

The Genetic Aspect of Non-Syndromic Cleft Lip and Palate towards Candidate Genes in the Etiology : A literature Review

Mala Kurniati^{1,2}, RM Coen Pramono D³, Agung Sosiawan⁴, Ni Wayan Tirtaningsih⁵

¹Post Graduate, Doctoral Program, Faculty of Medicine, Universitas Airlangga, Surabaya-Indonesia, ²Lecturer, Department of Biology, Faculty of Medicine, Universitas Malahayati, Lampung, Indonesia, ³Professor, Department of Oral & Maxillofacial Surgery, Faculty of Dental Medicine, Universitas Airlangga, Surabaya, Indonesia, ⁴Senior Lecturer, Department of Dental Public Health, Faculty of Dental Medicine, Universitas Airlangga, Surabaya, Indonesia, ⁵Senior Lecturer, Department of Anatomy and Histology, Faculty of Medicine, Universitas Airlangga, Surabaya, Indonesia

Abstract

Cleft lip and cleft palate (CL/P) is a cleft lip deformity indicated by an opening or an uncommon cleft in the lip or roof of the mouth (palate). The differences in ethnicity, gender, and the correlation with genetic factors influence the prevalence of Non-syndromic CL/P. This study was conducted through a literature review on genes that were allegedly associated with Non-syndromic CL/P. Genetics play a role, to a greater or lesser extent, in all diseases. Besides, palatogenesis involves many diverse genes in a complex process. In this case, oral cleft phenotypes develop when this process is disrupted in some manner because of gene dysfunction. Various genetic approaches, including genome-wide and candidate gene association studies as well as linkage analysis, have been undertaken to identify etiologic factors, but results have often been inconclusive or contradictory. Therefore, it concludes that the genetic basis of CL/P is still controversial because of the genetic complexity of clefting.

Keywords: Cleft Lip, Cleft Palate, Genetic

Introduction

Cleft lip and cleft palate or orofacial cleft is a cleft lip deformity indicated by an opening or an uncommon cleft in the lip or the palate (roof of the mouth). There are three primary types of orofacial cleft, namely cleft lip (CL) that is a cleft appeared only in the lip, cleft palate (CP) that is a cleft in the palate area, and cleft lip palate (CLP) that is a cleft appeared from the palate through the lip.¹ Non syndromic cleft lip with or without a cleft palate or abbreviated as *NSCL/P* is a type of deformity that can happen separately or simultaneously and it

strongly influences a child's growth and development as a series of cause and effect, such as speech problems, hearing problems, dental and oral problems, feeding problems and malnutrition, respiratory disorders, psychological disorders, and facial aesthetics.^{2,3} The proportion of genetic and non-genetic contributions for some types of disease or abnormality is various. However, the proportion of variants of each observed disease in a population caused by variants in genetic factors is known as heritability and it can be calculated through several methods, for example, by knowing the concordance rate.⁴ The genetic aspects are known for playing a vital role in *NSCL/P*. However, up until now, the specific genes have not been fully identified due to a high number of gene interactions that are involved.

Method

The method of this study used a literature review with an electronic database. This study used secondary data, namely, articles collected from a search engine in

Corresponding author:

Prof. RM Coen Pramono D, drg., SU., Sp.BM (K)
Professor, Department of Oral & Maxillofacial Surgery,
Faculty of Dental Medicine, Universitas Airlangga, Jl.
Mayjen Prof. Dr. Moestopo No.47, Pacar Kembang,
Surabaya City, East Jawa, Indonesia 60132
Email: profcpd@gmail.com

some publication sources. During the search, the author used several criteria for keywords as follows: “cleft palate”, “facial clefts”, “genetic”, “cleft lip”, dan “non syndromic”. In addition, a combination of the following terms was used: “Genetic cleft lip palate”, “Genetic cleft lip palate” and “non-syndrome”.

