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Review Article

Genetic Basis of Dental Agenesis: Non-Syndromic Hypodontia

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Abstract

Tooth agenesis or hypodontia is one of the most prevalent developmental anomalies of the human dentition which affects up to 8% of the Caucasian population. It is a quite heterogenous condition which describes the congenital absence of one or more teeth and can occur either with a syndrome (syndromic hypodontia) or without (non-syndromic hypodontia). Hypodontia still constitutes a challenging clinical problem. Our insight on the cause of tooth agenesis is increasing as a result of recent advances in the field of molecular biology and human genetics. Further research is needed to establish a genotype phenotype correlation and to fully understand the pathogenesis of tooth agenesis. This review presents the genes and signaling pathways associated with non-syndromic hypodontia, based on the most current literature and provides an overview of novel genes that seem to contribute to dental agenesis.

Keywords: Dental agenesis; Oligodontia; Hypodontia; Genetics; Molecular biology

Introduction

Tooth development begins during embryonic day 9-11, from the oral epithelium under the governance of signal transduction pathways [1,2]. These pathways establish a crosstalk between two adjacent tissues, the mesenchymal stem cells originating from cranial neural crest cells and the primitive epithelium lining the stomatodeum. Genes associated with dental development encode signal transducers, transcription and decisive factors of cell proliferation and differentiation [3]. This constant mesenchyme-epithelium interaction throughout tooth development regulates growth, determines differentiation and pattern formation [4,5]. The type of genes involved as well as their expression affect tooth primordium in such a way that its development may be arrested at the bud or cap or bell stage, or even be altogether abolished [1].

Among dental abnormalities of human dentition, tooth agenesis is the most common, defined as absence of development of one or more teeth [2,6,7]. In this congenital condition, permanent dentition is not fully formed to an extent that varies from 2.2% to 10.1%, depending on factors such as race and nationality. Third molars are the most affected. Upper lateral incisors, mandibular second premolars and maxillary second premolars come next. Agenesis involving first and second molars is very rare [8]. One of the most frequently presented forms of hypodontia is maxillary lateral incisors agenesis [9].

Hypodontia is a clinically challenging problem. One to six teeth are absent with considerable variation in both expressivity and penetrance. Oligodontia represents the congenital absence of six or more teeth and anodontia refers to absence of all teeth [4,6,7,10]. To this classification absence of third molars is not considered.

Up to 8% of Caucasian population have tooth agenesis, mostly in incisors and/or premolars, only 0.25% have oligodontia, whereas anodontia is very rare [10,11]. Hypodontia mainly appears due to genetic causes and can occur, either along with other genetic conditions due to a clinical syndrome or as a non-syndromic form [1]. However, other factors may also be involved in tooth agenesis pathogenesis such as infectious diseases, trauma of the dental region, drug use during pregnancy and chemo or radiotherapy [12]. Non- syndromic hypodontia may be sporadic, or familial, concerning mostly the secondary dentition [4,13-15]. It is inherited in an autosomal-dominant, autosomal recessive, or X-linked mode [6,7,10,16]. Environmental and genetic factors may cause sporadic cases of 1 to 3 missing teeth, again not considering third molars. Hypodontia may be the only abnormality attributed also to non-sporadic cases [4]. Familial non-syndromic hypodontia is presented with significant heterogeneity [6]. More specifically, family members with the condition may be characterized with different location, symmetry and number of teeth involved [16]. Known mutations influence all major signaling transduction pathways and typically entail characteristic phenotypes in teeth affected [2].

Genes in Teeth Development

Tooth morphogenesis involves more than 300 genes. The number and structure of teeth is determined to a great extent by genetic and environmental factors. These factors are multilevel and progressive [7]. Although hypodontia is not considered a wide public health problem, it may be responsible for speech and masticatory difficulties, esthetic problems and malocclusion [17]. Advancements in molecular biology such as the now complete human genome sequences and gene mapping techniques on families known to have hypodontia/oligodontia have contributed a lot in the detection of the genetic factors associated with tooth agenesis [16].

The genes' regulatory role throughout the development of the tooth organ and the relation with all major signal transduction

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cascades and transcription factors mediating these signaling pathways has been identified [1,18]. Changes in the nucleotides in the identified genes lead to malfunctions in proteins as well as altered structure and/or interactions. As a result, tissues may not play their role correctly and lead to missing teeth, due to the breakdown in the signaling cascade [4]. Several genes have a pronounced effect on tooth development (MSX1, PAX9, WNT10, EDA, AXIN2, LEF1, LTBP3), while others' influence is moderate (DLX1, DLX2, GLI2, GLI3) [1].

