion of the elastin gene (ELN) on long arm of chromosome 7. Thai patients were previously reported by clinical diagnosis. This study reports the first two cases of WS with ELN deletion diagnosed by fluorescent *in situ* hybridization (FISH) technique. Clinically, hyperacusis is a common finding in WS associated with otitis media. Neither of the patients had hyperacusis, but one of them had bilateral sensorineural hearing loss, which to our knowledge, has never been reported.

Key word : Williams Syndrome, Elastin Gene, Hearing Loss

Williams syndrome had been described by Williams et al[1] long before a microdeletion of the elastin gene at chromosome 7q11.23 was reported to be the genetic etiology in 1993[2,3]. The clinical manifestations include some dysmorphic facial features, cardiovascular abnormalities, developmental delay, infantile hypercalcemia and other relatively less common features including short stature, kyphoscoliosis, inguinal or umbilical hernia, etc[4]. Hyperacusis and otitis media were also commonly reported in this syndrome[5], but to our knowledge, there has been no report of sensorineural hearing loss as an associated finding. These 2 cases of Williams syndrome were diagnosed by fluorescent *in situ* hybridization (FISH) technique. One of the patients had bilateral sensorineural hearing loss.

CASE REPORTS

Case I

HC was a 16-month-old girl referred to a developmental clinic for an evaluation of her developmental delay. She was a 2.7 kg product of uneventful term pregnancy. Her mother was 33 years old and father was 60 years old. She was born *via*

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caesarean section due to primigravida breech presentation. Apgar score was 3 and 7 at 1 and 5 minutes, respectively. The postnatal period was also remarkable for hyperbilirubinemia with maximal bilirubin level at 11mg/dl and a recognizable cardiac murmur. 2-D echocardiogram revealed ventricular septal defect and pulmonary stenosis. HC had been clinically stable without any other medical problems. She was described as an easy-going and pleasant infant. Developmentally, she reportedly sat without-support at 7 months, walked independently at 14 months, occasionally said nonspecific "mama" at 9 months. Her weight was 8.6 kg (10th percentile), length was 76 cm (between 25th and 50th percentile), and head circumference was 43.8 cm (just below 3rd percentile). Physical examination revealed a flat nasal bridge with an upturned nose, periorbital fullness, malar flattening, full cheeks and lips, a wide mouth, and long smooth philtrum. Other characteristics were remarkable for systolic cardiac murmur at left parasternal border and mild hyperreflexia, but normal muscle tone.

Serum calcium level was 10.2 mg/dl (upper normal range). The developmental assessment using Denver II revealed globally developmental delays. She was functioning at approximately 12-14 months of age, except for her language development, which was commensurate with 9 months. An audiogram (auditory brainstem evoked response, ABR) was performed due to her significant language delay. It showed that she had bilateral sensorineural hearing loss.

Case II

CN was a 15-month-old girl referred by a cardiologist due to poor weight gain and developmental delay. Her parents were in their early 30s. The perinatal period was remarkable for low birth weight (2,100 g). She had poor feeding, but slightly improved by the age of one year. Her cardiac defect was recognized a few months after birth, and later diagnosed as atrial septal defect and branch pulmonary stenosis. At 4 months, she had herniotomy for left inguinal hernia. Developmental delay was first noted by 8 months. Aside from her feeding difficulty, CN was a good-tempered infant. Her weight was 7.4 kg (below 3rd centile), length was 71 cm (10th centile), and head circumference was 43 cm (below 3rd centile). On examination (Fig. 1), CN was a pleasant child with a flat nasal bridge and an upturned nose, periorbital fullness, malar flattening, full cheeks and lower lip, a wide mouth, and long smooth philtrum. A cardiac murmur was noted at left upper sternal border area with fixed split of S2. Other characteristics were unremarkable. The serum calcium level was 10.8 mg/dl (upper normal range). Developmentally, her overall functioning level was at approximately 12 months, except for gross motor skill, which was at 9 month level.

Due to some clinical features, which were compatible with Williams syndrome, microdeletion of chromosome 7q 11.23 was investigated in both girls.

