Genome-wide Association Study of Blood Pressure and Hypertension in Six Population-based Cohort Studies

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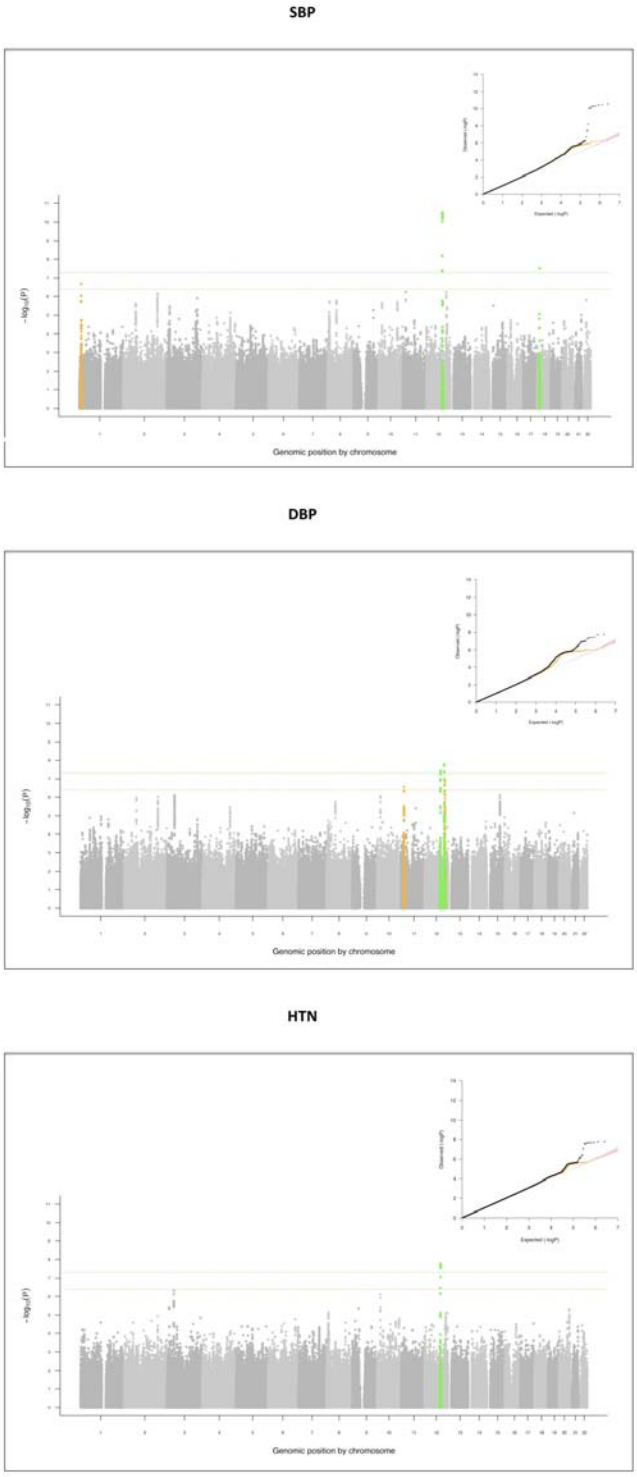
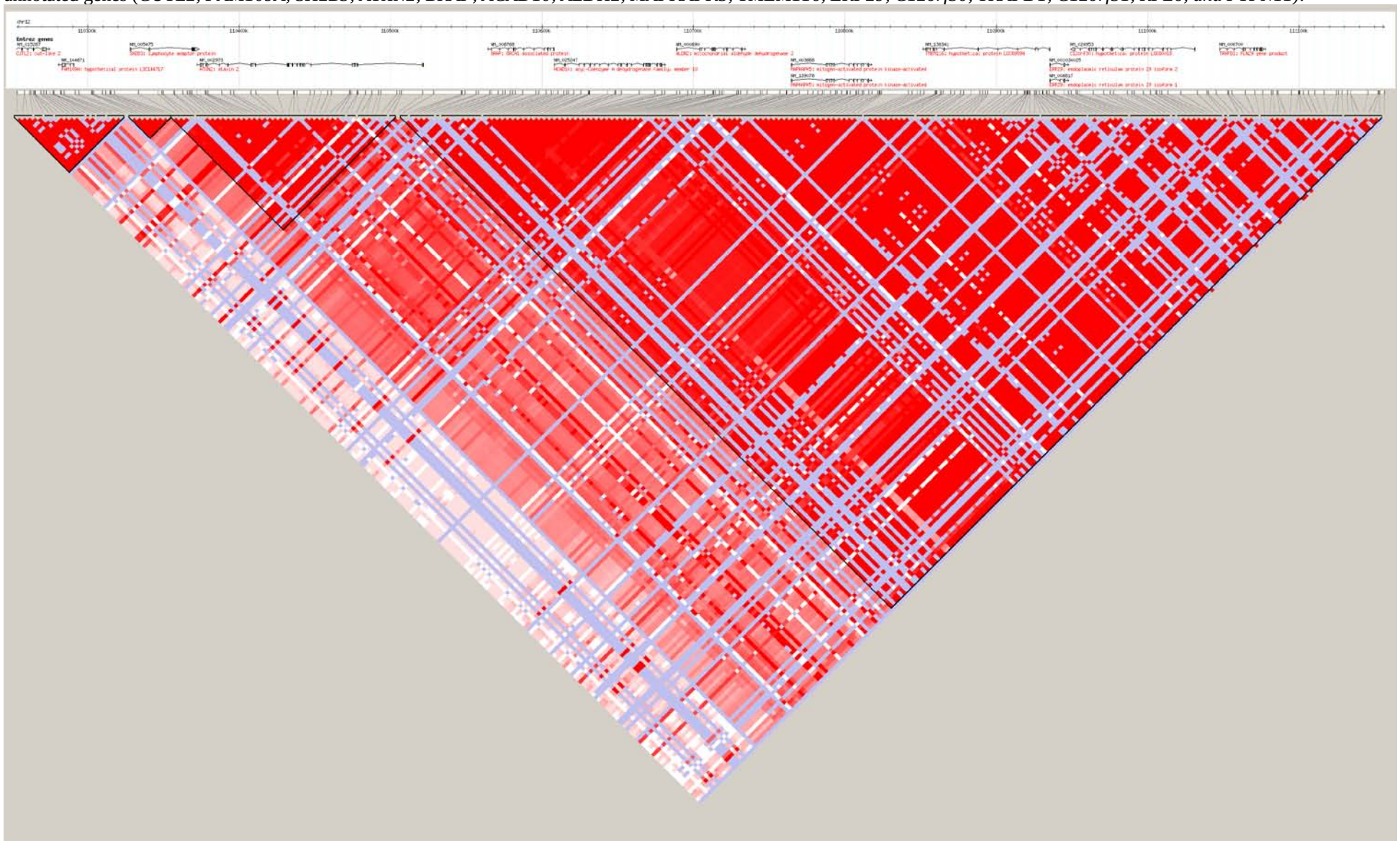
Supplementary Figure 1. QQ and $-\log_{10}$P plots of genome-wide association