Mutations in axis inhibition protein 2 (AXIN2), muscle segment homeobox 1 (MSX1), paired box gene 9 (PAX9) and wingless-type MMTV integration site family, number 10 (WNT10A) are identified to have a strong relationship with isolated tooth agenesis [4]. MSX1 and PAX9 were the first genes with detected mutations in isolated tooth agenesis [2,6]. Detection of similar dominant mutations in AXIN2 gene followed and lately, mutations in isolated tooth agenesis have been confirmed in EDA and WNT10A. EDA encodes a signal molecule ectodysplasin which plays a role in the epithelium. Mutations in WNT10A were detected for the first time in patients with recessive Odonto-Onycho-Dermal Dysplasia (OODD) and Schopf-Schulz-Passarge Syndrome (SSPS). WNT10A's impact on tooth agenesis depends on its expression in the epithelial signaling centers (Figure 1) [2]. Frameshift and nonsense mutations are mainly involved and lead to major modification of the primary structure of the protein.

MSX1

This gene codes for a DNA-binding protein which during embryogenesis intervenes in several epithelial-mesenchymal interactions that lead to organogenesis (Figure 1) [12]. At the early stages of tooth formation it is strongly expressed in the dental mesenchyme, mainly during bud and cap stages. It is classified in the muscle segment homeobox family [10,17,19], as a highly conserved sequence found on chromosome 4 comprised of two exons [12]. The homeodomain is a part of the second exon.

To this date fifteen different mutations in MSX1 have been confirmed. Several are in exon 2, while others in exon 1 and 2 are intronic. Most of the exon 2 mutations constitute missense mutations in the homeodomain, which points out to the importance and effect of the structural of integrity of this domain for MSX1 function [10,12]. MSX1 encodes a transcription factor controlling the expression of Bone Morphogenetic Protein 4 (BMP-4) throughout the bud and cap stages of tooth formation (Figure 1) [7]. There is no clear correlation between the severity of the identified missense mutations' consequences and the severity of hypodontia.

A new MSX1 frameshift insertion in the homeodomain of MSX1 gene was identified in DNA samples taken from two sisters with tooth agenesis, concerning the first and second premolars and first molars. It was postulated that this novel mutation plays a pivotal role in DNA binding [10]. Moreover, an aberrant homozygotic 11-nucleotide deletion in the 5' region of the intron, downstream exon 1, was identified in patients with oligodontia. If MSX-1 pre-mRNA canonical or alternative splicing were taken into consideration, the introns' mutations and their impact on tooth agenesis could be further enlightened (Figure 3) [12].

Bonczek and colleagues examined for mutations pertaining to PAX9, MSX1, AXIN2, EDA, EDAR and WNT10a genes. DNA samples from 60 child patients were analyzed by applying whole gene Next Generation Sequencing (NGS). In one female participant, a novel MSX1 mutation was detected causing premature stop codon in the second exon of the gene, at the position G8177G>T, coding now for a considerably truncated MSX1 protein Table 1. The woman exhibited oligodontia with 17 missing teeth [20]. It is considered that MSX1, as a homeobox protein, may have two roles , one in tooth

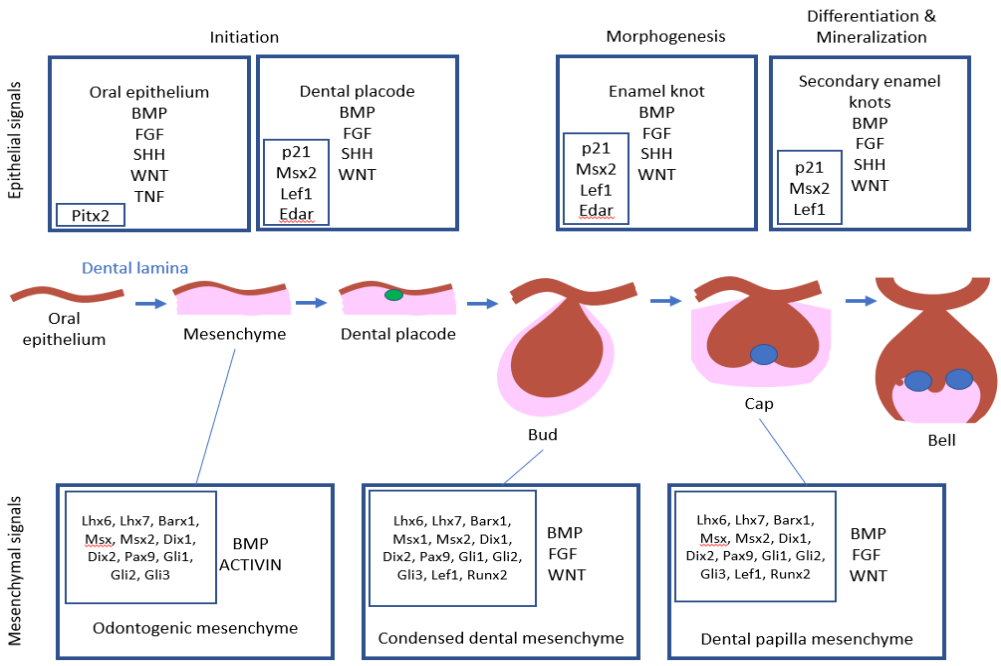

Figure 1: Epithelial-mesenchymal signaling in teeth development.

