CLINICAL REPORT

AMERICAN JOURNAL OF medical genetics PART A

Middle and Inner Ear Malformations in Mutation-Proven Branchio-Oculo-Facial (BOF) Syndrome: Case Series and Review of the Literature

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Manuscript Received: 30 October 2011; Manuscript Accepted: 1 April 2012

Hearing impairment is common in individuals with branchio-oculo-facial (BOF) syndrome. The majority of described individuals have conductive hearing impairment due to malformed ossicles and/or external canal stenosis or atresia, although a sensorineural component to the hearing impairment in BOF syndrome is increasingly being reported. Sophisticated computed tomography (CT) of the temporal bone has revealed middle and inner ear malformations in three previous reports. We present middle and inner ear abnormalities in three additional individuals with mutation-proven BOF syndrome. We suggest that temporal bone CT imaging be included in the medical workup of a child with BOF syndrome, in order to guide management. © 2012 Wiley Periodicals, Inc.

Key words: BOF syndrome; inner ear; middle ear; hemangioma; branchial arch; neural crest; temporal bone; computed tomography

How to Cite this Article:
Carter MT, Blaser S, Papsin B, Meschino W, Reardon W, Klatt R, Babul-Hirji R, Milunsky J, Chitayat D. 2012. Middle and inner ear malformations in mutation-proven branchio-oculo-facial (BOF) syndrome: Case series and review of the literature.
Am J Med Genet Part A 158A:1977–1981.

INTRODUCTION

Branchio-oculo-facial syndrome [BOFS (OMIM 113620)] is an autosomal dominant disorder primarily of the first and second pharyngeal arches. The main features of this syndrome are cutaneous defects in the cervical, infra-auricular and/or supra-auricular region ("branchio"); ocular anomalies including microphthalmia or anophthalmia, cataract, coloboma, strabismus, ptosis, and nasolacrimal duct obstruction ("oculo"); and characteristic appearance with malformed pinnae, broad nasal tip, pseudocleft or cleft lip and/or palate, and upslanting palpebral fissures ("facial") [Lin et al., 1995]. Other common but variable findings are scalp cysts, prematurely gray hair, ectopic dermal thymus, and congenital hearing impairment [Lin et al., 1995]. Recently, the gene associated with BOF syndrome was identified as *TFAP2A* [Milunsky et al., 2008]. The gene is expressed in cranial neural crest cells and their derivatives including the mesenchyme of the medial and lateral nasal prominences and the maxillary prominence. A mutation hotspot has been identified in exons 4 and 5; however, no genotype-phenotype correlations have yet emerged [Milunsky et al., 2011].

Hearing impairment in individuals with BOF syndrome is common, reported in approximately 70% of mutation-proven cases [Milunsky et al., 2011]. The majority of reported individuals are described as having conductive hearing impairment. However, a sensorineural component to the hearing impairment is increasingly being reported. Sophisticated computed tomography of the temporal bone (TB-CT) has revealed inner ear malformations in three reports of patients with BOF syndrome [Stoetzel et al., 2009; Tekin

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Article first published online in Wiley Online Library (wileyonlinelibrary.com): 18 June 2012
DOI 10.1002/ajmg.a.35436

et al., 2009; Thomeer et al., 2010]. We present three additional patients with classical BOF syndrome and *TFAP2A* mutations, all of whom had detailed TB-CT scans revealing middle and inner ear anomalies. Our study further delineates the variety of inner and middle ear abnormalities associated with BOFS.

PATIENT REPORTS

Patient 1

An 11-year-old boy was born to healthy, non-dysmorphic, non-consanguineous East-Indian parents. The pregnancy was uncomplicated and there was no history of maternal illness or exposures to teratogens. Delivery was spontaneous, vaginal, and uncomplicated and the birth weight was 3.5 kg (25–50th centile). At birth, bilateral cleft lip (with intact palate) was noted as well as "hemangiomatous" areas of skin behind both auricles. Moderate to severe conductive hearing impairment was diagnosed when he presented with speech delay as a toddler; air conduction hearing aids were prescribed with good effect. Myopia was diagnosed later in childhood. He attends regular school and performs well academically. On examination at 11 years, he had a short forehead, heavy eyebrows, broad nasal root and tip, bilateral scars of repaired cleft lip, thin upper lip vermilion with prominent lower lip, low-set ears, and bilateral hemangiomatous areas with atrophic skin behind both auricles (left>right), non-midline pseudocleft of the upper lip, and abnormal teeth (Fig. 1).

