

Genetic and Epidemiologic Assessment of Mandibular Cortical Indices and Bone

Mineral Density in Peripubertal Children: The Generation R Study

Clinical Oral Investigations

Vid Prijatelj^{a,b,c*}, (MPharm), Olja Grgic-Chavez^{a,b,c*}, (DMD, PhD), Justin van der Tas^{a,b}, (DMD, PhD), Constanza L. Andaur Navarro^a, (DMD, PhD), Andre G. Uitterlinden^{b,c} (PhD), Fernando Rivadeneira^{a,b,c} (MD, PhD), Eppo B. Wolvius^{a,b} (DMD, PhD), Carolina Medina-Gomez^{b,c,d}, (PhD)

m.medinagomez@erasmusmc.nl

SUPPLEMENT

Mandibular indices measurements

DPRs were performed during the same visit as DXA scans, using the orthopantomograph® OP200D (Oldelft Benelux B.V., Veenendaal, the Netherlands). Before scanning, participants were instructed to remove glasses, elastic bands from the hair, and any jewelry that may influence the scan. A chin rest was provided when participants were missing front teeth or had malocclusion. Otherwise, a bite block was used. Scans were then performed following the manufacturer's specifications. A trained dentist assessed the mandibular cortical thickness, providing bilateral measurements for mental index (MI) and panoramic mandibular indices (PMI superior and inferior) on DPRs. The arithmetic means for each of the three bilateral measurements were established when possible. When only unilateral measurements were available, those from the sinistral side of the mandible were used. MI was measured as the cortical thickness on an axis that crossed the approximate center of the mental foramen and formed a 90° angle with an axis tangential to the lower mandibular rim. The PMI superior was calculated as the ratio of the MI and the distance between the mental foramen's upper border and the mandible's lower border, measured on the same vertical axis. The PMI inferior was calculated in the same manner, only using the distance measured to the lower border of the mental foramen (instead of the upper one).

Genotyping, quality control and imputation of GENR4 dataset

Initial QC consisted of the exclusion of samples with a call rate below 97.5%, excess of heterozygosity (more than 4 standard deviations from the mean), or that were deemed to be sex mismatches and/or genetic duplicates. In addition, variants with a call rate below 97.5% and Hardy-Weinberg equilibrium (HWE) deviation ($p\text{-value} < 1 \times 10^{-7}$) were excluded. ZCall was used on previously uncalled genotypes aiming to improve the call rate of less-frequent to rare variants (Goldstein et al PMID: 22843986) after which filtering on missingness was more stringent ($>99\%$) for both variants and samples. Lastly, a comparison of reported familiar relationships based on questionnaires across both subsets of participants and genetically derived familiar relationships resulted in detection and exclusion of sample swaps. The first 20 genomic principal components were calculated for all participants of both datasets simultaneously. Roughly 27,000 independent and common SNPs (minor allele frequency > 0.05) that were in common between GENR3, GENR4, and the 1000 Genomes Project Phase III version 5 (1000 GP; 1000 Genomes Project Consortium PMID:26432245) data were used in the calculation. GENR4 genotypes were phased using ShapeIT v2 r904 (Delaneau PMID:25653097), followed by imputation using minimac4 (Das PMID:27571263) with reference to the 1000 GP using an in-house pipeline.

Supplementary GWAS results

Table S1. Genome-wide significant variants in the PMI-GWAS meta-analysis. All effect sizes (B) are reported for the effect allele (EA). Chr – Chromosome, BP – chromosomal position on genome build 37, OA – Other allele, Freq – Frequency of the effect allele, SE – standard error, P – p-value, N – sample size

