MOLECULAR GENETICS OF CLEFT LIP AND PALATE: A REVIEW

G.O. Oboli†, D.I. Chukwuma‡, O.F. Fagbule§, E.O. Abe∥ and A.O. Adisa†,∥

1. College of Medicine, University of Ibadan, Ibadan
2. Department of Periodontology and Community Dentistry, University College Hospital, Ibadan
3. Department of Oral Pathology and Oral Medicine, University College Hospital, Ibadan

Correspondence:
Dr. EO Abe
Department of Oral Pathology and Oral Medicine, University College Hospital, Ibadan, Nigeria.
Email: eoabe83@yahoo.co.uk

INTRODUCTION

Cleft lip with or without cleft palate (CLP) is a common congenital disability. They exist either in combination with one or more other anomalies (syndromic cleft) or in isolation (non-syndromic cleft). Non-syndromic CL/P is more common as it is present in about 70% of cases, out of which 80% are sporadic, and 20% are familial.1 CLP which is commoner in males, occurs in 1 out of 300 to 2500 births, while isolated cleft palate (CP) which occurs more frequently in females, occurs in 1 out of 1500 births.2,3 People with cleft lip and palate often require multidisciplinary care involving several surgical repairs commencing in the first year of life, orthodontic interventions for malocclusion, speech therapy, treatment of recurrent middle ear infections, and psychological interventions. These have been noted to contribute a significant burden to the patient, family, and society at large. Thus, an intense effort has been made to unravel its aetiology, which would be important in genetic counselling, risk prediction, and overall prevention of cleft lip and palate.4

Aetiology of Cleft Lip and Palate

Generally, cleft lip and palate is thought to result from interactions between genetic and environmental factors. Substantial pieces of evidence for the former have arisen from family, and twin studies which revealed high rates of familial aggregation and increased concordance rates in monozygous twins, compared with dizygous twins.5 For instance, studies by Sivertsen et al.6 and Grosen et al.7 showed that cleft palate has a relative risk of occurrence which is 15 to 56 times higher among first degree relatives. Although environmental factors such as maternal use of alcohol, cigarette and antiepileptic drugs have been identified as risk factors for CLP, recent studies have now revealed important genes either acting alone or within gene networks. Such cases are found as parts of Mendelian monogenic syndromes, chromosomal abnormalities, or otherwise unknown genetic syndromes.8 These identified genetic risk factors have shed more light on normal craniofacial development with some also implicated in non-syndromic CL/P. As an example of gene-environment interaction, Shaw et al.9 demonstrated a 3 to 8 fold increase in CLP in babies with lack of multivitamins in the first trimester of pregnancy and the TaqI C2 mutation in the Tgfa gene. The same mutation was shown to raise the risk of CLP by 6 to 8 times when co-existent with maternal smoking10, while Jugessur et al.11 found that combined mutations of the Tgfa and Msx1 genes cause an almost ten-fold increase in cleft lip and palate risk as an evidence of gene-gene interaction.

Genetic Regulation of Craniofacial Development

Craniofacial development is a complex event involving several transcription factors and molecular signals. Disruptions in the network of these proteins lead to the development of facial clefts. The diversity in the functions of these genes and their products shows the susceptibility of the craniofacial developmental pathways to form clefts4.

Facial development in humans begins in the fourth week of intrauterine life with the migration of cranial neural crest cells (CNC) from the rostral part of the neural tube to form the facial primordia and secondary palate. Genes such as Tgfb2, Hoxa2, Gli2, and Gli3 have been identified to play a role in CNC migration, mutations of which have been shown to contribute to cleft lip and palate in mice.12-14 Palatal shelves are subsequently derived from the secondary palate and undergo elevation to become horizontally apposed in the midline. Failure of apposition has been linked with mutations in the genes Mxc1, Pax9 and Lhx8 leading to CP.15-17 Furthermore, epithelial-mesenchymal interactions mediated by interrelated gene networks – sonic hedgehog (Shh), bone morphogenetic proteins
Table 1: Summary of molecular genetic mechanisms in syndromic cleft lip and palate

