Genetic Evaluation for Craniofacial Conditions

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KEYWORDS

• Genetic evaluation • Genetic counseling • Family history • Teratogens • Dysmorphology • Genetic testing • Cleft lip • Cleft palate

KEY POINTS

- Every child born with a craniofacial disorder should be evaluated by a clinical geneticist.
- Many craniofacial disorders have a genetic etiology, and large variety of genetic testing is available for testing affected individual and family members.
- Although many genetic disorders are common, many patients present with rare or unique conditions requiring specialized genetics evaluations and tests.
- All children with craniofacial disorders should be managed by an interdisciplinary craniofacial or cleft team.

INTRODUCTION

Congenital anomalies and disorders are those conditions that are present at birth and that require some level of medical intervention. These conditions occur in approximately 3% to 5% of all live births.[1] Craniofacial conditions, including orofacial clefts, craniosynostoses, the mandibulofacial dysostoses, and craniofacial macrosomia, are among the most common birth congenital anomalies. Many of these conditions have a genetic etiology (chromosomal, single-gene disorders, or epigenetic mutation) or may be caused by teratogens. Because of this, it is important for each child born with a craniofacial condition to be evaluated and followed by a medical geneticist. The American Cleft Palate-Craniofacial Association in their Standards for Cleft Palate and Craniofacial Teams states, "The Team also must demonstrate access to refer to a neurosurgeon, an ophthalmologist, a radiologist, and a geneticist."[2] The role of the medical geneticist is to assist in making a diagnosis of any known genetic disorder or syndrome, assist families and craniofacial team members in understanding the natural history of any syndrome, and ensure that additional medical evaluations and interventions are performed as indicated. There are thousands of different causes for craniofacial conditions. Identifying the etiologies is important for understanding the cause of a particular condition and influencing the management of a particular disorder. Also, craniofacial conditions are chronic conditions and follow-up evaluations with a medical geneticist should be encouraged.

THE GENETICS EVALUATION

The purpose of the genetics evaluation is to

- Make a diagnosis
- Characterize natural history
- Establish appropriate follow-up evaluations and testing
- Determine recurrence risk and potential genetic testing for family
- Provide genetic counseling for family

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Ideally, the genetics evaluation should be performed as early as possible, often soon after birth. Given the technical advances in prenatal diagnosis, prenatal genetic evaluation has become a common occurrence. The genetics evaluation differs from the typical medical evaluation with greater emphasis on prenatal and family histories.

Prenatal Evaluation

Congenital craniofacial conditions begin in utero. Therefore, obtaining a comprehensive pregnancy history is essential to understanding etiology, especially with regard to teratogen exposure, maternal illness, and prenatal testing. Teratogens are substances that interfere with normal embryologic and fetal development. Teratogens include medications and drugs, high-dose radiation, viruses, and maternal illnesses.

Maternal illnesses that are known to cause craniofacial anomalies are diabetes and maternal phenylketonuria. Women with diabetes, both type 1 diabetes mellitus and type 2 diabetes mellitus, have least a 2-fold risk for having a child with birth defects, the greatest risks associated with type 1 diabetes mellitus.[3] The major birth defects are renal, vertebral, brain, and craniofacial anomalies. Craniofacial anomalies include cleft lip, cleft palate (CP), and Pierre Robin sequence (PRS). In my institution, maternal diabetes is among the most common causes of cleft lip with or without CP (CLP) and CP. Women who have phenylketonuria are unable to properly metabolize the amino acid phenylalanine. If an affected woman does not follow a phenylalanine-restricted diet, the elevated levels of the metabolites of phenylalanine can cause multiple anomalies, including microcephaly, ear anomalies, congenital heart defects, and CP.[4] Maternal hyperthyroidism and Graves disease have been associated with neonatal craniosynostosis.[5]

Prenatal testing is commonly performed, especially fetal ultrasound. Ultrasound is performed in midtrimester in most pregnancies in the United States. Cleft lip can be identified with routine ultrasound in approximately 75% of cases[6] and diagnosis approaches 100% with high-resolution ultrasound.[7] It is more difficult to diagnose CP by ultrasound; however, micrognathia and PRS can be diagnosed prenatally.[8] For more complex cases, especially with those with multiple anomalies, fetal MRI scans are performed at several high-risk centers and can be useful for assessing severity of fetal structural and brain anomalies and have a direct impact on pregnancy management (**Fig. 1**).[9]

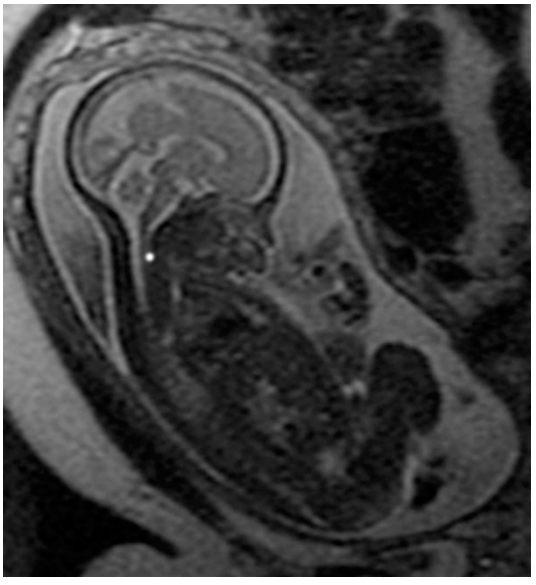

Fig. 1. Fetal MRI scan demonstrating severe micrognathia in a fetus with PRS.

If fetal anomalies are suspected, prenatal genetic testing should be considered. Invasive testing includes amniocentesis, which can be performed from 14 weeks' gestation to term, and chorionic villus sampling can be performed at 12 weeks' gestation. These procedures are usually

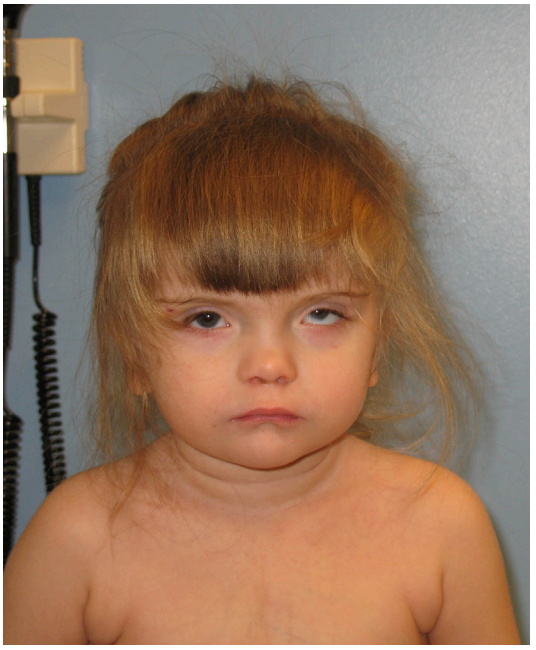

Fig. 2. Young girl with fetal valproate syndrome. Note the short nose, long philtrum, and up-slanting palpebral fissures.

performed to obtain chromosome analysis, chromosomal microarray, fluorescence in situ hybridization (FISH), or single-gene sequencing analysis.

Teratogens

Teratogens are those exogenous substances or physical agents, which, if there is fetal exposure, can cause birth defects. Many teratogens cause craniofacial anomalies. These include but are not limited to

- Physical agents – amniotic bands, radiation
- Infectious agents
- Medications
- Maternal illnesses
- Tobacco

Box 1
Clinical genetics history for the evaluation for craniofacial conditions

History of present illness

- Gestational age
- Type of delivery and complications
- Birth parameters: weight, length, and head circumference
- Other congenital anomalies or major illnesses
- Neonatal complications
- Early feeding and growth

Pregnancy history

- Maternal illnesses
- Maternal medications
- Exposure to other substance (alcohol, cigarettes, and history of substance abuse)
- Prenatal genetic testing (maternal screening tests, ultrasounds, and fetal chromosome or genetic testing)

Past medical history

- Major illnesses
- Hospitalizations
- Surgeries
- Feeding, nutrition, and growth
- Prior medical specialty evaluations

Comprehensive review of systems

- Ten-system review
- Overall health assessment

Developmental history

- Early developmental milestones
- Therapeutic interventions (early intervention, speech, physical, and occupational therapies)
- School performance
- Developmental and neuropsychological evaluations

Family history

- Four-generation pedigree
- Birth defects
- Pregnancy losses (miscarriages and stillbirths)
- Infant, childhood, and early adult deaths
- Infertility
- Consanguinity

- Alcohol
- Toluene (solvent for glues and spray paints)
- Cocaine

Alcohol is a commonly used and potent teratogen. Exposed children are at risk for many serious birth defects, the most common being developmental delay and intellectual disability.[10] Fetal alcohol syndrome and fetal alcohol spectrum disorder are associated with a large number of birth defects. Dysmorphic facial features are common as are microcephaly, brain anomalies, holoprosencephaly, limb anomalies, short stature, and behavior disorders. Craniofacial anomalies include CLP, CP, and PRS.[11,12]

Isotretinoin is a medication prescribed for cystic acne. Although this is an extremely effective medication for treatment of acne, it is a potent teratogen. Isotretinoin can cause multiple anomalies, including microcephaly, brain anomalies, microtia, absent auditory canal, hearing loss, and congenital heart defects.[13]

Valproic acid is an anticonvulsant with significant teratogenicity. This medication causes neural tube defects in approximately 1% of children exposed in utero. Other reported findings include facial dysmorphism, microcephaly, developmental delay and cognitive impairment, CP, and metopic craniosynostosis (**Fig. 2**).[14,15] Diphenyl hydantoin is another anticonvulsant that is teratogenic. It is associated with short stature, developmental disabilities, distal digital and nail hypoplasia, and craniofacial anomalies, including CLP and CP.[16]

Methotrexate is used to treat malignancies, autoimmune disorders, molar pregnancies, and tubal pregnancies. It acts as a folic acid antagonist and interferes with nucleic acid synthesis (thymidine) and, therefore, is highly cytotoxic. Because of this, methotrexate is a potent teratogen and can cause multiple birth defects, including CLP, CP, craniosynostosis, digital anomalies, microcephaly, brain anomalies, and developmental disabilities.[17]