Chr	BP	EA	OA	Freq	B	SE	P	N
11	196944	t	c	0.18	0.209	0.035	2.526e-09	3324

11	200343	t	c	0.83	-0.213	0.036	4.422e-09	3324
11	202812	a	g	0.18	0.191	0.033	1.051e-08	3324
11	205021	c	g	0.82	-0.191	0.033	1.061e-08	3324
11	205889	t	c	0.82	-0.191	0.033	1.069e-08	3324
11	202856	a	g	0.18	0.191	0.033	1.083e-08	3324
11	208235	a	g	0.18	0.191	0.033	1.091e-08	3324
11	225180	a	g	0.17	0.195	0.034	1.191e-08	3324
11	237737	a	t	0.18	0.189	0.033	1.621e-08	3324
11	237209	a	g	0.18	0.189	0.033	1.642e-08	3324
11	212408	t	c	0.18	0.188	0.033	1.664e-08	3324
11	229302	a	g	0.18	0.189	0.033	1.683e-08	3324
11	240754	a	c	0.82	-0.189	0.034	1.714e-08	3324
11	227798	a	t	0.18	0.189	0.033	1.729e-08	3324
11	230951	a	g	0.18	0.189	0.034	1.776e-08	3324
11	231305	a	g	0.18	0.189	0.034	1.786e-08	3324
11	226083	d	i	0.18	0.188	0.033	1.827e-08	3324
11	229811	t	g	0.82	-0.188	0.034	1.983e-08	3324
11	234349	t	c	0.82	-0.188	0.034	2.032e-08	3324
11	217171	t	c	0.18	0.188	0.034	2.034e-08	3324
11	242859	a	g	0.82	-0.185	0.033	2.306e-08	3324
11	247028	a	g	0.18	0.185	0.033	2.503e-08	3324
11	218640	a	g	0.22	0.173	0.032	4.45e-08	3324

Table S2. Genome-wide significant variants in the MI-GWAS meta-analysis. All effect sizes (B) are reported for the effect allele (EA). Chr – Chromosome, BP – chromosomal position on genome build 37, OA – Other allele, Freq – Frequency of the effect allele, SE – standard error, P – p-value, N – sample size

Chr	BP	EA	OA	Freq	B	SE	P	N
11	200343	t	c	0.83	-0.328	0.036	7.298e-20	3324
11	196944	t	c	0.18	0.315	0.035	1.034e-19	3324
11	205021	c	g	0.82	-0.298	0.033	1.792e-19	3324
11	205889	t	c	0.82	-0.298	0.033	1.792e-19	3324
11	208235	a	g	0.18	0.298	0.033	1.82e-19	3324
11	202812	a	g	0.17	0.298	0.033	1.862e-19	3324
11	202856	a	g	0.18	0.298	0.033	1.889e-19	3324
11	212408	t	c	0.18	0.296	0.033	2.999e-19	3324
11	227798	a	t	0.18	0.296	0.033	4.086e-19	3324
11	217171	t	c	0.18	0.296	0.033	4.13e-19	3324
11	229302	a	g	0.18	0.296	0.033	4.213e-19	3324
11	230951	a	g	0.18	0.296	0.033	4.657e-19	3324
11	226083	d	i	0.18	0.295	0.033	4.733e-19	3324
11	231305	a	g	0.18	0.296	0.033	4.927e-19	3324
11	225180	a	g	0.17	0.302	0.034	5.293e-19	3324
11	229811	t	g	0.82	-0.295	0.033	5.779e-19	3324
11	234349	t	c	0.82	-0.295	0.033	6.629e-19	3324
11	269129	t	c	0.21	0.277	0.031	6.811e-19	3324
11	237737	a	t	0.18	0.291	0.033	1.52e-18	3324

