Bioinformatic analysis of genetic variation and functional effects of WNT10A/LRP6 rare variants associated with tooth agenesis

Laboratory UR 2496 Pathologies, Imaging and Orofacial Biotherapy - Molecular diagnosis of isolated dental structure abnormalities

ABSTRACT

Dental inherited rare anomalies are usually classified as structural, developmental and eruption anomalies. All of them can be inherited either as a non-syndromic trait or a as a phenotype part of a larger syndrome. Molecular basis of those anomalies are not still well understood but knowledges are daily improving. In France, the “Filières Santé Maladie Rares”(FSMR) organization has been structuring the diagnosis pathway for rare diseases since 2008 and this includes molecular biology analysis. Many FSMR (“Tête et Cou”, “OSCAR”,“FIMARAD” and others) propose targeted genetic analyses for syndrome or biological pathways including teeth (and/or jaws) anomalies, most of them base on Next Generation Sequencing (NGS) techniques. More recently, genetic approaches to help diagnosing isolated dental anomalies have been developed (IGBMC, Strasbourg, and URP2496, Paris). The most frequent phenotype among dental rare anomalies is tooth agenesis.

Tooth agenesis of more than 6 teeth (excluding wisdom teeth) is considered as a developmental rare disease named oligodontia. Monogenic inheritance (AR, AD, X-linked) has first been described but more recently some authors have reported the combination of two single nucleotide variation (SNV) or single nucleotide polymorphism (SNP) and SNV supporting this phenotype.

A cohort of 181 patients presenting tooth agenesis have been analyzed at the service of “Médecine Génomique des Maladies de Système et d’Organe” (Hôpital Cochin, APHP Centre-UPC). 173 of them have given consent for further research analysis.

The first aim of the project is to identify SNV/SNV or SNV/SNP co-existence in the isolated tooth agenesis cohort and correlate those digenic inheritance/co-inheritance (this term is precisely to be discussed) with accurate phenotyping.

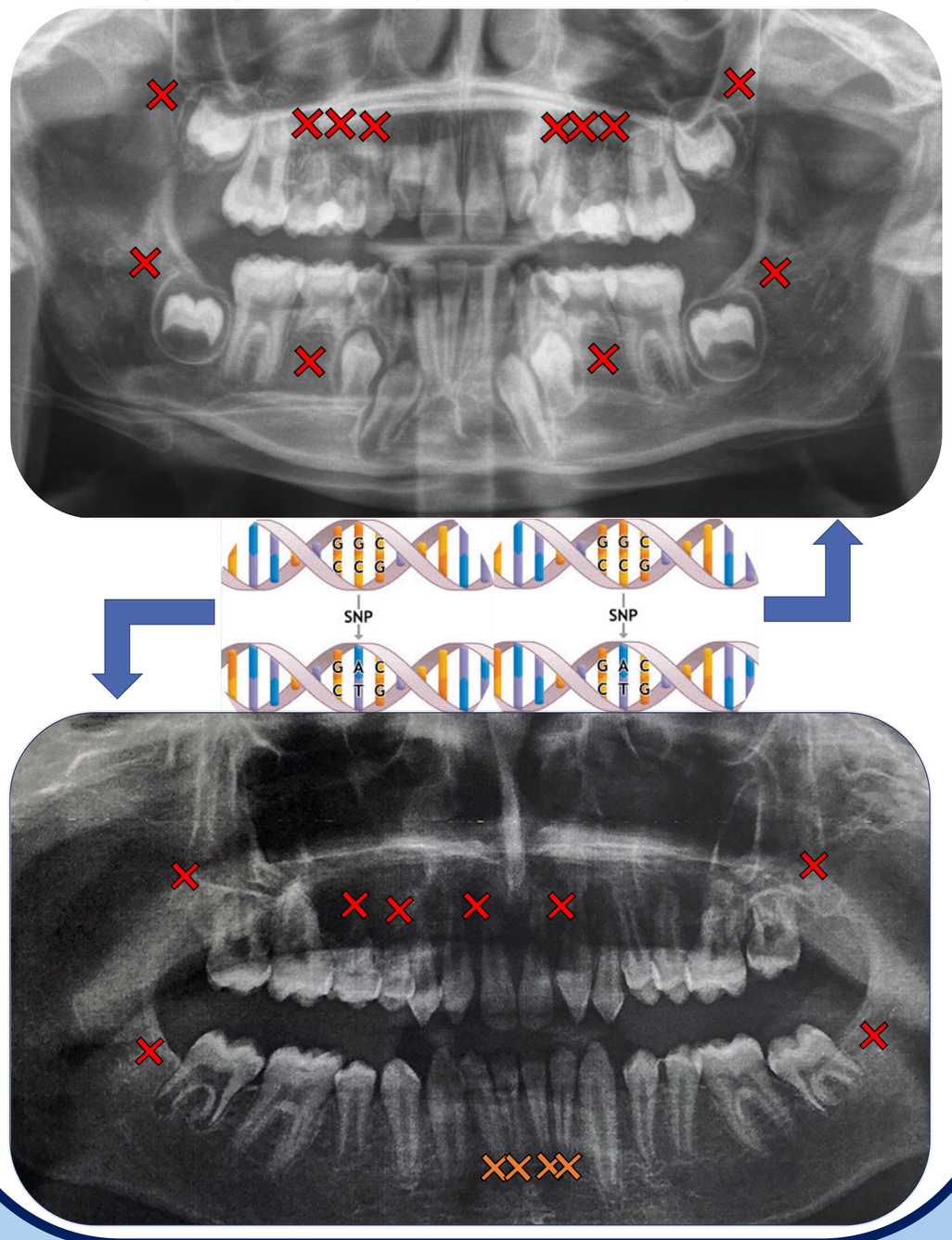
The second aim is to propose a model for functional analysis of WNT10A/LRP6 genetic association reported in the cohort.