TB-CT showed deficient long process of the right incus, bony atresia plate covering left oval window (Fig. 1), and enlarged vestibular aqueducts (EVAs) measuring 2.9×1.4 mm on the left and 2.1×1.0 mm on the right (measured halfway between the porus and vestibule, an EVA is greater than 1.5 mm in any dimension [Propst et al., 2005]). Karyotype was 46,XY. Mutation analysis of the *TFAP2A* gene revealed a missense change in exon 4 (NM_003220.2:c.760C>T; p.R254W) [Milunsky et al., 2011].

Patient 2

A 10-year-old boy was born to healthy, non-dysmorphic, non-consanguineous, Iranian parents. Two older brothers were also healthy. The patient had moderate bilateral mixed-type hearing impairment, nasolacrimal duct stenosis, myopia, velopharyngeal insufficiency, and skin defects superior to the malformed pinnae bilaterally. He also had a left accessory nipple, dysplastic fingernails,

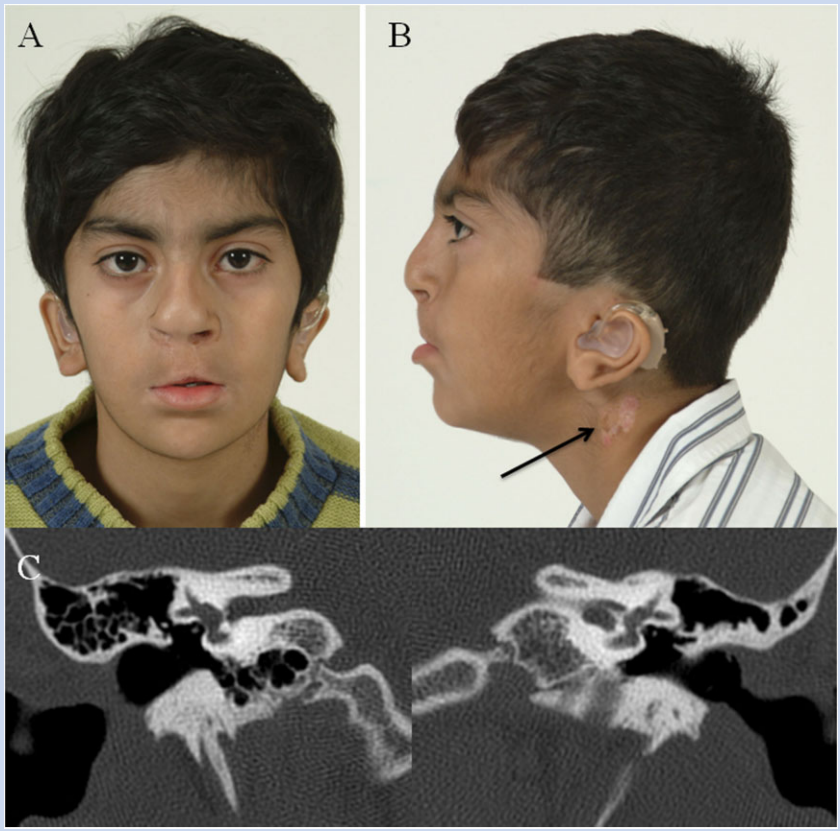

FIG. 1. A, B: Facial appearance of patient 1. Note low anterior hair line with temporal hair extending down almost to the eyebrows, synophrys, scars of bilateral repaired cleft lip, short columella, long philtrum with prominent ridges, flattened tip of the nose, micrognathia, low-set and posteriorly rotated ears with hearing aids, and posterior auricular hemangioma which turned into a hypopigmented scar (arrow). C: TB-CT findings in this patient. Coronal unenhanced petrous CT demonstrates a bony atresia plate over the left oval window (arrow). The right side is shown for comparison. Other anomalies included deficient long process of the right incus, and mild enlargement of the left vestibular aqueduct (not shown).