The statistical significance value across the 22 autosomes of each SNP association with systolic blood pressure (top panel), diastolic blood pressure (middle panel), and hypertension (top panel) are plotted as $-\log_{10}$(p-value) by meta-analysis across the six studies using inverse variance weighting. We filtered out SNPs for minor allele frequency <0.01 and adjusted values by genomic controls within study and across the meta-analysis (λ = 1.057 for systolic blood pressure, 1.049 for diastolic blood pressure, and 1.037 for hypertension prior to adjustment). Two thresholds are displayed by horizontal lines: $4x10^{-7}$ (orange) and $5x10^{-8}$ (green). SNPs within loci with p values $<5x10^{-8}$ are displayed in green; those with p values between $4x10^{-7}$ and $5x10^{-8}$ are displayed in orange. Embedded within each panel are QQ plots of observed vs. expected $-\log_{10}$P values. The black QQ lines represent the observed vs. expected associations for all SNPs; the orange QQ lines represent the observed vs. expected associations after omitting all loci with $p<4x10^{-7}$.



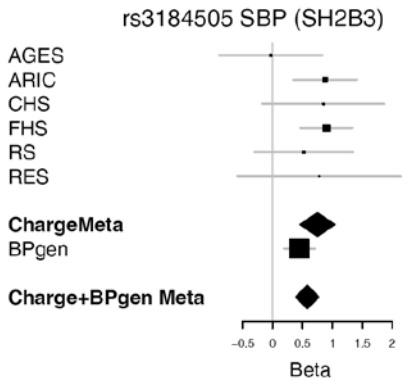
Supplementary Figure 2: Linkage dysequilibrium across chromosome 12q24

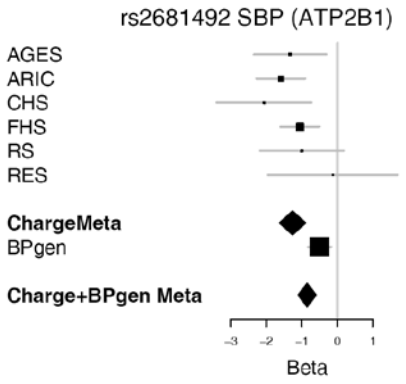
Linkage disequilibrium for a region on chromosome 12 showing genome-wide significant association with systolic and diastolic blood pressure. This block encompasses at least 15 annotated genes (*CUTL2, FAM109A, SH2B3, ATXN2, BRAP, ACAD10, ALDH2, MAPKAPK5, TMEM116, ERP29, C12orf30, TRAFD1, C12orf51, RPL6, and PTPN11).*

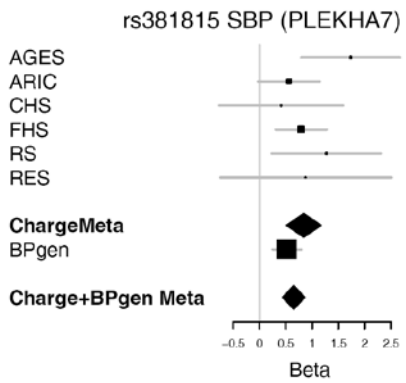


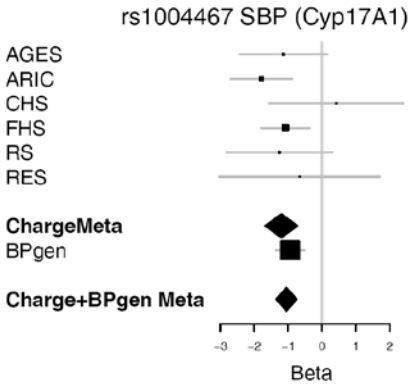
Supplementary Figure 3: Forest plots for systolic blood pressure loci

Forest plots for 4 SNPs with genome-wide significant association in the combined systolic blood pressure meta-analysis of CHARGE and Global BPgen. The four SNPs are rs3184504 (*SH2B3), rs2681492 (ATP2B1), rs381815 (PLEKHA7), and rs1004467 (CYP17A1).* Regression coefficients (beta in mm Hg per variant allele) and confidence intervals are provided for the 6 studies within CHARGE, meta-analysis across CHARGE, Global BPgen meta-analysis, and meta-analysis of CHARGE and Global BPgen.