11	237209	a	g	0.18	0.291	0.033	1.551e-18	3324
11	201584	a	t	0.16	0.313	0.036	1.581e-18	3324
11	247028	a	g	0.18	0.288	0.033	1.718e-18	3324
11	240754	a	c	0.82	-0.291	0.033	1.744e-18	3324
11	242859	a	g	0.82	-0.286	0.033	2.237e-18	3324
11	270330	a	t	0.80	-0.280	0.032	2.564e-18	3324
11	268927	t	c	0.20	0.276	0.032	2.934e-18	3324
11	268940	t	c	0.80	-0.275	0.032	3.352e-18	3324
11	261209	t	c	0.20	0.273	0.032	4.992e-18	3324
11	262930	t	c	0.20	0.273	0.032	5.193e-18	3324
11	252851	a	g	0.80	-0.272	0.032	5.25e-18	3324
11	256978	t	c	0.20	0.272	0.032	5.562e-18	3324
11	270715	t	c	0.80	-0.269	0.031	8.025e-18	3324
11	274181	a	c	0.19	0.284	0.033	8.068e-18	3324
11	268718	a	g	0.78	-0.257	0.030	1.853e-17	3324
11	280464	c	g	0.81	-0.291	0.034	2.534e-17	3324
11	247905	a	c	0.17	0.344	0.041	4.454e-17	3324
11	217814	t	g	0.16	0.300	0.036	4.759e-17	3324
11	267568	a	g	0.22	0.254	0.030	6.022e-17	3324
11	218640	a	g	0.22	0.261	0.031	7.719e-17	3324
11	268628	a	g	0.22	0.253	0.030	7.752e-17	3324
11	249408	a	g	0.18	0.268	0.032	1.416e-16	3324
11	249131	a	g	0.18	0.268	0.032	1.55e-16	3324

11	256976	a	c	0.22	0.250	0.030	1.97e-16	3324
11	267250	a	g	0.75	-0.245	0.030	2.045e-16	3324
11	266957	t	g	0.25	0.245	0.030	2.175e-16	3324
11	266921	a	g	0.75	-0.245	0.030	2.218e-16	3324
11	266804	t	c	0.25	0.245	0.030	2.269e-16	3324
11	268430	c	g	0.77	-0.247	0.030	2.594e-16	3324
11	265777	t	g	0.25	0.244	0.030	2.661e-16	3324
11	266248	t	c	0.75	-0.244	0.030	2.67e-16	3324
11	266315	t	c	0.75	-0.244	0.030	2.915e-16	3324
11	265602	a	g	0.75	-0.243	0.030	3.213e-16	3324
11	263868	d	i	0.25	0.242	0.030	4.395e-16	3324
11	218055	d	i	0.95	-0.713	0.088	7.212e-16	3324
11	240844	t	c	0.78	-0.244	0.031	3.31e-15	3324
11	283415	a	g	0.17	0.294	0.038	5.456e-15	3324
11	268183	t	c	0.20	0.265	0.034	6.391e-15	3324
11	243268	t	g	0.78	-0.236	0.030	9.623e-15	3324
11	242126	d	i	0.21	0.245	0.032	1.133e-14	3324
11	268020	a	g	0.72	-0.217	0.029	1.427e-13	3324
11	262191	a	c	0.68	-0.205	0.028	5.234e-13	3324
11	254421	a	g	0.71	-0.192	0.028	5.121e-12	3324
11	279214	c	g	0.27	0.201	0.031	4.409e-11	3324
11	270071	t	c	0.53	-0.156	0.025	5.759e-10	3324
11	269147	t	c	0.51	-0.151	0.025	1.09e-09	3324

11	244552	a	g	0.43	0.156	0.026	1.4e-09	3324
11	279798	a	g	0.35	0.218	0.036	1.71e-09	2370
11	259575	t	c	0.09	0.292	0.048	1.724e-09	3324
11	249105	a	g	0.05	0.455	0.076	2.303e-09	3324
11	280000	a	g	0.35	0.214	0.036	3.92e-09	2370
11	198986	t	c	0.55	-0.148	0.026	6.365e-09	3324
11	247998	t	c	0.10	0.261	0.045	8.427e-09	3324
11	253072	t	c	0.57	-0.145	0.025	1.111e-08	3324
11	247906	c	g	0.54	0.185	0.033	2.455e-08	3324
11	211015	t	c	0.41	-0.140	0.026	4.871e-08	3324