shawl scrotum, broad thumbs, and great toes. Renal ultrasound showed structurally normal kidneys. A skull radiograph at 8 months of age showed wormian bones along the lambdoid sutures. TB-CT scan showed EVAs (4.1 × 1.1 mm on the left, 2.4 × 1.0 mm on the right, measured at the midportion of the vestibular aqueduct), thin bony plate over the right oval window and misshapen ossicles bilaterally (Fig. 2). Sequence analysis of *TFAP2A* revealed a missense change in exon 4 (NM_003220.2:c.709C>G; p.R237G) [Milunsky et al., 2011].

Patient 3

An 11-year-old Caucasian girl previously reported as an infant [Raveh et al., 2000], prior to knowledge of the causative gene for BOF syndrome. She was born to healthy, non-dysmorphic, nonconsanguineous parents. She has one sister who is healthy. A previous pregnancy 5 years earlier was terminated because the fetus had bilateral multicystic dysplastic kidneys and anhydramnios. During the pregnancy with the patient, the 18-week fetal anatomy ultrasound scan detected a left multicystic dysplastic kidney. She was delivered by cesarean at 38 weeks because of asymmetrical intrauterine growth restriction. Birth weight was 1.67 kg (<3rd centile), birth length was below the 3rd centile, and head circumference was at the 10th centile. At birth she was noted to have subcutaneous scalp cyst, upslanting palpebral fissures, bilateral lacrimal duct obstruction, pseudo-cleft lip, malformed pinnae, cutaneous defects superior and inferior to the pinnae bilaterally, bilateral supernumerary nipples, and left 2–3 toe syndactyly. Postnatal abdominal ultrasound confirmed left multicystic kidney on the left and the ureteral pelvic junction obstruction on the right. Her karyotype was 46,XX. At 11 years old, she had mixed-type hearing impairment with a bone-anchored hearing aid on the right. She had right facial nerve palsy, bilateral atresia of the lacrimal ducts, and high myopia. She had several small or missing teeth, malar hypoplasia, a short anteverted nose, short philtrum, pseudo-cleft lip, and abnormal pinnae with deficiency of the superior helices. There was evidence of scarring inferior to the ears and neck bilaterally. She had a dermoid cyst removed from her scalp at 12 months of age. Mild dilatation of the right renal collecting system was stable. Her postnatal growth improved; her height and weight at 11 years of age were at 10th–25th centile and her head circumference was at the 40th centile. She has done well in school.

TB-CT showed narrow external auditory canals and cochlear dysplasia, small horizontally oriented semicircular canals, small bone islands, and an accessory right facial nerve canal with ectopic origin (Fig. 2). Sequence analysis of *TFAP2A* revealed a missense change in exon 4 (NM_003220.2:c.760C>G; p.R254G) [Milunsky et al., 2011]. The mutation was inherited from her father whose only clinical features of BOF syndrome were a right-sided preauricular pit, a single supernumerary nipple and a white forelock.

DISCUSSION

The branchio-oculo-facial (BOF) syndrome is characterized by branchial [cervical (90%) or infra- or supra-auricular (60%)] skin defects that range from barely identifiable thin skin or hair patch to "hemangiomatous" lesions; ocular anomalies that can include microphthalmia, anophthalmia, coloboma, and nasolacrimal duct stenosis/atresia; and facial anomalies that can include ocular hypertelorism or telecanthus, broad nasal tip, upslanted palpebral fissures, and cleft lip or pseudocleft lip with or without cleft palate [Lin et al., 1995]. Intellect is usually normal [Lin et al., 1995]. The only gene known to be associated with BOF syndrome is the *TFAP2A* gene [Kaiser et al., 2007; Milunsky et al., 2008].

