REVIEW ARTICLE

Etiopathogenesis of orofacial clefting revisited

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ABSTRACT
The cleft anomaly may be more ancient than the man himself. It is one of the most common developmental malformations reported in the literature. There are number of intriguing theories regarding its etiopathogenesis, each of which has some evidence in its favor. This review highlights all the genetic and environmental etiologic factors and focuses on its pathogenesis.
Key words: Cleft lip, cleft palate, etiopathogenesis.

INTRODUCTION

A 'cleft' is a congenital abnormal space or gap in the upper lip, alveolus or palate.[1] The formation of the face and oral cavity is complex in nature and involves the development of multiple tissue processes that must merge and fuse in a highly orchestrated fashion. Disturbances in the growth of these tissue processes or their fusion may result in the formation of orofacial clefts.[2] Cleft lip (CL) and cleft palate (CP) are the single most common congenital deformity affecting the orofacial structures in man and constitute about 13% of all reported anomalies.[3,4]

CL and CP are complex multi-factorial disorders, where in both genetic and/or environmental factors are responsible for its occurrence.[5] Although the mode of inheritance of cleft lip-palate (CLP) has been investigated for many years, the results appear to be controversial.[4] It is said that genetic and/or environmental factors that inhibit the flow of neural crest cells or decrease their number so that contact between the facial prominences is inadequate or impossible, resulting in clefts.[6]

The zones affected by common orofacial clefts are upper lip, alveolar ridge, hard palate, soft palate, nose (not so common) and eyes (not so common).[7] The cleft may be syndromic, where the patient has more than one malformation or involving more than one developmental field and non-syndromic, if there is only a single malformation or if multiple anomalies are limited to a single developmental field.[8] In this treatise, an attempt has been made to review the etiopathogenesis of CL and CP.

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FORMATION OF OROFACIAL CLEFTING

Precious[9] summarizes the major processes and events whose disruption, if not compensated for, would lead to the development of clefts. They are as follows: abnormal migration of neural crest cells, altered rates of cell division and/or cells death, abnormal localization of post-migratory neural crest cells, defective epithelial-mesenchymal interactions, gene defects in the production of cell specific molecules such as collagen types I and II or cartilage-specific proteoglycan, defective synthesis, deposition, and/or degradation of extracellular matrices, inability to maintain the differentiated cell state, impaired interactions between skeletal, muscular, nervous, and/or vascular components (impaired functional matrices) because of defective development in one or more of these systems, abnormal hormonal environment because of either defective production of a hormone or impaired receptor function, delays or accelerations in the timing of development, leading to uncoupling of normally coupled processes.[10]

Cohen,[11] summarizes embryonic basis of orofacial clefting may because of the following factors: genetic and/or environmental factors that inhibit the flow of neural crest cells or decrease their number may affect their masses so that contact between the facial prominences are inadequate or impossible, the epithelium that covers the mesenchyme may not undergo programmed cell death so that fusion cannot take place, any change in the position of nasal placodes or abnormal direction of growth of the facial prominences may result in CL. It has been suggested that the tip of the tongue becomes wedged in the labial cleft, thus not allowing the tongue to drop, which would inhibit palatal contact and fusion, resulting in CL and CP.

The embryological development of the face and more specifically the oral region relies on the interplay of different types of cells (ectoderm and endoderm) in a vast range of factors encompassing cell differentiation, growth, apoptosis, cell-cell adhesion and inter- and intra-cellular signaling. So the etiology of clefting may be due to disruption of a gene controlling one or more of these factors, inhibition of cell function by environmental teratogens and combination of the two. Thus CL, CP and CLP are complex, multi-factorial in nature and are likely outcome of several developmental and biochemical events.[12]

Genetics

Much of the evidences support the view that genetic factors are associated with orofacial clefting. Sibling risk for CLP is approximately 30 times higher than that for the normal population prevalence. In twins with CLP, concordance is far greater for monozygotic twins (40%) than for dizygotic twins (4.2%).[8,12,13] Thus orofacial clefting is heterogeneous and variation in liability is probably determined by a number of factors such as: major genes, minor genes, environmental factors and a developmental threshold.[14]

Genetic hypotheses for the etiology of CL and CP have evolved considerably, despite numerous extensive investigations; however, no simple Mendelian pattern of inheritance has been readily apparent. This has led to the proposal by different authors of a variety of genetic modes of inheritance including: dominance, recessiveness, sex linkage and various modifying conditions such as incomplete penetrance and variable expressivities.

