HHS Public Access

Author manuscript

Circ Cardiovasc Genet. Author manuscript; available in PMC 2018 December 01.

Published in final edited form as:

Circ Cardiovasc Genet. 2017 December ; 10(6): . doi:10.1161/CIRCGENETICS.117.001811.

Genome-Wide Gene Potassium Interaction Analyses on Blood Pressure: The GenSalt Study

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Abstract

Background—Gene environmental interaction analysis can identify novel genetic factors for blood pressure. We performed genome-wide analyses to identify genomic loci that interact with potassium to influence blood pressure (BP) using single marker (one and two degrees of freedom [DF] joint tests) and gene-based tests among Chinese participants of the Genetic Epidemiology Network of Salt-Sensitivity (GenSalt) study.

Methods and Results—Among 1,876 GenSalt participants, the average of three urine samples was used to estimate potassium excretion. Nine BP measurements were taken using a random-zero-sphygmomanometer. A total of 2.2 million SNPs were imputed using Affymetrix 6.0 genotype data and the Chinese Han of Beijing and Japanese of Tokyo HapMap reference panel. Promising findings ($P<1.00\times10^{-4}$) from GenSalt were evaluated for replication among 775 Chinese participants of the Multi-ethnic Study of Atherosclerosis (MESA). SNP and gene-based results were meta-analyzed across the GenSalt and MESA studies to determine genome-wide significance. The one DF tests identified interactions for *ARL15* rs16882447 on systolic BP

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Disclosures: None

(P=2.83×10^{-9}) and *RANBP3L* rs958929 on pulse pressure (P=1.58×10^{-8}). The two DF tests confirmed the *ARL15* rs16882447 signal for systolic BP (P=1.15×10^{-9}). Genome-wide gene-based analysis identified *CC2D2A* (P=2.59×10^{-7}) at 4p15.32 and the *BNC2* (P=4.49×10^{-10}) at 9p22.2 for systolic BP, *GGNBP1* (P=1.18×10^{-8}) and *LINC00336* (P=1.36×10^{-8}) at 6p21 for diastolic BP, *DAB1* (P=1.05×10^{-13}) at 1p32.2 and *MIR4466* (P=5.34×10^{-8}) at 6q25.3 for pulse pressure. *BNC2* (P=3.57×10^{-8}) gene was also significant for mean arterial pressure.

Conclusions—We identified 2 novel BP loci and 6 genes through the examination of SNP- and gene-based interactions with potassium.

Keywords

potassium; genome-wide analysis; blood pressure; interaction; gene-based analysis

Journals Subject Terms

Genetic; Association Studies; High Blood Pressure; Diet and Nutrition

Introduction

Blood pressure (BP) is determined by genetic factors, environment factors, and their interactions. Genome-wide association studies (GWAS) have identified many genetic loci that are robustly associated with BP.[1–5] However, these findings together only explain a small proportion of the inter-individual variation of BP.[6] A large number of genetic loci are yet to be identified [6]. It has been proposed that exploring the interaction between genes and environmental risk factors for BP may help to identify novel genetic loci underlying BP regulation.[7–9] In addition, gene-based analysis methods testing the joint contributions of single SNPs with modest effect may have higher power to detect BP loci.[10, 11]

Dietary potassium intake has been demonstrated to decrease BP in clinical trials.[12] Since genetic factors may modify the effects of potassium on BP, examination of gene-dietary potassium intake interactions may help to identify novel genetic variants and genes underlying BP regulation.[13] However, few studies have explored gene-potassium interactions using single variant and gene-based analyses.[14, 15] The objective of this analysis was to identify novel genetic variants and genes influencing BP regulation by conducting genome-wide SNP-based and gene-based gene-potassium interaction analyses of systolic BP (SBP), diastolic BP (DBP), mean arterial pressure (MAP), and pulse pressure (PP) phenotypes among 1,876 Chinese participants of the Genetic Epidemiologic Network of Salt Sensitivity (GenSalt) study.

Methods

The data, analytic methods, and study materials will be made available to other researchers for reproducing the results or replicating the procedure upon request, discussion, and agreement with the investigators.