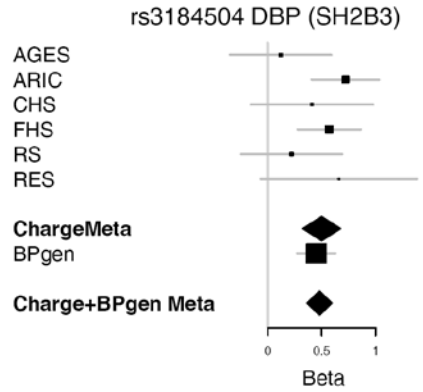


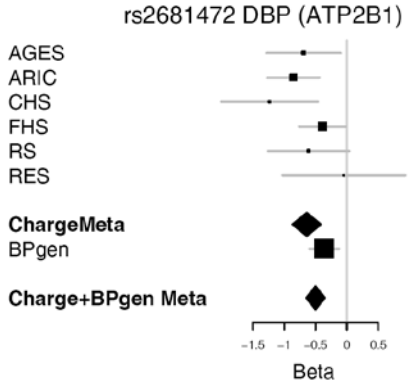


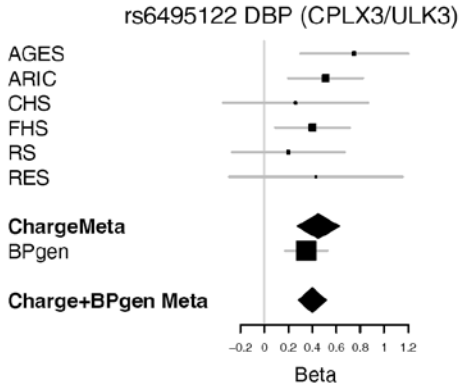


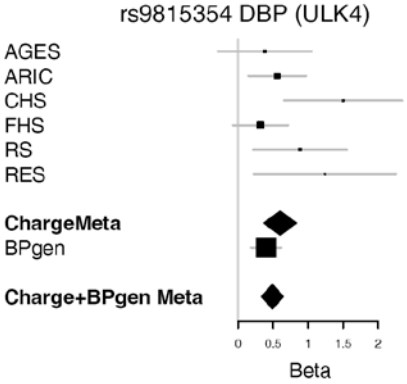
Supplementary Figure 4: Forest plots for diastolic blood pressure loci

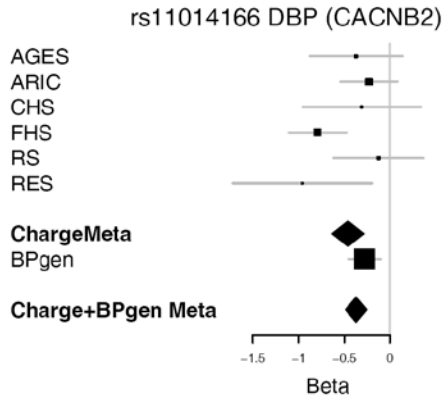
Forest plots for 6 SNPs with genome-wide significant association in the combined diastolic blood pressure meta-analysis of CHARGE and Global BPgen. The six SNPs are rs3184504 (*SH2B3*), rs2681472 (*ATP2B1*), rs6495122 (*CSK*), rs9815354 (*ULK4*), rs11014166 (*CACNB2*), and rs2384550 (*TBX3/TBX5*). Regression coefficients (beta in mm Hg per variant allele) and confidence intervals are provided for the 6 studies within CHARGE, meta-analysis across CHARGE studies, Global BPgen meta-analysis, and meta-analysis of CHARGE and Global BPgen.

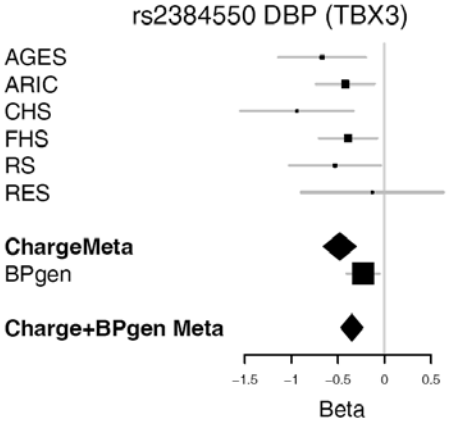












Supplementary Table 1:
Clinical Characteristics of Study Participants

	AGES	ARIC	CHS	FHS	RS	RES
Number of individuals in GWAS	3219	8047	3277	8096	4737	1760
Percent women	58	53	61	54	60	56
Mean age (yrs)	51 (6)	54 (6)	72 (5)	38 (9)	68 (8)	64 (7)
Mean body mass index (kg/m^2)	25.2 (3.5)	27.0 (4.9)	26.3 (4.4)	25.9 (4/9)	26.2 (3.6)	27.2 (4.2)
Mean observed systolic blood pressure (mm Hg)	132 (17)	118 (17)	135 (21)	119 (15)	139 (22)	143 (21)
Mean observed diastolic blood pressure (mm Hg)	83 (10)	72 (10)	70 (11)	77 (10)	74 (11)	79 (11)
Mean systolic blood pressure with addition of 10 mm Hg for treatment effect	133 (18)	120 (18)	139 (22)	120 (15)	141 (24)	145 (22)
Mean diastolic blood pressure with addition of 5 mm Hg for treatment effect	84 (10)	73 (10)	74 (13)	77 (10)	75 (13)	80(11)
Percent treated for hypertension	7	20	35	5	18	22
Percent hypertensive	35	27	53	17	53	60
Years of baseline examinations	1968-1991	1987-1989	1989-90	1948-1951* 1971-1974^ 2002-2005#	1990-1993	2000-2001
Years of DNA collection	2002-2006	1987-1998	1989-1990	1996-1999* 1996-1999^ 2002-2005#	1990-1993	2000-2001

