Nine Novel *PAX9* Mutations and a Distinct Tooth Agenesis Genotype-Phenotype

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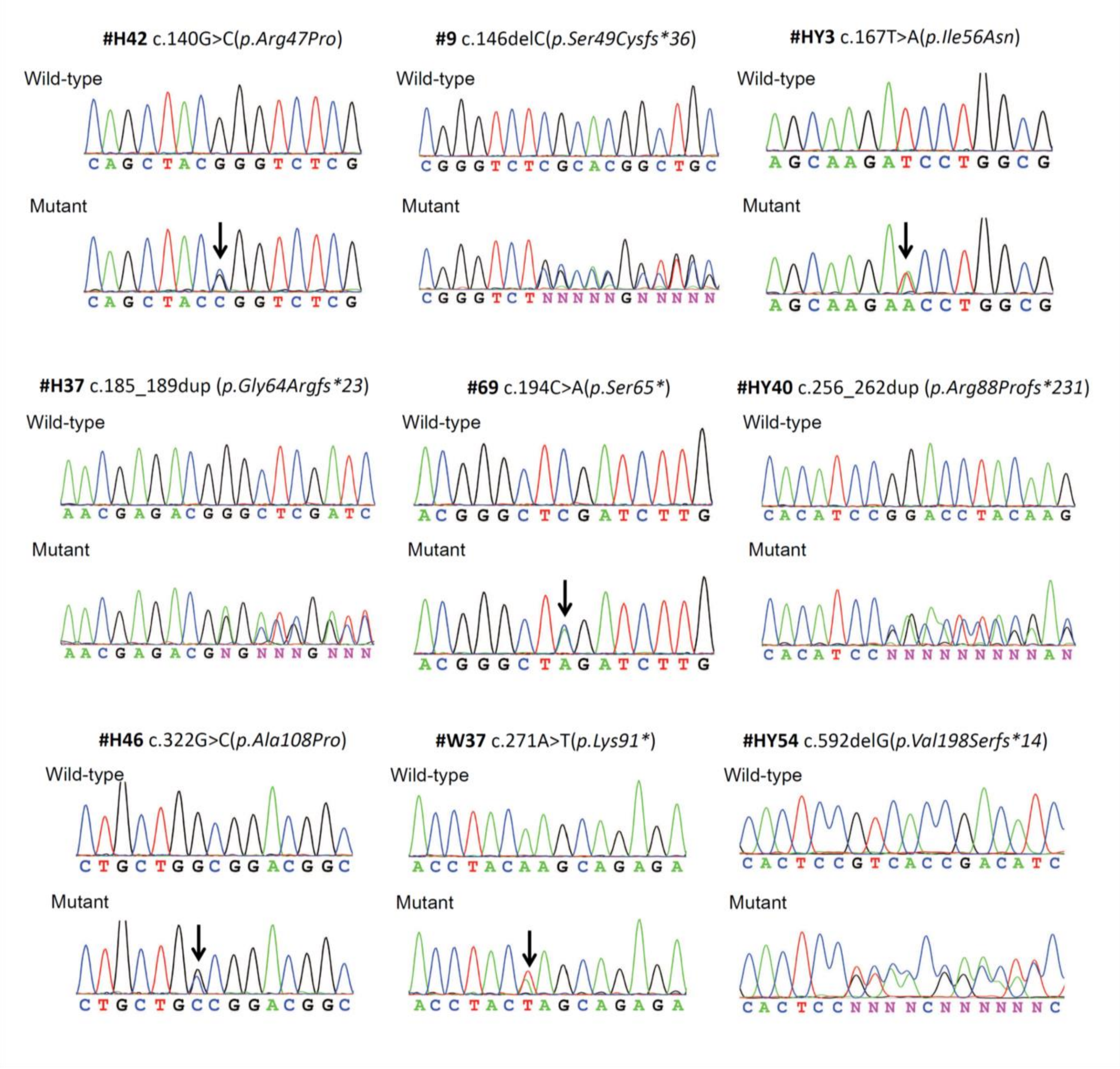
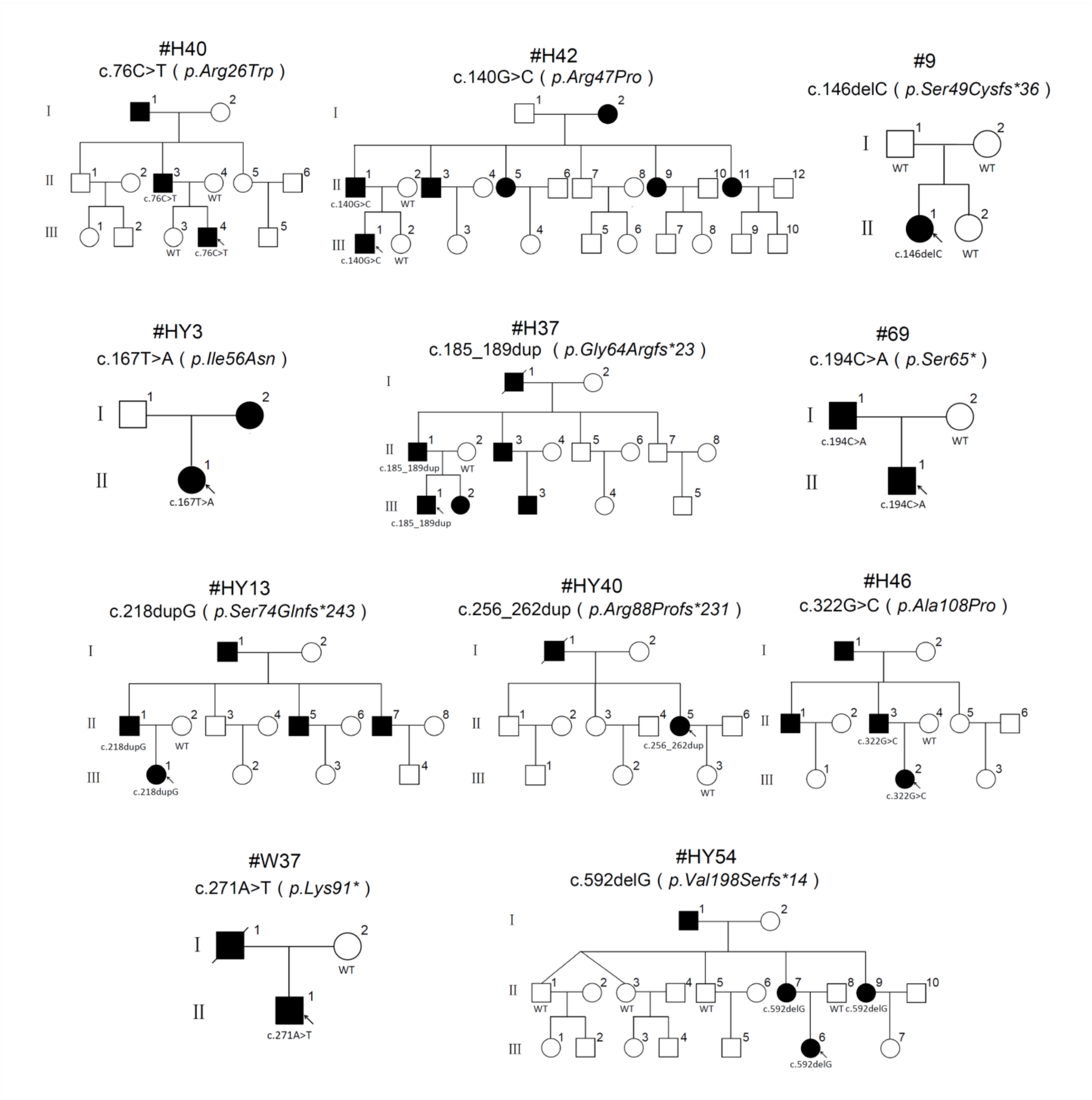
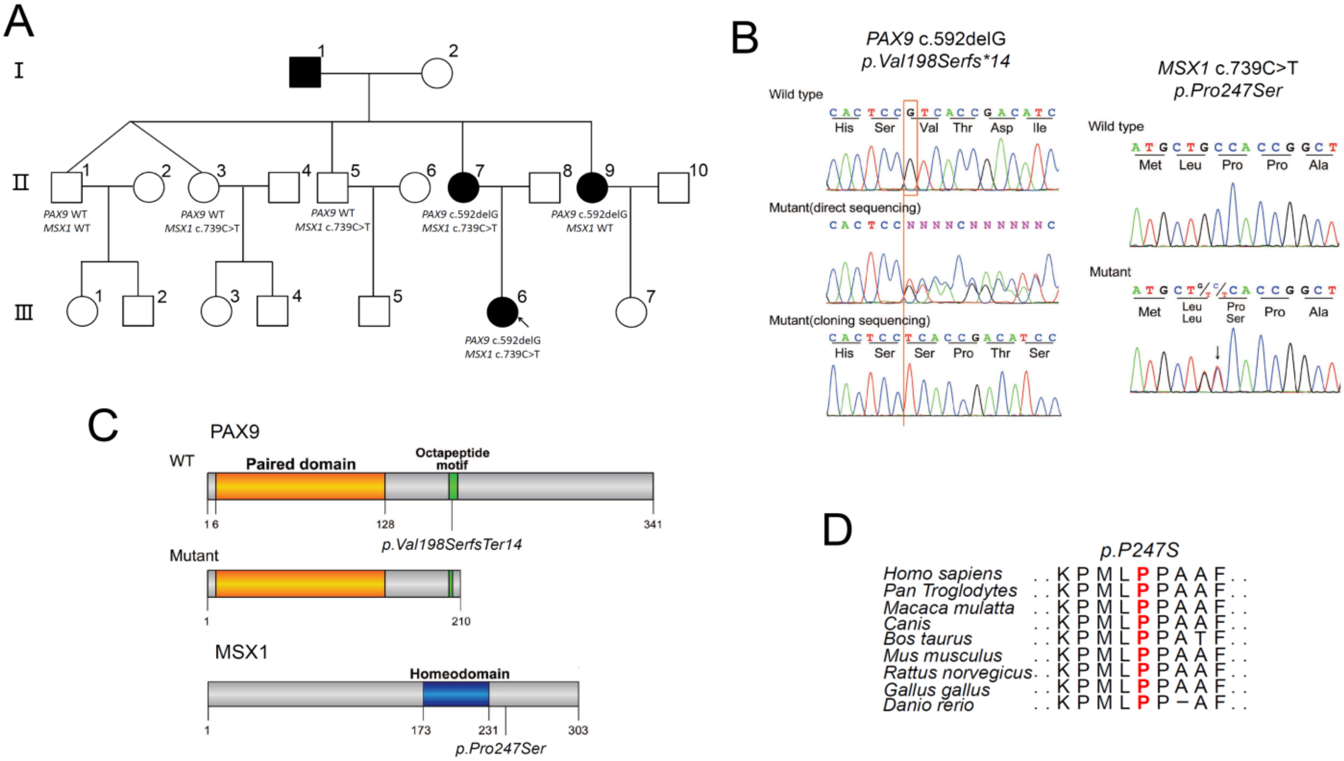
Appendix