Study population

GenSalt is a family-based dietary intervention study designed to identify genes that influence BP responses to environmental risk factors for hypertension in human populations. The study includes 1,906 Han Chinese participants aged 16 years and older from 633 families recruited from 6 study sites in rural, north China.[16] In the current study, genotypes, BP, and covariates at baseline are available for a total of 1,876 participants (98.4%).

Institutional review boards at the Tulane University Health Sciences Center, Washington University School of Medicine, University of Texas School of Public Health, Fu Wai Hospital and Chinese National Human Genome Center at Beijing, and Chinese Academy of Medical Sciences approved the GenSalt study. Written informed consents for the baseline observation were obtained from each participant.

Genotyping and Genotype Imputation

A total of 868,158 autosomal SNPs across the entire genome were genotyped using the Affymetrix 6.0 platform for each participant. After excluding monomorphic SNPs and SNPs with Hardy-Weinberg Equilibrium $P<1\times10^{-6}$, missing rate>25% or MAF<1%, a total of 820,017 genotyped SNPs remained for genotype imputation. Imputation of 2,416,663 SNPs from the HapMap release 22 build 36 Chinese Han of Beijing and Japanese of Tokyo (CHB +JPT) reference panel was conducted using MACH software (version 1.0).[17] After imputation, SNPs with $r^2<0.30$, MAF<1%, or HWE $P<1\times10^{-6}$ were removed, and a total of 2,216,774 SNPs, with fractional values ranging from 0 to 2, remained for the analysis.

Urinary Potassium and Covariates Measurement

Dietary potassium intake was measured from 24-hour urinary potassium level. During the three-day baseline examination, each GenSalt participant collected three urine samples, one 24-hour urine sample and 2 overnight urine samples. Among a random sample of 238 participants, 24-hour urine samples were collected in two containers, one for day time and the other for overnight. Using this random sample, an equation was generated to estimate 24-hour urinary potassium level from the overnight urine sample, and this equation was applied to the remaining overnight urine samples to estimate 24-hour urinary potassium levels. The mean of the one measured 24-hour urinary potassium level and two estimated 24-hour urinary potassium levels was used in the current analysis.

Demographic variables, including age and gender, and medication use were collected using standard questionnaires by trained study staff. Body weight and height were measured twice during the baseline examination with the participants in light indoor clothing without shoes. Body mass index (BMI) was calculated as kilograms per meters squared.

BP Measurement and Imputation

SBP, DBP, MAP and PP phenotypes were assessed as outcomes in the current analyses. BP was measured three times at the same time each morning during the three-day baseline examination by trained and certified observers using a random zero sphygmomanometer according to standard protocol. For participants taking anti-hypertensive medications, BP was imputed by adding 10 mm Hg to systolic BP (SBP) and 5 mm Hg to diastolic BP

(DBP).[18] After imputation, the mean of the 9 SBP and DBP measures were used in the current analysis. The mean SBP and DBP values were also used to calculate PP (SBP-DBP) and MAP (SBP/3 + 2*DBP/3) phenotypes.

Replication Study and Genotype Imputation

We attempted to replicate promising GenSalt findings ($P<1\times10^{-4}$) among Chinese participants of the Multi-Ethnic Study of Atherosclerosis (MESA). MESA participants were recruited between July 2000 and August 2002 from 6 field centers around the US. Participants were aged 45–84 and free of cardiovascular diseases at the baseline examination. Phenotype and Affymetrix 6.0 genotype data of MESA participants were made publicly available through the NCBI's database of Genotypes and Phenotypes (dbGaP) and were downloaded for the current analysis. Among 777 Chinese MESA participants, genotype, urinary potassium, BP, and all covariables at baseline were available for 775 (99.7%) participants. Similar to GenSalt, we imputed BP for participants taking antihypertensive medication by adding 10 and 5 mm Hg to SBP and DBP, respectively. Since potassium was measured from spot urine using baseline urine samples, 24-hour urine potassium was estimated from spot urine potassium levels using Tanaka's equation.[19]

A total of 909,622 autosomal SNPs across the entire genome were genotyped using the Affymetrix 6.0 platform for each participant. We excluded SNPs with Hardy-Weinberg Equilibrium $P<1\times10^{-6}$, missing rate>5% and MAF<1%. Minimac software[20] was used to perform targeted imputation of the 3 Mb region surrounding each identified SNP based on the ALL ancestry panel from the 1000G Phase I Integrated Release Version 3 Haplotypes, which contains haplotypes of 1,092 individuals of all ethnic background. [21] After imputation, SNPs with $r^2<0.30$, MAF<1%, or HWE $P<1\times10^{-6}$ were removed.