Cigarette smoking, in addition to causing intrauterine growth restriction, can also cause birth defects. There is an association between cigarette smoking and gastroschisis.[18] Cigarettes have also been shown to cause CLP and CP. It is estimated that 6.1% of oral clefts can be attributed to smoking during pregnancy.[19–21] In addition, several genes have been identified, which have been associated with risk for CLP in women who smoke during pregnancy.[22]

Medical History

Obtaining a comprehensive history is essential for genetic diagnosis. Data should include birth history, including length of gestation, birth weight, length, and head circumference. Any

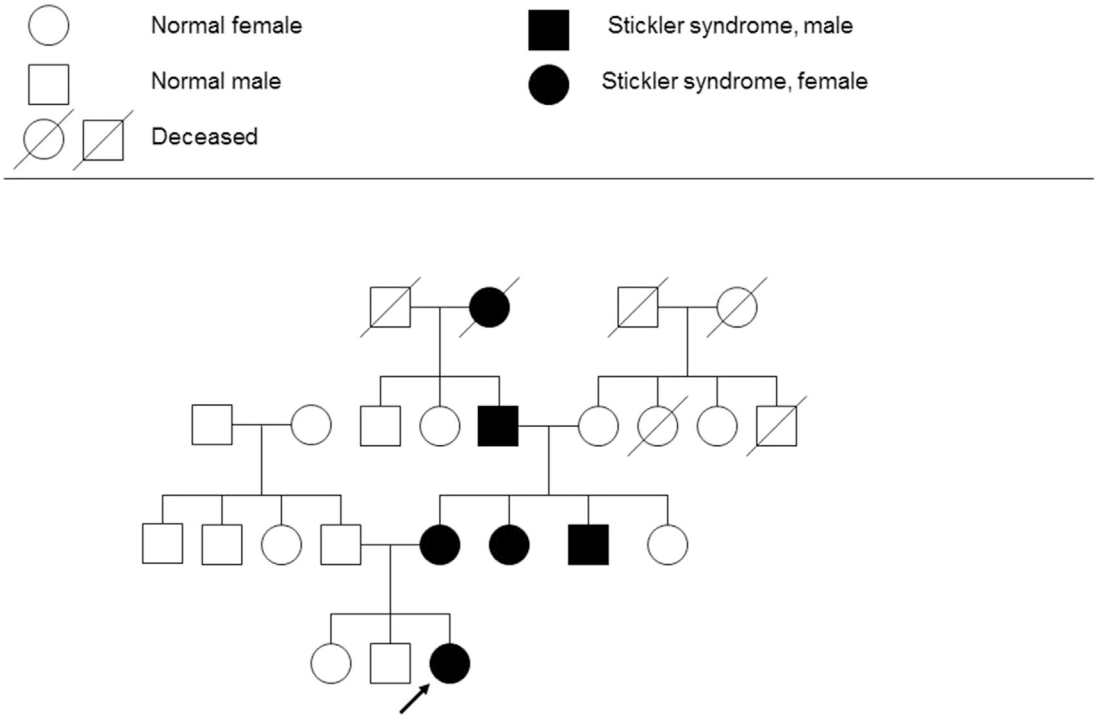

Fig. 3. Four-generation pedigree in a family with an autosomal dominant disorder, in this case Stickler syndrome.

Box 2
Craniofacial genetics physical examination

- Growth parameters – including *z* scores and growth trends on standardized charts
  - Height
  - Weight
  - Head circumference
  - Arm span
  - Upper to lower segment ratios (for disproportionate short stature)
- Skin
  - Birth marks
  - Hemangiomas
  - Hyperpigmented or hypopigmented macules
  - Hair – alopecia, texture, or hypertrichosis
  - Nails – missing nails or dysplastic nails
- Head/craniofacial
  - Cranial shape – evidence of craniosynostosis, ridging of sutures, or plagiocephaly
  - Fontanelles
  - Inner canthal, interpupillary, and outer canthal distances (hypertelorism and hypotelorism)
  - Facial asymmetry
  - Palpebral fissures – length and epicanthic folds
  - Ear position – low set and posteriorly rotated
  - Ear shape, microtia, and anotia
  - External auditory canal (stenosis and atresia)
  - Preauricular skin tags or fistulae (ear pits)
  - Eye examination – red reflex, iris colobomas, epibulbar dermoids, extraocular movements, nystagmus, and ptosis
  - Nose – short, anteverted nares, shape of nasal tip, and flat or prominent nasal bridge
  - Upper lip – clefting, unilateral, bilateral, and midline
  - Lower lip – clefting and lip pits
  - Palate – clefting (V-shaped or U-shaped), bifid uvula, and SMCP
  - Dentition – abnormally shaped teeth and missing teeth
  - Tongue – lobulations, microglossia or macroglossia, and asymmetry
  - Oral synechiae
  - Mandible – micrognathia, asymmetry, and ankylosis
- Neck
  - Masses
  - Torticollis
  - Branchial clefts or cysts
- Chest
  - Symmetry
  - Chest size and shape
  - Lung auscultation
  - Intercostal and subcostal retractions
  - Pectus deformities

- Cardiovascular
  - Heart murmurs
  - Pulses (upper and lower extremities)
- Abdomen
  - Organomegaly
  - Masses
  - Bowel sounds
- Genitalia — male
  - Penis size
  - Hypospadias
  - Testes – cryptorchidism and testicular size
- Genitalia – female
  - Labial adhesions and fusion
  - Vaginal discharge
- Musculoskeletal
  - Limb deformities
  - Brachydactyly
  - Clinodactyly (incurving of fifth finger)
  - Contractures
  - Joint hypermobility
  - Syndactyly
  - Polydactyly
  - Broad thumbs and halluces
  - Scoliosis
- Neurologic
  - Muscle tone (hypertonia or hypotonia)
  - Strength
  - Gait
  - Cranial nerve abnormalities (facial palsy, hearing loss, abnormal eye movements)

hospitalizations and surgeries should also be recorded as well as significant illnesses. Information regarding feeding and growth is also important, especially to establish if feeding problems are related to a CP or craniofacial anomaly or perhaps caused by underlying neurologic or other structural anomalies, such as a heart defect. Many genetic disorders, especially chromosomal conditions, are associated with poor growth.

Developmental history also gives important clues to diagnosis and management. Major parameters of development include speech and language development, gross motor skills, fine motor skills and personal–social development. For patients with craniofacial disorders, speech and language delays may indicate hearing deficits, whether from middle ear effusions with recurrent otitis media or possibly other structural neurologic problems causing conductive and/or sensorineural deafness. See **Box 1** for essential components of the genetics medical history.

Family History

A family history is an essential component of a genetics evaluation. Information from family history can provide information regarding hereditary disorders and birth defects. A pedigree is constructed, which is a pictorial representation of the family history. Usually information for at least 3 generations is obtained (**Fig. 3**). Family history should include information about

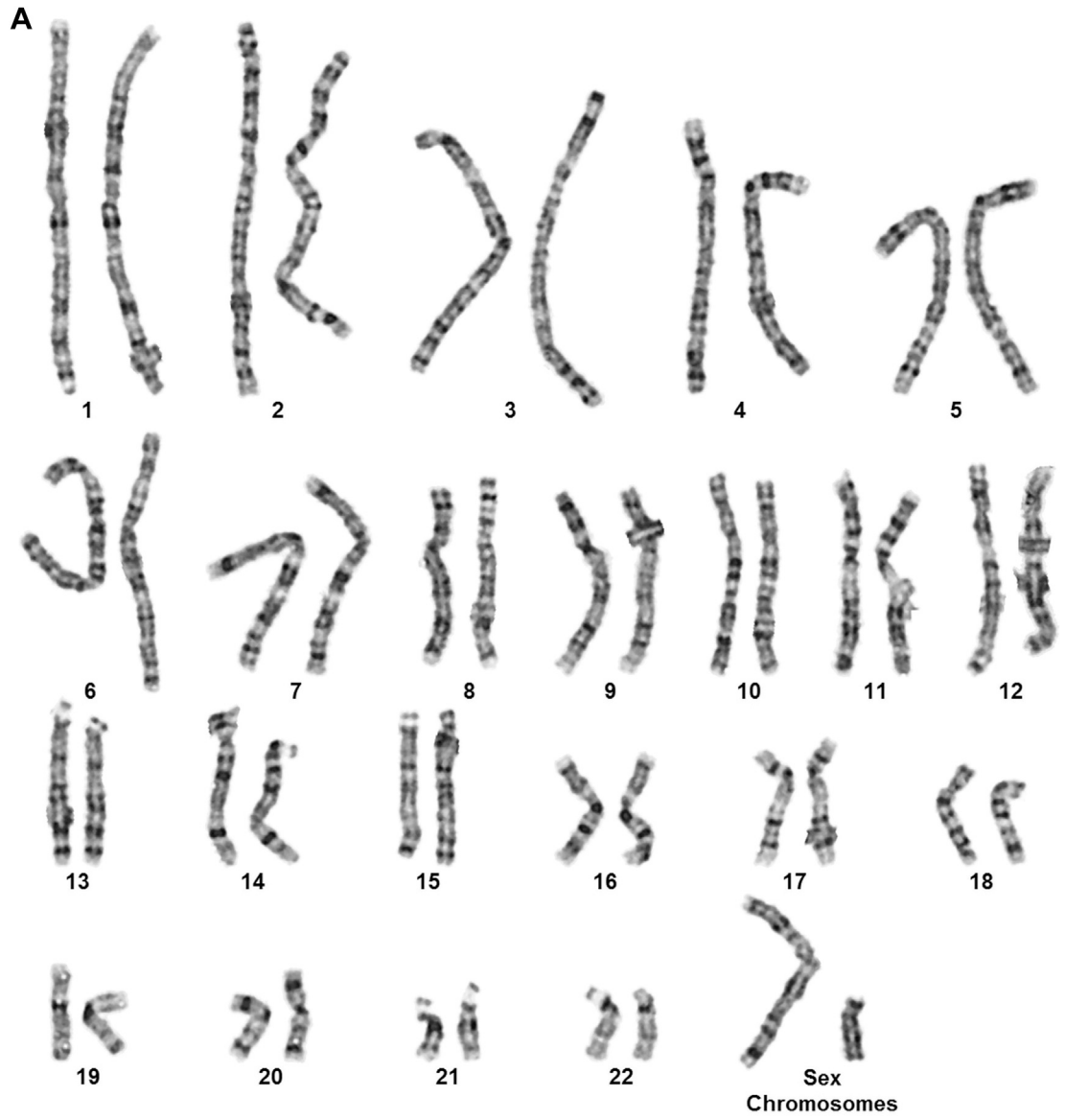

Fig. 4. Chromosome karyograms. On the left is a normal male karyogram with a 46,XY karyotype (*A*). On the right is an abnormal male karyogram with 47 chromosomes and trisomy 21 consistent with a diagnosis of Down syndrome (*B*).