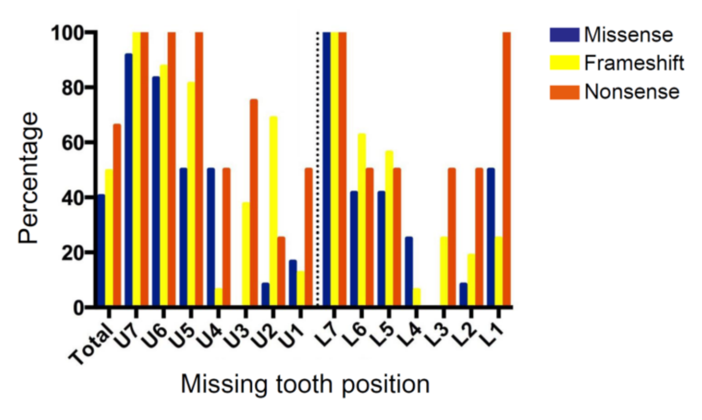
Appendix Figure 1. Nine novel *PAX9* mutations found in patients with tooth agenesis. DNA sequencing chromatograms of the novel *PAX9* mutations. Arrowheads indicate point mutations.



Appendix Figure 2. Pedigrees of tooth agenesis patients with a *PAX9* mutation. Filled circles and squares indicate affected females and males family members, respectively. Arrows indicates the proband of a family.



Appendix Figure 3. The tooth agenesis patient with *PAX9* and *MSX1* digenic mutations. (A) The pedigree of the patient with digenic mutations. (B) DNA sequencing chromatograms of *PAX9* c.592delG and *MSX1* c.739C>T mutations. PCR products harboring the c.592delG frameshift mutation were cloned into pGEM-T Easy Vectors (Tiangen, Beijing, China) to exclusively sequence the mutant allele. (C) Schematic pictures of mutant PAX9 and MSX1. (D) Conservation analysis of residue 247 in MSX1 among nine different vertebrate species.



Appendix Figure 4. The prevalence of missing teeth in patients with different types of *PAX9* mutations (missense, frameshift and nonsense mutations).