Statistical Analysis

To accommodate familial relationships in both GenSalt (discovery) and MESA (replication) studies, linear mixed effect models were used to examine single SNP- potassium interactions on BP, after adjustment for age, gender, and BMI. Principal components analysis revealed population substructure in MESA (but not GenSalt). Therefore, ancestry was also accounted for in MESA by adjusting for the first three principal components. Both 1 degrees of freedom (df) interaction and 2 df joint tests were explored. The 2 df joint test has higher power to identify variants with both moderate main effect and moderate interaction effect. [22] As shown in supplementary figures S1 – S4, genomic inflation was minimal with lamda values ranging from 1.029 for DBP in the 1 df interaction analyses to 1.075 for pulse pressure in the 2 df joint tests. Still, genomic control was applied to single SNP-based analyses results before gene-based analyses and meta-analyses. After genomic control, lead SNPs with interaction term $P<1.0\times10^{-4}$ or joint test $P<1.0\times10^{-4}$ from independent loci in the discovery stage analyses were further evaluated for replication in Chinese MESA participants. For the 1 df interaction test, inverse-variance-weighted meta-analysis was conducted to combine results from GenSalt and MESA using METAL software.[23] For the 2 df joint test, meta-analysis was performed using methods described by Manning and colleagues, which were implemented in METAL software [23] with their patch source code.[24] After ensuring the effect direction of interaction terms were consistent, SNPs with

replication stage $P<0.05$ and meta-analysis $P<5.0\times10^{-8}$ were considered significant for the 1 df interaction test. SNPs with consistent effect directions in both the main effect and interaction term, replication stage $P<0.05$, and joint meta-analysis $P<5.0\times10^{-8}$ were considered significant for the 2 df joint test. We also conducted a sensitivity analysis restricting the replication study to Chinese MESA participants not taking any antihypertensive medication.

For the gene-based analyses, SNPs within the 5 kilo bp flanking regions of a gene were first mapped to the gene according to physical positions. SNPs within 5 kilo bp flanking regions of two genes were assigned to both genes. *P* values of both 1 df interaction and 2 df joint tests in single marker analyses were used to generate gene-based *P* values using the extended Simes procedure (GATES) method for gene-based association testing.[10] Genes with $P<1.0\times10^{-4}$ in the discovery stage gene-based interaction analysis were further evaluated for replication among Chinese MESA participants. Specifically, SNPs from promising genes were tested for SNP-potassium interactions using the methods described in the above single marker analysis, and *P* values of these SNPs were used to generate gene-based p values using GATES method.[10] Fisher's method was applied to combine gene-based p values across GenSalt and MESA.[25] Genes with replication stage $P<0.05$ and combined $P<5.0\times10^{-6}$ were considered significant. Gene-based analysis combines contributions of all variants within a gene, therefore, has higher power to detect genes interacting with dietary potassium intake on BP regulation compared to single marker analyses.

Results

As shown in Table 1, participants of both GenSalt and the MESA replication sample had, on average, optimal BP and BMI levels. However, the Chinese MESA participants were older, had a higher proportion with hypertension, and had relatively higher potassium intake. Discovery stage genome-wide analysis results using both 1 df interaction and 2 df joint tests are shown in Figures S5–S8. A total of 5,331 variants from 1092 loci ($r^2<0.3$) achieved $P<1\times10^{-4}$ and were tested for replication among MESA participants.