Mean values (standard deviation); *Original cohort; ^Offspring cohort; #Third Generation cohort

Supplementary Table 2. Individual Study Results for Blood Pressure and Hypertension Associations with Meta-analysis $p<1x10^{-6}$

SNP	Chr	AGES			ARIC			CHS			FHS			RS			RES		
Systolic Blood Pressure																			
		Beta	SE	p	Beta	SE	p	Beta	SE	p	Beta	SE	p	Beta	SE	p	Beta	SE	p
rs2681492	12	-1.33	0.52	0.01	-1.59	0.35	5.8E-06	-2.06	0.68	2.4E-03	-1.05	0.28	1.4E-04	-1.00	0.60	0.10	-0.13	0.93	0.89
rs2681472	12	-1.47	0.54	6.0E-03	-1.81	0.36	5.2E-07	-1.97	0.68	3.6E-03	-0.96	0.28	7.7E-04	-1.07	0.62	0.09	0.16	0.95	0.87
rs11105354	12	-1.48	0.54	5.7E-03	-1.80	0.36	5.4E-07	-2.22	0.75	3.1E-03	-0.95	0.28	7.7E-04	-1.07	0.62	0.09	0.12	0.95	0.90
rs11105364	12	-1.50	0.54	5.2E-03	-1.81	0.36	4.5E-07	-2.68	0.95	4.9E-03	-0.94	0.28	8.6E-04	-1.16	0.63	0.07	0.08	0.96	0.94
rs17249754	12	-1.50	0.54	5.3E-03	-1.81	0.36	4.0E-07	-2.67	0.95	5.0E-03	-0.95	0.28	8.0E-04	-1.09	0.62	0.08	0.08	0.96	0.93
rs11105368	12	-1.50	0.54	5.2E-03	-1.80	0.36	5.0E-07	-2.70	0.96	4.9E-03	-0.94	0.28	9.0E-04	-1.18	0.63	0.06	0.07	0.96	0.94
rs12579302	12	-1.50	0.54	5.4E-03	-1.80	0.36	5.1E-07	-2.65	0.94	4.9E-03	-0.95	0.28	7.9E-04	-1.08	0.62	0.08	0.09	0.95	0.93
rs12230074	12	-1.51	0.54	4.9E-03	-1.84	0.37	5.0E-07	-3.58	1.30	5.9E-03	-0.95	0.29	1.1E-03	-1.23	0.63	0.05	0.07	0.96	0.94
rs11105378	12	-1.51	0.54	5.0E-03	-1.83	0.37	5.0E-07	-3.50	1.27	5.9E-03	-0.94	0.29	1.0E-03	-1.23	0.63	0.05	0.07	0.96	0.94
rs4842666	12	-1.52	0.56	6.7E-03	-1.69	0.37	4.2E-06	-2.13	1.01	0.03	-0.85	0.30	3.8E-03	-0.99	0.67	0.14	0.20	1.02	0.85
rs8096897	18	-14.61	7.84	0.06	NA	NA	NA	NA	NA	NA	-13.83	3.33	3.4E-05	-15.69	4.82	1.0E-03	2.77	7.39	0.71
rs11105328	12	-1.33	0.56	0.02	-1.61	0.37	9.9E-06	-2.22	1.03	0.03	-0.79	0.29	6.0E-03	-1.03	0.66	0.12	0.42	1.01	0.68
rs880315	1	0.56	0.43	0.19	1.44	0.33	1.3E-05	1.60	0.59	6.2E-03	0.66	0.27	0.01	0.02	0.48	0.98	1.72	0.75	0.02
rs3184504	12	-0.03	0.44	0.95	0.88	0.27	1.2E-03	0.85	0.52	0.10	0.90	0.22	5.1E-05	0.52	0.45	0.25	0.78	0.70	0.27
rs381815	11	1.73	0.48	3.5E-04	0.56	0.30	6.2E-02	0.41	0.60	0.50	0.79	0.25	1.8E-03	1.27	0.53	0.02	0.88	0.83	0.29
rs7926335	11	1.73	0.48	3.6E-04	0.55	0.30	6.7E-02	0.46	0.62	0.46	0.79	0.25	1.8E-03	1.28	0.53	0.02	0.87	0.83	0.29
rs7571613	2	0.32	0.56	0.56	1.13	0.34	1.0E-03	0.57	0.67	0.40	1.11	0.29	1.4E-04	0.96	0.59	0.10	0.74	0.92	0.42
rs11895934	2	0.32	0.56	0.56	1.13	0.34	1.0E-03	0.57	0.68	0.40	1.11	0.29	1.4E-04	0.97	0.59	0.10	0.74	0.92	0.42
