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GENETIC VARIATION IN THE RENIN-ANGIOTENSIN-ALDOSTERONE SYSTEM:
ASSOCIATIONS WITH BLOOD PRESSURE DIPPING IN THE JACKSON HEART
STUDY

BY
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B.A., BEREA COLLEGE, 2007

A THESIS
SUBMITTED TO THE FACULTY OF THE SCHOOL OF PUBLIC HEALTH AND
INFORMATION SCIENCES
OF THE UNIVERSITY OF LOUISVILLE
IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE DEGREE OF

MASTER OF SCIENCE

DEPARTMENT OF EPIDEMIOLOGY AND POPULATION HEALTH
UNIVERSITY OF LOUISVILLE
LOUSVILLE, KY

AUGUST 2012

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DEDICATION

I dedicate this thesis to my wonderful parents

Cristobal Napoleon

and

Doris M. Napoleon

who have encouraged me and supported me
throughout my entire educational career.

ACKNOWLEDGEMENTS

I would like to thank deeply my professor, Dr. Kira Taylor, for guiding me and encouraging me every step of the way. I couldn't have done this without her. I would also like to thank my committee members, Dr. Richard Baumgartner and Dr. Guy Brock, for their helpful comments and their wonderful sense of humor. I would also like to thank all the members of the Jackson Heart Study who provided me with the opportunity to study this topic. I would like to give a special thanks to my beautiful sisters, Willkenia Napoleon, Johannil Napoleon, and Rosanna Napoleon. They were always ready to help me no matter the circumstances. I love you all dearly. Lastly, I would like to thank Christopher Henderson for his unwavering support throughout this journey. His words of wisdom and his continuous reminder of my purpose in this lifetime will stay with me forever.

ABSTRACT

GENETIC VARIATION IN THE RENIN-ANGIOTENSIN ALDOSTERONE SYSTEM:
ASSOCIATIONS WITH BLOOD PRESSURE DIPPING IN
THE JACKSON HEART STUDY

Jersy L. Napoleon

August 6, 2012

Cardiovascular diseases, including hypertension, are the leading causes of death in the United States. African-Americans carry the highest burden of disease with 32% of adults with hypertension. Blood pressure (BP) dipping, defined as a drop of >10% in blood pressure from daytime to nighttime, occurs less often in African-Americans and is a risk factor for CVD. BP dipping has a significant genetic component. This study focuses on the single nucleotide polymorphisms (SNPs) in the renin-angiotensin-aldosterone system that may influence BP dipping in this population. A total of 928 participants (266 dippers and 664 non-dippers) from the Jackson Heart Study (JHS) were analyzed. The study found that 37 common SNPs across three genes (*ACE*, *AGT*, and *AGTR1*) were not significantly associated with BP dipping in this population. Therefore, there are likely additional genes and possibly other SNPs within these genes that are responsible for the genetic contribution to BP dipping.

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INTRODUCTION

Cardiovascular diseases (CVD) are the leading cause of death in the country [1]. The prevalence of CVD is increasing each year and the profile of CVD patients is getting younger in age (under 65) [2]. Not only does CVD account for 17% of the national health expenditures, but costs are projected to rise 61% by 2030 [3, 4]. In 2008, heart disease and strokes accounted for over 470 million dollars in direct and indirect costs [5].

In the United States, hypertension or high blood pressure (HBP) is considered one of the major risk factors for CVD [6, 7]. Hypertension increases the incidence of ischemic and hemorrhagic stroke, coronary heart disease, left ventricular mass and renal disease. An individual can acquire either essential (primary) or secondary hypertension. Causes of secondary hypertension may be due to prescribed pharmaceuticals or complications of other diseases, such as preeclampsia, renal artery stenosis, endocrine disorders, and obstructive sleep apnea [8]. However, secondary hypertension accounts for a very small percentage of all hypertension cases in the United States. Ninety to ninety-five percent of hypertension cases are primary (or essential) [8]. Major risk factors for primary hypertension are age, ethnicity, gender, lack of physical activity, high salt intake, obesity, insulin resistance, high alcohol intake, socioeconomic status (SES) and family history [7, 8].