| Syndrome                                                                 | Inheritance | Gene  | Locus     | Function                                                                 | Gene also implicated in non-syndromic CL/P | References |
|--------------------------------------------------------------------------|-------------|-------|-----------|--------------------------------------------------------------------------|--------------------------------------------|------------|
| Cleft lip/palate ectodermal dysplasia syndrome (CL/PED)                  | AR          | Per1  | 11q23.3   | Encodes nectin-1 which plays a role in cell adhesion                    | Yes                                        | 28-30      |
| Acrofrontofacial dysostosis syndrome                                      | AR          | Nbas  | 2p24      | Skeletal morphogenesis, mediating Golgi-to-endoplasmic reticulum retrograde traffic. | -                                          | 31         |
| Popliteal pterygium syndrome (PPS)                                        | AD          | Irf6  | 1q32      | Mediates TGFβ3 activity in palatal fusion                               | Yes                                        | 25,27      |
| Van der Woude (VDW) syndrome                                              | AD          | Irf6  | 1q32      | Mediates TGFβ3 activity in palatal fusion                               | Yes                                        | 25,27      |
| Rapp-Hodgkin syndrome                                                    | AD          | Tp63  | 3q28      | Apoptosis. Also in establishment of enhancers needed for expression of genes important in craniofacial development such as Irf6 | Yes                                        | 32-24      |
| Roberts syndrome                                                         | AR          | Eso2  | 8p21      | Acetyltransferase activity necessary for sister chromatid cohesion needed for cell proliferation | -                                          | 35,36      |
| Hay-Wells syndrome                                                        | AD          | Tp63  | 3q28      | As for RHS                                                               | Yes                                        | 37         |
| Blepharocheidodontic syndrome                                             | AD          | Cdhh  | 16q22     | Cell adhesion molecule involved in the maintenance of epithelial cell morphology during embryonic development | -                                          | 38-39      |
| Thurston syndrome                                                        | AR          | Ddx59 | 1q32      | Ciliary SHH signaling                                                   | -                                          | 40         |
| Uvealcoloboma-cleft lip and palate-intellectual disability syndrome      | AD          | Yap1  | 11q22     | Activation of transcription factors important for apoptosis such as p73 | -                                          | 41,42      |
| Varadi-Papp syndrome                                                     | AR          | Cplane1 | 5p13   | Ciliary SHH signaling                                                   | -                                          | 43,44      |
| Left palate, cardiac defects and mental retardation (CPCR)               | AD          | Meis2 | 15q14     | Palatal fusion. Repression of SHH/FGF feedback loop.                    | -                                          | 45,46      |
| Vici syndrome                                                            | AR          | Epfg  | 1q32      | Autophagy during embryogenesis                                           | -                                          | 47         |
| Ectrodactyly, ectodermal dysplasia, and cleft lip/palate syndrome 3 (EED3) | AD          | Tp63  | 3q28      | As for RHS                                                               | -                                          | 48         |
| Brachiocephalic syndrome                                                  | AD          | Tcfap2a | 6q24   | Transcription activation necessary for formation of neural crest cells during embryogenesis | -                                          | 49-51      |
| Left palate with ankyloglossia, X-linked (CPX)                           | X-linked    | Tloc22 | Xq21     | Repressor of transcription, with an important role in horizontal elevation of palatal shelves | -                                          | 52,53      |
| Holoprosencephaly 2                                                       | AD          | Sic3  | 2p21      | Regulation of SHH expression                                             | -                                          | 54,55      |
| Optiz-Frias syndrome or (Optiz GBBB syndrome type II)                    | AD          | Spec1 | 22q11.23  | Regulates microtubule and actin organization for proper cell adhesion and migration | -                                          | 56,57      |
| Simpson-Golabi-Behmel syndrome type 1                                     | XLR         | Gpc3  | 2q26.2    | Regulation of SHH, FGF, and BMP activities                               | -                                          | 58,59      |
| Oral-facial-digital syndrome 1                                            | XLD         | Ojdh1 | Xp22.22   | Regulation of microtubule function                                       | -                                          | 60,61      |
| Gorlin-Goltz syndrome                                                    | AD          | Psh1, Psh2, Sufu | 9q22, 1p32, 10q24 | Regulation of SHH signaling                                             | -                                          | 62-64      |
| Waardenburg syndrome, type 1                                              | AD          | Pac3  | 2q36      | Transcription factor necessary for skeletal muscle formation            | -                                          | 65,66      |
| CHARGE syndrome                                                          | AD          | Cbl7  | 8q12      | Transcription factor necessary for neural crest cell migration          | -                                          | 67,68      |
| DiGeorge syndrome                                                        | AD          | Tbx1  | 22q11.21  | Regulator of BMP signaling                                              | -                                          | 69         |