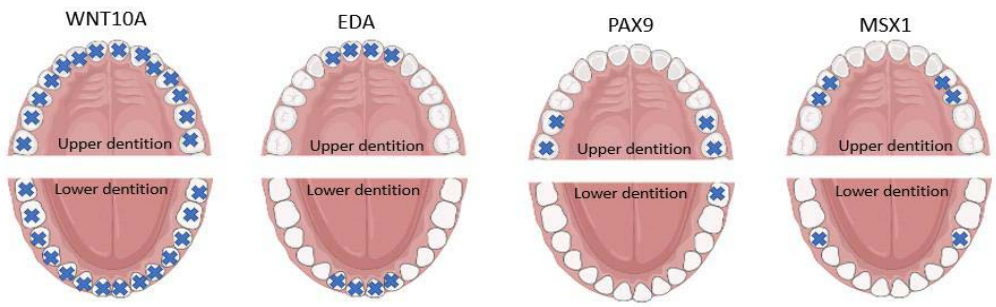

Figure 2: Teeth affected from WNT10A, EDA, PAX9 and MSX1 mutations in upper and lower dentition.

germ generation through the bud stage, as well as in multiplication and differentiation potential of the odontoblasts Table 1.

PAX9

It is a member of gene family encoding transcription factors that regulate cell migration and tissue formation during embryogenesis and are responsible for the epithelial and mesenchymal tissues' interaction (Figure 1) [1,7,14,21]. It is expressed at the future sites of all teeth prior to any signs of tooth development (Figure 2) [13]. Previous works have indicated that small nucleotide alterations at specific locations of the PAX9 gene were related to tooth agenesis and more specifically, to hypodontia in humans [14]. Another PAX family member, PAX6 can control the number of upper incisors.

PAX9 gene is sited at the long arm of chromosome 14 and it consists of 5 exons. The first exon is the 5' untranslated region. The pair-domain is encoded in the third exon. Missense and nonsense mutations have been connected to disruption of tooth formation, since the first identification of an insertion mutation in the paired domain of PAX9 [13,19]. At least 50 mutations in intron and exon regions have been found, twelve of which are located in the intron sequences and near regions of the PAX9 genes Table 1. Recent works showed that PAX9 mutations were detected on single nucleotide or amino acid alterations and premature stop codon with a shorter protein [22].

The majority of these are missense mutations or insertions/deletions in the paired domain [20]. Furthermore, seven missense and three frameshifts PAX9 mutations are related to hypodontia. Many PAX9 frameshifts, deletion and missense termination mutations lead to hypodontia in both the permanent and the primary dentitions, whereas missense substitution mutations concern the permanent dentition only [1]. A former research on non-syndromic type of tooth agenesis analyzed the genotype and phenotype of Chinese families and showed 9 novel and 2 previously known heterozygous mutations in the PAX9 gene among 120 probands [23].

Genetic heterogeneity of PAX9 is very high as, for example, the same mutation can lead to diverse outcomes in the phenotype. As a result, the involvement of PAX9 offers many opportunities for the various mutations to interfere with the fragile balance in teeth development. The nucleotide changes may not only influence PAX9 protein structure or function, but they might have impact on its quantity. Mutations located outside the protein-coding region could affect gene expression or regulation (Table 1) [24].

AXIN2

AXIN2 gene is extensively studied in tooth agenesis, as it determines tooth development to a great extent. Mutations of AXIN2 lead to reduced AXIN2 function and most likely represent mutations that regulate the activation of WNT signaling. AXIN2 inhibits the WNT signaling pathway and it will be discussed later in this section.

LTBP3 is a key regulator of the transforming growth factor beta (TGFB1, TGFB2 and TGFB3) that controls TGF-beta activation by maintaining it in a latent state during storage in extracellular space. It associates specifically via disulfide bonds with the Latency-Associated Peptide (LAP), which is the regulatory chain of TGF-beta, and regulates integrin-dependent activation of TGF-beta (Figure 3).

Lipoprotein Receptor-Related Protein 6 (a member of LDL receptor gene family) is a transmembrane protein [25] found at the long arm of chromosome 11 and expressed in both the mesenchymal and epithelial tissues. Moreover, LRP, being also a receptor for Transforming Growth Factor-b (TGF-beta), determines its bioavailability [1]. TGF-β is a molecule which participates in a signaling event cascade related to cell differentiation. Dental epithelium expresses TGF-β RNA strongly and dental mesenchyme is amplified at the bud and cap stages. Consequently, local TGF-β expression in the dental epithelial tissues leads to cell proliferation in the dental mesenchyme. On the other hand, TGF-β regulates the extracellular matrix formation. The effect of TGF-β gene on tooth agenesis is not widely investigated, so more studies are required [17].