- Birth defects
- Consanguinity
- Pregnancy loss (miscarriages and stillbirths)
- Developmental delay and intellectual disability
- Early or unexpected deaths and causes (if known)
- Mental illness and psychiatric disorders
- Early or unusual cancers
- Blindness
- Deafness
- Chromosome disorders

Physical Examination

A physical examination is an important component of any medical evaluation. The medical genetics physical examination differs from the typical physical examination because in addition to looking for typical findings, the genetics evaluation focuses on looking for atypical or dysmorphic physical features, which may give clues to a genetic or other syndromic disorder or possible etiology. **Box 2** outlines many of the features that may be seen. Any physical examination should include growth

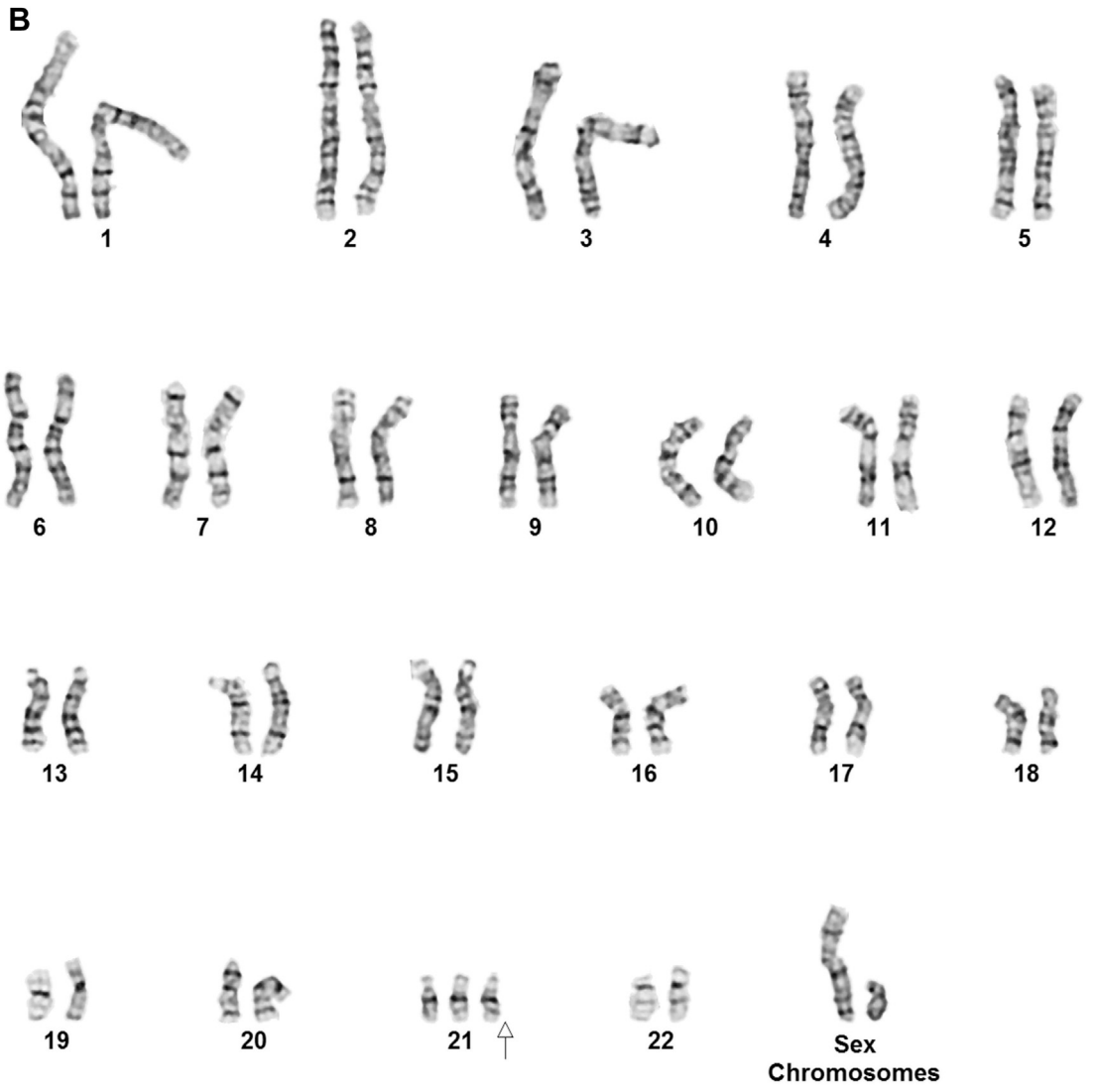

Fig. 4. (*continued*)

parameters, height, weight, and head circumference and should be accompanied by growth percentiles and *z* scores. These data are essential for diagnosis of short stature, microcephaly, and failure to thrive, all of which may give critical clues to causes of craniofacial disorders and diagnoses.

Laboratory Analysis

There are more than 1000 different disorders that can cause craniofacial anomalies, CLP, and CP. The prenatal, medical, and family histories and the physical examination often give clues as to the specific diagnosis; it then becomes important to confirm the diagnosis if a genetic etiology is suspected. In an analysis of children born with CLP or CP at Cincinnati Children's Hospital Medical Center, chromosomal anomalies were among the most common group of genetic conditions associated with orofacial clefts (**Fig. 4**). There was no clustering of specific chromosome disorders, but several chromosome conditions are associated with CLP and CP, including trisomy 13, trisomy 18, and Wolf-Hirschhorn syndrome (deletion of the short arm of chromosome 4). Velocardiofacial syndrome, or deletion 22q11.2 syndrome, is caused by a microdeletion of the long arm of chromosome 22 and can be diagnosed with a specific fluorescent-tagged DNA sequence, which hybridizes to the specific sequence of chromosome 22 by a test, FISH. In 22q11.2 deletion syndrome, 1 of the 2

chromosome 22 homologues has a deletion of this critical region (**Fig. 5**). Standard chromosome analysis is helpful for many conditions with additional or missing chromosomes or for identifying large chromosomal rearrangements. Chromosomal microarray is a more recently developed test that not only can identify large chromosomal rearrangements but also is able to identify small submicroscopic rearrangements.[23,24] Microarray has many advantages over routine chromosome analysis, including the ability to identify rare chromosomal rearrangements and increase yield of diagnoses.[24]

Many genetic disorders are caused by mutations in single genes. Some common craniofacial disorders and syndromes are single-gene disorders, such as Stickler syndrome and many of the craniosynostosis syndromes. More than 7000 single-gene disorders have been identified (https://globalgenes.org/rare-diseases-facts-statistics/).[25] Genetic testing is available for many of these disorders using gene sequencing. This testing, often called Sanger sequencing, uses polymerase chain reaction to sequence single genes. When ordering gene sequencing for specific genetic disorders, there must be a degree of suspicion of a diagnosis to determine which genetic test or tests to perform.

Many children have rare or even unique genetic disorders for which clinical genetic testing may not be available. These types of cases can now be evaluated using next-generation sequencing. This technology allows for massive parallel sequencing of the human genome. Because the approximately 20,000 human genes comprise only 1% to 2% of the entire human genome, however, only the coding portions of the genome (exons) are sequenced and analyzed and this test is called whole-exome sequencing.[26] Whole-exome sequencing has significantly increased the ability to diagnose genetic disorders, leading to a 25% to 40% increase in diagnostic yield.[27,28] Since the advent of whole-exome sequencing, hundreds of new genetic disorders have been identified.

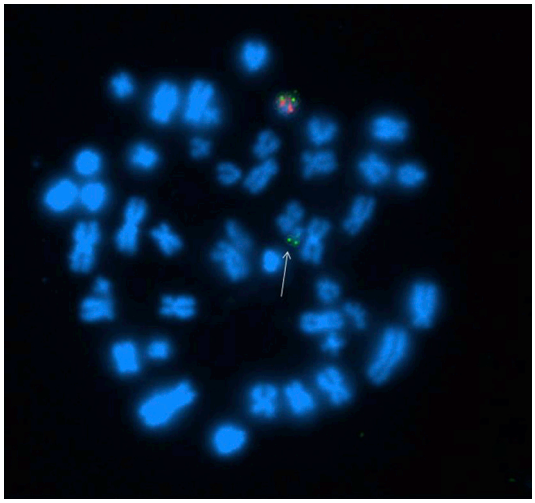

Fig. 5. FISH showing interstitial deletion of chromosome 22q11.2. The arrow points to the chromosome with the deletion; note the absence of the hybridization to the red fluorescence-tagged cDNA, which would hybridize to this region if not deleted.

Additional Evaluations

In addition to these studies, further clinical evaluation is often helpful, including additional medical evaluations, radiographs and other imaging studies, and audiology evaluation. Imaging studies are helpful to identify additional structural anomalies. Ophthalmology evaluation can identify myopia, a clue to Stickler syndrome or Marshall syndrome. Iris and retinal colobomas may be clues to multiple anomaly syndromes, such as CHARGE syndrome (colobomas, heart defects, atresia choanae, retarded growth and/or development, ear anomalies and/or deafness). Children with craniofacial macrosomia may have epibulbar dermoids. An otolaryngology evaluation is important for assessing ear anomalies and laryngotracheal anomalies that may be syndromic. Microtia and absent external auditory canals are seen in the mandibulofacial dysostoses, including Treacher Collins syndrome. Many cases of glottic webs are associated with 22q11.2 deletion syndrome.[29] Cardiology evaluation is essential for diagnosis and management of many syndromes associated with heart defects. The evaluation includes an echocardiogram. Approximately 75% of children with deletion 22q11.2 syndrome have a

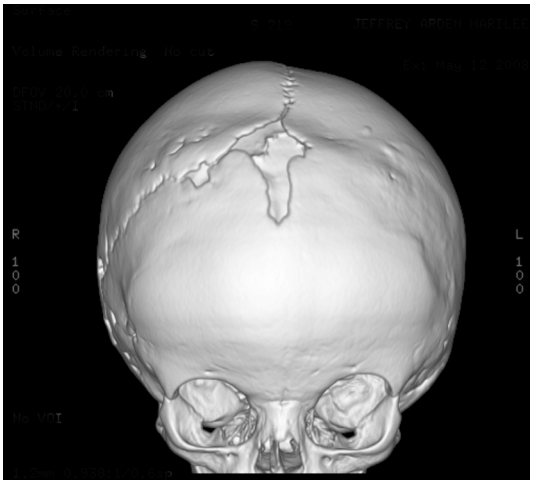

Fig. 6. CT scan with 3-D reconstruction demonstrating fusion of the left coronal suture.

congenital heart defect. Children with chromosome anomalies frequently have heart defects. Congenital heart defects are cardinal findings of CHARGE syndrome.