To do this, we will first perform bioinformatics analysis on genetic data in variant call format (VCF) from the cohort. In a second step, we will use cultures of wild and mutated dental pulp stem cells (DPSC) by CRISPR-cas9, and then quantify the expression by immunocytochemistry or western blot.

This work could enrich the field of genotype/phenotype correlation in isolated dental agenesis and contribute to a better understanding of the Wnt/Beta-Catenin pathway in dental development.

AIMS

1) Identify the coexistence of SNV/SNV or SNP/SNV pairs in a cohort of patients with dental agenesis, correlate these digenic inheritance and perform precise phenotyping
2) Propose a functional analysis model of the genetic association between WNT10A and LRP6 already identify in the cohort to assess the function of these genes/proteins, the effects on their signaling and the expression of other genes.



DESIGN AND METHODOLOGY

1) Bioinformatics analysis on genetic data in variant call format (VCF) from a cohort of 173 patients with dental agenesis (Department of Genetic Medicine of System and Organ Diseases, Cochin Hospital, AP-HP).

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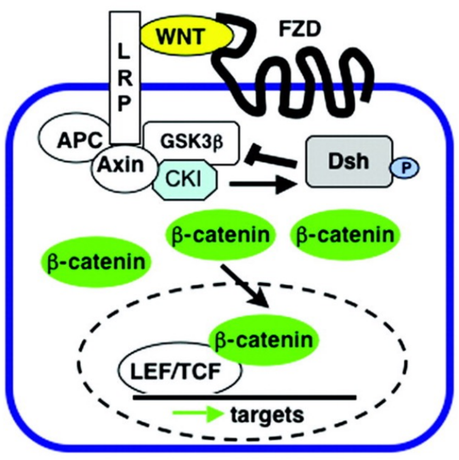
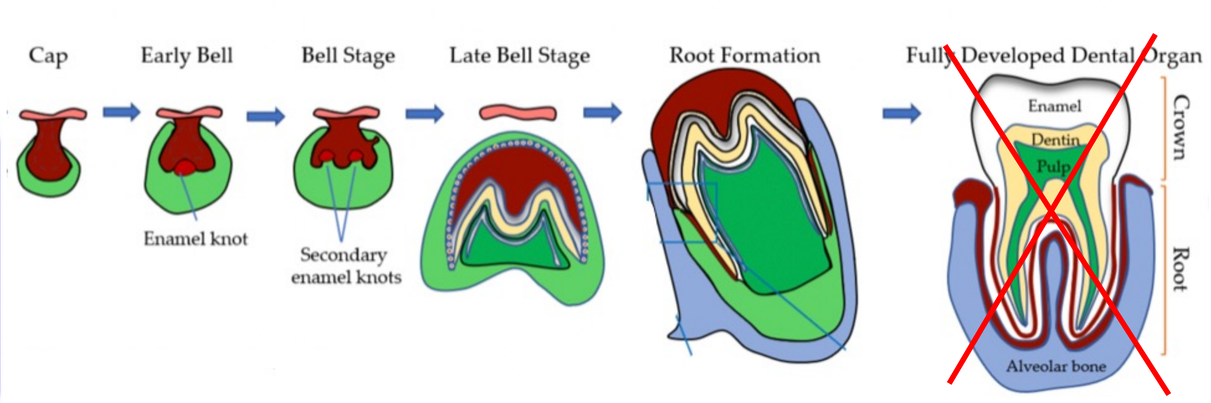
2  81171  .  G  A   .  .    AC=6;AN=7446   GT:DP:GQ   0/1:4:12    0/0:3:9
2  81182  .  A  G   .  .    AC=5;AN=7506   GT:DP:GQ   0/0:5:15    0/0:4:12
   81204  .  T  G   .  .    AC=2;AN=7542   GT:DP:GQ   1/0:5:15    0/0:9:2
```

2) Culture of wild and mutated dental pulp stem cells (DPSC) by CRISPR-cas9, then quantify the expression by immunocytochemistry or western blot.

SIGNIFICANCE, IMPACT AND RELEVANT BACKGROUND

1) The molecular basis of these abnormalities is not yet well understood. This work aims to enrich the field of genotype/phenotype correlation in isolated dental agenesis
2) Contribute to a better understanding of the Wnt/B-Catenin pathophysiological pathway which is an important part of dental development. Enable better interpretation of dental agenesis phenotypes during clinical diagnosis while providing important information for the development of future dental replacement therapies.





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