Phenotypic dissection of bone mineral density facilitates the identification of skeletal site specificity in the genetic regulation of bone mass attainment

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INTRODUCTION:

The heritability of bone mineral density (BMD) varies across skeletal sites, reflecting different relative contributions of environmental and genetic influences. Genetic studies using paediatric BMD measures may target factors preferentially involved in bone mass attainment.

AIM:

I: Quantify the degree to which common genetic variants and environmental factors influence BMD at different skeletal sites.

II: Identify genetic variants which are preferentially associated with the attainment of bone mass at one or more skeletal sites.

METHODS:

Phenotypes:

- BMD measured by whole-body DXA in 4890 ALSPAC children
- partition scans: upper-limb (UL), lower-limb (LL) and skull (S)

Genome wide complex trait analysis (GCTA):

- performed on each skeletal site using 500K genotyped SNPs
- quantify the narrow sense heritability (h^2)
- estimate pair wise genetic (r_g) and environmental (r_e) correlation

Genome-wide association meta-analysis (GWAMA):

- linear regression: BMD at each site and 2.5M imputed SNPs
- meta-analysis: ALSPAC (n=5299) and GEN-R (n=4098)

RESULTS I: GCTA ANALYSIS

Heritability estimates of BMD:

Skull (h^2=0.51, SE=0.07, P=2.0x10^{-13})
Lower limb (h^2=0.40, SE=0.07, P=8.0x10^{-9})
Upper limb (h^2=0.39, SE=0.07, P=2.0x10^{-8})

Genetic and Environmental correlations:

TRAIT 1	TRAIT2	r_g	SE	r_e	SE
LL-BMD	**UL-BMD**	**0.78**	0.067	0.55	0.055
	S-BMD	0.43	0.100	**0.20**	0.088
UL-BMD	**S-BMD**	0.58	0.091	0.24	0.085

Table 1: Estimates of the pair-wise genetic and environmental correlations between skeletal sites.

RESULTS II: GWAS ANALYSIS

Genome wide association:

- 12 previously published BMD-associated variants replicated

Site specific association:

- magnitude of effect size differed between skeletal sites **-Table 2**
- for example: *WNT16/CPED1* locus - **Figure 1**
- larger per allele effect for S- and/or UL-BMD vs. LL-BMD.

Novel association:

- *RIN3* and LL-BMD (rs754388: β=0.13, SE=0.02, P=1.4x10^{-10})
- *RIN3* previously associated with Pagets disease.