Imaging studies that can be helpful include radiographs, CT scans, and MRI scans. The radiographs are most helpful when looking for specific skeletal anomalies, including complete skeletal surveys looking for skeletal dysplasia syndromes. Vertebral anomalies are often seen with craniofacial macrosomia, Klippel-Feil syndrome, and diabetic embryopathy. Children with a large

Table 1
Syndromes associated with cleft lip with or without cleft palate

Disorder	Etiology	Gene	Inheritance	Clinical Features
Craniofacial microsomia (see **Fig. 8**)	Sporadic, possibly vascular disruption		Sporadic, Rarely AD	Facial asymmetry, microtia, vertebral anomalies, renal anomalies, heart defect
Amniotic bands (see **Fig. 9**)	Sporadic		Sporadic	CLP, digit or limb amputation, encephalocele
Van der Woude syndrome (**Fig. 7**)	Single-gene mutation	*IRF6*	AD	Lower lip fistulae (pits), CLP, CP, SMCP
Opitz syndrome (G syndrome; hypertelorism-hypospadias syndrome)	Single-gene mutation	*MID1* *SPECCIL*	XLR AD	Hypertelorism, hypospadias, CLP, congenital heart defects
Oral-facial-digital syndrome type I	Single-gene mutation	*OFD1*	XLD	CLP (midline), hyperplastic oral frenulae; tongue lobulations with hamartomas, digital anomalies, syndactyly
CHARGE association	Single-gene mutation	*CHD7*	AD, most cases de novo mutations	Heart defects, deafness, colobomas, genitourinary anomalies, choanal atresia, developmental disabilities
Smith-Lemli-Opitz syndrome	Single-gene mutation	*SLOS*	AR	Growth retardation, microcephaly, CLP, CP, heart defects, genital, syndactyly of toes 2 and 3, anomalies, developmental disabilities
Fetal alcohol syndrome	Teratogenic		Sporadic	Growth retardation, CLP, CP, heart defects, facial dysmorphism, developmental disabilities
Diabetic embryopathy	Teratogenic		Maternal diabetes	Heart defects, CLP, CP, vertebral anomalies, renal anomalies, brain anomalies
Deletion 1p36 syndrome	Chromosomal deletion		Sporadic	CL, CP, facial dysmorphism, speech apraxia, developmental disability
Wolf-Hirschhorn syndrome (4p- syndrome)	Chromosomal deletion		Sporadic	CLP, facial dysmorphism, hypotonia
Trisomy 13	Chromosomal, 3 copies of chromosome 13 (meiotic nondisjunction)		Sporadic (may be familial if there is a translocation)	CLP, CP, brain anomalies including holoprosencephaly, cutis aplasia of scalp, microphthalmia, heart defects, neural tube defects

Abbreviations: AD, autosomal dominant; AR, autosomal recessive; XLD, X-linked dominant; XLR, X-linked recessive.

Fig. 7. Infant with Van der Woude syndrome and bilateral cleft lip with CP. Note lip pits of lower lip.

number of genetic disorders may have renal anomalies, including dysplastic kidneys, hydronephrosis, and congenitally absent kidneys. Renal anomalies are common in craniofacial macrosomia, diabetic embryopathy, Klippel-Feil syndrome, and 22q11.2 deletion syndrome. Brain MRI is

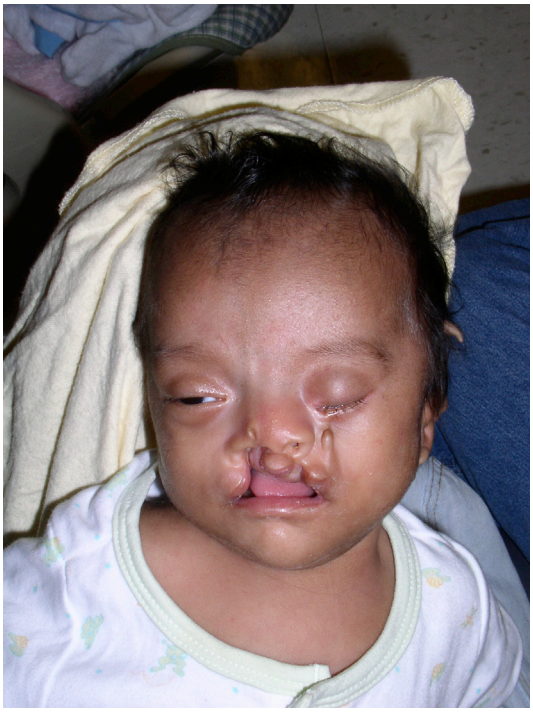

Fig. 9. Infant with cleft lip and CP secondary to amniotic band sequence. Note the remnant of the band below the left eye and lower lid coloboma caused by the band.

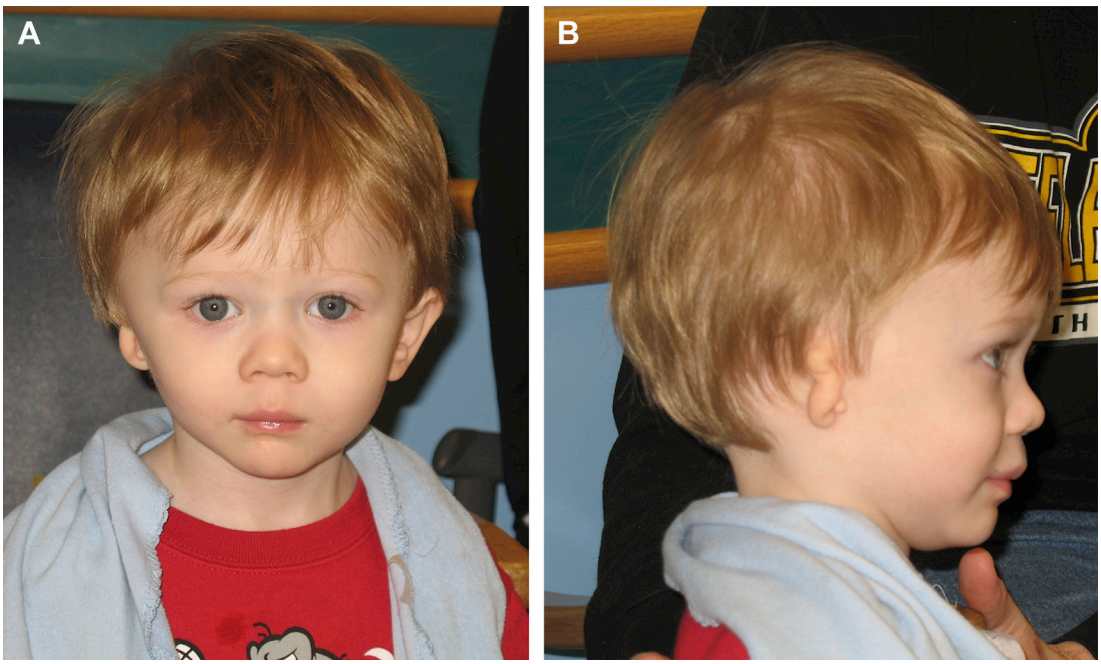

Fig. 8. Young boy with right craniofacial microsomia. Photo on right shows facial asymmetry (*A*) and photo on left shows microtia of right ear (*B*).

useful for identifying structural brain anomalies. It can be a useful test for diagnosis of neurofibromatosis type 1, in which optic pathway gliomas are commonly seen.[30] CT scans are helpful for looking at calcified tissues. This is an important test for evaluation of cranial structures, especially with 3-D reconstruction, and is the test of choice for diagnosing craniosynostosis (**Fig. 6**).

Genetic Counseling

Once all data are gathered and a diagnosis is made (and in many instances no diagnosis is made), it is important to discuss this information with the family. This becomes a time of educating the family regarding genetics and inheritance. Many genetic conditions have a low recurrence risks, either because these are de novo chromosomal anomalies, such as trisomy 13, or are de novo gene mutations for autosomal dominant disorders, such as CHARGE syndrome. Some disorders are inherited. Although 22q11.2 deletion syndrome is de novo in 90% of patients, in 10% of cases 1 of the parents also has a deletion of 22q11.2. Recurrence risk is 50% with each pregnancy.

Many genetic disorders have variable expressivity. For example, if an infant has Stickler syndrome with CP, the affected parent may have myopia as the sole manifestation of Stickler syndrome. Therefore, once a diagnosis is made, it is important to examine the parents and other family members and consider genetic testing of individuals at risk. Recurrence risk analysis should also include information regarding prenatal testing and reproductive options. Prenatal testing, with amniocentesis or chorionic villus sampling, is available for most chromosomal and single-gene disorders. Preimplantation testing using assisted reproductive technology can be an option for some conditions.

It is important to discuss the natural history of a genetic disorder or syndrome with the patient and family. Because most genetic disorders involve more than one organ system and frequently are associated with developmental and learning issues, medical and developmental interventions should be discussed. Because most craniofacial disorders are chronic conditions, long-term follow-up by a geneticist can be helpful for anticipatory management and to help address any additional medical issues that may arise.

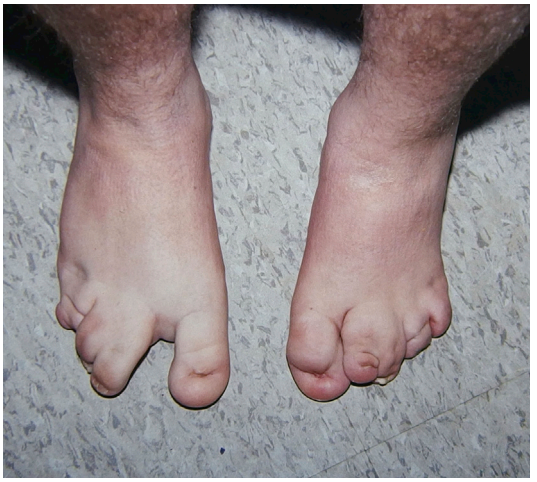

Fig. 10. Feet of a man with popliteal pterygium syndrome. There is triangular tissue over the left great toe and syndactyly.

