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ORIGINAL ARTICLE

# Novel genetic variations associated with salt sensitivity in the Korean population

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**Salt sensitivity is a risk factor for cardiovascular morbidity and mortality. To date, only a few genetic variations have been identified as being associated with salt sensitivity. This study aimed to estimate the prevalence of salt sensitivity in the Korean population and to identify genetic variants affecting its development. A total of 101 Korean participants consumed a low-salt diet for 7 days followed by a high-salt diet for 7 additional days. Salt sensitivity was determined by noting any significant elevation in the 24-h mean arterial blood pressure. To determine genetic variants affecting salt sensitivity, 36 single-nucleotide polymorphisms (SNPs) that were previously reported to be associated with hypertension were tested for any associations with salt sensitivity. Of the 101 subjects, 28 (27.7%) were determined to have salt sensitivity. Out of the 36 SNPs tested, four were significantly associated with salt sensitivity after adjusting for confounding factors: rs2681472 in ATPase, $Ca^{++}$ transporting, plasma membrane 1 (ATP2B1), rs7961152 in branched chain aminotransferase 1 (BCAT1), rs16998073 in fibroblast growth factor 5 (FGF5) and rs2398162 in LOC100132798. For rs3754777 in serine threonine kinase 39 (STK39) and rs1937506, associations with salt sensitivity were observed before adjusting for confounding factors. Haplotype analysis revealed that the A-C haplotype of rs3754777–rs6749447 in STK39 was more frequent in the salt-sensitive group compared with the salt-resistant group, and was associated with salt sensitivity. This study estimates the prevalence of salt sensitivity in the Korean population and demonstrates a novel association between salt sensitivity and the ATP2B1, BCAT1, FGF5, LOC100132798 and STK39 genetic variations.**
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## INTRODUCTION

Hypertension, a complex trait that is influenced by both genetic and environmental factors, is a serious public health burden that affects about one-third of adults worldwide,[1] and is associated with an increased risk of cardiovascular diseases. Although it has been suggested that dietary salt intake is strongly linked to high blood pressure,[2,3] studies of the effects of dietary sodium reduction on blood pressure often show that alterations in sodium intake lead to minimal reductions in blood pressure.[4,5] This relationship is likely because of the part to the heterogeneity of blood pressure responses to alterations in dietary sodium intake. Salt sensitivity, defined as significant changes in blood pressure on the depletion or repletion of dietary salt, accounts for about 50% of cases of essential hypertension.[6] Salt sensitivity has been associated with an increase in cardiovascular disease and the reduced survival of both normotensive and hypertensive individuals.[7]

The identification of common genetic variants affecting hypertension has been challenging because of its multifactorial etiology, complex pathophysiology and limited statistical power.[8] Previous genome-wide association (GWA) studies, including the Wellcome Trust Case Control Consortium study and the Framingham Heart Study 100K Project, have suggested that several genetic variants may be associated with hypertension. However, none of these studies have achieved genome-wide significance ($P<5\times10^{-7}$).[9,10] Recently, obstacles hindering the identification of genetic variants have been overcome through the use of larger sample sizes and advances in genotype-calling algorithms and analytical methods. Thus, a greater number of genetic variants that are highly associated with hypertension ($P<5\times10^{-7}$) have been found.[11,12] Given that numerous genes are reported to be associated with hypertension and that the prevalence of salt sensitivity in hypertensive subjects is relatively high, multiple genes with variant alleles associated with hypertension are believed to be linked to salt sensitivity.

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The goal of this study was to estimate the prevalence of salt sensitivity in the Korean population and to identify genetic variants affecting the development of salt sensitivity by analyzing 36 single nucleotide polymorphisms (SNPs) previously reported to be associated with hypertension.

## METHODS

### Subjects

The study subjects consisted of 101 volunteers between the ages of 18 and 65 years (mean age 46 ± 17 years); 51 of the participants were men (50%). We excluded volunteers with stage 2 hypertension (blood pressure ⩾160/100 mm Hg), secondary hypertension, angina pectoris, diabetes mellitus, chronic kidney disease (estimated glomerular filtration rate by the Modification of Diet in Renal Disease equation $<60\,ml\,min^{-1}$) and congestive cardiac failure, volunteers who had experienced a myocardial infarction or a stroke, and volunteers who were taking diuretics or any medications that affect blood pressure and could not be safely discontinued. Volunteers who were pregnant, lactating or who were engaged in heavy alcohol consumption were also excluded. The Dongguk University institutional review board approved the study protocol, and written informed consent was obtained from all participants.

