Genetic Analyses of Enamel Hypoplasia in Multiethnic Cohorts

Supplementary Materials

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Table S1. DNA collection, storage, Genotyping and dbGaP accessions numbers

	POFC	COHRA1	COHRA2 and Dental SCORE
DNA Collection	Saliva (Oragene)	Blood samples were the priority, but saliva (Oragene), cheek swab (Oragene), or mouthwash buccal cell samples were taken if blood samples were not possible	Saliva or Cheek Swabs (Oragene)
DNA Storage	Saliva (Oragene) samples stored at room temperature	Blood samples and mouthwash samples are refrigerated at 4 degrees Celcius. Saliva (Oragene) samples stored at room temperature	Saliva (Oragene) samples stored at room temperature
Genotyping Chip	Illumina HumanCore+Exome array	Illumina Human610-Quad Beadchip	Illumina Human610-Quad Beadchip
Genotyped SNPs	557,677 SNPs	620,901 SNPs	17,482,50 SNPs
Total SNPs	32,630,291 SNPs (Genotyped + imputed)	16,219,283 SNPs (Genotyped + imputed)	40,405,505 SNPs
dbGaP accession numbers https://www.ncbi.nlm.nih.gov/gap/accession	phs000774.v2.p1	phs000095.v3.p1	phs001591.v1.p1

Table S2. Data Collection

	POFC	**COHRA1, COHRA2 and Dental SCORE**
Questionnaire	Self-reported questionnaires	Self-reported questionnaires
Dental Exam	In-person dental exams Trained and calibrated dental examiners Intraoral photos: 5 photographs Trained calibrated examiner	In-person dental exams Trained and calibrated dental examiners
Years of Examination	2006-2016	COHRA1: 2003 – 2009 Dental Score: 2007 – 2010 COHRA2: 2011 and is ongoing
IRB	Pitt:CR19080127-00 Site:CR19030367-003	COHRA1: Pitt: CR19030392-001 Coordinating Site: CR19100225-001 WV : 1411480509A022 DentalSCORE: MOD19110210-001 COHRA2: Pitt: CR19110013-002 Coordinating Site: CR19080178-001 WV: 1411480509A022

Table S3 . Meta-analysis Results with Heterogenity Statistics

SNP	Chr	BP	Meta-*P*	Direction	Het I^2	Het Q	Het Df	Het *P**
rs1359694	9	9531084	5.86E-07	----	0	2.947	3	0.3999
rs12830414	12	78827822	6.07E-07	++++	32.6	4.45	3	0.2168
rs682846	19	50696240	6.36E-07	----	0	0.366	3	0.9471
rs4649222	1	233491437	1.83E-06	++++	0	1.107	3	0.7755
rs2840075	4	71992629	2.20E-06	----	50.2	6.021	3	0.1106
rs62196465	20	15611956	2.80E-06	----	40.7	5.063	3	0.1672
rs16968212	15	76724882	2.92E-06	++++	0	2.658	3	0.4473
rs9846530	3	122651543	3.69E-06	----	58.3	7.191	3	0.06606
rs7921002	10	80887721	4.10E-06	----	11.1	3.374	3	0.3375

Het I^2: measure of inconsistency
Het Q: Cochran's Q measure of heterogeneity
*Significant heterogeneity based on Q is p<0.10