KDF1

Immunohistochemical staining pointed out the importance of KDF1 gene (Keratinocyte Differentiation Factor 1) expression concerning tooth formation. KDF1 was strongly expressed in the dental epithelium at the bud stage, whereas it was absent in the mesenchymal cells. During the cap stage, KDF1 was expressed more in the peripheral region and less in the central in the dental epithelium. Finally, at the bell stage, KDF1 was predominately expressed in the inner enamel epithelium of the tooth bud. A novel pathogenic mutation c.G908C was found in KDF1 gene [26].

GREMLIN2

GREMLIN2 (GREM2) encodes a molecule which antagonizes and plays a role in the regulation of the BMPs (including BMP4) in tissue development. GREM2 mutations differ in expressivity and follow an autosomal dominant with incomplete penetrance type of

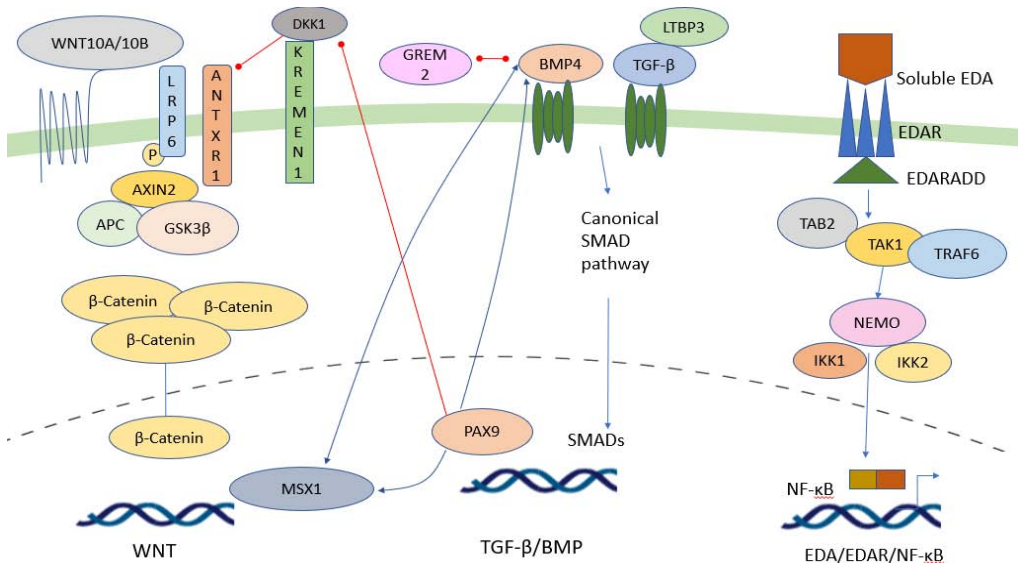

Figure 3: Tooth agenesis gene's interaction inWnt/β-catenin, TGF-β/BMP and EDA/EDAR/NF-κB signaling pathway, respectively.

Table 1: Detailed description of MSX1 and PAX9 gene mutations.

Defectivegene	Location	Mutations Associated with Agenesis	Defect-Mode of Transmission
MSX1	4p16.3-p16.1	M61K, S105X, Q187X, R196P & S202X	Hypodontia-Autosomal Dominant Hypodontia-Autosomal Recessive Oligodontia-Autosomal Dominant
PAX9	14q12-q13	K114X, L21P, R26W, R28P, G51S, K91E, G73fsX316, V265fsX316 & R59fsX177	Oligodontia-Autosomal Dominant Molar Hypodontia-Autosomal Dominant Peg Shaped Laterals-Autosomal Dominant

inheritance. Three mutations (c.38C>T, c.408G>T , c.226C>G) in this gene have been detected so far and lead to isolated tooth agenesis, microdontia, short tooth roots and taurodontism [15,21,27].

LAMA3

The LAMA3 gene provides information for making the alpha subunit of a protein called laminin 332. Researchers have proposed roles for laminin 332 in the clear outer covering of the eye (the cornea) and in the development of tooth enamel. Mutations in this gene are linked to oligodontia. The LAMA3 (c.2798G>T) polymorphism leads to a glycine to valine change (p.G933V), resulting in a destabilized disulfide which alters protein packing and structure [28].

Signaling Pathways

WNT signaling pathway and relevant genes

During embryonic development, WNT signaling pathway is very important for the proliferation and differentiation of various cell types and organs. WNT molecules act through the Frizzled transmembrane receptors and lipoprotein receptor-related protein 5/6 (LRP5/6). Co-receptors activate either the β-catenin dependent canonical pathway, or the β-catenin independent noncanonical pathways (Figure 3) [4,7,21].

Secretion of WNTs, including WNT4, 6, 10, plays a crucial role in tooth formation [21] and when the process is disrupted, the development stops at the early cap stage. WNT10's expression takes place in the dental epithelium and mesenchymal tissues (Figure 1). Many studies have worked on WNT10A and alterations in this gene tend to be found in more than 50% of the affected individuals. Over 50 WNT10A variants are found in 15.8% of patients with 1 to 3 missing teeth and in 52% with >4 missing teeth [28]. Novel Wnt10A mutations (c.521T>C and c.653T>G) were identified in families with specific cases of hypodontia [29]. Non-canonical WNT family includes WNT4, 5a, 11. Absence of WNT5a may have severe consequences in tooth development [4,28,30,31].