Etiology of Cleft lip and Palate

The incidence of *NSCL/P* is not surely known due to its multifactorial causes and it is influenced by several factors besides the genetic and non-genetic factors. In genetic aspects, there is a gene playing a role in the occurrence of *NSCL/P*. Non-genetic factors that play a role in cleft lip are nutritional deficiency (folic acid), chemical substances/drugs, rubella virus, radiation, infection of infectious disease, endocrine disorders, smoking, drinking alcohol, and trauma (mental trauma and physical trauma).⁵

Epidemiology of Cleft lip and Palate

The epidemiology of Cleft lip and Palate occurs two times higher in boys, while *Cleft palate* occurs two times higher in girls. In the Czech Republic, it has been found 2,147 babies with *Cleft lip* and or *Cleft palate*. The overall incidence is 1 out of 600 live births.⁶ Twelve studies conducted in several locations in North America reported that the level of prevalence for white people, black people, Hispanic, and American Indians was around 0.6 to 3.92 per 1,000 births. Six studies conducted in several locations in Europe reported that the level of prevalence for white people and Arabs was around 1.02 – 1.94 per 1,000 births. There is a difference in the prevalence of births with orofacial clefts (*OFC*) among ethnicities. American Indians had the highest level of prevalence followed by Japan, Chinese, and white people, while black people had the lowest level of prevalence.⁷ The data of global prevalence for orofacial clefts are not fully completed and, up until now, the international level of prevalence has not been established yet.

Genes Contributed to Cleft

1. Transforming growth factor – alpha (TGFA)

The *TGFA* gene is located at the 2p13 chromosome. *TGFA* plays a role in the regulation process of palate development. The previous genetic studies showed a significant correlation between the transforming growth

factor-alpha (*TGFA*) and *CL/P*.^{8,9}

2. Transforming growth factor - beta 3 (TGFβ3)

Transforming Growth Factor-Beta 3 (*TGF-β3*) is one of the strongest candidate genes for cleft lip and palate in humans.^{10,11} *TGF-β3* (located at the 14q24 chromosome) has a wide spectrum of biological activities and they are known to be able to induce palatal fusion¹²; over the last few years, several studies had been conducted to explain the correlation between *TGF-β3* and cleft lip and palate.^{13,14,15}

3. Muscle Segment Homobox Gene 1 (MSX1)

MSX1 is at the 14p16.1 position to code protein from 297 amino acids that are functioned as the transcriptional repressor during embryogenesis. *MSX1* plays a role as a strong candidate causing *NSCL/P*, based on cleft palate and the complete failure of the development of incisor teeth.¹⁶

4. Interferon regulatory factor 6 (IRF6)

The human interferon regulatory factor 6 gene (*IRF6*) located at the 1q32.2 chromosome is responsible for the majority of patients with Van der Woude syndrome. The risk of *NSCL/P* relapse has been reported higher to infect an individual with the C allele, namely single nucleotide polymorphism (SNP) rs2235371, that encodes the Val274 amino acid from the product of *IRF6* gene.¹⁷

5. T-box-containing transcription factor 22 (TBX22)

Mutation in the *TBX22* gene is correlated with the causes of a syndrome related to the X chromosome with cleft palate and ankyloglossia. The T-box *TBX22* transcription factor plays an important role in the normal craniofacial development as what has been reported in a finding related to non-sense mutation, frameshift, splice-site, or missense in patients with CPX. The *TBX22* mutation is reported to occur in around 4%-8% of patients with non syndromic cleft palate.¹⁸

6. Retinoic Acid Receptor α (RARA)

The retinoic acid receptor α (*RARA*) is one of the candidate genes for causing *NSCLP*.¹⁹ *RARA* gene is located in the 17q12-21 chromosome region. The *RARA*

gene plays an important role in the development stage that its functions are mediated by retinoic acid receptor alpha (RAR- α), including the regulation of development, differentiation, apoptosis, granulopoiesis. A study in a transgenic rat that has been knocked out showed the role of the RARA gene in facial development.²⁰

7. Poliovirus receptor-related 1 (PVRL1)

The poliovirus receptor related-1 (PVRL1) gene is located in the 11q23.3 chromosome region that encodes the nectin-1 and cell adhesion molecules (CAMs) (OMIM #600644). An experiment in a rat reveals that mRNA PVRL1 is expressed in the medial edge epithelium (MEE) of the palate that is developing, it showed that there is an involvement of normal PVRL1 gene in the fusion of palatal shelves during palatogenesis.^{21,22,23}