LABORATORY METHOD

For detection of deletion, a digoxiginin labeled cosmid probe, D72427 (Oncor^{R}) was used. This probe was located at 7q11.23 in the region of elastin gene.

FISH was performed on metaphase chromosomes from each patient. Prehybridization treatment and hybridization condition was carried out as described[6]. The digoxiginin probe was detected by mouse antidigoxiginin and antimouse FITC (Sigma).

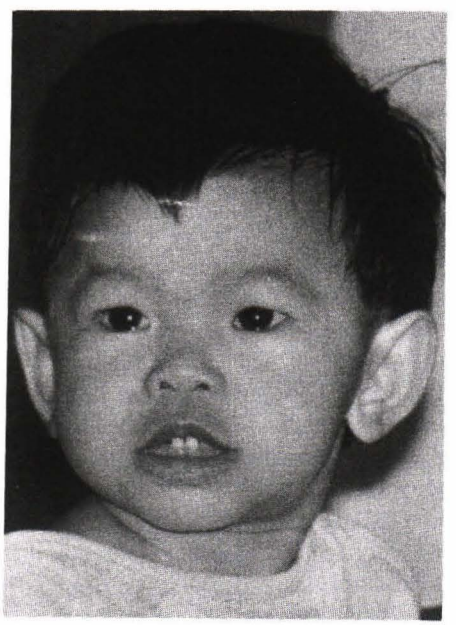

Fig. 1. The typical facial features of the patient (case II) include a flat nasal bridge and an upturned nose, periorbital fullness, malar flattening, full cheeks, a wide mouth, and a long smooth philtrum.

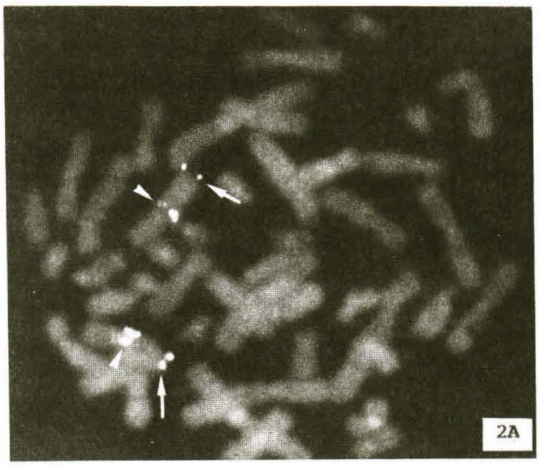

Fig. 2A. Metaphase spreads obtained from normal control were hybridized with 7q11.23 probe (control probe, arrow) and elastin gene probe (arrowhead). Both probes were detected on both chromosomes 7.

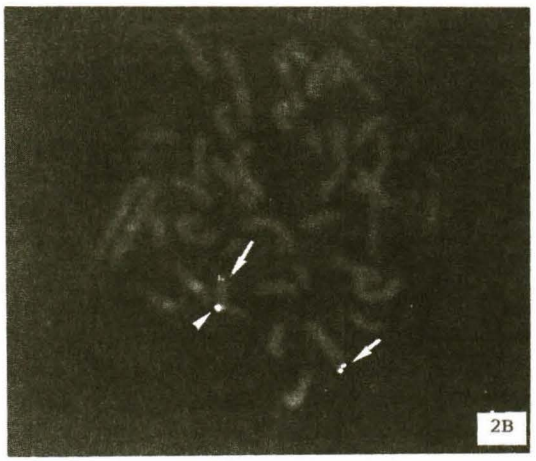

Fig. 2B. Hybridization of the same set of probe on metaphase chromosomes from Williams syndrome patient, hybridization signal of probe 7q11.23 (arrow) were detected on both chromosomes 7. Whereas only one chromosome 7 of each index case was labeled with elastin gene probe (arrowhead).

About 25 metaphases were assessed under fluorescence microscope (Nikon, Labophot) and photographs were taken from digital image printing (Metasystem).

The result was shown in Fig. 2.