Two novel loci which achieved genome-wide significance in the combined analyses using the 1 df interaction test are presented in Table 2 and supplementary Figures S9–12. Novel *ARL15* gene variant rs16882447 interacted with potassium on SBP (GenSalt $P=1.99\times10^{-7}$, MESA $P=2.28\times10^{-3}$, Meta-analysis $P=2.83\times10^{-9}$). In addition, significant gene-potassium interaction on pulse pressure was identified for *RANBP3L* gene variant rs958929 (GenSalt $P=1.79\times10^{-7}$, MESA $P=1.66\times10^{-2}$, Meta-analysis $P=1.58\times10^{-8}$). Variant rs16882447 was also identified in the 2 df joint test (GenSalt $P=9.79\times10^{-9}$, MESA $P=8.19\times10^{-3}$, Meta-analysis $P=1.15\times10^{-9}$) as shown in Table 3. Regional association plots for the 2 novel loci achieving genome-wide significance in meta-analyses are presented in Figure 1. Results of sensitivity analyses conducted among Chinese MESA participants not taking antihypertension medication were similar to those conducted among the overall replication sample (supplementary Table S1).

Discovery stage gene-base analysis results are shown in Figures S13–S16. Genome-wide significant findings from meta-analysis of both the 1 df and 2 df tests are shown in Table 4.

In the gene-based analysis of the 1 df test, significant interactions between potassium and the *BNC2* and *CC2D2A* genes on SBP, *GGNBP1* and *LINC00336* on DBP, *BNC2* gene on mean arterial pressure, and *DAB1* and *MIR4466* on pulse pressure were identified. Gene-based analysis of the 2 df joint test confirmed the gene-BP associations for *BNC2*, *CC2D2A*, *GGNBP1*, and *DAB1* genes, and additionally identified novel gene-potassium interactions for *RANBP3L* on PP.

Discussion

In the first ever genome-wide gene-potassium interaction study in a Chinese population, we identified 1 novel locus, *ARL15*, that interacted with potassium to influence the BP phenotypes using both the 1 df interaction test and 2 df joint test. The 1 df interaction test also provide direct evidence of a previously reported BP locus at *RANBP3L*. Furthermore, gene-based analyses using both the 1 df and 2 df joint tests revealed 6 additional genes at 5 novel loci that were associated with BP, including: *CC2D2A* at 4p15.32 and *BNC2* at 9p22.2 for SBP, *GGNBP1* and *LINC00336* at 6p21 for DBP, *DAB1* at 1p32.2 and *MIR4466* at 6q25.3 for pulse pressure. *BNC2* gene was also significant for mean arterial pressure. These findings contribute to our understanding of the biological mechanisms underlying BP regulation.

Novel *ARL15* marker rs16882447 was identified by both the 1 df interaction test and the 2 df joint test. The 2 df joint test signal was mainly driven by the interaction effect. The *ARL15* gene encodes ADP ribosylation factor like GTPase 15.[26] This gene has been previously identified by GWAS of related cardiometabolic traits including diabetes.[27] rheumatoid arthritis.[28] high density lipoprotein cholesterol.[29] and adiponectin.[30] More importantly, a recent GWAS by Gorski and colleagues identified that the *ARL15* gene was associated with kidney function. [31] Considering the importance of kidney function in BP regulation and potassium filtration, further investigation into this gene is warranted. While the functional relevance of *ARL15* to BP is unclear, this signal could reflect other genes at this locus. For example, the nearby *HSPB3* gene, encoding the heat shock protein family B (small) member 3 [32] represents a biologically plausible candidate for hypertension. The Hspb3 gene is highly expressed in heart and skeletal muscles [33]. Animal studies showed that cardiac overloading related to hypertension increased expression of the hspb3 gene in rat models.[34] Furthermore, in Bnp knockout rats, Hspb3 was overexpressed in hypertrophic left ventricular mass prior to the development of adult-onset hypertension [35]. Future studies with higher genotyping resolution at this locus are warranted to identify the causal variant(s) that interact with potassium intake on hypertension.