(Bmp), and fibroblast growth factors (Fgf) – are essential in normal palatal development\(^1\). For example, expression of Shh in the palatal epithelium is regulated by Bmp4 in the mesenchyme. Shh then regulates Bmp2 in the mesenchyme, which is essential for mesenchymal proliferation\(^1,2\). A positive feedback loop also exists.
between the fibroblast growth factor Fgf10 and Shh expression in the palatal mesenchyme and epithelium, respectively\textsuperscript{19,21}. Also, the homeobox gene Msx1 further modulates the expression of the genes Bmp4, Shh, and Bmp2 above. At week 12, development of the palate is completed in humans.

Genetic Analysis of Cleft Lip and Palate

Almost 500 syndromes have been identified in syndromic cleft lip and palate\textsuperscript{22}, although not all have been linked to specific genes. Cohen\textsuperscript{23} published a review of 154 of these syndromes with their clinical features to aid diagnosis. However, recent molecular genetic analysis has identified the loci of these mutations and functions of the implicated genes. For example, popliteal pterygium and Van der Woude syndromes, the latter being the most common cause of syndromic cleft lip and palate\textsuperscript{24}, are both autosomal dominant conditions secondary to mutations in the interferon regulatory factor-6 (Irf6) gene on chromosome 1q32\textsuperscript{25}. Interestingly, mutations in Irf6 have also been found in non-syndromic cleft lip and palate\textsuperscript{26}. The protein product IRF6 is now known to be a transcription factor up-regulated by TGFb3 protein in palatal fusion during embryonic development in humans\textsuperscript{27}. In syndromic cleft lip and palate, a given gene may be affected by several different mutations, which accounts for the varied phenotypes that may be observed\textsuperscript{4}. For instance, mutations of the C-terminus of the protein TP63 results in cleft lip or cleft palate, whereas mutations of the conserved DNA binding region at the N-terminus results in cleft lip and palate\textsuperscript{8}.

We conducted a search on the Online Mendelian Inheritance in Man (OMIM) database with keywords ‘cleft lip’ and ‘cleft palate’ which produced over 1500 results. Table 1 summarizes genes implicated in some syndromic cleft lip and palate.

Detection of genes in non-syndromic cleft lip and palate (summarized in Table 2) has been done in recent decades by various methods including linkage analysis, candidate gene approach, and genome-wide association studies (GWAS), with the discovery of shared genetic lesions between syndromic and non-syndromic cleft lip and palate\textsuperscript{70}.

CONCLUSION

There has been some success in elucidating the genetic basis of cleft lip and palate with the identification of numerous susceptibility genes. However, this number is bound to increase, revealing the overall genetic complexity of craniofacial clefts. Given the role of environmental factors, studies that further explore feto-maternal genetics together with exposure to different environmental factors could aid in the development of a weighted genetic risk assessment for cleft lip and palate which in turn would better inform genetic counselling and prescription of preventive measures.

REFERENCES

1. Basha M, Benedicte D, Revencu N, et al. Whole exome sequencing identifies mutations in 10% of familial non-syndromic cleft lip and/or palate in genes mutated in well-known syndromes. J Med Genet. 2018;55(7):449–458.
2. Fraser F. The Genetics of Cleft Lip and Cleft Palate. Am J Hum Genet. 1970;22(3):336–352.
3. Wyszynski DF, Beatty TH, Maestri NE. Genetics of Nonsyndromic Oral Clefts Revisited. Cleft Palate-Craniofacial J. 1996;33(5):406–417.
4. Kohli SS, Kohli VS. A comprehensive review of the genetic basis of cleft lip and palate. J Oral Maxillofac Pathol. 2012;16(1):64–72.
5. Mitchell L, Risch N. Mode of inheritance of nonsyndromic cleft lip with or without cleft palate:

| Non-syndromic CLP | Inheritance | Gene | Locus | Function | References |
|-------------------|-------------|------|-------|----------|------------|
| Orofacial cleft 1 | AD          | Ofc1 | 6p24  | -        | 71         |
| Orofacial cleft 5 | -           | Msx1 | 4p16  | Homeobox gene controlling expression of downstream genes, involved in patterning of the face and palatal midline apposition. | 72-73 |
| Orofacial cleft 6 | AD          | Irf6 | 1q32  | As in VDW and PPS | 74 |
| Orofacial cleft 8 | -           | Tp63 | 3q28  | Apoptosis. Also in establishment of enhancers needed for expression of genes important in craniofacial development such as Irf6 | 32-34 |
| Orofacial cleft 11| -           | Bmp4 | 14q22 | Encodes Bmp4 which up-regulates Msx1 and SHH for palatal fusion. | 19,75,76 |