CRANIOFACIAL DISORDERS

Cleft Lip with or Without Cleft Palate

CLP is among the most common birth defects. In the United States, between the years 2004 and 2006, 4437 infants were born with CLP with an incidence of 1 in 940 live births.[31] The prevalence of CLP depends on race, gender, and socioeconomic factors. Native Americans have the highest prevalence, with 3.6 cases of CLP per 1000 births, compared with Asians, with 1.7 to 2.1 cases per 1000 births, with Africans and African Americans having approximately 1 in 2500 births.[32,33] Boys are more likely to be affected than girls by a 2:1 ratio.[34]

Approximately 30% of cases of CLP are associated with an underlying syndrome or multiple anomalies disorders.[35] The remaining 70% of cases are nonsyndromic. Inheritance is multifactorial, meaning that the condition is caused by both genetic and nongenetic factors. Nongenetic factors may include maternal environment, fetal environment, teratogenic exposures, placental factors, maternal nutritional factors, and as yet several undefined factors.[36] The importance of genetic factors in isolated multifactorial CLP is supported by racial differences in prevalence. In addition, recurrence risk for first-degree relatives of individuals with CLP is elevated to approximately 3% to 5%.[37] This recurrence risk is further elevated as the number of affected individuals in the family increases. In families with 2 affected first-degree relatives, recurrence risk is increased to 5% to 10%. There have been several genes that have been implicated as predisposing to isolated nonsyndromic CLP. In 1 review, 17 genes are listed, including *TGF-A*, *TGF-B3*, *MSX1*, and *IFR6.*[38] Perhaps one of the better studied genes has been *IRF6*, which causes Van der Woude syndrome and popliteal pterygium syndrome. A recent study has demonstrated that 0.24% to

Table 2
Syndromes associated with cleft palate

Disorder	Etiology	Gene	Inheritance	Clinical Features
Craniofacial microsomia (see **Fig. 8**)	Sporadic, possibly vascular disruption		Sporadic, rarely AD	Facial asymmetry, microtia, vertebral anomalies
Fetal alcohol syndrome	Teratogenic		Environmental	Growth retardation, CLP, CP, heart defects, facial dysmorphism, developmental disabilities
Fetal valproate syndrome (see **Fig. 2**)	Teratogenic		Environmental	CP, metopic craniosynostosis, microcephaly, spina bifida, developmental disabilities, facial dysmorphism
Van der Woude syndrome	Single-gene mutation	*IRF6*	AD	Lower lip fistulae (pits), CLP, CP, SMCP
Stickler syndrome (**Fig. 11**)	Single-gene mutation (genetic heterogeneity)	*COL2A1* *COL11A1* *COL11A2*	AD AD AD	Micrognathia, CP, SMCP, myopia, vitreoretinal degeneration (COL2A1 and COL11A1), sensorineural hearing loss, early adult osteoarthritis, facial dysmorphism
Treacher Collins syndrome (**Fig. 12**)	Single-gene mutation (genetic heterogeneity)	*TCOF1* *POLR1D* *POLR1C*	AD AD AR	Micrognathia, hypoplastic zygomas, CP, conductive hearing loss, lower eyelid colobomas
22q11.2 Deletion syndrome (**Fig. 13**)	Chromosomal deletion		AD	Facial dysmorphism, CP, CLP, SMCP, heart defects, developmental disabilities, renal anomalies, psychiatric disorders, hypocalcemia, immunodeficiency
Smith-Lemli-Opitz syndrome	Single-gene mutation	*SLOS*	AR	Growth retardation, microcephaly, CLP, CP, heart defects, genital, syndactyly of toes 2 and 3, anomalies, developmental disabilities
Diabetic embryopathy	Teratogenic		Maternal diabetes	Heart defects, CLP, CP, vertebral anomalies, renal anomalies, brain anomalies
Deletion 1p36 syndrome	Chromosomal deletion		Sporadic	CL, CP, facial dysmorphism, speech apraxia, developmental disability
Branchio-oto-renal syndrome	Single-gene mutation (genetic heterogeneity)	*EYA1* *SIX1* *SIX5*	AD AD AD	Branchial cleft cysts, preauricular fistulae (pits), dysplastic ears, conductive hearing loss, sensorineural hearing loss, CP, SMCP, renal dysplasia, renal cysts, small kidneys
Cornelia de Lange syndrome	Single-gene mutation (genetic heterogeneity)	*NIPBL* *SMC1A* *HDAC8* *RAD21* *SMC3*	AD XLD XLD AD AD	Growth restriction, microcephaly, CP, facial dysmorphism, micrognathia, synophrys, small mouth, limb and digital anomalies, clinodactyly, brachydactyly, oligodactyly, developmental disabilities
Nager syndrome (**Fig. 14**)	Single-gene mutation	*SF3B4*	AD	Growth restriction, micrognathia, CP, conductive hearing loss, hypoplastic zygoma, microtia, thumb aplasia
Loeys-Dietz syndrome (**Fig. 15**)	Single-gene mutation	*TGFBR1* *TGFBR2*	AD AD	Aortic root dilation, scoliosis, joint hypermobility, pectus deformity, narrow arched palate, SMCP, bifid uvula craniosynostosis, mitral valve prolapse

Abbreviations: AD, autosomal dominant; AR, autosomal recessive; XLD, X-linked.

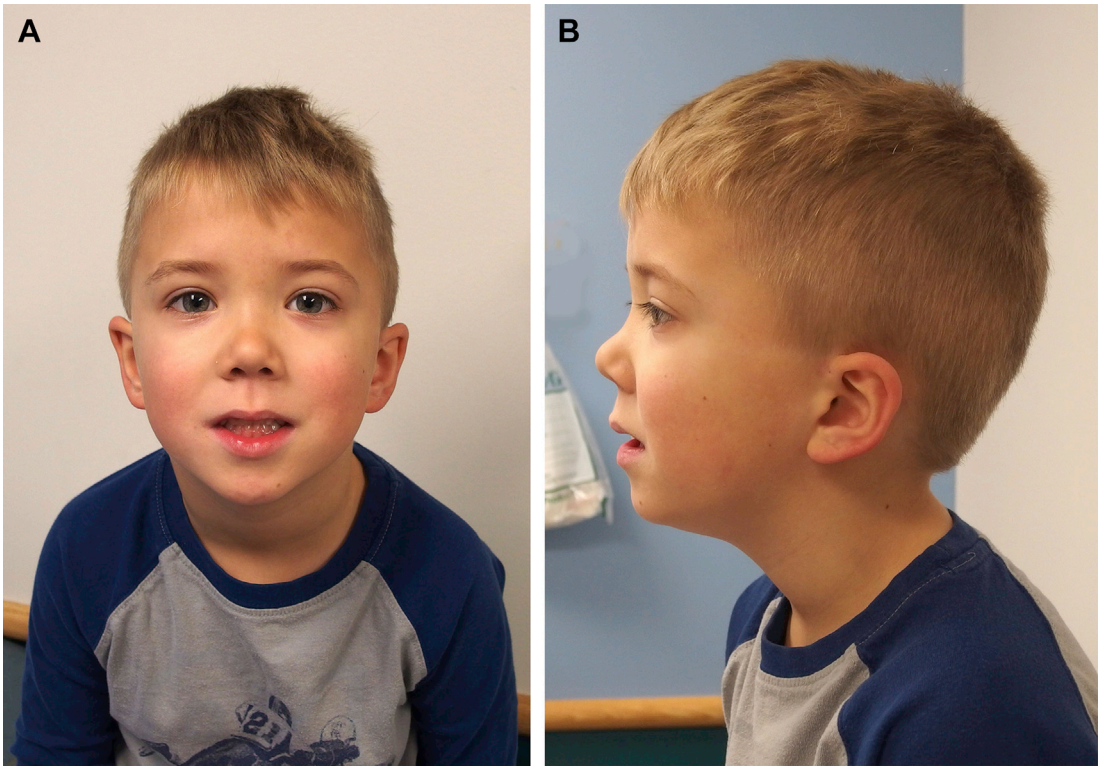

Fig. 11. Young boy with Stickler syndrome. He has a short upturned nose, flat nasal bridge (*A*), and a flat facial profile (*B*).

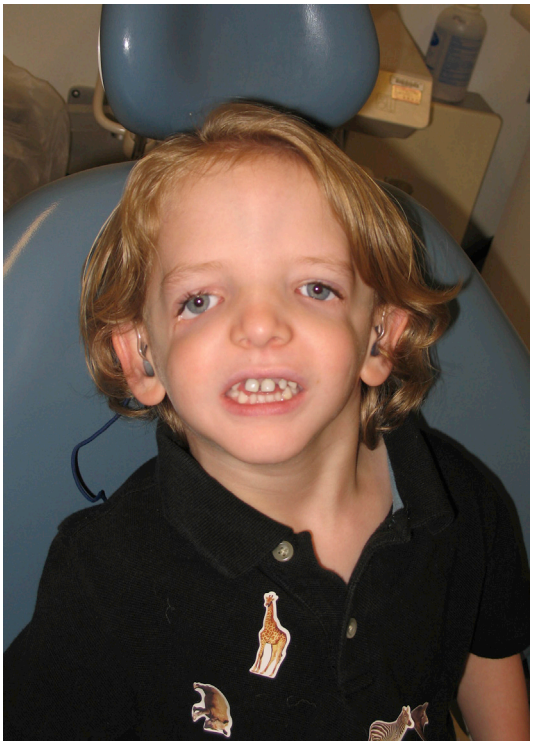

Fig. 12. Young boy with Treacher Collins syndrome caused by a mutation of *TCOF1*. He has micrognathia, down-slanting palpebral fissures, ectropion of lateral lower lids, and low-set ears with conductive hearing loss.