### Determination of salt sensitivity

After the eligibility of each subject was confirmed, all participants were asked to discontinue any medications, including antihypertensives that affect blood pressure and urinary electrolyte excretion, for at least 2 weeks before to the study. All eligible participants were hospitalized and administered a low-salt diet (100 mmol NaCl per day) for 7 days followed by a high-salt diet (300 mmol NaCl per day) for 7 additional days. All three meals consumed by participants each day were cooked by trained dieticians based on the hospital's standard dietary plan. Dietary compliance was verified by two trained dieticians. Participants who could not consume >90% of meals were dropped from the study; dietary compliance was 99.8%. In addition, all participants were hospitalized, their daily activity was monitored and their access to other food in addition to study meals was prohibited. The 24-h ambulatory blood pressure for each volunteer was measured on the last day of each diet period using an automated, non-invasive oscillometric device (P6 Pressurometer; Del Mar Reynolds, Irvine, CA, USA) attached to the left upper arm. Salt sensitivity was defined as an increase of >4 mm Hg ($P<0.05$) in the mean arterial pressure (MAP) in response to the high-salt diet. The MAP was calculated with the following equation: MAP=(2 diastolic blood pressure+systolic blood pressure)/3. Blood samples were obtained before the initial 2-week test period, and at the conclusion of the low-salt and high-salt diet periods. All blood samples were taken after an overnight fast. Glucose, triglyceride, total cholesterol, high-density lipoprotein cholesterol, aspartate aminotransferase and alanine aminotransferase were measured in each blood sample. Low-density lipoprotein cholesterol levels were calculated according to the following equation: low-density lipoprotein cholesterol=total cholesterol−high-density lipoprotein cholesterol−(triglyceride/5).

### Selection of SNPs and genotyping

Candidate SNPs were selected from recent GWA and large candidate gene association studies.[9,11–17] After excluding SNPs with a minor allele frequency <0.01 or a Hardy–Weinberg equilibrium *P*-value <0.001, a total of 36 SNPs were analyzed for an association with salt sensitivity. Genotyping was accomplished via a single base primer extension assay using an ABI PRISM SNaPshot Multiplex kit (Applied Biosystems, Foster City, CA, USA) according to the manufacturer's instructions. Briefly, the genomic DNA flanking each SNP was amplified by PCR with forward and reverse primer pairs and standard PCR reagents in a 10 μl reaction volume containing 10 ng of genomic DNA, 0.5 pM of each oligonucleotide primer, 1 μl of 10× PCR buffer, 250 μM deoxynucleotide triphosphate (2.5 mM each) and 0.25 U i-StarTaq DNA Polymerase ($5\,U\,\mu l^{-1}$, iNtRON Biotechnology, Sungnam, Gyeonggi-Do, Korea). PCR was performed as follows: 10 min at 95 °C for 1 cycle, followed by 35 cycles at 95 °C for 30 s, $T_m$ °C for 1 min, 72 °C for 1 min and 1 cycle at 72 °C for 10 min. After amplification, the PCR products were treated with 1 U each of shrimp alkaline phosphatase (SAP; USB Corporation, Cleveland, OH, USA) and exonuclease I (USB Corporation) at 37 °C for 75 min and 72 °C for 15 min to purify the amplified products. For the primer extension reaction, 1 μl of the purified amplification products was added to a SNaPshot Multiplex Ready reaction mixture (Applied Biosystems) containing 0.15 pmol of genotyping primer. The primer extension reaction was performed for 25 cycles at 96 °C for 10 s, 50 °C for 5 s and 60 °C for 30 s. The reaction products were treated with 1 U of SAP at 37 °C for 1 h and 72 °C for 15 min to remove excess fluorescent dye terminators. Aliquots of 1 μl of the final reaction sample containing the extension products were added to 9 μl of Hi-Di formamide (Applied Biosystems). The mixture was incubated at 95 °C for 5 min, held in ice for 5 min and analyzed by electrophoresis using an ABI Prism 3730×l DNA analyzer. Genemapper software (version 4.0; Applied Biosystems) was used for the analysis.