Table S4 . Lead SNPs from all Cohorts in the Meta-analysis Results

	SNP					POFC		COHRA1		COHRA2		Dental SCORE	
Cohort		**Chr**	**BP**	**Meta-*P***	**Direction**	***P***	**MAF**	***P***	**MAF**	***P***	**MAF**	***P***	**MAF**
POFC	rs12043922	1	154118497	0.03004	--+-	**7.45E-08**	0.06949	0.14154593	0.05223	0.00494044	0.05761	0.76416127	0.05761
	rs9616163	22	47328584	0.0391	+--+	**3.22E-07**	0.06117	0.26296264	0.09908	0.54246182	0.09425	0.45641165	0.09425
	rs55726235	13	34022292	0.156	-+++	**5.12E-07**	0.06841	0.43690116	0.09641	0.20530252	0.1146	0.88820188	0.1146
COHRA1	rs62522905	8	136395352	0.06302	+-++	0.69309218	0.1259	**5.04E-08**	0.1001	0.42707949	0.0912	0.58722706	0.0912
	rs80311444	4	18789773	0.0001425	--+-	0.3047191	0.06305	**1.26E-07**	0.1122	0.90275869	0.1029	0.15611722	0.1029
	rs3888090	19	54317914	0.0001074	++-+	0.11281692	0.09921	**2.37E-07**	0.06076	0.97033179	0.1045	0.30281975	0.1045
	rs1893781	18	42443643	0.0032	+---	0.38305603	0.183	**3.44E-07**	0.1197	0.89445325	0.121	0.12407199	0.121
COHRA2	rs11232439	11	80743780	0.05053	--+-	0.46788552	0.1418	0.37690971	0.08351	**1.24E-08**	0.08397	0.86215315	0.08397
	rs57555457	14	48260934	0.001391	-+++	0.39836976	0.07178	0.06628699	0.09913	**1.80E-07**	0.06224	0.85547366	0.06224
	rs62575356	9	118497768	0.2283	-++-	0.09401534	0.06158	0.40673299	0.05443	**2.77E-07**	0.06036	0.05959045	0.06036
	rs6539378	12	108242099	0.2427	--+-	0.63903735	0.1705	0.19056993	0.06643	**7.15E-07**	0.06561	0.3994382	0.06561
	rs11074423	16	20000021	0.001965	+++-	0.26231365	0.1003	0.73307598	0.05054	**1.43E-06**	0.0654	0.92729066	0.0654
	rs34438514	11	4744156	0.001782	-+++	0.57463329	0.2526	0.13573498	0.2111	**2.56E-06**	0.2252	0.53846048	0.2252
Dental SCORE	rs36010081	1	212423782	0.004492	----	0.66250882	0.09574	0.98660941	0.1798	0.93293181	0.1465	**2.67E-07**	0.1465
	rs7780210	7	139231244	0.1624	-++-	0.38514078	0.07205	0.97561258	0.125	0.00155775	0.1464	**3.06E-07**	0.1464
	rs13416055	2	236558145	0.04821	+---	0.1055595	0.2635	0.66633677	0.2987	0.86679214	0.2783	**6.68E-07**	0.2783
	rs3010205	1	11440957	0.009802	-+--	0.77400975	0.4632	0.21716486	0.369	0.24720619	0.3665	**7.22E-07**	0.3665
	rs7181456	15	59577736	0.1845	--++	0.63914636	0.2661	0.0661771	0.1804	0.97442933	0.1764	**8.30E-07**	0.1764
	rs2395706	6	38749903	0.02698	--++	0.6657175	0.3147	0.34641182	0.2935	0.37651867	0.3131	**8.97E-07**	0.3131
	rs182056767	23	127462931	0.007964	+---	0.05878762	0.05319	0.26404266	0.0598	0.23916044	0.07259	**9.44E-07**	0.07259

Table S5. Information on SNPs from GWAS catalog.

SNP	chr	bp	rsID	PMID	Trait	FirstAuth	Date	P
rs60167359	17	57885709	rs7212590	27863252	Monocyte count	Astle WJ	2016-11-17	2E-30
rs2840075	4	71991184	rs11729023	30595370	Heel bone mineral density	Kichaev G	2018-12-27	2E-19
rs60167359	17	57885709	rs7212590	27863252	Monocyte percentage of white cells	Astle WJ	2016-11-17	4E-19
rs2840075	4	71991184	rs11729023	30048462	Heel bone mineral density	Kim SK	2018-07-26	8E-19
rs2840075	4	71991184	rs11729023	30598549	Heel bone mineral density	Morris JA	2018-12-31	1E-18
rs60167359	17	57885709	rs7212590	27863252	Granulocyte percentage of myeloid white cells	Astle WJ	2016-11-17	5E-15
rs60167359	17	57934654	rs58681483	29403010	Monocyte count	Kanai M	2018-02-05	1E-12
rs2840075	4	71991184	rs11729023	28869591	Heel bone mineral density	Kemp JP	2017-09-04	5E-10
rs60167359	17	57934654	rs58681483	30696823	Chronotype	Jones SE	2019-01-29	1E-9
rs1359694	9	9550389	rs58823963	26545240	Pelvic organ prolapse (moderate/severe)	Giri A	2015-11-06	9E-7

Table S6. Gene Set Enrichment Analyses Results

Category	GeneSet	Ngenes	N_overlap	p	adjP	genes
Chemical_and_Genetic_pertubation	FARMER_BREAST_CANCER_CLUSTER_5	19	3	7.95E-07	0.00262618	VMP1:TUBD1:RPS6KB1
Positional_gene_sets	chr17q23	56	4	2.46E-07	6.75E-05	VMP1:TUBD1:RPS6KB1:RNFT1
Positional_gene_sets	chr15q24	65	4	4.52E-07	6.75E-05	ISL2:SCAPER:RCN2:PSTPIP1
Curated_gene_sets	FARMER_BREAST_CANCER_CLUSTER_5	19	3	7.95E-07	0.0043751	VMP1:TUBD1:RPS6KB1
TF_targets	PAX4_02	235	4	7.42E-05	0.04528779	ZMIZ1:SLC6A5:SEMA5B:ELMO1