The 1 df test also provided the first robust evidence for the relevance of the *RANBP3L* locus in BP regulation. A previous study utilizing a two-marker association testing approach identified a SNP pair approximately 200 kb downstream from *RANBP3L* for hypertension.[36] However, the study did not replicate their association signals in an independent sample. Our study identified a significant interaction between intronic *RANBP3L* variant rs958929 and dietary potassium intake on pulse pressure, which was replicated among MESA findings. Three variants in the *RANBP3L* gene were positively associated with urinary potassium levels (Supplementary Table S2). Therefore, the

involvement of this gene in blood pressure regulation may be mediated through its role in potassium metabolism. Our finding is further supported by a recent functional study, in which extracellular osmolality within the renal medulla up-regulated the expression of *RanBP3L*.[37] The *RANBP3L* signal may have also reflect other genes in this locus. For example, a nearby gene, *SKP2* encoding S-phase kinase associated protein 2, interacts with p27 and promotes p27 degradation [38]. Skp2 knockout mice showed p27 accumulation in cells [38, 39], and subsequently inhibited renal tubular epithelial cell proliferation and reduced renal damage. [40] Considering the importance of kidney function in blood pressure regulation and potassium filtration, this locus warrants further investigation.

Gene-based analysis of the 1 df interaction test and 2 df joint test identified 6 additional genes (*DAB1*, *CC2D2A*, *GGNBP1*, *LINC00336*, *MIR4466*, *BNC2*) at 5 novel loci that were associated with BP. The 6 genes were not reported in previous GWAS of cardiovascular related phenotypes. However, the *GGNBP1* gene was previously reported to associate with SBP in a secondary analysis using the genetic pleiotropy-informed conditional false discovery rate method.[41] In addition, mutations in the *CC2D2A* gene have been shown to cause Joubert Syndrome, a disorder characterized by renal impairment and hypertension.[42] The functional relevance of the other identified genes on BP regulation is unclear. Future studies of these genes are warranted.

Our study has several strengths. First of all, stringent quality control measures were employed for genotyping, data cleaning, covariable collection, BP measurement, and urinary potassium measurement. Second, since we limited our analysis to Chinese participants, population stratification should be minimized. Third, GenSalt was a family based study, and dietary potassium intake should have strong familial resemblance, therefore, 24-hour urinary potassium levels should have smaller variability, and subsequently we should have stronger statistical power to test gene-potassium interactions on blood pressure. Finally, a total of 9 BP were measured using random zero sphygmomanometer at the same time during the 3 day baseline examination in GenSalt. Using multiple BP measures should greatly reduce measurement error and increase statistical power to detect genetic variants for BP. However, certain limitations should also be addressed. The Chinese MESA replication sample was small, and we may not have had enough power to replicate all promising findings from the GenSalt study. In addition, 24-hour potassium level was estimated from spot urine using Tanaka's equation in MESA. Although the method has been validated previously, any measurement error could further reduce statistical power for replicating genetic variants identified in GenSalt. In addition, MESA had a high proportion of participants taking antihypertensive medication. We imputed BP by adding 10 and 5 mm Hg to SBP and DBP, respectively. Although this approach is widely applied to genetic studies of BP, [2, 5, 43] any resulting inaccuracy may dilute associations between genetic factors and BP. Furthermore, additional factors such as age and urbanization level differ between the GenSalt and MESA samples. If these factors influence gene-potassium interactions on BP, our power to detect (or replicate) SNP-BP associations could again be reduced. Imputation quality of the significant SNPs in MESA was not very high. Thus, future replication studies with larger replication sample sizes, more homogeneous populations, better measurement of urinary potassium levels, and higher imputation quality or higher genotyping resolution are

warranted. Finally, GenSalt study did not collect data on serum potassium level, so we cannot assess correlations between daily potassium intake and serum potassium levels.