Primary hypertension is especially a concern in African-Americans because they carry a disproportionately higher burden of the disease than other ethnic groups. Unlike

other ethnic groups, hypertension is more common and severe in African-Americans. African-Americans also have a higher of occurrence of coronary heart disease and are also more like to die from CVD than their white counterparts [9]. According to the Center for Disease Control and Prevention (CDC), between 2005 and 2008, an estimated 18% of US adults 18 years and older in the United States were considered pre-hypertensive, while 30% of US adults had hypertension due to a high systolic or diastolic BP. In addition, African-Americans are more likely than white Americans to develop hypertension (32% vs. 25%) [10].

THE BIOLOGY OF HYPERTENSION

Hypertension is a failure of the body's attempt to normalize blood pressure. Blood pressure is mainly regulated by the renin-angiotensin-aldosterone system (RAAS) (Fig. 1). RAAS is a cascade of enzymatic and hormonal chemical reactions involving the kidneys and the sympathetic nervous system [11]. RAAS regulates blood volume, cardiac output, vasoconstriction in the body, sodium and water balance. Angiotensinogen, released by the liver, interacts with the renal enzyme renin to produce angiotensin I (Ang I), which in turn reacts with the angiotensin-converting enzyme (ACE) to produce angiotensin II (Ang II) [12, 13]. Angiotensin II, a highly vasoactive peptide, causes vasoconstriction in the blood vessels, which increases blood pressure. Angiotensin II stimulates the adrenal glands to secrete aldosterone, which increases salt and water retention and blood volume, raising the blood pressure [14].

RAAS works in a circadian rhythm, which allows blood pressure to naturally fluctuate [15]. RAAS activity generally decreases at nighttime and increases again during the daytime. However, for some individuals RAAS remains very active during both day and night. As a result, the adrenal glands consistently stimulate a release of renin and the levels of ACE, angiotensinogen, Ang I, Ang II and aldosterone, remain high [14]. An insufficient drop of blood pressure from day to night has been associated with CVD [11, 14, 16, 17].

HYPERTENSION AND RAAS GENES

Studies have shown that hypertension may be a multi-factorial disease where there is an interaction between environmental and genetic factors [7]. The angiotensinogen (*AGT*), angiotensin II type 1-receptor *AGTRI* (also called *ATRI*,) and the angiotensin I-converting-enzyme (*ACE*) genes in chromosomes 1, 3, and 17 respectively, are three genes that have been associated with hypertension and RAAS [18-20].

The CORGENE study, which compared coronary atherosclerosis in 156 Caucasians with post-myocardial infarctions (MI) and 307 healthy patients with no history of MI, was one of the firsts to report association with an *AGT* polymorphism. The study found that *AGT* M235T, located in chromosome 1 position 235, where there is a methionine to threonine substitution, was associated with hypertension [21]. Likewise, in a cohort study of 916 black and 771 white post-myocardial infarction patients, Goldenberg and company found that the *M235T* polymorphism in the *AGT* gene was associated with increased levels of angiotensinogen and high blood pressure. The frequency of the *T* allele was substantially more common in black patients (82%) than in white (44%) patients. Black patients with essential hypertension who were homozygous for the *T* allele (TT) were over 3 times more likely to experience a cardiac event [22]. Moreover, a meta-analysis of 11 studies published between 1992 and 1996, including 14 different populations with 5,493 individuals, reported that the *AGT* M235T polymorphism was significantly associated with hypertension (Odds Ratio (OR): 1.20;

95% Confidence Interval (CI): 1.11-1.29; (P-value) $P < .0001$) and the association increased with a family history of hypertension (OR: 1.42; 95% CI: 1.25 -1.61, $P < .0001$) [23].

The *AGTR1* gene has also been linked to essential hypertension in different populations. Five genetic variants of the *AGTR1* gene were found in 239 hypertensive and 371 normotensive Mexican individuals [24]. However, the study found that participants who had *rs5182* CC genotype were more at risk of hypertension (OR=1.83, $p=0.009$). A recent meta-analysis, which examined 24 different populations in 22 case-control studies published through February 2010 and included 8,249 hypertensive individuals and 8,225 controls, reported that *AGTR1* +1166 C allele was associated with essential hypertension (OR:1.14; 95%CI, 1.00-1.30; $P=0.05$) [25].