### Statistical analysis

PASW Statistics 17 software (SPSS, Chicago, IL, USA) was used to perform all statistical analyses. Data are expressed as the mean ± s.d. for normally distributed variables, and as a median and interquartile range for variables that are not normally distributed. The significance of differences between the groups was evaluated using a Student's *t*-test, one-way analysis of variance, Mann–Whitney test or Kruskal–Wallis test, depending on whether the data were normally distributed. The differences in frequencies between groups were tested for statistical significance with $\chi^2$-tests. The association between genotypes and the incidence of salt sensitivity was analyzed by multivariate logistic regression analysis after adjusting for age and sex. The association between each SNP and salt sensitivity was examined through the use of four different models (minor allele dominant, minor allele recessive, minor allele additive and minor allele frequency models). For each SNP, the model with the highest likelihood of developing salt sensitivity in the logistic regression model was selected. Taking into account the fact that 36 SNPs were tested in parallel, a Bonferroni's correction was performed for the associations between SNPs and salt sensitivity to correct for multiple testing. Haplotypes were generated using PHASE v2.1 (available at http://www.stat.washington.edu/stephens/phase/download.html),[18] and haplotype statistics were determined using Haploview v4.1.[19] A multifactor dimensionality reduction analysis was done with multifactor dimensionality reduction software package (available at http://www.multifactordimensionalityreduction.org) to detect gene–gene interactions. Values of $P<0.05$ were considered to be statistically significant.

## RESULTS

### Basic characteristics

A total of 28 participants (27.7%) were determined to have salt sensitivity. The baseline characteristics of salt-sensitive and salt-resistant subjects are summarized in Table 1. Sex distribution, body weight, body mass index and metabolic parameters (fasting glucose, triglyceride, total cholesterol, low-density lipoprotein cholesterol, high-density lipoprotein cholesterol, aspartate aminotransferase and alanine aminotransferase) did not differ between the groups. The mean age of the participants with salt sensitivity was significantly higher than the mean age of those who remained salt resistant. This result is consistent with previous findings in normotensive and hypertensive subjects.[2] The overall height was also significantly different between groups.

### SNPs significantly associated with the development of salt sensitivity

All 36 SNPs tested were in Hardy–Weinberg equilibrium ($P>0.05$); four were significantly associated with salt sensitivity in a multivariate logistic regression analysis adjusted for confounding factors: rs2681472 in ATPase, $Ca^{++}$ transporting, plasma membrane 1 (ATP2B1), rs7961152 in branched chain aminotransferase 1 (BCAT1), rs16998073 in fibroblast growth factor 5 (FGF5) and rs2398162 in LOC100132798 (see Table 2). The minor allele dominant model showed that rs7961152 in BCAT and rs2398162 in LOC100132798 had the highest likelihood of developing salt sensitivity, whereas the minor allele recessive model showed that

**Table 1 Subject characteristics**

| | *Salt resistant* (n=*73*) | *Salt sensitive* (n=*28*) | P-*value* |
|---|---|---|---|
| Age (years) | 43 ± 17 | 54 ± 13 | 0.001 |
| Sex (male/female) | 40/33 | 11/17 | 0.163 |
| Height (cm) | 165 ± 9 | 159 ± 8 | 0.005 |
| Weight (kg) | 66.3 ± 13.1 | 64.5 ± 13.1 | 0.534 |
| BMI ($kg\,m^{-2}$) | 24.1 ± 3.6 | 25.3 ± 4.0 | 0.181 |
| Fasting blood glucose ($mmol\,l^{-1}$) | 5.2 ± 0.6 | 5.3 ± 0.5 | 0.162 |
| Triglyceride ($mmol\,l^{-1}$) | 1.6 ± 0.7 | 1.8 ± 0.8 | 0.275 |
| Total cholesterol ($mmol\,l^{-1}$) | 4.9 ± 1.0 | 5.1 ± 0.9 | 0.439 |
| LDL cholesterol ($mmol\,l^{-1}$) | 3.0 ± 0.9 | 3.1 ± 0.8 | 0.617 |
| HDL cholesterol ($mmol\,l^{-1}$) | 1.2 ± 0.3 | 1.2 ± 0.4 | 0.934 |
| AST ($U\,l^{-1}$) | 19.3 ± 7.3 | 20.8 ± 8.7 | 0.372 |
| ALT ($U\,l^{-1}$) | 19.5 ± 12.9 | 21.0 ± 14.5 | 0.609 |