The AXIN2 gene downregulates the standard WNT signaling cascade through β-catenin decomposition. Nucleotide alterations in AXIN2 are associated with tooth agenesis as well as other human cancers [4,15,19,21]. LRP6 is a Wnt-associated molecule which encodes part of the co-receptor complex for the transmission of Wnt/β-catenin signaling pathway (Figure 3). Eleven different mutations were acknowledged in patients with non-syndromic tooth agenesis. Transfected LRP6 mutants in mammalian cells led to the failure of Wnt activation. Frameshift, missense, and splice-site LRP6 variants were noted in patients with hypodontia. In a recent study, a novel LRP6 splice-site variant (c.3607+3_6del) was identified, increasing the tooth agenesis cases linked to LRP6. This variant may alter RNA splicing resulting in mRNA susceptible to nonsense- mediated decay [21,25].

Moreover, recessive mutations of the ANTXR1 gene (coding a transmembrane protein) have been identified. In the beginning of tooth formation ANTXR1 was localized in maxillary and mandibular processes, along with the dental epithelium and mesenchyme. Consequently, ANTXR1's expression was localized in the enamel and dental papilla, and then expanded towards the ameloblasts and differentiating odontoblasts. However, the exact contribution of ANTXR1 in tooth agenesis is still unestablished [15]. Novel variants

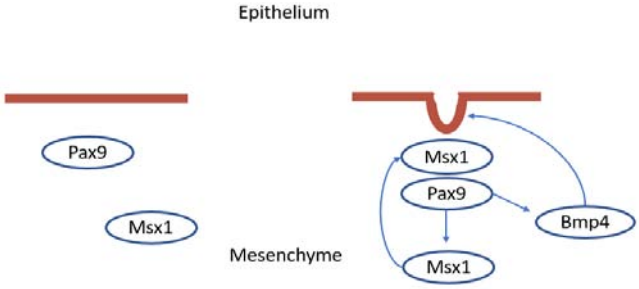

Figure 4: Genes' interaction during teeth morphogenesis.

were found in Dickkopf WNT Signaling Pathway Inhibitor 1 (DKK1) and Kringle Containing Transmembrane Protein 1 (KREMEN1) that are probably pathogenic. It is worth mentioning that scarce mutations of KREMEN1 were recently discovered by WES analysis as variants playing a role in isolated tooth agenesis or oligodontia. KREMEN1 codes for a transmembrane receptor that functionally collaborates with DKK1 to prevent Wnt/β-catenin signaling. More investigation is needed to assess the importance of DKK1 and KREMEN1 variants in tooth agenesis [28].

NF-κB/EDA-EDAR-EDARADD signaling pathway

Nuclear factor NF-κB controls the rate of transcription of genetic information at the cellular level. Members of the TNFR superfamily lead to its activation. The EDA- EDAR-EDARADD signaling pathway is crucial for tooth development when it comes to its growth and morphogenesis (Figure 4). Ectodysplasin- A is a trimeric membrane protein of the TNF-related ligand family which, with the proper stimuli, is attached to its receptor EDAR, which in turn binds to the adaptor EDARADD [7]. After that, TRAF6's interaction with EDARADD results in protein kinase complex activation (including transforming growth factor-β), activated kinase 1 (TAK1) and the activation of TAK1-binding protein (TAB) [4]. Mutations in EDA, EDAR, EDARADD, TRAF6 genes are mainly associated with syndromic tooth agenesis and provoke Hypohidrotic Ectodermal Dysplasia (HED), which includes tooth, hair and sweat gland abnormalities [4,6]. EDA can lead to X-linked HED (XLHED), while EDAR and EDARADD gene mutations are characterized by the autosomal type of heredity. The type of mutation and location in EDA3 are not usually affected by tooth agenesis' severity (number of teeth missing and their position). In the EDA gene, over 210 mutations have been found and most of them might provoke XLHED. On the other hand, the exome sequencing can result in single-nucleotide variants and small insertion/deletion variants. Lately, missense mutations in the EDA gene have provoked familial non-syndromic hypodontia. Moreover, a novel nonsense mutation in the TNF homology domain of ectodysplasin-A, was identified [6]. This mutation prevents specified X-Linked Hypohidrotic Ectodermal Dysplasia (XLHED) [29]. The difference in phenotype between XLHED and tooth agenesis could be associated with the various results of the mutations on EDA.

A recent research study referred to three EDA, one EDAR and two WNT10A mutations in two patients with X-Linked Hypohidrotic Ectodermal Dysplasia (XLHED) and four patients with non-syndromic tooth agenesis. EDA mutation c.1051G>T (p.Val351Phe) and EDAR mutation c.73C>T (p.Arg25*) were observed for the first time. These results expand the existing knowledge concerning the morbidity of mutations in WNT10A and EDAR genes [18]. To be specific, mutations in XLHED lead to the complete loss of EDA expression and, as a result, stop the signaling pathway. On the other hand, mutations in dental agenesis lead to reduced EDA affinity for their target receptors and weak signaling activity.