8. Methylenetetrahydrofolate reductase (MTHFR)

MTHFR gene is a gene playing a role in coding for making *methylenetetrahydrofolate reductase* enzyme. The enzyme functions to folic metabolism and it has been learned in an abnormality, such as NSCL/P. This reaction is needed for an advanced process in changing amino acid types, namely homocysteine into methionine that functions to produce protein and other important compounds for the metabolism. MTHFR gene has a sequential order of complete DNA of 20,373 bp in length consisting of 11 exons, located in the 1p36.22 chromosome (at the chromosome's long arm of 1, area 3, band 6, sub-band 2).^{24,25,26}

9. Bone Morphogenetic Protein 2 (BMP2)

BMP2 gene encodes the ligand secreted from the TGF- β superfamily. The ligand from this superfamily binds several TGF- β receptors that play a role in the activation of a transcription factor, especially SMAD protein, in the regulation of gene expression. BMP2 gene is located in the 20p12.3 chromosome (the chromosome's short arm of 20, area 1, band 2, sub-band 3). It is recorded that a significant association between the polymorphism of *BMP2* rs235768 A>T with the incidence of CB/L as a risk factor causing CB/L in the Iranian population.²⁷

10. Bone Morphogenetic Protein 4 (BMP4)

BMP4 gene with a sequential order of complete

DNA and 11.2 kb in length consists of four exons and is located at 14q22.2 chromosome (at the chromosome's long arm of 14 and area 2, band 2, sub-band 2). BMP4 gene encodes the ligand secreted from the TGF- β superfamily. Mutation in this gene is correlated with orofacial disorder and microphthalmia.^{28,29}

Conclusion

A genetic study is needed to provide an understanding of the pathophysiology of NSCLP. It has been an effort to identify the genetic disease related to NSCL/P to allow disease prevention and provide suggestions in the clinical management for controlling the risk factors.

Ethical Clearance: This study was approved by the ethical committee of the Faculty of Dentistry, Universitas Airlangga, number 606/HRECC.FODM/IX/ 2019.

Conflict of Interest: There was no conflict of interests regarding the publication of this study.

Source of Funding: The funding source of this study was supported by Directorate of Research and Community Services, Deputy of Research and Development Reinforcement, Ministry of Research and Technology/ National Agency for Research and Innovation.

References

1. Savitha S, Sharma S, Veena S. BMP4 Gene and Pathophysiology of Cleft Lip and Palate. *J Dent Sci.* 2014;2(1):92–6.
2. Muhamad A, Azzaldeen A, Watted N. Cleft Lip and Palate ; a Comprehensive Review. *Int J Basic Appl Med Sci.* 2014;4(1):338–55.
3. Kati FA. Cleft Lip And Palate : Review Article. *World J Pharm Med Res.* 2018;4(7):155–63.
4. Hussain S. A new conceptual framework for investigating complex genetic disease. *Front Genet.* 2015;6(11):1–9.
5. Wong F, Hagg U. An update on the aetiology of orofacial. *Hong Kong Med J.* 2004;10(5):331–6.
6. McGarry A. The influence of genetics on syndromic and non-syndromic cases of cleft lip and cleft palate. Washington; 2017.
7. Panamonta V, Pradubwong S, Panamonta M, Chowchuen B. Global Birth Prevalence of Orofacial Clefts: A Systematic Review. *J Med Assoc Thai.*