Table 2: Summary of molecular genetic mechanisms in non-syndromic cleft lip and palate
a reanalysis. Am J Hum Genet. 1992;51(2):323–332.
6. 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.
7. Grosen D, Chevrier C, Skytte A, et al. A cohort study of recurrence patterns among more than 54,000 relatives of oral cleft cases in Denmark: support for the multifactorial threshold model of inheritance. J Hum Genet. 2010;47(3):162–168.
8. 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(1):73–81.
9. Shaw GM, Wasserman CR, Murray JC, Lammer EJ. Infant TGF-Alpha Genotype, Orofacial Clefts, and Maternal Periconceptional Multivitamin Use. Cleft Palate-Craniofacial J. 1998;35(4):366–370.
10. Hwang SJ, Beatty TH, Panny SR, et al. Association study of transforming growth factor alpha (TGF alpha) Taq1 polymorphism and oral clefts: indication of gene-environment interaction in a population-based sample of infants with birth defects. Am J Epidemiol. 1995;141(7):629–636.
11. Jugessur A, Lie RT, Wilcox AJ, et al. Variants of developmental genes (TGFA, TGFB3, and MSX1) and their associations with orofacial clefts: A case-parent triad analysis. Genet Epidemiol. 2003;24(3):230–239.
12. Mo R, Freer A, Zynyk D, et al. Specific and redundant functions of Gli2 and Gli3 zinc finger genes in skeletal patterning and development. Development. 1997;124(13):113–123.
13. Rijli FM, Mark M, Lakkaraju S, et al. A homeotic transformation is generated in the rostral branchial region of the head by disruption of Hoxa-2, which acts as a selector gene. Cell. 1993;75(7):1333–1349.
14. Sanford PL, Ormsby I, Gittenberg-de Groot AC, et al. TGFbeta2 knockout mice have multiple developmental defects that are non-overlapping with other TGFbeta knockout phenotypes. Development. 1997;124(13):2659–2670.
15. Peters H, Neubuser A, Kratochwil K, Balling R. Pax-9 deficient mice lack pharyngeal pouch derivatives and teeth and exhibit craniofacial and limb abnormalities. Genes Dev. 1998;12(17):2735–2747.
16. Satokata I, Maas R. Msx1 deficient mice exhibit cleft palate and abnormalities of craniofacial and tooth development. Nat Genet. 1994;6(4):348–356.
17. Zhao Y, Guo YJ, Tomac A, et al. Isolated cleft palate in mice with a targeted mutation of the LIM homeobox gene lhx8. Proc Natl Acad Sci USA. 1999;96(26):15002–15006.
18. Burg ML, Chai Y, Yao CA, Iii WM, Figueiredo JC. Epidemiology, Etiology, and Treatment of Isolated Cleft Palate. Front Physiol. 2016;7 (March) :1–16.
19. Chai Y, Masson R. Recent advances in craniofacial morphogenesis. Dev Dyn. 2006;235(9):2353–2375.
20. Smith T, Lozanoff S, Iyyanar P, Nazarali A. Molecular signaling along the anterior-posterior axis of early palate development. Front Physiol. 2012;3(488):1–14.
21. Zhou J, Gao Y, Lan Y, et al. Pax9 regulates a molecular network involving Bmp4, Fd10, Shh signaling and the Osr2 transcription factor to control palate morphogenesis. Development. 2013;140(23):4709–4718.
22. Venkatesh R. Syndromes and anomalies associated with cleft. Indian J Plast Surg. 2009;42(Suppl):S51–55.
23. Cohen M. Syndromes with cleft lip and cleft palate. Cleft Palate J. 1978;15(4):306–328.
24. Burdick A. Genetic epidemiology and control of genetic expression in van der Woude syndrome. J Craniofac Genet Dev Biol Suppl. 1986; 2: 99–105.
25. Kondo S, Schutte B, Richardson R, et al. Mutations in IRF6 cause Van der Woude and popliteal pterygium syndromes. Nat Genet. 2002;32(2):219.
26. Pegelow M, Koilininen H, Magnusson M, et al. Association and Mutation Analyses of the IRF6 Gene in Families with Nonsyndromic and syndromic Cleft Lip and/or Cleft Palate. Cleft Palate-Craniofacial J. 2014;51(1):49–55.
27. Ke C-Y, Xiao W-L, Chen C-M, et al. IRF6 is the mediator of TGFB3 during regulation of the epithelial mesenchymal transition and palatal fusion. Sci Rep. 2015;5(12791).
28. Suzuki K, Hu D, Bustos T, et al. Mutations of PVRL1, encoding a cell-cell adhesion molecule/ herpesvirus receptor, in cleft lip/palate-ectodermal dysplasia. Nat Genet. 2000;25:427–430.
29. Takahashi K, Nakanishi H, Miyahara M, et al. Nectin/PRR: An Immunoglobulin-like Cell Adhesion Molecule Recruited to Cadherin-based Adherens Junctions through Interaction with Afadin, a PDZ Domain-containing Protein. J Cell Biol. 1999;145(3):539–549.
30. Sozen M, Suzuki K, Tolarova M, et al. Mutation of PVRL1 is associated with sporadic, non-syndromic cleft lip/palate in northern Venezuela. Nat Genet. 2001;29:141–142.
31. Palagano E, Zuccarini G, Pfrontner P, et al. Mutations in the Neuroblastoma Amplified Sequence gene in a family affected by Acrofrontofacioanal Dysostosis type 1. Bone. 2018;114:125–136.
44. *Toriyama M*, Lee C, Taylor S, et al. The ciliopathy-associated CPLANE proteins direct basal body recruitment of intraflagellar transport machinery. Nat Genet. 2016;48(6):648–656.