Nucleotide alterations in the EDA's TNF homology domain could have a connection with familial non- syndromic tooth agenesis. However, the development of anterior teeth requires EDA-EDAR signaling pathway of great quality. Individuals carrying EDA mutations have a significant variation in clinical expression of tooth agenesis [6].

Gene Interactions

PAX9 and MSX1 gene products are transcription factors that interact throughout tooth formation (Figure 4) [12]. PAX9 triggers MSX1 expression and they form a heterodimeric protein complex that transactivates both the MSX1 and the mesenchymal BMP4 gene expression. In the bud stage, BMP2 and BMP4 act in epithelial and mesenchymal dental tissues through MSX1, while in the cap stage, MSX1 boosts the mesenchyme and ensures that odontoblasts do not differentiate, through the prevention of BMP2 and BMP4 expression. In dental mesenchyme, BMP4 signaling promotes tooth development with the aid of MSX1 [15,21,27]. The regulation and the maintenance of the mesenchyme through BMP4 expression and signaling is necessary for the transition from bud to cap stage (Figure 1 and 4) [1,7,12,16].

Mutations in PAX9, MSX1 can affect the physiological protein-protein interactions that hinder normal tooth development processes. Tooth growth is blocked during the bud stage in PAX9 and MSX1 mutant mice [1]. In addition, BMP signaling is very important for MSX1 expression in tooth mesenchyme [13]. On the other hand, WNT/βcatenin signaling aims at MSX1, which then controls the activity of the pathway. Previous works have studied the interaction of TGF-β1 and MSX1 genetic polymorphisms in relation to hypodontia, but their exact role is not understood as yet [17]. Also, PAX9 requires FGF signaling for activating and regulating mesenchymal-epithelial interactions involving BMP4 signaling (Figure 3 and Table 1).

ANTXR1 was considered to modify the normal Wnt signaling pathway concerning the development of the vessels and thus the authors suggested an interaction between ANTXR1, LRP6 and beta-catenin stabilization [15].

The EDA-EDAR-EDARADD- NF-κB signaling pathway contributes significantly to the migration of neural crest cells in the beginning of tooth morphogenesis. EDA controls FGF signaling so that MSX1 would end up being expressed to a great extent. Consequently, EDA mutations impairing EDA-NF-κB-BMP interactions with MSX1, and FGF signaling might disrupt the tooth development [6].

An EDARADD mutation was detected in subjects who were heterozygous when it comes to WNT10A and EDAR2. The majority of these were presented with more severe clinical symptoms than those with only the EDAR or WNT10A allele. As a consequence, a

combination of 2 mutations affecting the EDA pathway probably has a more pronounced impact compared with a single mutation. The alleles leading to common hypodontia are present in an heterozygous state (dominant inheritance), whereas alleles responsible for severe tooth agenesis are usually characterized with biallelic recessive genotypes or allele combinations of different genes [2]. Gene interactions have become a matter of research as they are significant to determine the susceptibility to various diseases [9].

Runx2 is a gene of the odontogenic mesenchyme of the bud stage in odontogenesis (Figure 1). Its nucleotide alterations could cause the creation of spare teeth in the syndrome called human cleidocranial dysplasia. It was found to inhibit the impact of Wnt inhibitors Axin2 and Drapc1 on dental mesenchymal tissue. Evidence shows that the level of mesenchymal Wnt/β-catenin signaling was modulated by Axin2/Runx2 antagonistic interactions [31]. Moreover, EDA is induced by WNT6 and several factors such as WNT10A and WNT10B are its targets. A feedback circuit between BMP and WNT pathways has been confirmed [4].

Maxillary Lateral Incisors Agenesis (MLIA)

In MLIA, deciduous or permanent upper lateral incisors are absent. PAX9, EDA, SPRY2, SPRY4 and WNT10A have a protective influence or increase the risk for MLIA and they actively interact with each other. Interactions were found between TGFA-AXIN2 (indicative of the WNT pathway involvement), MSX1-TGFA and SPRY2-SPRY49 [9].

Missing Teeth

Mutations in MSX1 and PAX9 genes affect mainly the posterior teeth. To be specific, MSX1 mutations concern all second premolars and third molars, whereas PAX9 mutations affect mostly the permanent molars (Figure 2). WNT10A mutations apply to all teeth. More specifically˙ the third molars are the ones that are usually affected, then the second premolars, the maxillary lateral and the mandibular incisors. However, canines, first premolars and second permanent molars are frequently missing in individuals with biallelic WNT10A mutations [2]. In EDA mutations, central/lateral incisors and canines of the maxilla and/or mandible are frequently absent [6].