Abbreviations: ALT, alanine aminotransferase; AST, aspartate aminotransferase; BMI, body mass index; HDL, high-density lipoprotein; LDL, low-density lipoprotein.
Data are expressed as mean ± s.d.

rs2681472 in ATP2B1 had the highest likelihood of developing salt sensitivity. The SNP rs16998073 in FGF5 exhibited the highest likelihood of developing salt sensitivity in the minor allele additive model. The association with salt sensitivity did not remain significant at a conservative Bonferroni-corrected level. For rs3754777 in serine threonine kinase 39 (STK39) and rs1937506, associations with salt sensitivity ($P$=0.040 and 0.006, respectively) were observed before adjusting for confounding factors. However, the association was not significant after adjusting for age and sex. Details regarding the associations between other SNPs and salt sensitivity are shown in Table 2.

In addition, an association analysis was performed to identify SNPs associated with changes in MAP between low-salt and high-salt diets. The SNPs rs2398162 in LOC100132798 and rs1937506 were significantly associated with changes in MAP between low-salt and high-salt diets (Table 3). These SNPs were the ones significantly associated with salt sensitivity before adjusting for confounding factors, and the association for rs2398162 remained significant even after adjusting for confounding factors.

The genotype frequencies of the four significant SNPs are shown in Table 4. Although there were no significant changes with respect to the genotype in the 24-h systolic blood pressure when going from a low- to a high-salt diet, salt-induced changes measured by the 24-h diastolic blood pressure and 24-h MAP were significantly higher for GT and TT in rs7961152 (BCAT), TT in rs16998073 (FGF5) and GA in rs2398162 (LOC100132798; see Table 4). This finding suggests that these alleles are regulatory alleles that control blood pressure.

### Association of STK39 haplotype with salt sensitivity

Haplotype association with salt sensitivity was tested on four haplotype groups generated by PHASE v2.1: (1) rs1378942 in c-src tyrosine kinase-rs6495122, (2) rs4532-rs5326-rs265981 in type 1 dopamine receptor, (3) rs1024323-rs1801058-rs2960306 in G protein-coupled receptor kinase type 4 and (4) rs3754777–rs6749447 in STK39. Among the tested haplotype groups, the A-C haplotype of rs3754777–rs6749447 in STK39 occurred more frequently than other haplotypes in the salt-sensitive group compared with the salt-resistant group (Table 5), and showed a significant association with salt sensitivity (odds ratio=5.3, $P$=0.034). In addition, the multifactor dimensionality reduction method was used for a gene–gene interaction analysis;[20] however, no significant gene–gene interactions were identified for salt sensitivity.

## DISCUSSION

Previous studies suggest that salt sensitivity is a risk factor for developing hypertension and other cardiovascular diseases, and that genetic factors contribute to salt sensitivity susceptibility.[7,14,21–22] Given both the high prevalence of salt sensitivity (about 50%) in cases of essential hypertension and the emergence of publications showing the influence of SNPs on the development of hypertension, it has been hypothesized that many hypertension-associated SNPs may influence the development of salt sensitivity. In this work, four SNPs and one haplotype were found to be associated with the development of salt sensitivity: rs2681472 in ATP2B1, rs7961152 in BCAT1, rs16998073 in FGF5, rs2398162 in LOC100132798 and the A-C haplotype of rs3754777–rs6749447 in STK39.