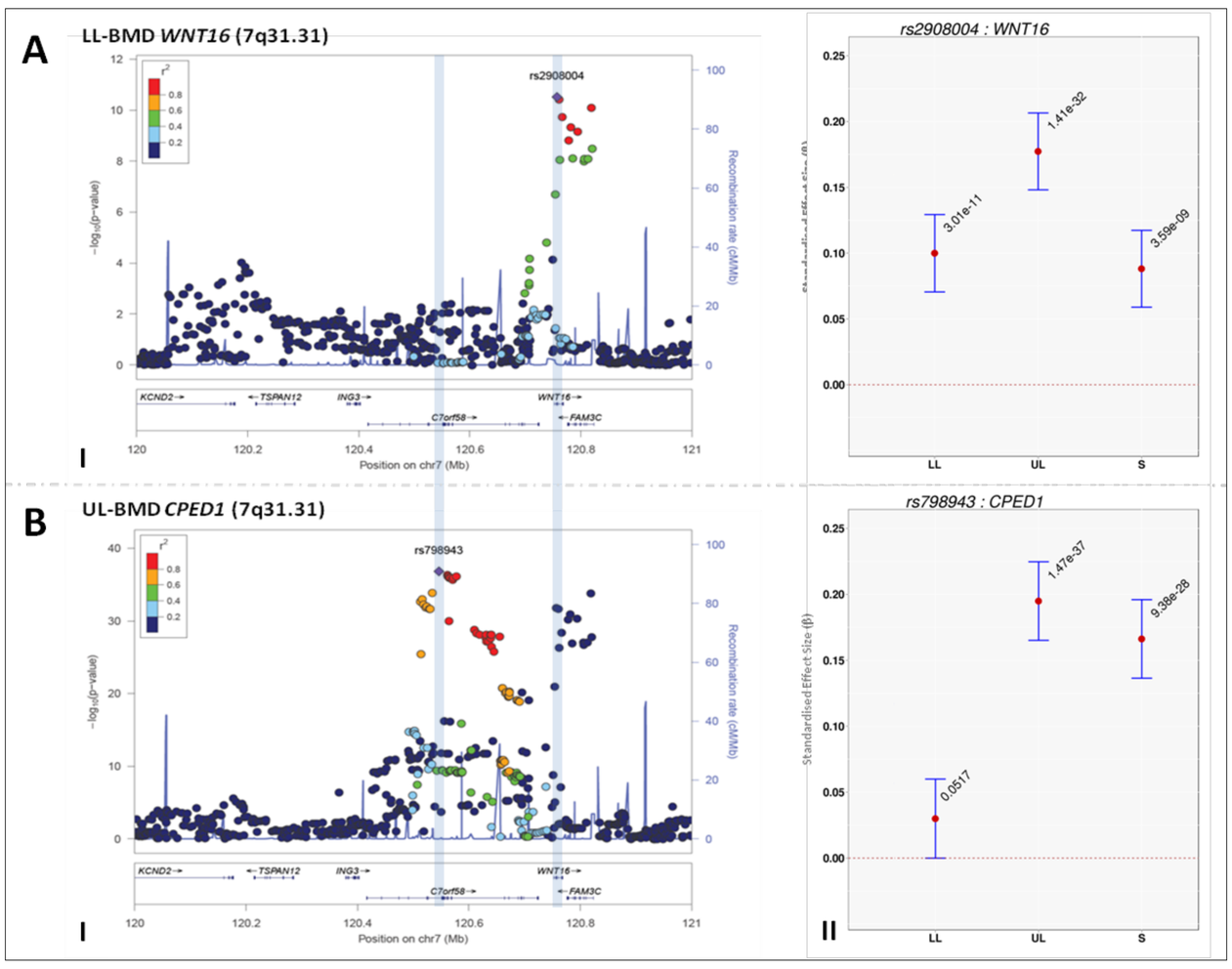

Figure 1: Regional association plots of the top SNPs associated with (A) LL- and (B) UL-BMD at the *WNT16* locus; in addition to (II): a comparison of the effect size of the top site specific SNP described by (I) on BMD at each skeletal sites.