The different expression levels and the various roles of these genes in normal development are probably liable for varying clinical symptoms. Posterior dentition may depend on the role of MSX1 and PAX9, while anterior dentition requires the proper EDA signaling to determine the size of the ectodermal signaling centers. However, the extent of agenesis and the affected tooth types vary to a great extent among patients [2]. Also, when there is incomplete penetrance and phenotypic variation in female carriers, the random X-chromosome inactivation is considered [12].

Novel Genes

Of the 9 novel TA risk variants, 4 are located in or close to the genes ASCL5/CACNA1S, ARHGAP15, NOL11, and FAM49A, which have not been connected to tooth agenesis before. Gene expression data support ASCL5, whereas other studies propose CACNA1S as the tooth agenesis gene. For the other gene in this region, CACNA1S, the Cacna1sMDG/MDG mouse model frequently comprises secondary cleft palate as well as micrognathia. The ASCL5/CACNA1S locus has been associated with timing of eruption of the primary dentition. The function of ASCL5 is not fully understood [32]. ARHGAP15 is regarded to have a crucial role in variant signaling mechanisms as a Rac-specific GTPase-activating protein. A known TA gene, PITX2 affects the Rac-specific GTPase signaling pathway, suggesting a related function for ARHGAP15 [33]. The FAM49A locus confers risk of Orofacial Clefts (OFC) and agenesis of maxillary lateral incisors. The function of FAM49A is not fully understood; however, a recent work measured FAM49A expression during mouse development in the palatal mesenchymal cells and epithelium cells [32]. Moreover, various dental anomalies were detected in predominance in children with OFC. The maxillary lateral incisors are more commonly missing from subjects with OFC than from controls [34].

The demonstration of the phenotypic traits could even depend on other mechanisms, such as small and long non-coding RNAs and epigenetic alterations [7,24]. Whole Exome Sequencing (WES) may be an efficient way to find defective genes associated with tooth agenesis, especially in sporadic and family cases of oligodontia with big severity [7,10].

To conclude, the specification of the genetic etiology of tooth agenesis is crucial when it comes to genetic counseling to anticipate and minimize issues associated with the clinical management of dental anomalies. A better comprehension of genetic mechanisms and mutations that provoke tooth agenesis will provide accurate means prediction and possible alternative treatment plans in the future.

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References

1. Chhabra N, Goswami M, Chhabra A. Genetic basis of dental agenesis - Molecular genetics patterning clinical dentistry. Med Oral Patol Oral Cir Bucal. 2014; 19: 112-119.

2. Arte S, Parmanen S, Pirinen S, Alaluusua S, Nieminen P. Candidate Gene Analysis of Tooth Agenesis Identifies Novel Mutations in Six Genes and Suggests Significant Role for WNT and EDA Signaling and Allele Combinations. PLoS One. 2013; 8: 1-12.

3. Wen B, Lampe JN, Roberts AG, Atkins WM, Rodrigues AD, Nelson SD. NIH Public Access. 2007; 454: 42-54.

4. Yin W, Bian Z. The Gene Network Underlying Hypodontia. J Dent Res. 2015; 94: 878-885.

5. Brook AH. Multilevel complex interactions between genetic, epigenetic and environmental factors in the aetiology of anomalies of dental development. Arch Oral Biol. 2009; 54: 1-23.

6. Nikopensius T, Annilo T, Jagomägi T, Gilissen C, Kals M, Krjutškov K, et al. Non-syndromic tooth agenesis associated with a nonsense mutation in ectodysplasin-A (EDA). J Dent Res. 2013; 92: 507-511.

7. Salvi A, Giacopuzzi E, Bardellini E, Amadori F, Ferrari L, De Petro G, et al. Mutation analysis by direct and whole exome sequencing in familial and sporadic tooth agenesis. Int J Mol Med. 2016; 38: 1338-1348.

8. Erwin WG, Corkern RW. A pedigree of partial anodontia. J Hered. 1949; 40: 215-218.

9. Alves-Ferreira M, Pinho T, Sousa A, Sequeiros J, Lemos C, Alonso I. Identification of genetic risk factors for maxillary lateral incisor agenesis. J Dent Res. 2014; 93: 452-458.

10. Abid MF, Simpson MA, Petridis C, Cobourne MT, Sharpe PT. Non-syndromic

severe hypodontia caused by a novel frameshift insertion mutation in the homeobox of the MSX1 gene. Arch Oral Biol. 2017; 75: 8-13.

11. Bergendal B, Klar J, Stecksén-Blicks C, Norderyd J, Dahl N. Isolated oligodontia associated with mutations in EDARADD, AXIN2, MSX1, and PAX9 genes. Am J Med Genet Part A. 2011; 155: 1616-1622.

12. Pawlowska E, Janik-Papis K, Wisniewska-Jarosinska M, Szczepanska J, Blasiak J. Mutations in the human homeobox MSX1 gene in the congenital lack of permanent teeth. Tohoku J Exp Med. 2009; 217: 307-312.

13. Pawlowska E, Janik-Papis K, Poplawski T, Blasiak J, Szczepanska J. Mutations in the PAX9 gene in sporadic oligodontia. Orthod Craniofacial Res. 2010; 13: 142-152.