Finally, the Framingham Heart Study reported an association between common variants of the *ACE* gene and essential hypertension and high levels of angiotensinogen [26]. The study looked at 276 men and 333 women with hypertension and found that men with the *ACE* DD allele were at a higher risk of developing hypertension than women.

HYPERTENSION AND RASS GENES IN AFRICAN-AMERICANS

Fewer studies of genes associated with hypertension have specifically targeted African-Americans compared to their white counterparts. However, studies like the International Collaborative Study on Hypertension (ICSHIB) examined the relationship between hypertension and environment in African descendants by comparing seven populations representing Nigeria, rural and urban areas of Cameroon, St. Lucia, Barbados, Jamaica and Chicago, Illinois. The study found an increase in prevalence of hypertension in participants who lived in urban (19.1%) and rural areas (15.1%) of Cameroon. Despite the variation of hypertension in West Africa, the results concluded that as the geographical region moved from east to west there was an increase in hypertension, with Nigeria having the lowest prevalence at 14.5% versus the US at 32.6% [27]. The same pattern was observed when the participants were stratified by gender: US males and females were more likely to be hypertensive than West Africans and Caribbean blacks, with US females displaying the highest prevalence over all [27].

Adeyemo and colleagues conducted a genome wide association study (GWAS) of hypertension in over 1,000 African-Americans residing in Washington, DC. They examined 800,000 single nucleotide polymorphisms (SNPs). The study found that (SNPs) were associated with biological systems and networks important to the regulation of blood pressure. Single nucleotide polymorphisms, such as PMS1, SLC24A4, YWHA7, IPO7, and CACANA1H were specifically found to be associated with systolic blood pressure [28].

BLOOD PRESSURE DIPPING

Accumulating evidence indicates that a decline in nocturnal blood pressure relative to daytime blood pressure, also known as blood pressure (BP) dipping, is an independent predictor of hypertension. BP dipping is defined as having a change of 10% or more in blood pressure from awake to sleeping hours [29]. In a study of blood pressure heritability in European-American and African-American twins, Wang et al. suggested that there may be distinct genes that influence nocturnal vs. diurnal blood pressure [30].

In the Wisconsin Sleep Cohort Study, investigators examined the influence of specific genetic variants on BP dipping in 497 non-hypertensive European Americans ages 30-60 years, not on hypertensive medication [16]. Blood pressure dipping was defined as having a 10% or greater change in blood pressure from daytime to nighttime. Single nucleotide polymorphisms (SNPs) in two types of beta-adrenergic receptors (β 1-AR and β 2-AR) were examined. These adrenergic receptors, which are proteins associated with the sympathetic nervous system, control the heart rate, and cardiac contractility, and conduction of cardiac muscle [31]. β 2-AR, specifically, is involved in the release of renin. Renin affects blood pressure by stimulating the release of aldosterone in the body [14, 16]. This study found that beta-adrenergic receptors particularly affect nighttime BP, which is the blood pressure mostly associated to BP dipping [11, 16]. The study found that β 2-AR rs2400707 was statistically significant with systolic (SBP) and

diastolic (DBP) blood pressure dipping. Those who were homozygous for A/A haplotypes were more likely to display a non-dipping pattern (less than 10% BP dipping) than those with A/G haplotypes (OR: 2.0; 95% CI, 1.0-3.8 vs. OR: 1.6; 95% CI, 0.9-2.8; $P=0.04$) [16].

Julve and colleagues reported an association between BP dipping and the insertion and deletion polymorphisms of the *ACE* gene in a sample of 99 previously-untreated hypertensive Caucasians less than 50 years old. The study analyzed three *ACE* gene genotypes (*II*, *ID*, and *DD*) and discovered that those who were homozygous for the D allele had less nocturnal fall of SBP than the other genotypes [32]. Only two other studies have shown that there is an association between BP dipping and the D alleles of the *ACE* gene. One study reported that parents of 65 Chinese hypertensives who had the D allele had a higher average 24-hr BP and a higher nocturnal BP [33]. In the other study, 235 untreated elderly Japanese hypertensives with the D allele had higher nighttime BP [16, 34].