Autosomal dominant gene

If the gene is dominant, half the offspring of the affected will display the condition, this pattern may continue through many generations, and the proportion displaying the condition in the pedigree will follow the pattern of ½ in the first degree relatives (siblings and children), ¼ in the second degree relatives (aunts, uncles, nephews and nieces).

Autosomal recessive gene

If the mutant gene is recessive, it is not uncommon for the affected brother or sister, for the recessive condition would manifest only in homozygous cases, born of the heterozygous parents.

Sex linked gene: If the mutant X-linked gene is contributed by the mother, half the daughters are affected – in case of a recessive sex-linked gene as in hemophilia, only half the sons are affected, and the daughters are the carriers. If the mutant gene, on the other hand, is contributed by father all his daughters but none of his sons are affected.[6] Many genes for orofacial clefting have been suggested by allelic association and by linkage analysis [Table 1].[15]

Syndromic vs. non-syndromic cleft

A cleft is called syndromic, if the patient has more than one malformation involving more than one developmental field. The causes for syndromic clefting include the following: chromosomal abnormalities, single gene disorders, teratogenic syndromes and syndromic clefting with unknown cause. Genetic aspects of some syndromes with clefting are reviewed in Table 2.[11]

Non-syndromic cleft

A cleft is said to be non-syndromic, if there is only single malformation, or when if there are multiple anomalies that are resultant of a single initiating event/primary malformation involving more than one developmental field.

Genetic hypothesis for the non-syndromic clefting have evolved considerably, and become more complex with time. In the first large scale study, Fogh-Anderson[16] concluded that isolated CP was inherited as a simple dominant trait with greatly reduced penetrance. Whereas, CLP was possibly transmitted by a gene of variable penetrance that could act as either as a recessive or a dominant gene, depending on the genetic background of the individual.

Table 3 summerizes the proposed genetic models for CL with or without CP.[4,8,17,18]

Table 1: Genes associated with orofacial clefting[15]

Candidate gene	Gene map locus	Name of gene	Type of cleft associated
TGF-α	2p13	Transforming growth factor α	CL/Cp, CPO
TGF-β	19q13.1-q13.3	Transforming growth factor β	CL/CP
MSX-1	4p16.1	Homeobox gene (HOX7)	CL/CP
RAR-α	17q12	Retinoic acid receptor α	CL/CP
DLX-2	2q32	Distal-less homeobox 2	CPO
BCL-3	19q13.1	B-cell leukemia/ lymphoma 3	CL/CP
	2q32	Unknown gene but different than DLX2; possible candidate genes include FN1, IHH, IGFBP2, and IGFBP5	CPO
	4q25-q31.1	Unknown gene	CL/CP
	6p23	Unknown gene	CL/CP
	17p11.1-p11.2	Unknown gene that may increase cleft palate susceptibility or work synergistically to increase susceptibility in van-der Woude syndrome that maps to 1q32-q41	CPO

CL/CP - Cleft lip with or without cleft palate, CPO - CP only.

Table 2: Genes involved in syndromic clefting[4,11]

Syndrome	Gene map locus	Comments
Van-der woude syndrome	1q32-q41 17p11.1–p11.2	Autosomal dominant inheritance; gene maps to 1q32 – q41; one large Brazilian family maps to 17p11.1 – p11.2; either the syndrome is genetically heterogeneous or the gene at both loci work synergistically, 17p11.1-p11.2 increasing the risk of CL with or without cleft palate.
Treacher collins syndrome	5q32-q33.1	Treacle gene mutations cause premature termination of the protein
del(22q11.2) syndrome (Velocardiofacial syndrome or DiGeorge syndrome)	22q11.1	Disruption of gene UFD1L alone or in combination with gene CDC45L and/or HIRA has been suggested as the most likely etiology and the role of the dHand-UFD1L pathway have been discussed; however, many patients do not have mutations in these genes. Thus, the genetic etiology is not resolved currently.
X-linked CP with or without ankyloglossia	(1) Xq21.3-q22 (2) Xq13-q21.31	Genetically heterogeneous is found in: (1) German and Icelandic families, (2) British Columbia families.