Appendix Table 1

Schematic presentation of congenital missing permanent teeth/microdontia in patients with a *PAX9* mutation

■ = the missing tooth; △ = microdontia; □ = unaffected teeth

Family/ Patient #	Genotype	Mutation	Right quadrants								Left quadrants						
			Max	7	6	5	4	3	2	1	1	2	3	4	5	6	7
			Mand	7	6	5	4	3	2	1	1	2	3	4	5	6	7
H40 III:4	*PAX9* c.76C>T	Missense		■	■	□	□	□	△	□	□	■	□	□	■	■	■
				■	□	□	□	□	□	■	■	□	□	■	□	□	■
H40 II:3	*PAX9* c.76C>T	Missense		■	■	□	□	□	□	□	□	□	□	□	□	■	■
				■	■	□	□	□	□	□	□	□	□	□	□	■	■
H42 III:1	*PAX9* c.140G>C	Missense		■	■	■	■	△	□	□	□	□	△	■	■	■	■
				■	■	■	■	□	□	■	■	■	□	■	■	■	■
H42 II:1	*PAX9* c.140G>C	Missense		■	□	□	■	□	□	■	■	□	□	■	□	□	■
				■	□	□	□	□	□	□	□	□	□	□	■	■	■
9 II:1	*PAX9* c.146delC	Frameshift		■	■	■	□	■	■	△	△	△	△	□	■	■	■
				■	■	△	□	□	■	■	■	□	□	□	△	■	■
HY3 II:1	*PAX9* c.167T>A	Missense		■	■	■	■	□	□	□	△	□	□	■	■	■	■
				■	□	■	□	□	□	□	□	□	□	□	■	□	■
H37 III:1	*PAX9* c.185_189dup	Frameshift		■	■	□	□	□	■	△	△	■	□	□	■	■	■
				■	■	□	□	□	■	□	□	■	□	□	□	□	■
H37 II:1	*PAX9* c.185_189dup	Frameshift		■	■	■	□	■	■	■	△	■	□	□	■	■	■
				■	■	■	■	□	□	□	■	□	□	□	■	■	■
69 II:1	*PAX9* c.194C>A	Nonsense		■	■	■	■	■	△	△	△	■	■	■	■	■	■
				■	■	■	□	□	□	■	■	□	□	□	■	■	■
HY13 III:1	*PAX9* c.218dupG	Frameshift		■	■	■	■	△	■	△	□	■	△	■	■	■	■
				■	△	■	△	■	□	□	□	□	■	△	■	△	■
HY13 II:1	*PAX9* c.218dupG	Frameshift		■	△	□	□	■	□	□	□	□	■	□	□	△	■
				■	△	□	□	■	□	□	□	□	■	□	□	△	■
HY40 II:5	*PAX9* c.256_262dup	Frameshift		■	■	■	□	□	□	□	□	□	□	□	■	■	■
				■	■	■	□	□	□	□	□	□	□	□	□	□	■
H46 III:2	*PAX9* c.322G>C	Missense		□	■	□	□	□	△	△	△	△	□	□	■	■	■
				■	□	□	□	□	□	■	■	□	□	□	□	□	■
W37 II:1	*PAX9* c.271A>T	Nonsense		■	■	■	□	■	△	■	■	■	△	□	■	■	■
				■	□	□	□	■	■	■	■	■	■	□	□	□	■
HY54 III:6	*PAX9* c.592delG *MSX1* c.739C>T	Frameshift		■	■	■	□	□	■	□	□	■	□	□	■	■	■
				■	■	■	□	□	□	□	■	□	□	□	■	■	■
HY54 II:7	*PAX9* c.592delG *MSX1* c.739C>T	Frameshift		■	■	■	□	■	■	■	□	■	■	□	■	■	■
				■	■	■	□	□	□	□	□	□	□	□	■	■	■

Appendix Table 2. Percentage of missing teeth and microdontia in different types of *PAX9* mutation

	Missense	**Frameshift**	**Nonsense**	***P* Value**
Missing tooth	40.5%	49.6%	66.1%	0.004
Microdontia	4.76%	9.80%	9.80%	<0.001

Chi-square test was used to determine the differences among groups.

Appendix Table 3. Comparison of the taste score between patients with a PAX9 mutation and controls

	Patient	**Control**	***P* value**
	Mean (SD)	**Mean (SD)**	
Sweet	3.67 (0.52)	3.70 (0.43)	0.91
Bitter	2.33 (0.82)	3.35 (0.87)	0.024
Salty	3.67 (0.52)	3.45 (0.68)	0.55
Sour	3.50 (0.84)	3.60 (0.57)	0.78
Total score	26.33 (1.52)	28.20 (3.48)	0.39

Three available patients with *PAX9* mutations (c.140G>C, c.140G>C and c.322G>C) were recruited to participate in this experiment; age-match healthy control N=10. Mann–Whitney test was used to determine the differences between groups.