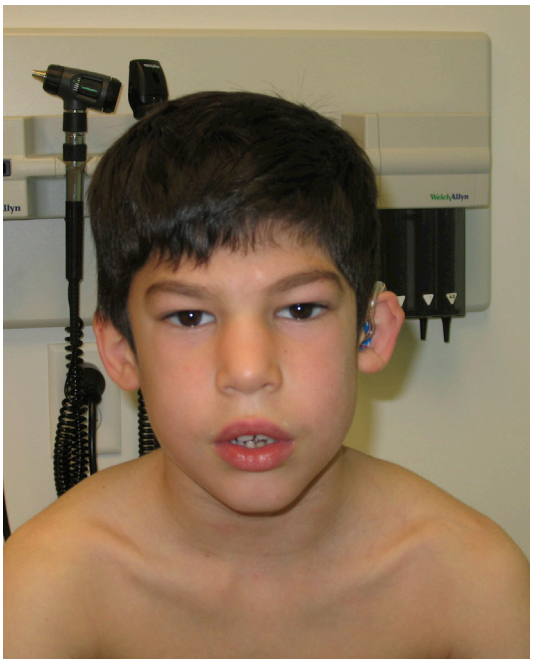

Fig. 13. The 22q11.2 deletion syndrome in a boy. He has an oval-shaped face with prominent nasal tip, prominent nasal pyramid, dysplastic ears, and up-slanting palpebral fissures.

0.44% of isolated multifactorial CLPs are associated with *IRF6* mutations.[39]

Many syndromes have been identified as being associated with CLP. The Online Mendelian Inheritance in Man (OMIM), a catalog of genetic disorders, lists 389 single-gene disorders with CP and 295 disorders with CLP.[40] At Cincinnati Children's Hospital Medical Center, the most common group of disorders associated with CLP is chromosome disorders followed by Opitz syndrome, craniofacial microsomia, diabetic embryopathy, fetal alcohol syndrome and less common disorders such as amniotic band syndrome (**Table 1**, **Figs. 8** and **9**).

Among the most common syndromes associated with both CLP and CP is Van der Woude syndrome, which is the most common single-gene cause of CLP, responsible for 2% of all CLP cases.[41,42] The classic clinical feature of Van der Woude syndrome is the presence of congenital, bilateral, and paramedian lower lip fistulae (pits) (see **Fig. 7**). Other clinical findings may include elevated mounds of the lower lip with a sinus tract leading from a mucous gland of the lip, CLP, CP, or submucous CP (SMCP).[42] Van der Woude syndrome is one of the few genetic disorders in which affected individuals can have CLP or CP. The penetrance of lip pits in Van der Woude syndrome is 86%.[42]

Popliteal pterygium syndrome is a genetic disorder also caused by IRF6 mutations.[42,43] In addition to having lip pits with CLP or CP, individuals with popliteal pterygium syndrome have popliteal pterygia, syndactyly, abnormal external genitalia, intraoral adhesions, and pyramidal skin on the hallux (**Fig. 10**).[42]

Cleft Palate

In the United States, between the years 2004 and 2006, the annual incidence of CP was 1 in 1574 live births.[31] Unlike CLP, there is no racial or ethnic predisposition for CP. CP is more likely to be associated with syndromes and multiple anomaly disorders than CLP. Approximately 50% of individuals with CP have an underlying syndrome or multiple anomaly disorder.[34,35] Although some conditions with CP are common, diagnosis of an underlying syndrome with CP can often be challenging because many of these disorders are rare. SMCP is a microform of CP. In SMCP, there is incomplete fusion of the muscular layers of the

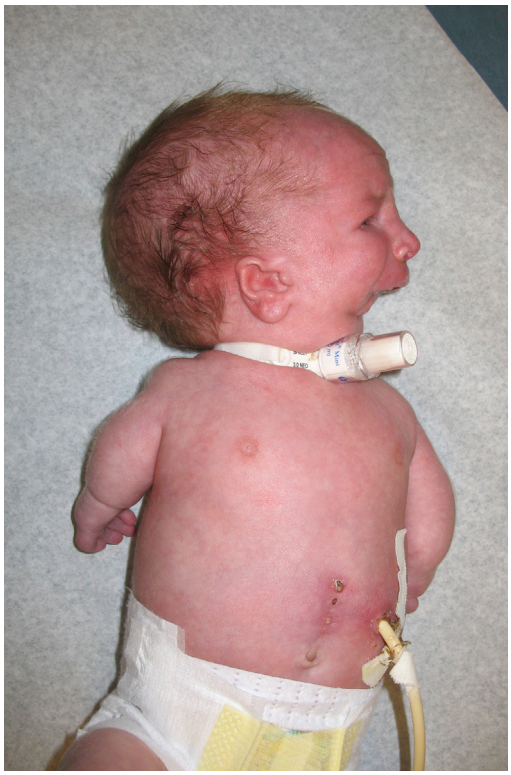

Fig. 14. Infant with Nager syndrome. He has severe micrognathia with PRS and absence of the radii and thumbs.

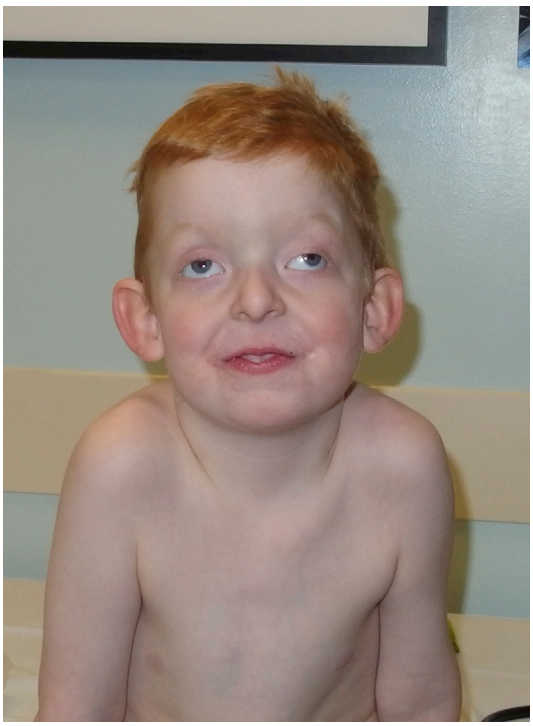

Fig. 15. Boy with Loeys-Dietz syndrome. Note his long oval-shaped face. Individuals with Loeys-Dietz syndrome are at risk for SMCP and craniosynostosis (usually sagittal).

velum (soft palate) with fusion of the overlying mucosa. Presenting features vary ranging from infants with feeding disorders to children with velopharyngeal dysfunction and hypernasal speech. At Cincinnati Children's Hospital Medical Center, the most common disorders associated with CP are Stickler syndrome, 22q11.2 deletion syndrome, fetal alcohol syndrome, and chromosome disorders. See **Table 2** for a list of common CP syndromes (see **Figs. 11** and **13**).

The syndrome most commonly associated with CP is Stickler syndrome. This disorder is genetically heterogeneous, with 6 genes implicated, 3 with autosomal dominant inheritance and 3 with autosomal recessive inheritance.[44] Most cases are autosomal dominantly inherited and associated with mutations in genes for type 2 collagen, *COL2A1*, *COL11A1*, or *COL11A2*. Types I and II Stickler syndrome are caused by mutations in *COL2A1* and *COL11A1*, respectively, and have a similar presentation. The classic features of types I and II Stickler syndrome are micrognathia, RPS, CP, myopia, vitreoretinal degeneration, elevated risk for retinal detachment, early onset of osteoarthritis, and sensorineural hearing loss. Facial features are characterized by micrognathia in infancy with growth of the mandible as the child gets older, flat midface, shallow orbits, flat nasal bridge, and short upturned nose (see **Fig. 11**). Stickler syndrome type III is caused by mutations in *COL11A2* and has similar clinical presentation as types I and II Stickler syndrome with the exception of ocular involvement.[44]

The 22q11.2 deletion syndrome is a common genetic disorder also called velocardiofacial syndrome and DiGeorge syndrome. This disorder is caused by an interstitial deletion of the long arm of chromosome 22.[45] The incidence is approximately 1 in 4000 live births.[46] CP and SMCP are common, seen in approximately 27% of patients. Velopharyngeal dysfunction is also common, even in the absence of CP or SMCP.[47] This condition is also associated with multiple additional anomalies. Congenital heart defects are common, seen in more than 70% of affected individuals. Other significant anomalies are immunodeficiency related to thymus hypoplasia, hypercalcemia secondary to hypoparathyroidism, psychosis and schizophrenia, and developmental disabilities.[48,49] Many patients have typical facial features of narrow face, prominent nasal tip, small mouth, and dysplastic ears (see **Fig. 13**).

Pierre Robin Sequence

PRS is defined by the classic triad of micrognathia, glossoptosis, and obstructive apnea PRS. Patients with PRS frequently have a CP as well (**Fig. 16**). Previous studies have shown that PRS is associated with an underlying syndrome or

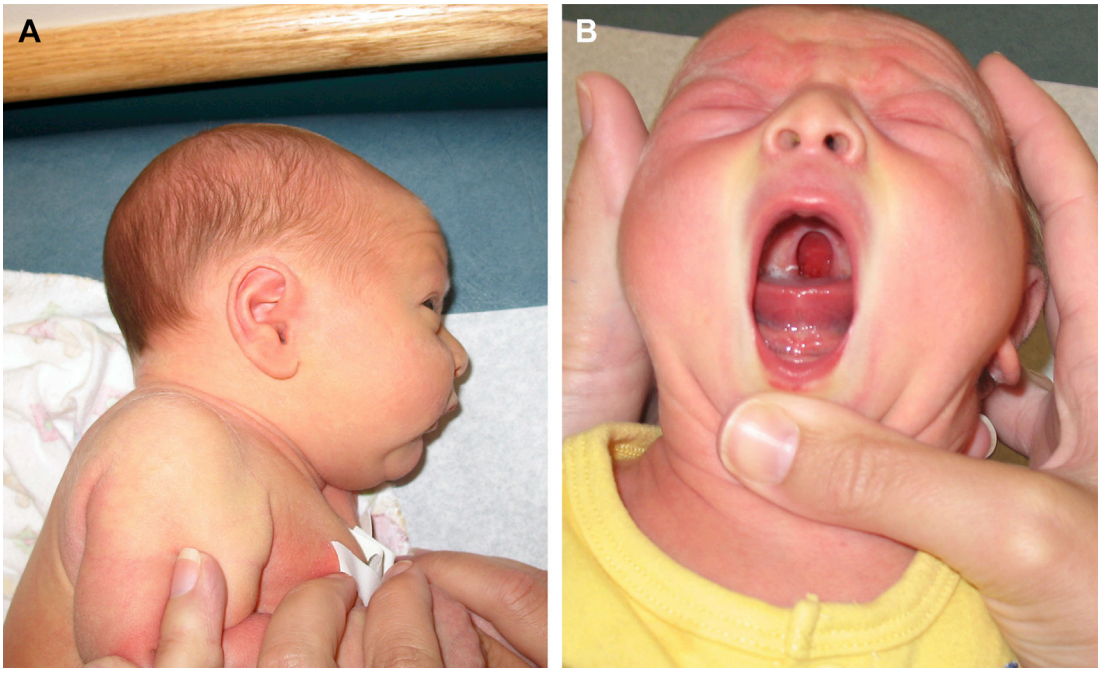

Fig. 16. Infant with PRS. Lateral view shows micrognathia (*A*) and there is a U-shaped cleft of the secondary palate (*B*).