rs7564968	2	0.32	0.56	0.57	1.12	0.34	1.1E-03	0.55	0.68	0.42	1.11	0.29	1.3E-04	0.96	0.59	0.11	0.75	0.92	0.42
rs653178	12	-0.04	0.43	0.93	0.87	0.27	1.2E-03	0.81	0.52	0.12	0.89	0.22	6.0E-05	0.48	0.45	0.29	0.75	0.71	0.29
rs284277	1	0.38	0.47	0.41	1.10	0.28	6.9E-05	1.77	0.65	6.4E-03	0.60	0.23	0.01	0.01	0.52	0.99	1.69	0.82	0.04
Diastolic Blood Pressure																			
		Beta	SE	p	Beta	SE	p	Beta	SE	p	Beta	SE	p	Beta	SE	p	Beta	SE	p
rs3184504	12	0.12	0.24	0.63	0.72	0.16	7.4E-06	0.41	0.29	0.16	0.57	0.15	1.4E-04	0.22	0.24	0.34	0.66	0.37	0.07
rs653178	12	0.12	0.24	0.63	0.70	0.16	9.5E-06	0.40	0.29	0.17	0.58	0.15	1.3E-04	0.21	0.24	0.37	0.67	0.37	0.07
rs2681472	12	-0.69	0.30	0.02	-0.85	0.21	5.3E-05	-1.23	0.39	1.4E-03	-0.39	0.19	0.04	-0.61	0.33	0.06	-0.05	0.50	0.92
rs4766578	12	0.12	0.24	0.62	0.70	0.16	1.1E-05	0.41	0.30	0.17	0.57	0.15	1.3E-04	0.15	0.24	0.53	0.62	0.37	0.09
s10774625	12	0.12	0.24	0.62	0.70	0.16	1.1E-05	0.41	0.30	0.17	0.58	0.15	1.4E-04	0.15	0.24	0.53	0.62	0.37	0.09
rs2681492	12	-0.55	0.29	0.06	-0.75	0.21	2.7E-04	-1.25	0.39	1.3E-03	-0.46	0.19	0.01	-0.57	0.32	0.07	-0.24	0.48	0.61
rs11105354	12	-0.70	0.30	0.02	-0.85	0.21	5.8E-05	-1.31	0.43	2.3E-03	-0.38	0.19	0.05	-0.62	0.33	0.06	-0.07	0.50	0.89
rs17630235	12	0.15	0.26	0.56	0.73	0.16	4.2E-06	0.60	0.56	0.28	0.52	0.15	6.5E-04	0.19	0.24	0.43	0.49	0.37	0.19
rs17249754	12	-0.71	0.30	0.02	-0.85	0.21	4.8E-05	-1.55	0.55	4.5E-03	-0.38	0.19	0.05	-0.62	0.33	0.06	-0.06	0.50	0.90
rs11066188	12	0.16	0.26	0.54	0.74	0.16	4.1E-06	0.60	0.56	0.28	0.52	0.15	7.3E-04	0.20	0.24	0.42	0.48	0.37	0.20
rs11105364	12	-0.71	0.30	0.02	-0.85	0.21	4.7E-05	-1.53	0.55	5.3E-03	-0.37	0.19	0.06	-0.66	0.33	0.05	-0.05	0.50	0.92
rs11105368	12	-0.71	0.30	0.02	-0.86	0.21	4.6E-05	-1.53	0.55	5.7E-03	-0.36	0.19	0.06	-0.67	0.33	0.04	-0.03	0.50	0.95
rs12579302	12	-0.71	0.30	0.02	-0.84	0.21	5.8E-05	-1.53	0.54	4.7E-03	-0.38	0.19	0.05	-0.62	0.33	0.06	-0.07	0.50	0.88
rs2384550	12	-0.67	0.24	0.01	-0.42	0.16	9.5E-03	-0.94	0.31	2.6E-03	-0.39	0.16	0.01	-0.53	0.25	0.03	-0.13	0.39	0.73
rs1991391	12	-0.67	0.24	0.01	-0.42	0.16	9.9E-03	-0.95	0.31	2.5E-03	-0.39	0.16	0.01	-0.53	0.25	0.03	-0.13	0.39	0.73
rs6489992	12	-0.60	0.25	0.02	-0.39	0.16	0.01	-0.99	0.33	2.8E-03	-0.46	0.16	3.5E-03	-0.50	0.25	0.04	-0.09	0.38	0.82
rs11065987	12	0.04	0.25	0.86	0.70	0.16	1.6E-05	0.60	0.44	0.18	0.56	0.16	3.6E-04	0.19	0.24	0.43	0.53	0.37	0.15
rs11024074	11	0.71	0.26	0.01	0.34	0.17	0.04	0.61	0.34	0.07	0.56	0.17	8.0E-04	0.72	0.26	0.01	-0.15	0.42	0.73