The SNP rs2681472 is located in ATP2B1, a plasma membrane calcium ATPase that is associated with the homeostasis of cellular calcium ion levels and the control of vascular reactivity and blood pressure.[23] A GWA study of blood pressure and hypertension in the Cohorts for Heart and Aging Research in Genome Epidemiology study and the Global Blood Pressure Genetics Consortium demonstrated a strong association of rs2681472 with blood pressure and the development of hypertension.[11] The functional importance of genetic variations in ATP2B1 with regard to the regulation of blood pressure was highlighted by the findings of several other GWA studies that identified additional significant SNPs in or near ATP2B1. These SNPs included rs2681492, rs11105354 and rs17249754.[11,13,24,25] Regardless of the involvement of these SNPs in hypertension, their association with salt sensitivity has not yet been documented. In this study, a significant association between rs2681472 in ATP2B1 and the development of salt sensitivity was demonstrated.

The strongest association between a SNP and salt sensitivity was observed for rs7961152, which is located in cytosolic BCAT1, an enzyme that catalyzes the reversible transamination of branched-chain α-keto acids to branched-chain L-amino acids. BCAT1 is known to have a role in cell growth and apoptosis.[26,27] Although the functional involvement of rs7961152 in the development of salt sensitivity and hypertension has not yet been reported, previous studies have shown the association of rs7961152 with systolic and diastolic blood pressure,[28] as well as with hypertension.[9]

FGF5 is a member of the FGF family that mediates a variety of biological processes, including embryonic development, cell growth, morphogenesis, tissue repair, and tumor growth and invasion.[29,30] A recent GWA study that focused on blood pressure revealed eight blood pressure-associated genetic variants, including rs16998073 in FGF5, suggesting a possible role in blood pressure control.[12] Regarding rs2398162 in LOC100132798, an association with hypertension has been suggested in a Wellcome Trust Case Control Consortium study.[9] However, a replication study in a Korean sample failed to confirm this association.[28] In this work, the finding that two SNPs (rs16998073 and rs2398162) were significantly associated with salt sensitivity suggested that further studies are needed to investigate functions related to blood pressure control with regard to the two genetic variations.

In addition to the above results, rs3754777 in STK39 exhibited a possible association with salt sensitivity only when analyzed before the adjustment of confounding factors. Moreover, haplotype analysis revealed a significant association between the STK39 haplotype and salt sensitivity. The strong association between salt sensitivity and two SNPs (rs3754777 and rs6749447) in the *STK39* gene, which encodes the Ste29-related proline–alanine-rich kinase protein, was reported in a GWA study in the Old Order Amish population and was confirmed in a separate meta-analysis.[16] In the aforementioned paper, the authors suggested that STK39 may regulate blood pressure