Table 3
Craniosynostosis syndromes

Syndrome	Inheritance	Gene(s)	Chromosome Location	Clinical Features
Antley-Bixler syndrome	AR	*POR*	7q11.23	Coronal and lambdoid synostosis, radiohumeral synostosis, genital anomalies, developmental disabilities
Apert syndrome (**Fig. 17**)	AD	*FGFR2*	10q26.13	Craniosynostosis, syndactyly of hands and feet, midface hypoplasia, developmental disabilities
Baller-Gerold syndrome	AR	*RECQLR*	8q24.3	Craniosynostosis, radial aplasia
Carpenter syndrome	AR	*RAB23*	6p22.1-p11.2	Acrocephaly, craniosynostosis, brachydactyly, syndactyly, preaxial polydactyly, developmental disabilities
Craniofrontonasal dysplasia	XLD	*EFNB1*	Xq13.1	Female: craniofrontonasal dysplasia, craniofacial asymmetry, bifid nasal tip. Male: hypertelorism
Craniosynostosis FGFR3 mutation (Muenke syndrome)	AD	*FGFR3*	4p16.3	Coronal craniosynostosis
Crouzon syndrome (**Fig. 18**)	AD	*FGFR2*	10q26.13	Coronal synostosis, maxillary hypoplasia, mandibular prognathism, exophthalmos
Crouzon syndrome with acanthosis nigricans	AD	*FGFR3*	4p16.3	Coronal synostosis, maxillary hypoplasia, mandibular prognathism, exophthalmos, acanthosis nigricans
Fetal methotrexate syndrome	Environmental Teratogenic	NA	NA	Craniosynostosis, cleft lip, CP, limb anomalies, syndactyly, brain anomalies, developmental disabilities
Fetal valproate syndrome (see **Fig. 2**)	Environmental Teratogenic	NA	NA	Metopic suture synostosis, spina bifida, microcephaly, CP, developmental disability
Hyperthyroidism	Environmental Teratogenic	NA	NA	Craniosynostosis. May be neonatal (maternal hyperthyroidism or Graves disease) or acquired hyperthyroidism
Hypophosphatasia, perinatal and infantile	AR	*ALPL*	1p36.12	Poorly mineralized bone, coronal synostosis, sagittal synostosis, metopic synostosis, limb deformities, narrow thorax, respiratory distress
Jackson-Weiss syndrome	AD	*FGFR2*	10q26.13	Craniosynostosis, flat midface, hypertelorism, exophthalmos, broad deviated great toe
Loeys-Dietz syndrome (see **Fig. 15**)	AD	*TGFBR1* *TGFBR2*	9q22.33 3p24.1	Aortic root dilation, scoliosis, joint hypermobility, pectus deformity, narrow arched palate, SMCP, bifid uvula craniosynostosis, mitral valve prolapse

(*continued on next page*)

Table 3
(*continued*)

Syndrome	Inheritance	Gene(s)	Chromosome Location	Clinical Features
Pfeiffer syndrome	AD	*FGFR1* *FGFR2*	8p11.23 1026.13	Type 1: craniosynostosis (coronal), broad thumbs and great toes, maxillary hypoplasia. Type 2: cloverleaf skull, broad thumbs and great toes. Type 3: craniosynostosis (coronal), ankyloses of elbows, tracheobronchial anomalies.
Saethre-Chotzen syndrome (**Fig. 19**)	AD	*TWIST*	7p21.1	Craniosynostosis, ptosis, folded ear pinna, broad great toes
Shprintzen-Goldberg syndrome	AD	*SKI*	1p36.33-p36.32	Craniosynostosis, marfanoid body habitus, aortic root dilation, scoliosis, pectus deformity, developmental disabilities

Abbreviations: AD, autosomal dominant; AR, autosomal recessive; NA, not applicable; XLD, X-linked dominant.

multiple anomaly disorder in more than 50% of cases.[50,51] A recent study has shown that in patients with PRS both with and without CP, 54% had syndromes.[52] The most common syndrome was Stickler syndrome, followed by Treacher Collins syndrome (see **Fig. 12**), arthrogryposis multiplex congenital, and chromosome disorders.

Craniosynostosis

Craniosynostosis is the premature fusion of 1 or more of the cranial sutures.[53] The incidence is between 1 in 2000 to 2500.[54,55] Craniosynostosis can be syndromic or nonsyndromic. Approximately 85% of all cases of craniosynostosis are nonsyndromic. The etiologies of nonsyndromic craniosynostoses are complex and these seem to be heterogeneous.[55] Multiple genetic and teratogenic causes of craniosynostosis, however, have been identified. The OMIM recognizes 183 single-gene disorders associated with craniosynostosis.[56] Teratogenic causes include valproate

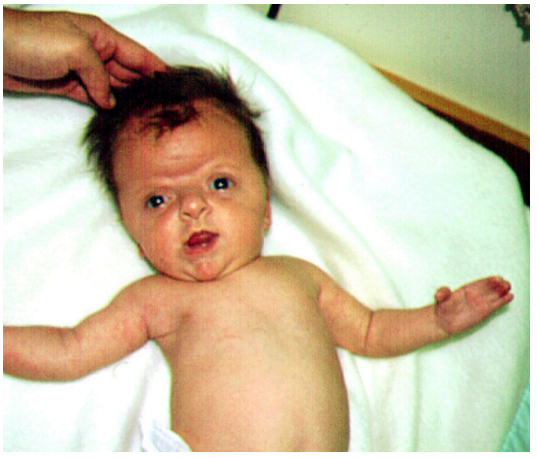

Fig. 17. Infant with Apert syndrome. She has bicoronal craniosynostosis with high forehead, depressed nasal bridge, and beaked nose. She also has the typical syndactyly with deviated thumb.

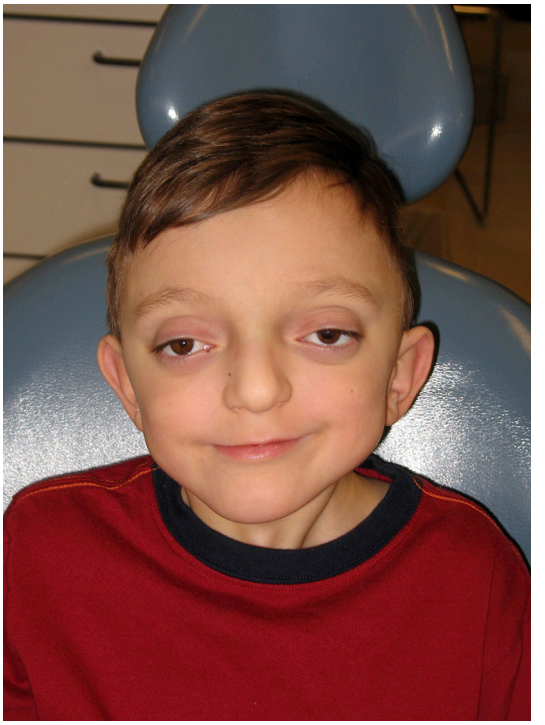

Fig. 18. Boy with Crouzon syndrome. He has shallow orbits and exophthalmos.

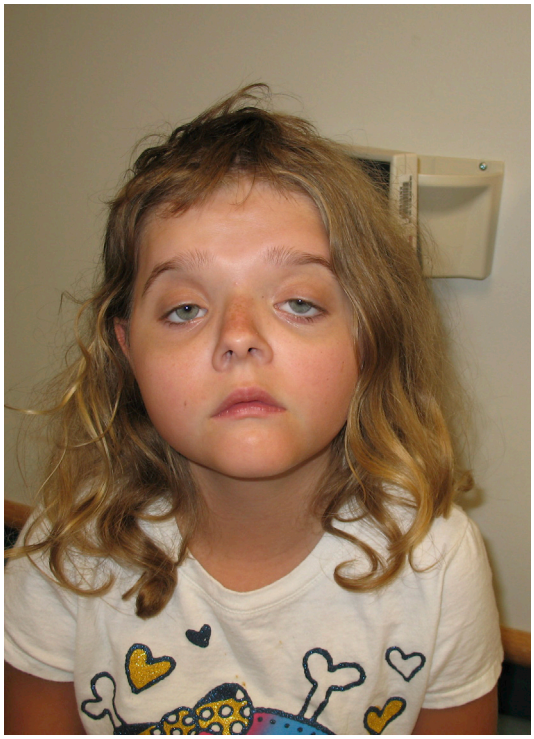

Fig. 19. Girl with Saethre-Chotzen syndrome who was born with bicoronal craniosynostosis. Note the downslanting palpebral fissures, posteriorly rotated ears, and upturned nose.

embryopathy,[57] which causes metopic suture synostosis, and hyperthyroidism, both neonatal and acquired. Because there is variable expression among the different craniosynostosis syndromes, genetic testing is often required to confirm diagnosis and to give accurate recurrence risks. See **Table 3** for a list of the common craniosynostosis syndromes (see **Figs. 17–19**).