rs11105378	12	-0.72	0.30	0.02	-0.87	0.21	5.0E-05	-1.78	0.73	0.01	-0.35	0.19	0.07	-0.69	0.33	0.04	-0.00	0.50	1.00
rs12230074	12	-0.72	0.30	0.02	-0.87	0.21	5.0E-05	-1.82	0.75	0.01	-0.35	0.20	0.07	-0.69	0.33	0.04	-0.00	0.50	1.00
rs7963771	12	-0.59	0.27	0.03	-0.50	0.17	2.8E-03	-0.77	0.37	0.04	-0.48	0.21	0.02	-0.55	0.27	0.04	-0.39	0.42	0.35
rs381815	11	0.71	0.27	0.01	0.34	0.18	0.05	0.52	0.35	0.13	0.61	0.17	4.0E-04	0.72	0.28	0.01	-0.11	0.43	0.80
rs4842666	12	-0.75	0.31	0.02	-0.82	0.22	1.5E-04	-1.72	0.57	2.5E-03	-0.35	0.20	0.08	-0.47	0.35	0.18	-0.33	0.54	0.54
rs7926335	11	0.71	0.27	0.01	0.33	0.18	0.06	0.54	0.36	0.13	0.61	0.17	4.0E-04	0.71	0.28	0.01	-0.11	0.43	0.80
rs11105328	12	-0.73	0.31	0.02	-0.78	0.21	2.6E-04	-1.83	0.58	1.8E-03	-0.32	0.19	0.10	-0.56	0.34	0.10	-0.43	0.53	0.42
rs17696736	12	0.12	0.26	0.63	0.70	0.16	8.9E-06	0.61	0.57	0.28	0.50	0.15	8.8E-04	0.08	0.24	0.73	0.49	0.37	0.18
rs10744835	12	-0.71	0.28	0.01	-0.45	0.17	9.1E-03	-1.36	0.43	1.4E-03	-0.39	0.16	0.02	-0.52	0.26	0.05	-0.04	0.40	0.92
rs7977406	12	-0.71	0.28	0.01	-0.44	0.17	9.4E-03	-1.37	0.44	1.8E-03	-0.39	0.16	0.02	-0.52	0.26	0.05	-0.07	0.40	0.86
rs9815354	3	0.38	0.34	0.27	0.56	0.21	8.7E-03	1.50	0.43	4.3E-04	0.32	0.20	0.11	0.88	0.34	0.01	1.24	0.52	0.02
rs6495122	15	0.75	0.23	1.3E-03	0.51	0.16	1.4E-03	0.26	0.31	0.41	0.40	0.16	0.010	0.20	0.24	0.41	0.43	0.37	0.25
rs11014166	10	-0.37	0.26	0.15	-0.23	0.16	0.16	-0.31	0.33	0.36	-0.79	0.16	5.1E-07	-0.13	0.25	0.61	-0.96	0.39	0.01
rs6768438	3	0.39	0.34	0.25	0.56	0.21	8.4E-03	1.60	0.46	5.6E-04	0.37	0.20	0.06	0.75	0.33	0.02	1.08	0.50	0.03
rs9852991	3	0.39	0.34	0.25	0.56	0.21	8.5E-03	1.59	0.46	5.5E-04	0.37	0.20	0.06	0.75	0.33	0.02	1.08	0.50	0.03
rs13401889	2	0.59	0.30	0.05	0.50	0.19	8.7E-03	0.30	0.42	0.46	0.56	0.19	2.9E-03	0.75	0.30	0.01	0.41	0.46	0.38
rs9816772	3	0.39	0.34	0.25	0.56	0.21	8.5E-03	1.60	0.46	5.6E-04	0.37	0.20	0.06	0.75	0.33	0.02	1.08	0.50	0.03
Hypertension																			
		Beta	SE	p	Beta	SE	p	Beta	SE	p	Beta	SE	p	Beta	SE	p	Beta	SE	p
rs2681472	12	-0.21	0.07	3.7E-03	-0.17	0.05	1.4E-03	-0.17	0.07	0.01	-0.13	0.07	0.05	-0.17	0.06	4.9E-03	-0.02	0.10	0.82
rs11105354	12	-0.21	0.07	3.5E-03	-0.16	0.05	1.4E-03	-0.19	0.08	0.01	-0.13	0.07	0.05	-0.17	0.06	4.4E-03	-0.02	0.10	0.82
rs11105364	12	-0.21	0.07	3.4E-03	-0.16	0.05	1.8E-03	-0.24	0.10	0.01	-0.13	0.07	0.05	-0.18	0.06	3.2E-03	-0.03	0.10	0.80
rs17249754	12	-0.21	0.07	3.4E-03	-0.16	0.05	1.5E-03	-0.24	0.10	0.01	-0.13	0.07	0.05	-0.17	0.06	3.7E-03	-0.02	0.10	0.80
rs11105368	12	-0.21	0.07	3.4E-03	-0.16	0.05	2.0E-03	-0.25	0.10	0.01	-0.13	0.07	0.05	-0.18	0.06	3.0E-03	-0.03	0.10	0.79
rs12579302	12	-0.21	0.07	3.4E-03	-0.16	0.05	1.5E-03	-0.24	0.09	0.01	-0.13	0.07	0.05	-0.17	0.06	3.9E-03	-0.02	0.10	0.81
rs11105378	12	-0.21	0.07	3.3E-03	-0.16	0.05	2.2E-03	-0.33	0.13	9.0E-03	-0.13	0.07	0.05	-0.18	0.06	2.7E-03	-0.03	0.10	0.78
rs12230074	12	-0.21	0.07	3.3E-03	-0.16	0.05	2.2E-03	-0.34	0.13	9.0E-03	-0.13	0.07	0.05	-0.18	0.06	2.7E-03	-0.03	0.10	0.78
rs2681492	12	-0.17	0.07	0.02	-0.13	0.05	7.5E-03	-0.17	0.07	0.01	-0.16	0.06	0.01	-0.17	0.06	3.9E-03	-0.00	0.10	0.99
rs4842666	12	-0.21	0.08	5.7E-03	-0.17	0.05	1.0E-03	-0.20	0.10	0.04	-0.11	0.07	0.10	-0.16	0.06	0.01	0.02	0.11	0.88
rs7640747	3	0.04	0.06	0.50	0.14	0.04	6.3E-04	0.08	0.07	0.30	0.17	0.05	1.3E-03	0.06	0.05	0.18	0.22	0.08	6.0E-03
rs11105328	12	-0.19	0.08	0.01	-0.16	0.05	1.9E-03	-0.21	0.10	0.04	-0.09	0.07	0.17	-0.18	0.06	3.5E-03	0.01	0.10	0.90
rs743395	3	0.04	0.06	0.50	0.14	0.04	1.1E-03	0.08	0.07	0.30	0.17	0.05	1.3E-03	0.06	0.05	0.18	0.22	0.08	5.0E-03
rs11014166	10	-0.08	0.06	0.20	-0.07	0.04	0.06	-0.14	0.06	0.01	-0.21	0.06	1.9E-04	-0.07	0.05	0.12	-0.14	0.08	0.07