DISCUSSION

Williams syndrome (WS), or Williams-Beuren syndrome (WBS), was described as a syndrome of supravalvular aortic stenosis, mental retardation and peculiar facial features by Williams et al[1] in 1961. Some additional findings including dental anomalies and peripheral pulmonary artery stenosis were described by Beuren et al[7] in 1962. Ten years before that, some infants, who were believed to have this syndrome, presented with poor feeding, constipation, vomiting, failure to thrive and idiopathic hypercalcemia[8,9]. The etiology had not been identified until 1993, when Ewart et al[2,3] demonstrated deletions on the locus called elastin gene (ELN) on the long arm of chromosome 7. The deletion of elastin gene is hypothesized to affect connective tissue causing abnormalities of vascular system and skin. However, mental retardation, atypical behaviors, and hypercalcemia still have unknown mechanism of pathogenesis. Deletions of some other genes flanking ELN are being studied[10].

WS is generally sporadic, with an estimated incidence of 1 in 20,000 live-births, although rare familial (autosomal dominant) cases have been reported[11]. Common phenotypic features include typical facial features, mental retardation or developmental delay, growth failure, congenital heart disease (mostly supravalvular aortic stenosis), and inguinal hernia[4,12]. Hypercalcemia, which is mostly transient, is not so common as originally thought[13]. The key features of dysmorphic face evolve with age. They are a broad forehead, periorbital fullness, strabismus, stellate iris pattern, flat nasal bridge, malar flattening, full cheeks and lips, a long smooth philtrum, a rather pointed chin, and a wide mouth. Our 2 patients have typical facial features, particularly with "unusually friendly" behavior, which is another typical characteristic of this syndrome[14]. Even almost all patients have developmental delay or mental retardation, they usually

function better in the area of language than fine motor. These 2 girls had globally developmental delays without any significant discrepancy between the areas. With their delayed growth, one of them reportedly had poor feeding, as previously reported in some infants[15].

Hyperacusis is a common finding in WS, but neither of the patients were reportedly sensitive to sound[16,17]. Morris et al[12] reported abnormal hearing acuity in two patients associated with otitis media. To our knowledge, there has never been sensorineural hearing loss reported in WS. The first case had bilateral sensorineural hearing loss, and no known etiology could be identified.

WS has been a known syndrome in Thailand for many years, but there was only one report of 2 cases, which were clinically diagnosed[18]. Currently, the elastin gene probe is commercially available, so a definite diagnosis can be made in suspected cases. Children with typical facial features, developmental delay or mental retardation, and supravalvular aortic or pulmonary stenosis warrant investigation.