REFERENCES

1. Leppig KA, Werler MM, Cann CI, et al. Predictive value of minor anomalies. I. Association with major malformations. J Pediatr 1987;110(4):531–7.
2. American Cleft Palate –Craniofacial Association. Standards for Cleft Palate and Craniofacial Teams. 2016. Available at: http://www.acpa-cpf.org/team_care/standards.
3. Liu S, Rouleau J, Leon JA, et al. Impact of prepregnancy diabetes mellitus on congenital anomalies, Canada, 2002-2012. Health Promot Chronic Dis Prev Can 2015;35(5):79–84.
4. Kesby G. Repeated adverse fetal outcome in pregnancy complicated by uncontrolled maternal phenylketonuria. J Paediatr Child Health 1999;35(5):499–502.
5. Zimmerman D. Fetal and neonatal hyperthyroidism. Thyroid 1999;9(7):727–33.
6. Wayne C, Cook K, Sairam S, et al. Sensitivity and accuracy of routine antenatal ultrasound screening for isolated facial clefts. Br J Radiol 2002;75(895):584–9.
7. Maarse W, Berge SJ, Pistorius L, et al. Diagnostic accuracy of transabdominal ultrasound in detecting prenatal cleft lip and palate: a systematic review. Ultrasound Obstet Gynecol 2010;35(4):495–502.
8. Lind K, Aubry MC, Belarbi N, et al. Prenatal diagnosis of Pierre Robin Sequence: accuracy and ability to predict phenotype and functional severity. Prenat Diagn 2015;35(9):853–8.
9. Saleem SN. Fetal MRI: An approach to practice: a review. J Adv Res 2014;5(5):507–23.
10. Dorrie N, Focker M, Freunscht I, et al. Fetal alcohol spectrum disorders. Eur Child Adolesc Psychiatry 2014;23(10):863–75.
11. Manning MA, Eugene Hoyme H. Fetal alcohol spectrum disorders: a practical clinical approach to diagnosis. Neurosci Biobehav Rev 2007;31(2):230–8.
12. Munger RG, Romitti PA, Daack-Hirsch S, et al. Maternal alcohol use and risk of orofacial cleft birth defects. Teratology 1996;54(1):27–33.
13. Lammer EJ, Chen DT, Hoar RM, et al. Retinoic acid embryopathy. N Engl J Med 1985;313(14):837–41.
14. Schorry EK, Oppenheimer SG, Saal HM. Valproate embryopathy: clinical and cognitive profile in 5 siblings. Am J Med Genet Part A 2005;133A(2):202–6.
15. Jentink J, Dolk H, Loane MA, et al. Intrauterine exposure to carbamazepine and specific congenital malformations: systematic review and case-control study. BMJ 2010;341:c6581.
16. Hanson JW, Smith DW. The fetal hydantoin syndrome. J Pediatr 1975;87(2):285–90.
17. Hyoun SC, Obican SG, Scialli AR. Teratogen update: methotrexate. Birth Defects Res A Clin Mol Teratol 2012;94(4):187–207.
18. Lam PK, Torfs CP. Interaction between maternal smoking and malnutrition in infant risk of gastroschisis. Birth Defects Res A Clin Mol Teratol 2006;76(3):182–6.
19. Honein MA, Paulozzi LJ, Watkins ML. Maternal smoking and birth defects: validity of birth certificate data for effect estimation. Public Health Rep 2001;116(4):327–35.
20. Honein MA, Rasmussen SA, Reefhuis J, et al. Maternal smoking and environmental tobacco smoke exposure and the risk of orofacial clefts. Epidemiology 2007;18(2):226–33.
21. Carmichael SL, Ma C, Rasmussen SA, et al. Craniosynostosis and maternal smoking. Birth Defects Res A Clin Mol Teratol 2008;82(2):78–85.
22. Beaty TH, Taub MA, Scott AF, et al. Confirming genes influencing risk to cleft lip with/without cleft palate in a case-parent trio study. Hum Genet 2013;132(7):771–81.

23. Manning M, Hudgins L. Array-based technology and recommendations for utilization in medical genetics practice for detection of chromosomal abnormalities. Genet Med 2010;12(11):742–5.
24. Hayeems RZ, Hoang N, Chenier S, et al. Capturing the clinical utility of genomic testing: medical recommendations following pediatric microarray. Eur J Hum Genet 2015;23(9):1135–41.
25. Genes G. 2015. Available at: https://globalgenes.org/rare-diseases-facts-statistics/. Accessed February 1, 2016.
26. Biesecker LG, Green RC. Diagnostic clinical genome and exome sequencing. N Engl J Med 2014;370(25):2418–25.
27. Sawyer SL, Hartley T, Dyment DA, et al. Utility of whole-exome sequencing for those near the end of the diagnostic odyssey: time to address gaps in care. Clin Genet 2016;89(3):275–84.
28. Yang Y, Muzny DM, Reid JG, et al. Clinical whole-exome sequencing for the diagnosis of mendelian disorders. N Engl J Med 2013;369(16):1502–11.
29. Miyamoto RC, Cotton RT, Rope AF, et al. Association of anterior glottic webs with velocardiofacial syndrome (chromosome 22q11.2 deletion). Otolaryngol Head Neck Surg 2004;130(4):415–7.
30. Prada CE, Hufnagel RB, Hummel TR, et al. The use of magnetic resonance imaging screening for optic pathway gliomas in children with neurofibromatosis type 1. J Pediatr 2015;167(4):851–6.e1.
31. Parker SE, Mai CT, Canfield MA, et al. Updated National Birth Prevalence estimates for selected birth defects in the United States, 2004-2006. Birth Defects Res A Clin Mol Teratol 2010;88(12):1008–16.
32. Croen LA, Shaw GM, Wasserman CR, et al. Racial and ethnic variations in the prevalence of orofacial clefts in California, 1983-1992. Am J Med Genet 1998;79(1):42–7.
33. Suleiman AM, Hamzah ST, Abusalab MA, et al. Prevalence of cleft lip and palate in a hospital-based population in the Sudan. Int J Paediatr Dent 2005;15(3):185–9.
34. Tolarova MM, Cervenka J. Classification and birth prevalence of orofacial clefts. Am J Med Genet 1998;75(2):126–37.
35. Jugessur A, Farlie PG, Kilpatrick N. The genetics of isolated orofacial clefts: from genotypes to subphenotypes. Oral Dis 2009;15(7):437–53.
36. Stanier P, Moore GE. Genetics of cleft lip and palate: syndromic genes contribute to the incidence of non-syndromic clefts. Hum Mol Genet 2004;13(Spec No 1):R73–81.
37. Sivertsen A, Wilcox AJ, Skjaerven R, et al. Familial risk of oral clefts by morphological type and severity: population based cohort study of first degree relatives. BMJ 2008;336(7641):432–4.
38. Rahimov F, Jugessur A, Murray JC. Genetics of non-syndromic orofacial clefts. Cleft Palate Craniofac J 2012;49(1):73–91.
39. Leslie EJ, Koboldt DC, Kang CJ, et al. IRF6 mutation screening in non-syndromic orofacial clefting: analysis of 1521 families. Clin Genet 2015;90(1):28–34.
40. Online Mendelian Inheritance in Man, OMIM®. McKusick-Nathans Institute of Genetic Medicine, Johns Hopkins University (Baltimore, MD), January 5, 2016. Available at: http://omim.org/.
41. Murray JC, Daack-Hirsch S, Buetow KH, et al. Clinical and epidemiologic studies of cleft lip and palate in the Philippines. Cleft Palate Craniofac J 1997;34(1):7–10.
42. Schutte BC, Saal HM, Goudy S, et al. IRF6-Related Disorders. 2003 Oct 30 [Updated 2014 Jul 3]. In: Pagon RA, Adam MP, Ardinger HH, et al, editors. GeneReviews® [Internet]. Seattle (WA): University of Washington, Seattle; 1993–2016. Available from: http://www.ncbi.nlm.nih.gov/books/NBK1407/.
43. Lees MM, Winter RM, Malcolm S, et al. Popliteal pterygium syndrome: a clinical study of three families and report of linkage to the Van der Woude syndrome locus on 1q32. J Med Genet 1999;36(12):888–92.
44. Robin NH, Moran RT, Ala-Kokko L. Stickler Syndrome. 2000 Jun 9 [Updated 2014 Nov 26]. In: Pagon RA, Adam MP, Ardinger HH, et al, editors. GeneReviews® [Internet]. Seattle (WA): University of Washington, Seattle; 1993–2016. Available from: http://www.ncbi.nlm.nih.gov/books/NBK1302/.
45. Goldmuntz E, Driscoll D, Budarf ML, et al. Microdeletions of chromosomal region 22q11 in patients with congenital conotruncal cardiac defects. J Med Genet 1993;30(10):807–12.
46. Goodship J, Cross I, LiLing J, et al. A population study of chromosome 22q11 deletions in infancy. Arch Dis Child 1998;79(4):348–51.
47. McDonald-McGinn DM, Kirschner R, Goldmuntz E, et al. The Philadelphia story: the 22q11.2 deletion: report on 250 patients. Genet Couns 1999;10(1):11–24.
48. Bassett AS, McDonald-McGinn DM, Devriendt K, et al. Practical guidelines for managing patients with 22q11.2 deletion syndrome. J Pediatr 2011;159(2):332–9.e1.
49. Hacihamdioglu B, Hacihamdioglu D, Delil K. 22q11 deletion syndrome: current perspective. Appl Clin Genet 2015;8:123–32.
50. Tomaski SM, Zalzal GH, Saal HM. Airway obstruction in the Pierre Robin sequence. Laryngoscope 1995;105(2):111–4.
51. Izumi K1 KL, Mitchell AL, Jones MC. Underlying genetic diagnosis of Pierre Robin sequence: retrospective chart review at two children's hospitals and a systematic literature review. J Pediatr 2012;160(4):645–50.

52. Weaver KN, Zhang X, Bender PL, et al. Diagnosis, treatment and outcomes of Robin sequence at Cincinniati Children's. American Cleft Palate-Craniofacial Association Annual Meeting. Scottsdale, AZ, April 24, 2015.
53. Cohen MM, MacLEan RE. Craniosynostosis: diagnosis, evaluation, and management. 2nd edition. New York: Oxford University Press; 2000.
54. Boulet SL, Rasmussen SA, Honein MA. A population-based study of craniosynostosis in metropolitan Atlanta, 1989-2003. Am J Med Genet Part A 2008;146A(8):984–91.
55. Heuze Y, Holmes G, Peter I, et al. Closing the gap: genetic and genomic continuum from syndromic to nonsyndromic craniosynostoses. Curr Genet Med Rep 2014;2(3):135–45.
56. Online Mendelian Inheritance in Man, OMIM®. Baltimore, MD: McKusick-Nathans Institute of Genetic Medicine; Johns Hopkins University; 2016. Available at: http://omim.org/.
57. Jentink J, Loane MA, Dolk H, et al. Valproic acid monotherapy in pregnancy and major congenital malformations. N Engl J Med 2010;362(23):2185–93.