In conclusion, in the first ever genome-wide gene-potassium interaction analyses of BP in a Chinese population, utilizing both single marker and gene-based analyses, we identified 6 novel loci that interacted with dietary potassium intake on SBP, and provided first robust evidence of the relevance of the *RANBP3L* locus in BP regulation. Both 1 df interaction and 2 df joint tests of single markers identified novel locus *ARL15* rs16882447. Gene-based analyses provided consistent support for the *RANBP3L* gene, and identified an additional 6 genes at 5 novel loci, including *DAB1*, *CC2D2A*, *MIR4466*, *GGNBP1*, *LINC00336* and *BNC2*. Such findings highlight the importance of examining gene-potassium interactions and conducting gene-based analysis to identify novel BP loci. Further, these findings contribute to understanding the mechanisms of BP regulation. Sequencing along with functional studies are needed to help delineate causal variants underlying the strong signals identified here.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

The Genetic Epidemiology Network of Salt Sensitivity (GenSalt) is supported by a cooperative agreement project grant (U01HL072507, R01HL087263, and R01HL090682) from the National Heart, Lung and Blood Institute, National Institutes of Health, Bethesda, MD. Research reported in this publication was supported by the National Institute of General Medical Sciences of the National Institutes of Health under Award Number P20GM109036, and by the NHLBI, NIH under Award Number 5R01HL111249-02. Support for MESA is provided by contracts N01-HC-95159, N01-HC-95160, N01-HC-95161, N01-HC-95162, N01-HC-95163, N01-HC-95164, N01-HC-95165, N01-HC-95166, N01-HC-95167, N01-HC-95168, N01-HC-95169 and CTSA UL1-RR-024156. Funding for SHARe genotyping was provided by NHLBI Contract N02-HL-64278.

Sources of Funding: The Genetic Epidemiology Network of Salt Sensitivity (GenSalt) is supported by a cooperative agreement project grant (U01HL072507, R01HL087263, and R01HL090682) from the National Heart, Lung and Blood Institute, National Institutes of Health, Bethesda, MD. Research reported in this publication was supported by the National Institute of General Medical Sciences of the National Institutes of Health under Award Number P20GM109036, and by the NHLBI, NIH under Award Number 5R01HL111249-02.

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Clinical Perspective

Dietary potassium intake has been shown to decrease blood pressure, and blood pressure responses to dietary potassium intake vary across individuals. We conducted the first genome-wide interaction analyses to identify genes and genomic loci that modified correlations between dietary potassium intake and blood pressure in a Chinese population. The current study identified one novel locus, *ARL15* rs16882447 that interacted with dietary potassium intake on systolic blood pressure in single-marker analysis. We also provided first robust evidence of the relevance of the previously reported *RANBP3L* rs958929 locus in blood pressure regulation in the single-marker analysis. Gene-based gene-potassium interaction analyses provided consistent support for the *RANBP3L* gene, and identified an additional 6 genes at 5 novel loci, including *DAB1*, *CC2D2A*, *MIR4466*, *GGNBP1*, *LINC00336* and *BNC2* for blood pressure phenotypes. Such findings provide strong evidence for the genetic background of the effect of dietary potassium intake on blood pressure regulation, and highlight the importance of examining gene-potassium interactions and conducting gene-based analysis to identify novel blood pressure loci. Further, these findings contribute to understanding the mechanisms of blood pressure regulation. However, Future sequencing and functional studies will be needed to better understand the causal mechanism underlying the identified interactions and to identify the causal variants underlying the gene-based signals.