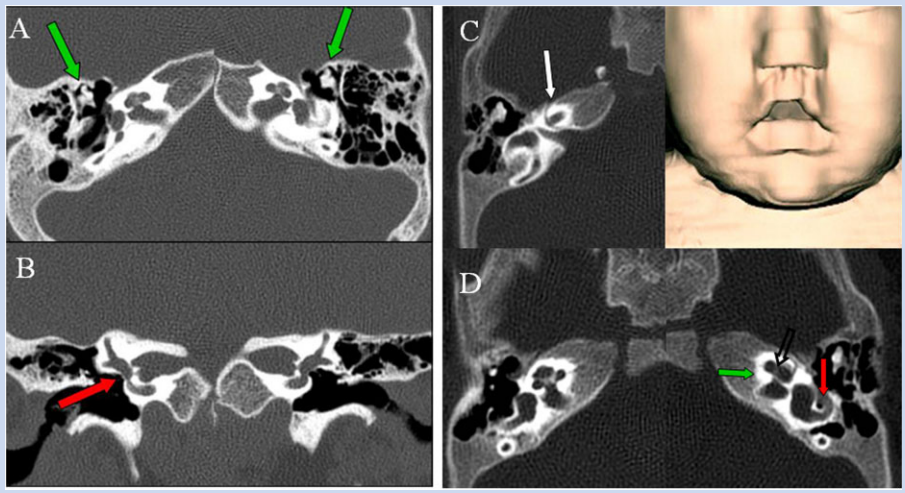

FIG. 2. A, B: TB-CT findings in patient 2. Axial CT images demonstrate misshapen ossicles (arrows), while coronal views reveal a thin bony plate over the right oval window (arrow). C, D: TB-CT findings in patient 3. 3D surface reconstruction CT demonstrates upper lip defect. Axial images reveal accessory facial nerve canal (arrow in C), small cochlear nerve canal (1.4 mm) (horizontal arrow in D), hypoplastic bone island of the lateral semicircular canals (vertical arrow in D) and hypoplastic modiolus (open arrow in D).

TABLE I. Middle and Inner Ear Anomalies and the *TFAP2A* Mutations Previously Reported in Mutation-Proven BOF Syndrome

Report	*TFAP2A* mutation	Temporal bone CT scan anomalies
Tekin et al. [2009]	p.276_281delLPAGRRinsRI	Bilateral cochlear dysplasia (Mondini type), enlarged vestibule, and vestibular aqueduct
Stoetzel et al. [2009]	p.S239P	Thin long process of incus, malformed stapes, small round and oval windows, deformed vestibules, thin left facial nerve canal
Thomeer et al. [2010]	p.R236P	Narrow external auditory canals, bony atretic plate with soft tissue at the medial end of the canals, bilateral external fixation of the malleus

Hearing impairment in BOF syndrome is well recognized, but temporal bone abnormalities are likely underreported [Milunsky et al., 2011]. Raveh et al. [2000] were the first to report inner ear dysplasia detected by TB-CT in a patient with BOF syndrome; case 1 in their report is the same girl as patient 3 in this series. Only a few case reports since then have detailed the middle and inner ear malformations associated with mutation-proven BOF syndrome; these are summarized in Table I.

Tekin et al. [2009] reported on a 4-year-old girl with BOF syndrome and bilateral profound sensorineural hearing loss. TB-CT showed bilateral cochlear dysplasia (Mondini type), enlarged vestibule, and EVA. Cochlear implantation was performed. Stoetzel et al. [2009] reported on a three-generation family, all of whom had similar temporal bone abnormalities: Oblique external auditory canals, deformed long processes of the incus, deformed stapes, small round and oval windows, deformed vestibules, and normal cochlea. One individual had a thin left labrinthine facial nerve canal. All three had conductive hearing loss; one of the three had a sensorineural component as well. Thomeer et al. [2010] presented an 8-year-old girl with BOF syndrome and conductive hearing impairment, with TB-CT findings of narrow external ear canals, and a possible bony atretic plate at the medial end of the ear canals. A bony external fixation of the malleus was discovered bilaterally during exploratory tympanotomy. No inner ear malformations were reported. The patient was treated with bone-anchored hearing aid.