**Table 2 Multivariate logistic regression analysis of SNPs associated with salt sensitivity**

| *SNP* | *Gene* | *Alleles 1/2* | *MAF* | *Odds ratio* | *95% CI* | P-*value* | *Model* |
|---|---|---|---|---|---|---|---|
| rs12946454 | *ACBD4* | T/A | 0.187 | 0.7 | 0.3–1.6 | 0.389 | T/A |
| rs4341 | *ACE* | C/G | 0.376 | 0.7 | 0.2–2.7 | 0.550 | CC+CG/GG |
| rs4961 | *ADD1* | T/G | 0.371 | 1.4 | 0.5–3.6 | 0.530 | TT/TG+GG |
| rs699 | *AGT* | C/T | 0.168 | 1.8 | 0.7–4.8 | 0.250 | CC/CT/TT |
| rs5186 | *AGTR1* | A/C | 0.021 | 2.7 | 0.3–22.4 | 0.360 | AA/AC/CC |
| rs2681472 | *ATP2B1* | T/C | 0.410 | 3.8 | 1.1–13.7 | 0.040 | (TT+TC)/CC |
| rs6594013 | *ATP2B4* | A/T | 0.369 | 1.4 | 0.4–5.3 | 0.650 | AA+AT/TT |
| rs7961152 | *BCAT1* | G/T | 0.105 | 4.9 | 1.5–15.5 | 0.007 | GG/(GT+TT) |
| rs1530440 | *C10orf107* | C/T | 0.175 | 0.7 | 0.3–1.8 | 0.495 | C/T |
| rs11014166 | *CACNB2* | A/T | 0.116 | 1.0 | 0.3–3.2 | 0.946 | AA/AT/TT |
| rs1378942 | *CSK* | C/A | 0.150 | 0.6 | 0.2–1.7 | 0.343 | CC/CA+AA |
| rs1799998 | *CYP11B2* | T/C | 0.340 | 1.1 | 0.5–2.1 | 0.860 | T/C |
| rs1004467 | *CYP17A1* | T/C | 0.305 | 0.7 | 0.3–1.8 | 0.502 | TT/TC+CC |
| rs4532 | *DRD1* | A/G | 0.119 | 1.3 | 0.4–3.7 | 0.680 | AA/AG+GG |
| rs5326 | *DRD1* | G/A | 0.273 | 0.7 | 0.3–1.7 | 0.404 | GG/GA+AA |
| rs265981 | *DRD1* | C/T | 0.110 | 1.5 | 0.5–4.4 | 0.508 | CC/CT+TT |
| rs16998073 | *FGF5* | A/T | 0.405 | 2.1 | 1.0–4.2 | 0.042 | AA/AT/TT |
| rs5443 | *GNB3* | T/C | 0.495 | 1.5 | 0.5–4.3 | 0.500 | TT/TC+CC |
| rs1024323 | *GRK4* | C/T | 0.18 | 0.6 | 0.3–1.5 | 0.300 | C/T |
| rs1801058 | *GRK4* | T/C | 0.431 | 1.4 | 0.7–2.8 | 0.280 | T/C |
| rs2960306 | *GRK4* | G/T | 0.115 | 0.8 | 0.3–2.2 | 0.660 | G/T |
| rs2398162 | *LOC100132798* | G/A | 0.380 | 3.5 | 1.2–10.4 | 0.023 | GG/(GA+AA) |
| rs17367504 | *MTHFR* | T/C | 0.091 | 1.5 | 0.5–4.7 | 0.519 | TT/TC/CC |
| rs11110912 | *MYBPC1* | C/G | 0.081 | 2.4 | 0.5–10.8 | 0.273 | CC/CG/GG |
| rs4149601 | *NEDD4 L* | G/A | 0.082 | 0.4 | 0.1–1.5 | 0.174 | G/A |
| rs11191848 | *OBFC1* | A/G | 0.360 | 0.5 | 0.2–1.0 | 0.054 | A/G |
| rs381815 | *PLEKHA7* | G/A | 0.190 | 1.9 | 0.7–5.0 | 0.174 | GG/GA+AA |
| rs3754777 | *STK39* | G/A | 0.225 | 5.0 | 0.5–51.2 | 0.178 | GG+GA/AA |
| rs6749447 | *STK39* | C/A | 0.310 | 0.9 | 0.5–1.9 | 0.830 | C/A |
| rs16948048 | *ZNF652* | T/C | 0.205 | 3.3 | 0.5–24.1 | 0.235 | TT+TC/CC |
| rs1937506 | | G/A | 0.130 | 1.9 | 0.8–4.8 | 0.153 | G/A |
| rs2384550 | | G/A | 0.105 | 2.2 | 0.8–6.3 | 0.152 | GG/GA/AA |
| rs2820037 | | A/T | 0.061 | 0.6 | 0.2–2.4 | 0.483 | A/T |
| rs6495122 | | A/C | 0.155 | 0.5 | 0.2–1.4 | 0.177 | AA/AC/CC |
| rs6997709 | | G/T | 0.095 | 0.3 | 0.1–1.5 | 0.136 | GG/GT+TT |
| rs9815354 | | G/A | 0.210 | 0.7 | 0.1–7.4 | 0.752 | GG+GA/AA |