Table S7. Enamel Genes in Meta-analysis Results

Gene	SNP	Chr	BP	SNP-count	*P*	Type	Sig
ENAM	**rs2840075**	**4**	**71992629**	**7836**	**2.20E-06**	**Imputed**	**yes**
TUFT1	rs6675984	1	151498624	189	0.002073	Imputed	no
AMBN	rs34012013	4	71449613	302	0.02754	Imputed	no
TFIP11	rs9613203	22	26945541	314	0.04368	Imputed	no
AMELX	rs62590037	23	11259693	66	0.1152	Imputed	no

Li and Ji Method for Number of Independent Tests=0.05/279= 0.0001792115=1.79 $\times 10^{04}$

LeadSNP	chr	pos	P-value
rs1359694	9	9531084	5.859e-07
rs12830414	12	78827822	6.067e-07
rs682846	19	50696240	6.358e-07
rs56282801	12	125459514	1.004e-06
rs4649222	1	233491437	1.834e-06
rs2840075	4	71992629	2.196e-06
rs62196465	20	15611956	2.799e-06
rs16968212	15	76724882	2.921e-06
rs9846530	3	122651543	3.691e-06
rs7921002	10	80887721	4.096e-06

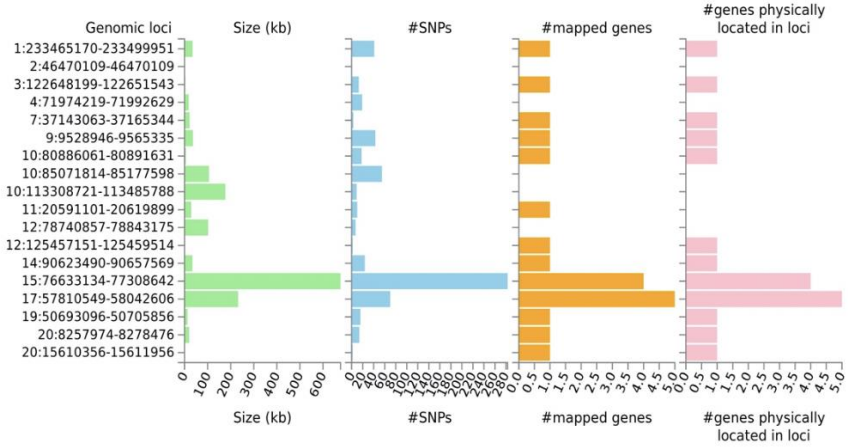


Figure S1. FUMA analysis of meta-GWAS results. GWAS risk loci ($P < 5.0 \times 10^{-6}$) with Lead SNPs and FUMA annotations.