Environmental factors

The majority of CL/CP cases are multi-factorial and a variety of environmental factors have been implicated. Most of these factors are yet to be studied in, regard to their mechanism of action. Animal experiments showed that these factors contribute in the development of cleft, but they couldn't reveal the exact mechanism of action of each factor in the cause of CL and CP. Various human teratogens involving oro-facial clefting are summarized by Cohen *et al* [Table 4].[11]

Bronsky *et al*.[19] revealed the mechanism of action of some environmental factors that inhibit the electron transport chain (ETC) and effects of hypoxia on facial prominence development in mice. Experimental results from these studies on animals showed that environmental factors inhibiting the ETCs were potent inducers of CL/CP in mammals and comparable clefts in chick embryo.

Maternal cigarette smoking

A meta-analysis was performed by Wyszynski *et al.*,[8] to estimate the association between maternal cigarette smoking and the risk of having a child with a non-syndromic oral cleft. This study suggested that maternal cigarette smoking during the first trimester of gestation is associated with a higher risk of having a child with a CL/ CP (11% of the CL/CP and 12% of the CP). It is thought that cigarette smoking may influence embryonic development by producing tissue hypoxia, which impedes tissue growth, particularly in the developing palate.[12] Hypoxia has been shown to induce cleft experimentally in mice. A human detoxification mechanism exists which is as follows:

Cytochrome p-450
↓
Oxidation of Polycyclic Aromatic Hydrocarbons
(Cigarette Smoke)
↓
Detoxification of Teratogens

This detoxification mechanism will be deficient in malformed

Table 3: Proposed genetic models for CLP[4,8,17,18]

Source	Type of cleft	Proposed model of inheritance
Fog-Anderson (1942)	CL /P CP	Single gene of variable penetrance, recessive or dominant, depending on the genetic background. Dominant trait with greatly reduced penetrance
Woolf *et al*, 1963	CL/P	Dominant gene in some families, interaction of polygenes and non-genetic factors in others and phenocopy cases.
Woolf, 1964 Tanaka *et al*, 1969 Carter, 1969 Fraser, 1970 Kogochi, 1975 Czeizel *et al*, 1984 Chung *et al*, 1986	CL/P	Multi-factorial with threshold
Carter, 1969 Bixler *et al*, 1971 Bear, 1976	CL/P	Polygenic
Marazita *et al*, 1984	CL/P	Major gene
Marazita *et al*, 1986	CL/P and CP	Autosomal major locus + Multi-factorial contributions
Rollnick, 1986	CP	Autosomal dominant in one family and X-linked recessive in the other two families.
Chung *et al*, 1986	CL/P	Autosomal recessive with low penetrance
Farrall and Holder,1992	CL/P	Major gene + Oligogenic background
Mitchell and Risch, 1992	CL/P	Either Multi-factorial with threshold or generalized single major locus
Murray *et al*, 1995	CL/P	Clefting locus on various Chromosomes like, 2p, 4q, 6p, 17q, 19q
Pirinan, 1998 Vieira *et al*, 2003	CL/P	Genes MSX-1 and TGF-β3 with a gene linkage disequilibrium strategy
Sozen *et al*, 2001	CL/P	Mutation in W185X PVRL1

CL/P - Cleft lip with or without cleft palate, CP - Isolated cleft palate.

Table 4: Teratogens involved in etiology of CL/CP[11]

Teratogen	Use	Frequency of Oro-facial clefting
Ethyl Alcohol	Recreational use or alcohol dependency	Occasional
Diphenylhydantoin	Anti-seizure drug	Occasional
Trimethadone	Anti-seizure drug	Occasional
Retinoids	Drugs to treat cystic acne	Occasional
Aminopterin, Methotrexate	Abortifacient	Occasional
Hyperthermia	From heat such as fever	Occasional
Smoking	Recreational	Occasional, may act alone or synergistically with TGF α

infants whose mothers were smokers.[12] According to Shaw *et al*.,[20] risk of clefting associated with maternal smoking can be increased in infants having TGF α mutation. So cigarette smoking can be correlated with clefting and may act alone/ synergistically with TGF α. According to Witter *et al.,*[21] serum folate may be decreased in cigarette smoking which may lead to form clefts as many other previous studies have shown that clefting is associated with folate deficiency.

Drugs

Cancer chemotherapeutic agents

Aminopterin, methotrexate, cyclophospamide, procarbazine and hydroxamic acid derivatives, interfere with DNA synthesis resulting in gross malformation in the fetus.