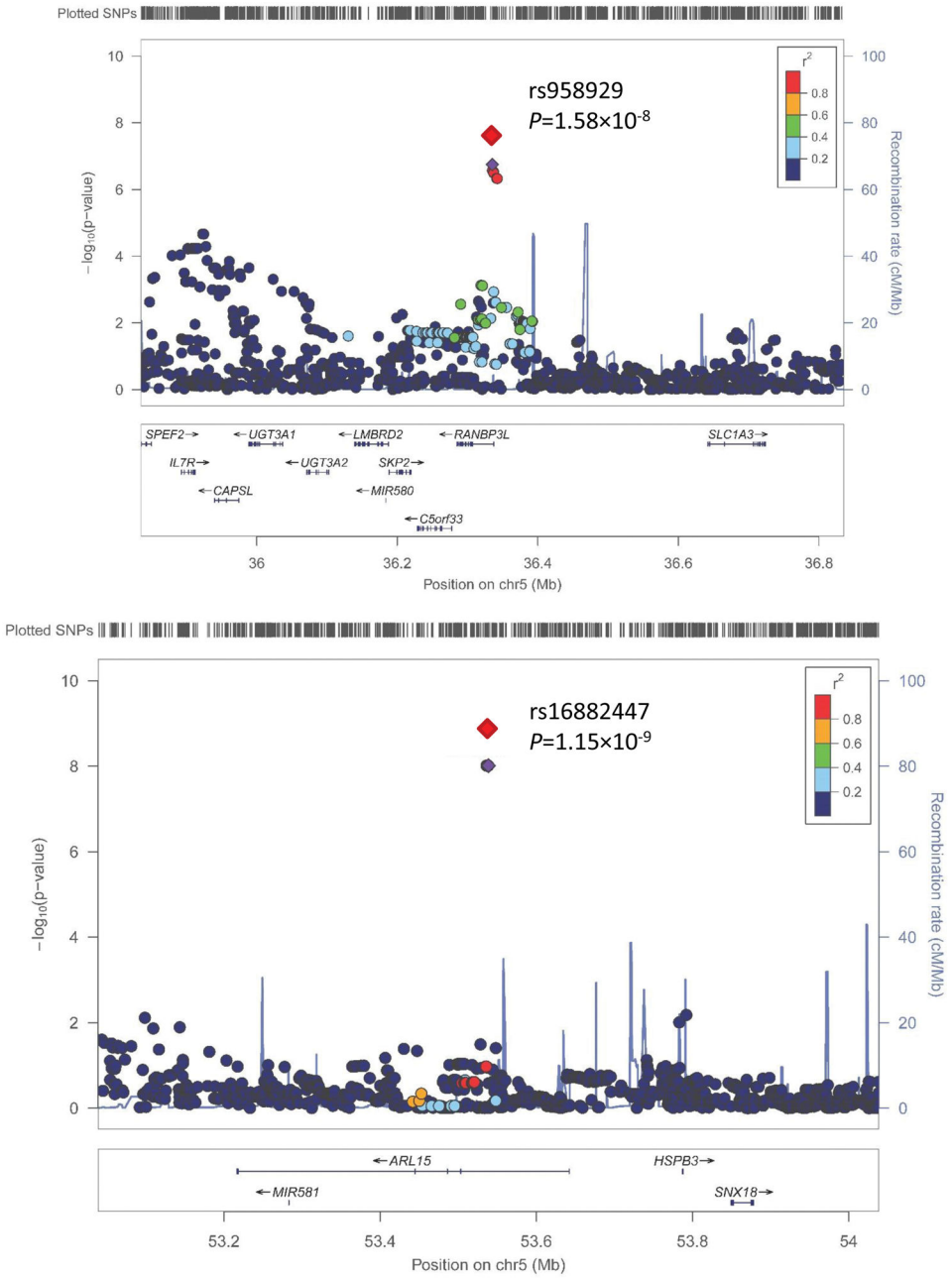

Figure 1.

Regional Association Plots for Loci Achieving Genome-Wide Significance in the Meta-Analysis of both 1 degrees of freedom (df) Interaction Test and 2 df Joint Test Results. For the SNPs associated with multiple phenotypes, the results for the most significant phenotype are shown. The index SNPs are shown in purple diamond; the correlation (r^2) of each of the surrounding SNPs to the index SNP are shown on a scale from minimal (blue) to maximal (red), The final P values for the index SNPs are shown and also indicated by the red diamonds. The genes in the 1 Mb regions around the index SNPs (500 kb on each side) are

indicated at the bottom, and recombination rates are shown in light blue line. The regional plots are drawn using LocusZoom online software

Table 1

Characteristics of GenSalt and Chinese MESA participants

	GenSalt (n=1876)	MESA (n=775)
Age, y, mean (SD)	38.7 (9.5)	62.4 (10.4)
Women, n (%)	883 (47.2)	394 (50.8)
Hypertension, n (%)	178 (9.5)	316 (40.8)
Antihypertensive medication use, n (%)	7 (0.4)	225 (29.0)
BMI, kg/m2, mean (SD)	23.3 (3.2)	24 (3.3)
24-h urinary K, mmol, mean (SD)	36.9 (9.6)	45.4 (7.8)
Baseline SBP, mmHg, mean (SD)	116.9 (14.2)	127.4 (23.8)
Baseline DBP, mmHg, mean (SD)	71.0 (9.7)	74.8 (12.0)