Supplementary Table 3
Technical Details of Genotyping, SNP Exclusion, and SNP Imputation Methods

Study	AGES	ARIC	CHS	FHS	RS	RES
Array type	Illumina 370CNV	Affymetrix 6.0	Illumina 370CNV	Affymetrix 500K and MIPS 50K combined	Illumina 550K	Illumina 550K
Genotype calling algorithm	Illumina BeadStudio	Birdseed	BeadStudio	BRLMM	Beadstudio	Beadstudio
Exclusion of SNPs used for imputation	call rate < 97%, HWE p<1e-6, MAF <1%,	Call rate <95% MAF < 1% pHWE < 10^{-5}	Call rate <= 95% HWE p< 10^{-5} >2 replicate errors or Mendelian inconsistencies; heterozygote frequency =0	Call rate < 97% HWE p <10^{-6} Mishap p < 10^{-9} Mendelian errors > 100	Call rate <98% HWE p <10^{-6} MAF < 0.01	Call rate <98% HWE p<= 10^{-6} MAF <= 0.01
Imputation method	MACH (version 1.0.16	MACH 1.0.16	BIMBAM10 v0.99	MACH 1.0.15	MACH 1.0.15	Mach 1.0.15
Imputation Backbone (NCBI build)	Hapmap CEU release 22 build 36	HapMap CEU release 21, build 35	HapMap CEU release 22, build 36	HapMap CEU release 22, build 36	Hapmap CEU release 22, build 36	Hapmap CEU release 22, build 36
Filtering of imputed genotypes	None	None	Variance of allele dosage < 0.005	None	None	None
Data handling and statistical tests	PLINK and R	PLINK, MACH2QTL	R	LMEKIN package in R – linear mixed effects model incorporating familial covariance based on degree of relatedness	ProbABEL	ProbABEL

Supplementary Methods

Study Samples and Blood Pressure Measurement

AGES-Reykjavik Study. The Reykjavik Study cohort originally comprised a random sample of 30,795 men and women born in 1907-1935 and living in Reykjavik in 1967. A total of 19,381 people attended, resulting in 71% recruitment rate. The study sample was divided into six groups by birth year and birth date within month. One group was designated for longitudinal follow up and was examined in all stages. One group was designated a control group and was not included in examinations until 1991. Other groups were invited to participate in specific stages of the study. Between 2002 and 2006, the AGES-Reykjavik study re-examined 5764 survivors of the original cohort who had participated before in the Reykjavik Study.[1] The midlife data blood pressure measurement was taken from stage 3 of the Reykjavik Study (1974-1979), if available. Half of the cohort attended during this period. Otherwise an observation was selected closest in time to the stage 3 visit. Participants came in a fasting state to the clinic. The supine blood pressure was measured twice by a nurse using a mercury sphygmomanometer after a 5-min rest. Blood pressure was measured according to World Health Organization recommendations.[2] Individuals with previous MI were excluded from the analyses (n=12). Successful genotyping was available for 3219 AGES participants who were eligible for this study. The AGES Reykjavik Study GWAS was approved by the National Bioethics Committee and the Data Protection Authority.

The ARIC study is a population-based prospective cohort study of cardiovascular disease sponsored by National Heart, Lung, and Blood Institute (NHLBI). ARIC included 15,792 individuals aged 45-64 years at baseline (1987-89), chosen by probability sampling from four US communities.[3] Cohort members completed four clinic examinations, conducted three years apart between 1987 and 1998. The data used in this study are from the first visit in 1987. A detailed study protocol is available on the ARIC study website (http://www.cscc.unc.edu/aric). Clinic examinations included assessment of cardiovascular disease risk factors, a detailed medical and psychosocial history, and measurement of various clinical and laboratory variables. The physical examination included measurements of weight and height from which the body mass index (BMI) was calculated. Blood pressure was measured using a standardized Hawskley random-zero mercury column sphygmomanometer with

participants in sitting position after a resting period of 5 minutes. The size of the cuff was chosen according to the arm circumference. Three sequential recordings for systolic and diastolic blood pressure were obtained and the mean of the last two measurements used in this analysis. Blood pressure lowering medication use was recorded from the medication history. Outliers (>4SD from the mean) with respect to the systolic or diastolic blood pressure distribution were excluded from the analysis. For this investigation we limited the sample to individuals of European descent by self report and in whom GWAS was carried out.

The CHS is a population-based cohort study of risk factors for cardiovascular disease in adults 65 years of age or older conducted across four field centers. The original predominantly white cohort of 5201 persons was recruited in 1989-1990 from random samples of the Medicare eligibility lists and an additional 687 African-Americans were enrolled in 1992-93 for a total sample of 5888. Details of the study design are summarized elsewhere.[4] A total of 1908 persons were excluded from the study sample due to prevalent coronary heart disease (n=1195), congestive heart failure (n=86), peripheral vascular disease (n=93), valvular heart disease (n=20), stroke (n=166) or transient ischemic attack (n=56). Participants with missing BMI (n=10) or BP measurements (n=8) were excluded. CHS participants completed standardized clinical examinations and questionnaires at study baseline and at nine annual follow-up visits. Research staff who received central training in blood pressure measurement assessed repeat right-arm seated systolic and diastolic blood pressure levels at baseline with a Hawksley random-zero sphygmomanometer. Means of the repeated blood pressure measurements from the baseline examination were used for GWAS analyses. Because the other cohorts were predominantly white, African American participants were excluded from this analysis. 3,277 CHS subjects contributed to this analysis.