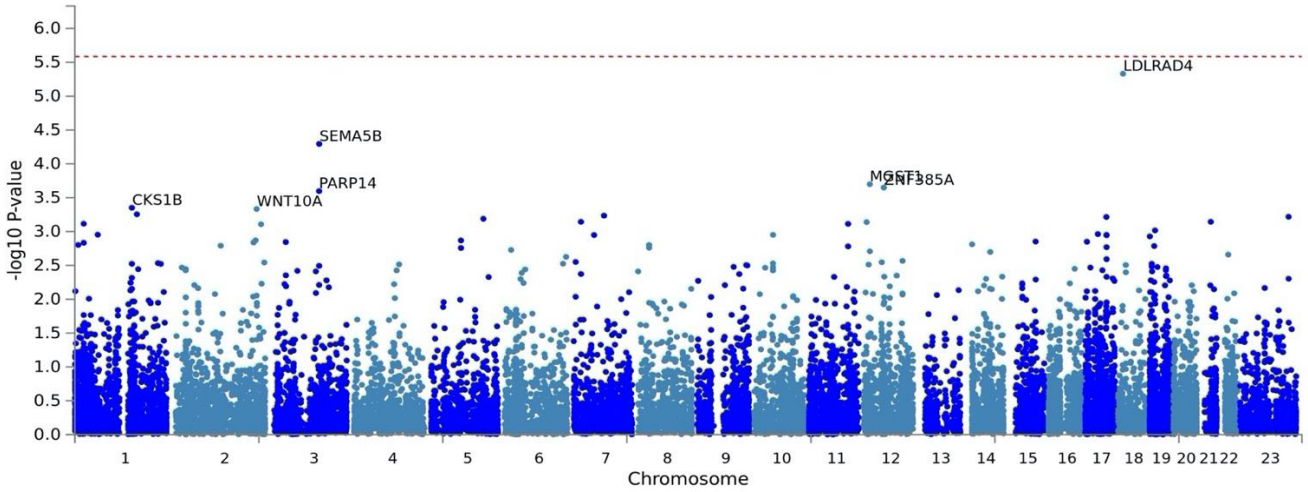


Figure S2. Manhattan Plot (gene-based test) of the meta-GWAS results

Visual Dictionary for Dental Anomalies

Brian J. Howe DMD, MS

Update 03/2016

General Rules for Scoring Photos

- When permanent tooth is erupting and remnants of primary tooth is still present = say that permanent tooth is present and not the primary tooth.
- If tooth is blurry, and unable to clearly see outline of tooth = not visible
- If teeth are not visible in photo = not visible
- If unable to tell what tooth is present (position of the tooth) = not visible, unable to tell position
- If root tips and unable to tell position of the tooth = not visible/unable to tell position, root tips
- If primary teeth present and permanent first molar and posterior have not erupted = not present – missing age appropriate
- If primary teeth present and permanent first molar erupted, but second permanent molar not erupted = not present – missing age appropriate
- If permanent second molar erupted and permanent canine not erupted = not present, impacted
- If 3rd molars are not present, with no history of 3rd molar extractions, = not visible

Hypoplasia

Pits, grooves, or larger areas of missing enamel

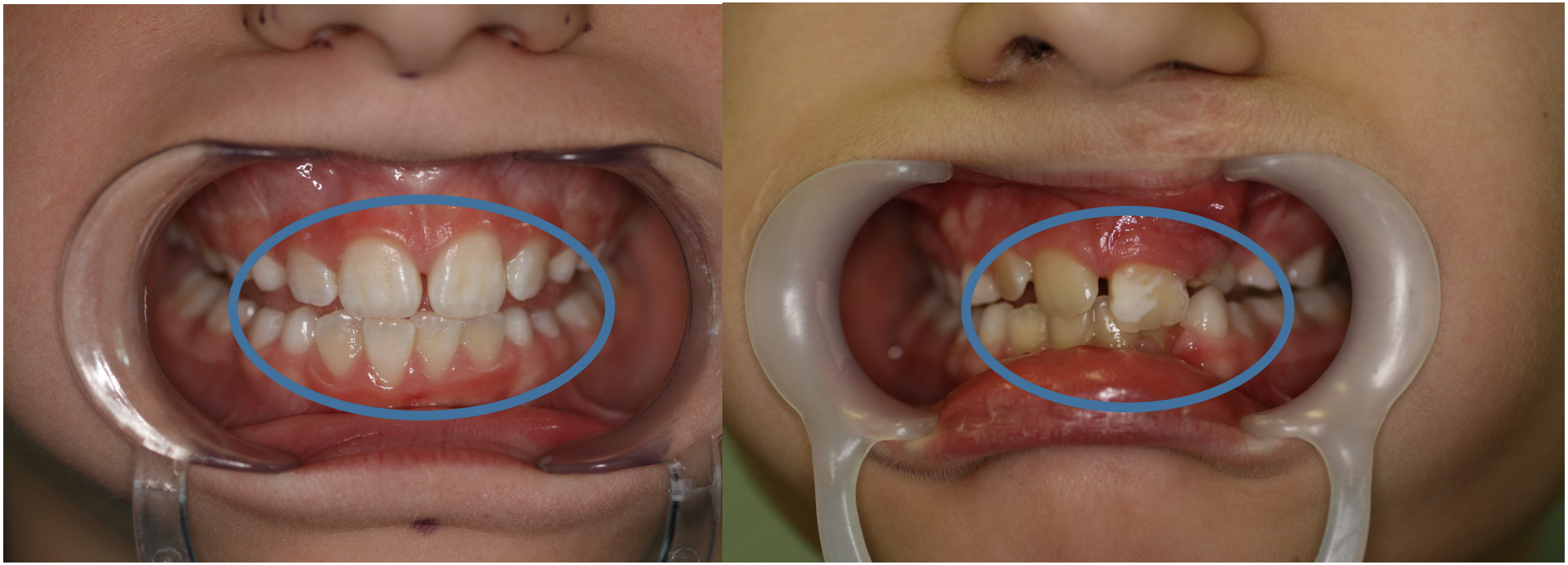
Hypoplasia: A defect involving the surface of the enamel associated with a reduced thickness of enamel. It can occur in the forms of pits, grooves, or large areas of missing enamel.

Hypoplasia

Hypoplasia

Hypocalcification

Areas of white opacity on the tooth, not associated or directly adjacent to areas of decay



Hypocalcification: defect in enamel characterized by reduced calcification

Hypocalcification

Hypocalcification