SD=standard deviation; SBP=systolic blood pressure; DBP=diastolic blood pressure; BMI=body mass index;

Table 2

Results Achieving Genome-wide Significance in Gene-Potassium Interactions Analysis Using 1 Degrees of Freedom Test

rsID	Chr	Position (build 36)	EA	EAF	Region	Genes	Studies	Imputation quality (r^2)	Beta	SE	*P*
Systolic Blood Pressure											
rs16882447	5	53538659	A	0.010	Intronic	*ARL15*	GenSalt	0.98	−0.82	0.16	1.99E-07
							MESA	0.51	−1.59	0.49	2.28E-03
							Meta-analysis		−0.89	0.15	2.83E-09
Pulse Pressure											
rs958929	5	36335304	C	0.340	Intronic	*RANBP3L*	GenSalt	0.83	−0.18	0.03	1.79E-07
							MESA	0.38	−0.37	0.15	1.66E-02
							Meta-analysis		−0.19	0.03	1.58E-08

Chr=chromosome; EA=effect allele; EAF=effect allele frequency; SE=standard error

Table 3

Results Achieving Genome-wide Significance in Gene-Potassium Interaction Analysis Using 2 Degrees of Freedom Joint Test

rsID	Chr	Position (Build 36)	EA	EAF	Function	Gene	Studies	SNP term		Interaction term		Covariance	Joint P
								Beta	SE	Beta	SE		
Systolic Blood Pressure													
rs16882447	5	53538659	A	0.010	Intron	*ARL15*	GenSalt	25.02	5.93	−0.82	0.15	−0.88	9.70E-09
							MESA	72.36	22.17	−1.59	0.49	−10.59	8.19E-03
							Meta-analysis	26.31	5.65	−0.83	0.15	−0.79	1.15E-09

Chr=chromosome; EA=effect allele; EAF=effect allele frequency; SE=standard error

Table 4

Genes Achieving Genome-wide Significance in Gene-based Analysis of Gene-Potassium Interactions

Gene	Chr	Start (Build 36)	Gene Length (bp)	GenSalt *P*	MESA *P*	Meta *P*
			1 DF Test			
Systolic Blood Pressure						
BNC2	9	16399500	461285	4.16E-06	1.51E-03	6.28E-09
CC2D2A	4	15080586	132105	1.16E-05	4.64E-02	5.38E-07
Diastolic Blood Pressure						
GGNBP1	6	33659453	5327	4.01E-07	2.94E-02	1.18E-08
LINC00336	6	33661860	7232	4.35E-07	3.14E-02	1.36E-08
Mean Arterial Pressure						
BNC2	9	16399500	461285	1.75E-05	2.04E-03	3.57E-08
Pulse Pressure						
DAB1	1	57236166	1255758	1.82E-05	7.14E-03	1.30E-07
MIR4466	6	1.57E+08	53	1.30E-06	4.11E-02	5.34E-08
			2 DF Test			
Systolic Blood Pressure						
BNC2	9	16399500	461285	3.97E-07	1.13E-03	4.49E-10
CC2D2A	4	15080586	132105	6.97E-05	3.71E-03	2.59E-07
Diastolic Blood Pressure						
GGNBP1	6	33659453	5327	6.86E-07	4.06E-02	2.79E-08
Mean Arterial Pressure						
BNC2	9	16399500	461285	5.35E-06	9.58E-03	5.13E-08
Pulse Pressure						

Gene	Chr	Start (Build 36)	Gene Length (bp)	GenSalt *P*	MESA *P*	Meta *P*
DAB1	1	57236166	1255758	3.29E-12	3.20E-02	1.05E-13
RANBP3L	5	36284860	54061	2.27E-05	3.36E-02	7.63E-07

DF=degrees of freedom;