- 2015;98(December):S11–21.
8. Lesmana S, Auerkari EI, Elza I. Genes Contributing in Cleft Lip and Cleft Palate: A Literature Review. *J Int Dent Med Res.* 2016;9 (Special Issue: Universitas Indonesia 1st):441–8.
 9. Rajion ZA, Alwi Z. Genetics Of Cleft Lip And Palate : A Review. *Malaysian J Med Sci.* 2007;14(1):4–9.
 10. Brunet CL, Sharpe PM, Ferguson MWJ. Inhibition of TGF- β 3 (but not TGF- β 1 or TGF- β 2) activity prevents normal mouse embryonic palate fusion. *Int J Dev Biol.* 1995;39(2):345–55.
 11. Vieira AR, Orioli IM, Castilla EE, Cooper ME, Marazita ML, Murray JC. MSX1 and TGFB3 contribute to clefting in South America. *J Dent Res.* 2003;82:289–92.
 12. Dudas M, Nagy A, Laping NJ, Moustakas A, Kaartinen V. Tgf- β 3-induced palatal fusion is mediated by Alk-5/Smad pathway. *Dev Biol.* 2004;266(1):96–108.
 13. Kim MH, Kim HJ, Choi JY, Nahm DS. Transforming growth factor- β 3 gene SfaN1 polymorphism in Korean nonsyndromic cleft lip and palate patients. *J Biochem Mol Biol.* 2003;36(533–7).
 14. Sun D, Vanderburg CR, Odierna GS, Hay ED. TGF β 3 promotes transformation of chicken palate medial edge epithelium to mesenchyme in vitro. *Development.* 1998;125(1):95–105.
 15. Taya Y, O’Kane S, Ferguson MWJ. Pathogenesis of cleft palate in TGF- β 3 knockout mice. *Development.* 1999;126(17):3869–79.
 16. Chandrasekharan D, Ramanathan A. Role of MSX1 Gene in Orofacial Clefting : A Systematic Review. *Biomed Pharmacol J.* 2017;10(3):1071–2.
 17. Pegelow M, Zucchelli M, Fransson I, Larson O, Kere J, Larsson C, et al. Familial non-syndromic cleft lip and palate — analysis of the IRF6 gene and clinical phenotypes. *Eur J Orthod.* 2008;30(January):169–75.
 18. Jiang RS, Zhao X, Liu R. Non-syndromic cleft palate: Analysis of TBX22 exon 5 gene mutation. *Arch Med Sci.* 2012;8(3):406–10.
 19. Kanno K, Suzuki Y, Yang X, Yamada A, Aoki Y, Kure S, et al. Lack of evidence for a significant association between nonsyndromic cleft lip with or without cleft palate and the retinoic acid receptor alpha gene in the Japanese population. *J Hum Genet.* 2002;47(6):269–74.
 20. Veros C, Iakovidou-kritsi Z. The genetic basis of cleft lip and cleft palate.
 21. Cheng HQ, Huang EM, Xu MY, Shu SY, Tang SJ. PVRL1 as a candidate gene for nonsyndromic cleft lip with or without cleft palate: No evidence for the involvement of common or rare variants in southern han chinese patients. *DNA Cell Biol.* 2012;31(7):1321–7.
 22. Gritli-linde A. Molecular control of secondary palate development. *Dev Biol.* 2007;301:309–26.
 23. Cobourne MT. The complex genetics of cleft lip and palate. *Eur J Orthod.* 2004;26(1):7–16.
 24. Rafik A, Nadifi S. Updating Genetics Polymorphisms of Non-Syndromic Clefts Lip-Palates. *Am J Mol Biol.* 2018;08(03):178–85.
 25. Sosiawan A, Kurniati M, Iskandar RPD, Furqoni AH, Nuraini I, Qurrota A, et al. An Analysis of the MTHFR Gene and Clinical Phenotypes in Familial Non-Syndromic Cleft Palate. *J Int Dent Med Res.* 2020;13(3):1160–4.
 26. Rochmah YS, Suwarsi L, Harumsari S, Sosiawan A, Muis SF, Faradz MS. Maternal Polymorphism MTHFR A1298C. *J Int Dent Med Res suppl.* 2018;11(1):120–3.
 27. Saket M, Saliminejad K, Kamali K, Moghadam FA, Anvar NE, Khorram Khorshid HR. BMP2 and BMP4 variations and risk of non-syndromic cleft lip and palate. *Arch Oral Biol.* 2016;72:134–7.
 28. Nie X, Luukko K, Kettunen P. BMP signalling in craniofacial development. 2006;521:511–21.
 29. Wang RN, Green J, Wang Z, Deng Y, Qiao M, Peabody M, et al. Bone Morphogenetic Protein (BMP) signaling in development and human diseases. *Genes Dis.* 2014;1(1):87–105.