Abbreviations: *ACBD4*, acyl-co-enzyme A-binding domain containing 4; *ACE*, angiotensin-converting enzyme; *ADD1*, adducin 1 (α); *AGT*, angiotensinogen; *AGTR1*, angiotensin II type 1 receptor; *ATP2B1*, ATPase, $Ca^{++}$ transporting, plasma membrane 1; *ATP2B4*, ATPase, Ca++ transporting, plasma membrane 4; *BCAT1*, branched chain aminotransferase 1; *C10orf107*, chromosome 10 open reading frame 107; *CACNB2*, calcium channel, voltage-dependent, β 2 subunit; CI, confidence interval; *CSK*, c-src tyrosine kinase; *CYP11B2*, cytochrome P450, family 11, subfamily B, polypeptide 2; *CYP17A1*, cytochrome P450, family 17, subfamily A, polypeptide 1; *DRD1*, type 1 dopamine receptor; *FGF5*, fibroblast growth factor 5; *GNB3*, G-protein β3 subunit; *GRK4*, G protein-coupled receptor kinase type 4; *MAF*, minor allele frequency; *MTHFR*, 5,10-methylenetetrahydrofolate reductase (nicotinamide adenine dinucleotide phosphate); *MYBPC1*, myosin-binding protein C, slow type; *NEDD4L*, neural precursor cell expressed, developmentally down-regulated 4-like; *OBFC1*, oligonucleotide/oligosaccharide-binding fold containing 1; *PLEKHA7*, pleckstrin homology domain containing, family A member 7; *SNP*, single-nucleotide polymorphism; *STK39*, serine threonine kinase 39; *ZNF652*, zinc-finger protein 652.
Multivariate logistic regression analysis was performed with adjustments for age and sex.

**Table 3 Association analysis between SNPs and changes in mean arterial pressure from low-salt diet to high-salt diet**

| *SNP* | *Gene* | *GG* | *GA* | *AA* | P *recessive* | P *dominant* |
|---|---|---|---|---|---|---|
| rs2398162 | *LOC100132798* | 0.3 ± 4.0 | 2.4 ± 4.2 | 1.3 ± 5.0 | 0.865 | 0.029 |
| rs1937506 | | 1.4 ± 4.4 | 1.0 ± 3.3 | 7.5 ± 3.5 | 0.014 | 0.725 |

Abbreviation: SNP, single-nucleotide polymorphism.
Data are expressed as mean ± s.d.

by increasing its expression and altering renal sodium excretion through its interaction with WNK kinase and cation-chloride cotransporters. The SNP rs1937506 was significantly associated with salt sensitivity and changes in MAP between low-salt and high-salt diets before adjusting for confounding factors. The SNP rs1937506 is not linked to any gene and, with the exception of the Wellcome Trust Case Control Consortium study,[9] no research explaining its function or association has been published.

Previous association studies have demonstrated that individuals with genetic variants in adducin 1 α, angiotensinogen, CYP11B2 (cytochrome P450, family 11, subfamily B, polypeptide 2), G-protein â3 subunit, G protein-coupled receptor kinase type 4 and neural precursor cell expressed developmentally downregulated 4-like are more susceptible to developing salt sensitivity.[14,15,21,22,31,32] Among these genetic variants, there is a growing body of evidence that Gly460Trp polymorphism (rs4961) in adducin 1 α, a heterodimeric cytoskeleton protein that is involved in cell signal transduction, the regulation of actin cytoskeleton and ion transport across the cell membrane, is associated with hypertension and salt sensitivity through the renal sodium retaining effect of the Trp allele.[22,33] However, in this paper, no significant association was observed between salt sensitivity and genetic variants in adducin 1α, angiotensinogen,