The FHS began in 1948 with the recruitment of an original cohort of 5,209 men and women who were 28 to 62 years of age (mean age 44 years; 55 percent women) at entry. In 1971 enrollment of a second generation of study participants took place; this cohort consisted of 5,124 children and spouses of children of the original cohort. The mean age of the offspring cohort was 37 years; 52 percent were women. A third generation cohort

of 4,095 children (mean age 40 years; 53 percent women) of offspring cohort participants was enrolled beginning in 2002. Details of study designs for the three cohorts are summarized elsewhere.[5,6,7] At each clinic visit, a medical history was obtained with a focus on cardiovascular content, and participants underwent a physical examination including measurement of height and weight from which BMI was calculated. Systolic and diastolic blood pressures were measured twice by a physician in the left arm of the resting seated participant using a mercury column sphygmomanometer. Pressures were recorded to the nearest even number. The means of two separate systolic and diastolic blood pressure readings at the first clinic examination of each of the three FHS cohorts were used for this GWAS analyses. For a subset of offspring cohort participants only one measurement was obtained. For the purposes of a blood pressure GWAS we excluded people who were under 20 years of age and those who had a myocardial infarction or congestive heart failure because those conditions may affect blood pressure levels.

The RS is a prospective population-based cohort study comprising 7,983 subjects aged 55 years or older. Participants completed an interview at home and at the research center, where participants were subsequently examined. Baseline data were collected between 1990 and 1993. In 1999, inhabitants who turned 55 years of age or moved into the study district since the start of the study were invited to participate in an extension of the RS (RES) of whom 3011 participated (67% response rate). The rationale and design of the RS have been described in detail elsewhere.[8] At the research center, we obtained two seated blood pressure measurements in the right brachial artery with a random zero sphygmomanometer. The mean of two consecutive measurements was used in association analyses. We excluded participants who were older than 85 years of age and those who had a history myocardial infarction or congestive heart failure, because of the impact of these conditions on blood pressure levels.

Genotyping

For AGES, DNA was genotyped in 3,219 participants who were eligible for this study using the Illumina 370CNV BeadChip array. Samples were excluded from the dataset based on sample failure, genotype

mismatch with reference panel, and sex mismatch. Standard protocols for working with Illumina data were followed. Prior to genotype imputation, SNPs were excluded using filters based on call rate (<97%), Hardy-Weinberg Equilibrium (<1e-6), mishap (<1e-9), and mismatched positions between Illumina, dbSNP and/or HapMap resulting in 325,094 SNPs passing all QC (of 353,202 prior to cleaning steps). Imputation was done using MACH against all the HapMap CEPH haplotypes (release 22/NCBI build 36) resulting in 2,533,153 total SNPs for analysis (Supplementary Table 3).

Genomic DNA was isolated from blood samples obtained on ARIC participants at their initial examination. Affymetrix 6.0 array genotypes were obtained in 8,861 self-identified whites of European descent: individuals were excluded for the following reasons: 1) discordance with previous genotype data, 2) genotypic sex mismatch, 3) first-degree relative of an included individual based on genotype data, 4) genetic outlier (as assessed by average identity by state (IBS) using PLINK[9] and >8 SD along any of 10 principal components in EIGENSTRAT[10] with 5 iterations. After these exclusions, 8,127 individual were eligible and 8,047 with successful genotyping and full covariate data were included in this GWAS; genotypes were available on 704,588 high quality SNPs before imputation (Supplementary Table 3).

DNA was extracted from blood samples drawn on all CHS participants at the baseline examination. In 2007-2008, genotyping was performed at the General Clinical Research Center's Genotyping Laboratory at Cedars-Sinai Medical Center using the Illumina HumanCNV370-Duo BeadChip system. The present report is based upon genotyping results from 3,397 CHS Caucasian participants, who were free of clinical cardiovascular disease at baseline, consented to genetic testing, and had DNA available for genotyping. Genotypes were called using the Illumina BeadStudio software. Genotyping was successful in 3,295 persons. Genotypes were called using the Illumina BeadStudio software. The following exclusions were applied to identify a final set of 306,655 autosomal SNPs used for imputation: call rate < 97%; HWE $P < 10^{-5}$; > 1 duplicate error or Mendelian inconsistency; heterozygote frequency = 0; SNPs not found in dbSNP. (Supplementary Table 3).

As part of the National Heart, Lung, and Blood Institute's SNP Health Association Resource (SHARe) project,[11] genotyping was conducted on 9,274 FHS participants using the Affymetrix 500K mapping array and the Affymetrix 50K gene-focused MIP array. We excluded individuals from association analyses when their call rate across all SNPs was less than 97 percent. We also restricted analyses to DNA samples derived from immortalized cell lines. Of those in whom genotyping was attempted, 92 percent (8508) had a within individual call rate of 97 percent or greater. After exclusions (Supplementary Table 3), there were 503,551 SNPs available for analysis. The sample size fulfilling eligibility criteria and with full covariate information and successful genotyping was 8,096.

The RS and RES used the Illumina 550K array to conduct the genotyping. The exclusion criteria for SNPs were minor allele frequency ≤1%, Hardy-Weinberg equilibrium (HWE) $p<10^{-6}$, or SNP call rate ≤90% and resulted in data on 530,683 SNPs in RS and 495,478 SNPs in RES. Imputation was done with reference to HapMap release 22 CEU using the maximum likelihood method implemented in MACH. After excluding 183 subjects with incomplete blood pressure data, 361 who were older than 85 years, 148 with prevalent heart failure, and 540 with prevalent myocardial infarction, there were 4,737 subjects for GWAS analysis in RS study. After excluding 5 subjects with incomplete blood pressure data, 33 who were older than 85 years, 36 with prevalent heart failure, and 62 with prevalent myocardial infarction, there were 1760 subjects for GWAS analysis in RES (Supplementary Table 3).

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