45. *Capdevila J*, Tsukui T, Esteban C, et al. Control of Vertebrate Limb Outgrowth by the Proximal Factor Meis2 and Distal Antagonism of BMPs by Gremlin. Mol Cell. 1999;4:839–849.

46. *Johansson S*, Berland S, Gradek G, et al. Haploinsufficiency of MEIS2 is associated with orofacial clefting and learning disability. Am J Med Genet. 2014;164(7):1622–1626.

47. *Cullup T*, Kho A, Dionisi-Vici C, et al. Recessive mutations in EPG5 cause Vici syndrome, a multisystem disorder with defective autophagy. Nat Genet. 2014;46(1):83–87.

48. *Celli J*, Duijf P, Hamel B, et al. Haplozygous germline mutations in the p53 homolog p63 are the cause of EEC syndrome. Cell. 1999;99:143–153.

49. *Milunsky J*, Maher T, Zhao G, et al. TFAP2A Mutations Result in Branchio-Oculo-Facial Syndrome. Am J Hum Genet. 2008; 82: 1171–1177.

50. *Reiber J*, Szajer Y, Posteguillo E, et al. Additional clinical and molecular analyses of TFAP2A in patients with the branchio-ocular-facial syndrome. Am J Med Genet. 2010;152(4):994–999.

51. *Zarelli V*, David I. Inhibition of neural crest formation by Kctd15 involves regulation of transcription factor AP-2. Proc Natl Acad Sci USA. 2013;110(8): 2870–2875.

52. *Andreu A*, Pauws E, Jones M, et al. TBX22 Missense Mutations Found in Patients with X-Linked Cleft Palate Affect DNA Binding, sumoylation, and Transcriptional Repression. Am J Hum Genet. 2013;110(8): 2870–2875.

53. *Braybrook C*, Doudney K, Marcano ACB, et al. The T-box transcription factor gene TBX22 is mutated in X-linked cleft palate and ankyloglossia. Nat Genet. 2001;29(2):107–109.

54. *Jeong Y*, Leskow F, El-Jaick K, et al. Regulation of a remote Shh forebrain enhancer by the SIX3 homeoprotein. NATURE Genet. 2008;40(11):1348–1353.

55. *Wallis D*, Roessler E, Hehr U, et al. Mutations in the homeodomain of the human SIX3 gene cause holoprosencephaly. Nat Genet. 1999;22:196–198.