				Lower limb-BMD				Upper limb-BMD				Skull-BMD			
LOCUS	**RSID**	**GENE**	**EA**	**β**	**CI-L**	**CI-U**	***P***	**β**	**CI-L**	**CI-U**	***P***	**β**	**CI-L**	**CI-U**	***P***
1p36.12	rs3765350	*WNT4*	A	0.10	0.06	0.13	**2.89x10^{-8}**	0.09	0.06	0.13	1.82x10^{-7}	0.12	0.08	0.15	**3.56x10^{-11}**
1p36.12	rs2235529	*WNT4*	C	0.11	0.07	0.15	3.27x10^{-7}	0.12	0.08	0.16	**1.21x10^{-8}**	0.14	0.10	0.18	**2.99x10^{-12}**
1p36.12	rs3920498	*WNT4*	G	0.08	0.04	0.11	8.41x10^{-5}	0.10	0.06	0.13	2.85x10^{-7}	0.13	0.10	0.17	**1.56x10^{-12}**
2q24.3	rs6726821	*GALNT3*	T	0.08	0.05	0.11	1.03x10^{-7}	0.08	0.05	0.11	**1.13x10^{-8}**	0.03	0.00	0.06	3.37x10^{-2}
6q22.32	rs1262476	*CENPW*	G	0.08	0.04	0.11	2.07x10^{-5}	0.10	0.07	0.14	**2.93x10^{-9}**	0.04	0.01	0.07	2.31x10^{-2}
6q22.32	rs2130604	*CENPW*	T	0.02	-0.02	0.05	2.82x10^{-1}	0.04	0.01	0.07	2.42x10^{-2}	**0.11**	**0.08**	**0.15**	**3.33x10^{-11}**
6q23.2	rs3012465	*EYA4*	G	0.02	-0.01	0.05	2.09x10^{-1}	0.05	0.02	0.08	7.45x10^{-4}	**0.13**	**0.10**	**0.16**	**8.29x10^{-17}**
7q31.31	rs13223036	*CPED1*	T	0.02	-0.01	0.05	2.02x10^{-1}	**0.19**	**0.16**	**0.22**	**1.25x10^{-34}**	**0.17**	**0.14**	**0.20**	**1.53x10^{-28}**
7q31.31	rs798943	*CPED1*	G	0.03	0.00	0.06	5.17x10^{-2}	**0.20**	**0.17**	**0.23**	**1.47x10^{-37}**	**0.17**	**0.14**	**0.20**	**9.38x10^{-28}**
7q31.31	rs2908004	*WNT16*	A	0.10	0.07	0.13	**3.01x10^{-11}**	**0.18**	**0.15**	**0.21**	**1.41x10^{-32}**	0.09	0.06	0.12	**3.59x10^{-9}**
7q31.31	rs7776725	*FAM3C*	C	0.10	0.07	0.14	**3.14x10^{-9}**	**0.19**	**0.16**	**0.23**	**1.67x10^{-28}**	0.10	0.07	0.13	**1.07x10^{-8}**
8q24.12	rs2450083	*COLEC10*	T	0.02	-0.02	0.05	3.38x10^{-1}	0.04	0.01	0.07	1.42x10^{-2}	**0.10**	**0.07**	**0.13**	**2.13x10^{-11}**
9q34.11	rs7466269	*FUBP3*	A	0.09	0.06	0.12	**1.51x10^{-8}**	0.08	0.05	0.11	3.67x10^{-7}	0.05	0.02	0.08	6.83x10^{-4}
11p14.1	rs10835187	*LIN7C*	C	0.05	0.02	0.07	3.10x10^{-3}	0.04	0.01	0.07	5.53x10^{-3}	**0.13**	**0.10**	**0.16**	**1.63x10^{-17}**
11q13.2	rs12272917	*PPP6R3*	T	0.07	0.03	0.10	1.38x10^{-4}	0.07	0.03	0.10	7.78x10^{-5}	0.11	0.08	0.14	**1.34x10^{-10}**
12p11.22	rs4420311	*KLHDC5*	G	0.09	0.06	0.12	**3.21x10^{-8}**	0.07	0.04	0.10	2.25x10^{-5}	0.04	0.01	0.07	1.58x10^{-2}
13q14.11	rs9525638	*TNFSF11*	C	0.06	0.04	0.09	2.09x10^{-5}	0.09	0.06	0.12	**2.47x10^{-9}**	0.06	0.03	0.09	1.43x10^{-4}
13q14.11	rs17536328	*TNFSF11*	T	0.07	0.04	0.10	1.19x10^{-5}	0.09	0.06	0.12	**3.08x10^{-9}**	0.06	0.03	0.09	2.03x10^{-4}
14q32.12	rs754388	*RIN3*	C	0.13	0.09	0.17	**1.40x10^{-10}**	0.10	0.06	0.14	3.13x10^{-7}	0.04	0.00	0.08	7.82x10^{-2}
18q21.33	rs884205	*TNFRSF11A*	C	0.00	-0.03	0.04	8.58x10^{-1}	0.02	-0.01	0.06	2.11x10^{-1}	**0.10**	**0.07**	**0.14**	**1.84x10^{-8}**

Table 2: Comparison of effect sizes and the strength of association of all variants which exceeded genome-wide significance at one or more skeletal sites. (GENE) = closest gene; (EA) = effect allele; (β) = estimates of effect size expressed as adjusted SD per copy of the effect allele (EA); (CI-L) = lower limit of the 95% confidence interval for β; (CI-U) = upper limit of the 95% confidence interval for β; (P) = P-value. Site specific effect sizes depicted in blue .

CONCLUSION:

Different skeletal sites as measured by whole body-DXA are to a certain extent under distinct environmental and genetic influences. Allowing for these differences may help to uncover new genetic influences on BMD, particularly those involved in bone mass attainment, for which S-BMD appears to be particularly well suited.

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