There are several genetic syndromes associated with both hearing impairment and temporal bone abnormalities. Pendred syndrome, an autosomal recessive condition caused by mutations in the *PDS* gene, was originally described with Mondini dysplasia, but more recent analyses have found that the most common radiologic anomaly is EVA [Phelps et al., 1998; Reardon et al., 2000]. Stoetzel et al. [2009] pointed out that the temporal bone abnormalities associated with BOF syndrome are distinct from those observed in CHARGE syndrome (aplastic semicircular canals) and overlap somewhat with those reported in branchio-oto-renal (BOR) syndrome. While BOF and BOR syndromes have overlapping clinical features (mixed hearing loss, pinna malformations, lacrimal duct stenosis, branchial sinus defects, and renal anomalies), they are genetically distinct [Kaiser et al., 2007]. BOR syndrome temporal bone anomalies include cochlear and vestibular malformations as well as malformations of the ossicles [Ceruti et al., 2002; Propst et al., 2005]. Based on their patient's findings and those of previous reports, Stoetzel et al. [2009] suggest that malformation of the malleus, cochlea, and vestibular aqueduct are features of BOR syndrome that distinguish it from BOF syndrome. However, two of our three patients had misshapen mallei; one had cochlear dysplasia and two of the three had EVA (Table II). Thus, it appears

TABLE II. Clinical, Molecular and Temporal Bone CT Scan Findings in Three BOF Syndrome Patients with Congenital Hearing Impairment

Patient	*TFAP2A* mutation	Type of hearing impairment and correction	Temporal bone CT scan abnormalities	
			Middle ear	Inner ear
Patient 1	p.R254W	Mixed (predominantly conductive), air conduction hearing aid	Deficient long process of the right incus, misshapen malleus, fusion of malleus and incus	Bony atresia plate covering left oval window, small bone islands, enlarged vestibular aqueducts
Patient 2	p.R237G	Conductive, air conduction hearing aid	Deficient long process of incus, misshapen malleus, fused malleus and incus	Enlarged vestibular aqueducts, small bone islands, fused foot plate
Patient 3	p.R254G	Mixed (predominantly conductive), bone-anchored hearing aid	Partial fusion of malleus and incus	Cochlear dysplasia, small horizontally-oriented semicircular canals, small bone islands, accessory right facial nerve canal with ectopic origin

that the range of temporal bone anomalies in BOR and BOF syndromes may overlap more than previously suspected. Thus, molecular analysis is important in clarifying the diagnosis in patients with overlapping features.

In total, six mutation-proven BOF syndrome patients have documented temporal bone abnormalities. Only two out of the six have a mutation in the same codon, patients 1 and 3 reported on herein (R254W and R254G, respectively). This codon in exon 4 of the *TFAP2A* gene appears to be the most commonly affected in probands with BOF syndrome [Milunsky et al., 2011]. Documentation of TB-CT anomalies in a larger cohort of mutation-positive BOF syndrome patients is needed before genotype-phenotype correlations can be determined regarding these malformations. It is important to catalogue the *TFAP2A* mutations associated with congenital deafness and middle and inner ear malformations in order to find clinically useful genotype-phenotype correlations.

BOF syndrome is associated with congenital hearing impairment in a large proportion of affected individuals. The type of hearing impairment is predominantly conductive, but mixed hearing impairment is also found. Those with a sensorineural component to their hearing impairment may have EVA or cochlear dysplasia. TB-CT is useful for evaluation of the bony structures of the middle and inner ear, as it can detect congenital malformations of the ossicular chain, cochlea, and semicircular canals. Proper determination of the etiology of hearing loss in these patients guides management. Therefore, all children with features of BOF syndrome and hearing impairment should have TB-CT scan performed as part of the diagnostic assessment to assist with management planning.

ACKNOWLEDGMENTS

The authors thank the patients' families for consenting to publish their clinical information and photographs.

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