56. *Kruszka P*, Li D, Harr M, et al. Mutations in SPECC1L, encoding sperm antigen with calponin homology and coiled-coil domains 1-like, are found in some cases of autosomal dominant Opitz G/BBB syndrome. J Med Genet. 2015;52(2):104–110.

57. *Saadi I*, Alkuraaya F, Gisselbrecht S, Goessling W, Cavallea C, Maas R. Deficiency of the cytoskeletal protein SPECC1L leads to oblique facial clefting. Am J Hum Genet. 2011;89(1):44–55.
58. **Filmus J, Capurro M, Rast J. Glypicans.** Genome Biol. 2008;9(5):224.
59. **Pilia G, Hughes-Benzie R, MacKenzie A, et al.** Mutations in GPC3, a glypican gene cause the Simpson-Golabi-Behmel overgrowth syndrome. Nat Genet. 1996;12:241–247.
60. **Emes R, Ponting C.** A new sequence motif linking lissencephaly, Treacher Collins and oral-facial-digital type 1 syndromes, microtubule dynamics and cell migration. Hum Mol Genet. 2001;10(24):2813–2820.
61. **Rakkolainen A, Ala-Mello S, Kristo P, Orpana A, Jarvela I.** Four novel mutations in the OFD1 (Cxorf5) gene in Finnish patients with oral-facial-digital syndrome 1. J Med Genet. 2002;39(4):292–296.
62. **Fan Z, Li J, Zhang H, et al.** A missense mutation in PTCH2 underlies dominantly inherited NBCCS in a Chinese family. J Med Genet. 2008;45(5):303–308.
63. **Johnson R, Rothman A, Xie J, et al.** Human homolog of patched, a candidate gene for the basal cell nevus syndrome. Science (80-). 1996; 272 (5268):1668–1671.
64. **Pastorino L, Ghiborzo P, Nasti S, et al.** Identification of a SUFU germline mutation in a family with Gorlin syndrome. Am J Med Genet. 2009; 149 (7):1539–1543.
65. **Ridgeway A, Skerjanc I.** Pax3 is essential for skeletal myogenesis and the expression of Six1 and Eya2. J Biol Chem. 2001;276:19033–19039.
66. **Tassabehji M, Read A, Newton V, et al.** Waardenburg’s syndrome patients have mutations in the human homologue of the Pax-3 paired box gene. Nature. 1992;355:635–636.
67. **Vissers L, van Ravenswaaij C, Admiral A, et al.** Mutations in a new member of the chromo-domain gene family cause CHARGE syndrome. Nat Genet. 2004;36:955–957.
68. **Bajpai R, Chen D, Rada-Iglesias A, et al.** CHD7 cooperates with PBAF to control multipotent neural crest formation. Nature. 2010; 463 (7283): 958–962.
69. **Chen L, Fulcoli F, Ferrentino R, et al.** Transcriptional control in cardiac progenitors: Tbx1 interacts with the BAF chromatin remodeling complex and regulates Wnt5a. PLOS Genet. 2012;8(3):e1002571.
70. **Leslie EJ, Marazita ML.** Genetics of Cleft Lip and Cleft Palate. Am J Med Genet C Semin Med Genet. 2013;163(4):246–258.
71. **Davies S, Wise C, Venkatesh B, et al.** Mapping of three translocation breakpoints associated with orofacial clefting within 6p24 and identification of new transcripts within the region. Cytogenet Genome Res. 2004;105:47–53.
72. **Deepak C, Ramanathan A.** Role of MSX1 Gene in Orofacial Clefting: A Systematic Review. Biomed Pharmacol J. 2017;10(3):1071–1072.
73. **van den Boogard M, Dorland M, Beemer F, van Amstel H.** MSX1 mutation is associated with orofacial clefting and tooth agenesis in humans. Nat Genet. 2000;24:342–343.
74. **Blanton S, Cortez A, Stal S, et al.** Variation in IRF6 contributes to nonsyndromic cleft lip and palate. Am J Med Genet. 2005;137(3):259–262.
75. **Suzuki S, Marazita M, Cooper M, et al.** Mutations in BMP4 are associated with subepithelial, microform, and overt cleft lip. Am J Hum Genet. 2009;84(3):406–411.
76. **Tucker A, Matthews K, Sharpe P.** Transformation of Tooth Type Induced by Inhibition of BMP Signaling. Science (80). 1998; 282(5391): 1136–1138.