Anticonvulsant drugs

Phenytoin, trimethadione, paramethadione, carbamazepine, valproic acid, mysoline and phenobarbital causes a deformity known as "anti-convulsant facies". These drugs are suspected of causing CL and CP in the offspring of epileptic mothers. Phenytoin is the most common drug among the CL/CP inducing environmental factors in which the pathogenesis has been studied. The incidence of CL/CP in the children of epileptic mothers receiving phenytoin appears to be approximately ten times more than in controls. After the treatment of pregnant mice with phenytoin, the overall growth of the embryo, including the facial prominences, is reduced, as reflected by a reduction in the rate of mesenchymal cell proliferation in facial prominences to approximately 50% that of controls.[22] The anticonvulsant drugs are thought to function therapeutically through interference with neurotransmitters (serotonin), and is possible that at least part of their teratogenic activities may result from interference with neurotransmitter regulation of development.

Folic acid antagonist

Frank *et al*.,[23] described the effects of 30 mg/kg of 3.3 dimethyl-1-phenyl triazene administered on day 12 of gestation in the rat. At external examination, they found 82% of cases with micrognathia and 100% of embryos with CP.

Radiation

Ionising radiation have induced microcephaly and CP in live born offsprings whose mothers had been exposed to therapeutic or accidental pelvic radiation during the first ten weeks of pregnancy.

Retinoids

Retinoid is the most commonly used for the treatment of cystic acne. It has the teratogenic effects such as abortion, hydrocephalus and retardation of cell growth. Occasionally, it can cause CL and CP.[23]

Other drugs inducing CP are

antiemetics, hydrocortisone analogues, opioid drugs, salicylates (asprin), diazepam, boric Acid.[23]

Folic acid

There is conclusive evidence for maternal folate supplementation in the prevention of CL/CP and neural tube defects, as both these tissues involved are derived from the neural crest lineage.[13] One of the most useful metabolic markers for folate deficiency is hyper homocysteinemia and evidence suggests that this may be a risk factor for oral clefting.[24] According to Tolarova *et al.*,[25] folic acid supplements taken by pregnant mothers might decrease their chance of having a second affected child with CL/CP by 25-65% and also neural tube defects. In human, drugs like phenytoin, which interfere with folate metabolism, are known to have teratogenic effects including oral clefts.[26,27] Currently, it is recommended that a supplement of 0.4 mg folic acid from pre-conception to the 12th week of pregnancy to prevent these malformation. It is also advised that women with previously affected pregnancy should take 4 mg of folic acid daily.[28]

Alcohol

Alcoholism and pregnancy are associated with a pattern of abnormalities in the offspring known as Fetal Alcohol syndrome (FAS). According to Cohen,[29] alcohol consumption by mother during pregnancy may cause teratogenic effects such as growth retardation, mental deficiency, cardio-vascular defects, etc. However, it is occasionally associated with oral clefting. It is believed that most of the children believed to have FAS have been born of frankly alcoholic women, whose alcohol intake is eight to ten drinks or more per day.[30]

Maternal Age

Older the age of pregnant mother, greater chances of incidence of Orofacial clefts, since there is a greater chance of a defective zygote.[7]

Other environmental factors

Other factors, such as deficiency of vitamin A and vitamin B, anorexia, stress and consanguineous marriages

are believed to have a significant influence on occurrence of congenital anomalies, but are occasionally associated with oral clefting.[31]

CONCLUSION

To conclude, formation of cleft is a complex multi-factorial in origin. Though various genes and environmental factors are thought to play an important role in its etiopathogenesis, it still remains controversial that what is the exact cause of this most common developmental anomaly. Research in this aspect is required further to locate precisely, the importance of all these entities which probably act together, leading to cleft formation.