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REFERENCES

1. Williams JCP, Barratt-Boyes BG, Lowe JB. Supravalvular aortic stenosis. Circulation 1961; 24:311-8.
2. Ewart AK, Morris CA, Ensing GJ, et al. A human vascular disorder, supravalvular aortic stenosis, maps to chromosome 7. Proc Natl Acad Sci USA 1993; 90:3226-30.
3. Ewart AK, Morris CA, Atkinson D, et al. Hemizygosity at the elastin locus in a developmental disorder, Williams syndrome. Nat Genet 1993; 5: 11-5.
4. Burn J. Williams syndrome. J Med Genet 1986; 23:389-95.
5. Klein AJ, Armstrong BL, Greer MK, Brown FR. Hyperacusis and otitis media in individuals with Williams syndrome. J Speech Hear Disord 1990; 55:339-44.
6. Tocharoentanaphol C, Cremer M, Schrock E, et al. Multicolor fluorescence in situ hybridization on metaphase chromosomes and interphase halo-preparations using cosmid and YAC clones for the simultaneous high resolution mapping of deletions in the dystrophin gene. Hum Genet 1994; 93:229-35.
7. Beuren AJ, Apitz J, Harmjanz D. Supravalvular aortic stenosis in association with mental retardation and certain facial appearance. Circulation 1962; 26: 1235-40.
8. Lightwood R. Idiopathic hypercalcemia with failure to thrive. Arch Dis Child 1952; 27:302.
9. Payne WW. The blood chemistry in idiopathic hypercalcemia. Arch Dis Child 1952; 27:302.
10. Wu YQ, Sutton VR, Nickerson E, et al. Delineation of the common critical region in Williams syndrome and clinical correlation of growth, heart defects, ethnicity, and parental origin. Am J Med Genet 1998; 78:82-9.
11. Morris CA, Thomas IT, Greenberg F. Williams syndrome : autosomal inheritance. Am J Med Genet 1993; 47:478-81.
12. Morris CA, Demsey SA, Leonard CO, Dilts CV, Blackburn BL. Natural history of Williams syndrome : physical characteristics. J Pediatr 1988; 113:318-26.
13. Lowery MC, Morris CA, Ewart A, et al. Strong correlation of elastin deletions, detected by FISH, with Williams syndrome : evaluation of 235 patients. Am J Hum Genet 1995; 57:49-53.
14. Dilts CV, Morris CA, Leonard CO. Hypothesis for development of a behavioral phenotype in Williams syndrome. Am J Med Genet 1990; 6:126-31.
15. Lenhoff HM, Wang PP, Greenberg F, Bellugi U. Williams syndrome and the brain. Sci Am 1997; 277:68-73.
16. Nigam A, Samuel PR. Hyperacusis and Williams syndrome. J Laryngol Otol 1994; 108:494-6.
17. Meyerson MD, Frank RA. Language, speech and hearing in Williams syndrome : intervention approaches and research needs. Dev Med Child Neurol 1987; 29: 258-70.
18. Jaruratanasirikul S, Soponthammarak S, Weeramonthanasak C, Sanguanchua P. Williams syndrome : a report of 2 cases and literature review. Thai J Pediatr 1995; 34:170-5.

กลุ่มอาการวิลเลียมส์ กับยีนส์อีลาสติน ในผู้ป่วยเด็กไทย

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กลุ่มอาการ Williams (WS) เป็นที่รู้จักกันมานานว่ามีลักษณะทางคลินิกซึ่งประกอบด้วยลักษณะใบหน้าที่ผิดปกติเล็กน้อย (บางครั้งถูกเรียกว่าใบหน้าของ elfin ซึ่งเป็นตัวละครในนิทานของประเทศทางตะวันตก) ภาวะปัญญาอ่อนหรือการเรียนผิดปกติ ผู้ป่วยจะมีบุคลิกภาพเฉพาะคือ ช่างพูดแต่ไม่ค่อยมีสาระ และมักมีอารมณ์ดี รวมทั้งมีความผิดปกติของหลอดเลือดชนิด supravalvular aortic stenosis ในปัจจุบันทราบว่า WS มีสาเหตุจากการขาดหายไปของยีนบางส่วนที่เรียกว่า elastin gene (ELN) บนแขนยาวของโครโมโซมคู่ที่ 7 ผู้วิจัยรายงานผู้ป่วยเด็ก 2 รายที่มาตรวจด้วยปัญหาพัฒนาการช้า และได้รับการวินิจฉัยว่าเป็นกลุ่มอาการ Williams โดยการตรวจโครโมโซมโดยเทคนิค fluorescent *in situ* hybridization (FISH) พบว่ามี ELN หายไป ในรายงานอื่น ๆ พบว่าผู้ป่วยกลุ่มนี้มักมีความไวต่อเสียงบางชนิดมากกว่าปกติ (hyperacusis) ซึ่งพบร่วมกับหูชั้นกลางอักเสบได้บ่อย อย่างไรก็ตามผู้ป่วยทั้งสองคนในรายงานนี้ไม่มีลักษณะดังกล่าว แต่พบว่าเด็ก 1 ใน 2 รายนี้มีการได้ยินผิดปกติชนิด sensorineural loss ของหูทั้งสองข้าง จากการทบทวนการศึกษาอื่น ๆ เท่าที่ผ่านมาไม่พบว่าเคยมีการรายงานลักษณะความผิดปกติดังกล่าวนี้ในกลุ่มอาการ Williams

คำสำคัญ : กลุ่มอาการวิลเลียมส์, ยีนส์อีลาสติน, หูตึง

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