14. Zhang W, Qu HC, Zhang Y. PAX-9 polymorphism may be a risk factor for hypodontia: a meta-analysis. Genet Mol Res. 2014; 13: 9997-10006.

15. Dinckan N, Du R, Akdemir ZC, Bayram Y, Jhangiani SN, Doddapaneni H, et al. A biallelic ANTXR1 variant expands the anthrax toxin receptor associated phenotype to tooth agenesis. Am J Med Genet Part A. 2018; 176: 1015-1022.

16. Vieira AR. Genetics of tooth agenesis: how to move the field forward. J Appl Oral Sci. 2013; 21.

17. Zhang W, Qu HC, Zhang Y. Association of MSX1 and TGF-β1 genetic polymorphisms with hypodontia: meta-analysis. Genet Mol Res. 2014; 13: 10007-10016.

18. Zeng B, Zhao Q, Li S, Lu H, Lu J, Ma L, et al. Novel EDA or EDAR mutations identified in patients with X-linked hypohidrotic ectodermal dysplasia or non-syndromic tooth agenesis. Genes (Basel). 2017; 8: 1-15.

19. Haddaji Mastouri M, De Coster P, Zaghabani A, Jammali F, Raouahi N, Ben Salem A, et al. Genetic study of non-syndromic tooth agenesis through the screening of paired box 9, msh homeobox 1, axin 2, and Wnt family member 10A genes: a case-series. Eur J Oral Sci. 2018; 126: 24-32.

20. Bonczek O, Bielik P, Krejčí P, Zeman T, Izakovičová-Hollá L, Šoukalová J, et al. Next generation sequencing reveals a novel nonsense mutation in MSX1 gene related to oligodontia. PLoS One. 2018; 13: 1-13.

21. Yu M, Wong SW, Han D, Cai T. Genetic analysis: Wnt and other pathways in nonsyndromic tooth agenesis. Oral Dis. 2019; 25: 646-651.

22. Fauzi NH, Ardini YD, Zainuddin Z, Lestari W. A review on non-syndromic tooth agenesis associated with PAX9 mutations. Jpn Dent Sci Rev. 2018; 54: 30-36.

23. Wong SW, Han D, Zhang H, Liu Y, Zhang X, Miao MZ, et al. Nine Novel PAX9 Mutations and a Distinct Tooth Agenesis Genotype-Phenotype. J Dent Res. 2018; 97: 155-162.

24. Bonczek O, Balcar VJ, Šerý O. PAX9 gene mutations and tooth agenesis: A review. Clin Genet. 2017; 2016.

25. Ockeloen CW, Khandelwal KD, Dreesen K, Ludwig KU, Sullivan R, Van Rooij IALM, et al. Novel mutations in LRP6 highlight the role of WNT signaling in tooth agenesis. Genet Med. 2016; 18: 1158-1162.

26. Zeng B, Lu H, Xiao X, Yu X, Li S, Zhu L, et al. KDF1 is a novel candidate gene of non-syndromic tooth agenesis. Arch Oral Biol. 2019; 97: 131-136.

27. Kantaputra PN, Kaewgahya M, Hatsadaloi A, Vogel P, Kawasaki K, Ohazama A, et al. GREMLIN 2 mutations and dental anomalies. J Dent Res. 2015; 94: 1646-1652.

28. Dinckan N, Du R, Petty LE, Coban-Akdemir Z, Jhangiani SN, Paine I, et al. Whole-Exome Sequencing Identifies Novel Variants for Tooth Agenesis. J Dent Res. 2018; 97: 49-59.

29. Zhao K, Lian M, Zou D, Huang W, Zhou W, Shen Y, et al. Novel mutations identified in patients with tooth agenesis by whole-exome sequencing. Oral Dis. 2019; 25: 523-534.

30. Tamura M, Nemoto E. Role of the Wnt signaling molecules in the tooth. Jpn Dent Sci Rev [Internet]. 2016; 52: 75-83.

31. Järvinen E, Shimomura-Kuroki J, Balic A, Jussila M, Thesleff I. Mesenchymal Wnt/β-catenin signaling limits tooth number. Development. 2018; 145: dev158048.

32. Jonsson L, Magnusson TE, Thordarson A, Jonsson T, Geller F, Feenstra B, et al. Rare and Common Variants Conferring Risk of Tooth Agenesis. J Dent Res. 2018; 97: 515-522.

33. Radu M, Rawat SJ, Beeser A, Iliuk A, Tao WA, Chernoff J. ArhGAP15, a Rac-specific GTPase-activating protein, plays a dual role in inhibiting small GTPase signaling. J Biol Chem. 2013; 288: 21117-21125.

34. Eerens K, Vlietinck R, Heidbüchel K, Van Olmen A, Derom C, Willems G, et al. Hypodontia and tooth formation in groups of children with cleft, siblings without cleft, and nonrelated controls. Cleft Palate Craniofac J. 2001; 38: 374-378.