**Table 4 Salt-induced blood pressure changes in different genotype carriers**

| | *rs2681472 genotype variants* | | | |
|---|---|---|---|---|
| | *TT (n=30)* | *TC (n=58)* | *CC (n=12)* | P *value; TT vs. TC or CC* |
| Δ24-h SBP (mm Hg) | 1.8 (−0.9 to 5.8) | 1.3 (−2.7 to 4.5) | 4.9 (−0.1 to 6.0) | 0.518 |
| Δ24-h DBP (mm Hg) | 1.8 (−1.5 to 4.7) | 0.4 (−1.5 to 2.9) | 4.8 (−1.0 to 6.6) | 0.242 |
| Δ24-h MAP (mm Hg) | 1.8 (−0.1 to 5.8) | 0.5 (−1.8 to 3.2) | 5.1 (−1.2 to 6.0) | 0.218 |
| | *rs7961152 genotype variants* | | | |
| | *GG (n=81)* | *GT (n=17)* | *TT (n=2)* | P *value; GG vs. GT or TT* |
| Δ24-h SBP (mm Hg) | 1.5 (−2.6 to 4.6) | 3.9 (1.5 to 5.8) | 5.4 (0.2 to 10.6) | 0.127 |
| Δ24-h DBP (mm Hg) | 0.4 (−1.8 to 3.4) | 3.2 (1.9 to 6.5) | 4.6 (0.2 to 9.0) | 0.013 |
| Δ24-h MAP (mm Hg) | 0.5 (−1.7 to 3.4) | 4.8 (2.0 to 5.8) | 3.6 (0.2 to 7.0) | 0.035 |
| | *rs16998073 genotype variants* | | | |
| | *AA (n=34)* | *AT (n=51)* | *TT (n=15)* | P *value; AA vs. AT or TT* |
| Δ24-h SBP (mm Hg) | 0.3 (−3.4 to 3.9) | 2.5 (−0.7 to 6.0) | 2.7 (1.4 to 4.5) | 0.077 |
| Δ24-h DBP (mm Hg) | −0.1 (−2.5 to 2.9) | 1.0 (−0.9 to 4.1) | 4.4 (1.0 to 5.0) | 0.007 |
| Δ24-h MAP (mm Hg) | −0.2 (−2.7 to 2.5) | 1.3 (−0.4 to 4.8) | 4.1 (1.1 to 5.0) | 0.035 |
| | *rs2398162 genotype variants* | | | |
| | *GG (n=37)* | *GA (n=54)* | *AA (n=9)* | P *value; GG vs. GA or AA* |
| Δ24-h SBP (mm Hg) | 1.1 (−1.2 to 4.5) | 2.7 (−2.0 to 6.7) | 0.1 (−3.2 to 3.6) | 0.073 |
| Δ24-h DBP (mm Hg) | 0.2 (−1.8 to 2.1) | 2.2 (−0.4 to 4.7) | −0.3 (−2.5 to 5.3) | 0.012 |
| Δ24-h MAP (mm Hg) | 0.5 (−1.9 to 2.4) | 2.5 (−0.2 to 5.3) | −0.1 (−2.5 to 4.7) | 0.042 |

Abbreviations: Δ24-h DBP, Δ24-h diastolic blood pressure; Δ24-h MAP, Δ24-h mean arterial blood pressure; Δ24-h SBP, Δ24-h systolic blood pressure.
Data are expressed as median (interquartile range). Blood pressure change in low- *vs.* high-salt intake.

**Table 5 STK39 rs3754777–rs6749447 haplotype frequencies in the study population**

| | *Frequency* | | |
|---|---|---|---|
| *Haplotype* | *Salt resistant (n=73)* | *Salt sensitive (n=28)* | P-*value* |
| G-C | 0.465 | 0.464 | 0.365 |
| G-A | 0.326 | 0.268 | 0.422 |
| A-C | 0.208 | 0.268 | 0.039 |
| A-A | 0 | 0 | |

Abbreviation: STK39, serine threonine kinase 39.

CYP11B2, G-protein β3 subunit, G protein-coupled receptor kinase type 4 and neural precursor cell expressed developmentally down-regulated 4-like. Such a discrepancy may be because of the differences in sample origin, methods of testing for salt sensitivity and the complexity and heterogeneity of salt sensitivity in relation to blood pressure.

To the best of our knowledge, this study is the first report regarding the details of salt sensitivity, its prevalence and related SNPs in a Korean sample. Although inconsistencies exist between previous reports and our findings, we demonstrated a novel association between salt sensitivity and genetic variations in ATP2B1, BCAT1, FGF5, LOC100132798 and STK39. Because the sample size of this cohort is relatively small and because certain aspects of hypertension were not considered, a more comprehensive study with a larger sample size and the consideration of prevalence, age of onset, severity and the complications of hypertension are required to replicate the associations detailed in this work. Furthermore, future research aimed at defining the functional role of each genetic variant identified in this paper on the risk of salt sensitivity will need to be performed. The accumulation of such evidence will allow for the use of diagnostic SNP screening tests for early detection of salt sensitivity and facilitate interventions to prevent the development of salt sensitive hypertension.

## CONFLICT OF INTEREST

The authors declare no conflicts of interest.

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