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REFERENCES

1. Peterson, Ellis, Hupp, Tucker. Contemporary oral and maxillofacial surgery. 4th ed. St. Louis: Mosby Co; 2003.
2. Neville BW, Damm DD, Allen CM, dan Bouquot JE. Oral and maxillofacial pathology. 2nd ed. Philadelphia: WB Saunders Company; 2002
3. Nanci A. Tencate's oral histology, development, structure, and function. 7th ed. St Louis: Mosby; 2010
4. Batra P, Duggal R, Parkash H. Genetics of cleft lip and palate revisited. J Clin Pediatr Dent 2003;27:311-20.
5. Langevin CJ, Gage E, Papay F. Craniofacial clefts and craniofacial syndrome. Plast Reconstr Surg 2010;4:253-64.
6. Bhatia SN. Genetics of cleft lip and palate. Br Dent J 1972:132;95-103
7. Malik NA. Textbook of oral and maxillofacial surgery. 1st ed. New Delhi: Jaypee Publications; 2002.
8. Wyszynski DF, Beaty TH, Maestri NE. Genetics of nonsyndromic oral clefts revisited. Cleft Palate Craniofac J 1996;33:406-17.
9. Precious DS**.** Cleft lip and palate. Oral and Maxillofacial Surgery. 1st ed. Philadelphia: WB Saunders Company; 2000.
10. Fonseca RJ. Oral Maxillofacial Surgery. 1st ed. Philadelphia: WB Saunders Company; 2000.
11. Cohen MM. Etiology and pathogenesis of orofacial clefting. Oral Maxillofac Surg Clin North Am 2000:12;379-96.
12. Prescott NJ, Winter RM, Malcolm S. Nonsyndromic cleft lip and palate: complex genetics and environmental effects. Ann Hum Genet 2001;65:505-15.
13. Cobourne MT. The complex genetics of cleft lip and palate. Eur J Orth 2004:26;7-16.
14. Fraser FC. Liability, thresholds, malformations and syndromes. Am J Med Genet 1996:66;75-6.
15. Diehl SR, Erickson RP. Genome scan for teratogen induced clefting susceptibility loci in the mouse, evidence of both allelic and locus heterogeneity distinguishing cleft lip and cleft palate. Proc Natl Acad Sci U S A 1997:94;5231-6.
16. Fogh-Anderson P. Inheritance of hare lip and cleft palate. Copenhagen: Arnold Busck, Nordisk Forlag; 1942.
17. Carinci F, Pezzetti F, Scapoli L, Martinelli M, Carinci P, Tognon M. Genetics of nonsyndromic cleft lip and palate: A review of international studies and data regarding the Italian population. Cleft Palate Crainiofac J 2000;37:33-40.
18. 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.
19. Bronsky PT, Johnston MC, Sulik KK. Morphogenesis of hypoxia-induced cleft lip in CL/Fr mice. J Craniofac Genet Dev Biol Suppl 1986;2:113-28.
20. Shaw GM, Wasserman CR, Lammer EJ, O'Malley CD, Murray JC, Basart AM, *et al*. Orofacial clefts, parental cigarette smoking, and transforming growth factor-alpha gene variants. Am J Hum Genet 1996;58:551-61.
21. Witter FR, Blake DA, Baumgardner R, Mellits ED, Niebyl JR. Folate, carotene, and smoking. Am J Obstet Gynecol 1982;144:857.
22. Sulik KK, Johnston MC, Ambrose LJ, Dorgan D. Phenytoin (dilantin)-induced cleft lip and palate in A/J mice: A scanning and transmission electron microscopic study. Anat Rec 1979;195:243-55.
23. Frank AA, Thompson DJ, Kazacos EA. Teratogenicity of 3,3-dimethyl-1-phenyltriazene in the rat: gross malformations including micrognathism. Teratology 1989;39:53-61.
24. Wong WY, Eskes TK, Kuijpers-Jagtman AM, Spauwen PH, Steegers EA, Thomas CM, *et al*. Nonsyndromic orofacial clefts: association with maternal hyperhomocysteinemia. Teratology 1999;60:253-7.
25. Tolarova M, Harris J. Reduced recurrence of orofacial clefts after periconceptional supplementation with high-dose folic acid and multivitamins. Teratology 1995;51:71-8.
26. Hartridge T, Illing HM, Sandy JR. The role of folic acid in oral clefting. Br J Orthod 1999;26:115-20.
27. McNulty H. Folate requirements for health in different population groups. Br J Biomed Sci 1995;52:110-9.
28. Czeizel AE, Toth M, Rockenbauer M. Population-based case control study of folic acid supplementation during pregnancy. Teratology 1996;53:345-51.
29. Cohen MM. The child with multiple birth defects. 2nd ed. New York: Oxford University Press; 1997.
30. Jones KL. Smith's recognizable patterns of human malformations**.** 5th ed. Philadelphia: WB Saunders Company; 1997.
31. Schubert J, Schmidt R, Raupach HW. New findings explaining the mode of action in prevention of facial clefting and first clinical experience. J Craniomaxillofac Surg 1990; 18:343-7

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