BLOOD PRESSURE DIPPING IN AFRICAN-AMERICANS

Most of the studies on BP dipping in African-Americans have focused mainly on the social and environmental factors affecting BP dipping. Hickson and colleagues explored the association between BP dipping and socioeconomic position (SEP), such as income and education, in the 837 eligible African-Americans in the Jackson Heart Study. Although the study concluded that a higher SEP was related to a greater BP dipping, the study also discovered that the prevalence of dippers was slightly higher in men than women (33% and 32%, respectively); the dippers were younger (57.4 vs. 60.1 years), had a lower BMI (30 vs. 31), consumed considerably more alcohol (6.1 vs. 2.5 grams/day), were less likely to be diabetic (16 vs. 22%) and to have CVD (6.4 to 11%) [35].

Barksdale and colleagues investigated the effects of stress on BP dipping in thirty non-hypertensive African-American women ages 26 to 51 years [36]. Ambulatory blood pressure monitoring (ABPM), the most common technique used to determine BP dipping status of individuals was performed and collected in the participants' homes.

Ambulatory blood pressure monitoring measures blood pressure variation during the normal, daily routines of the individuals and it's considered more accurate than BP clinical readings. Stress was assessed using a questionnaire that evaluated the participant's psychosocial factors (i.e. perceived stress and daily hassles) and personal factors (i.e. age, income, BMI). Using the same definition of BP dipping as the aforementioned research [16, 32, 35], the study observed that about 30% of the participants were non-dippers.

However, there was no significant association between the psychosocial and personal

factors and BP dipping in this population [36]. Similarly, Sherwood and colleagues investigated the associations between psychosocial and lifestyle factors and BP dipping, including sleep quality, in 56 African-Americans and 72 white Americans [17]. African-Americans were more likely to have a higher awake and sleep SBP and DBP and were less likely to be a dipper. Compared to white participants, African-Americans were also younger, had a higher BMI, a poorer sleep quality, and were at a higher risk of developing diabetes. There are other studies that have confirmed the influence of certain environmental factors on BP dipping [37, 38]; however, relatively little research to date has been directed at identifying genetic factors associated with BP dipping in African-Americans. This MS thesis will examine the influence of *AGT*, *AGTR1*, and *ACE* on BP dipping in African-Americans living in Mississippi who are participants in the Jackson Heart Study.

METHODS

TARGET POPULATION

Mississippi's population is just under 3 million [39]. The state's burden of diseases is among the highest in the nation. Of the total deaths in Mississippi, twenty-eight percent are due to heart disease [40]. According to a Vital Statistics report by the National Center for Health Statistics, hypertension in Mississippi is currently the highest in the nation at about 34 percent, which is approximately 700,000 individuals [40]; and African-Americans, who comprise 37% of the population in Mississippi, also carry the highest burden of disease in the state [39].

RECRUITMENT

The Jackson Heart Study is a community-based cohort study of African-Americans residing in Jackson, Mississippi [41]. The original study recruited 5,302 African-American men and women between the ages of 21 and 92 [42]. The JHS was a derivative of the Atherosclerosis Risk in Communities study (ARIC). The JHS was specifically designed to illuminate risk factors for CVD morbidity and mortality among African-Americans residing in Mississippi [43]. Eligible African-Americans from ARIC and the nearby community were recruited by random selection [41-43]. They were recruited from the three counties surrounding Jackson, Mississippi: Hinds, Madison, and Rankin. To determine the BP dipping status of this population, all the JHS participants were encouraged to participate in 24-hr ambulatory BP monitoring (ABPM), but data

collection was complete for only 1,150 participants [35]. The ABPM method eliminates the “white-coat effect”, which is the natural anxiety that occurs in patients when they are seen by healthcare providers in a clinical setting, by performing the readings in the comfort of the homes of each participant [42]. In this study, the JHS participants were taught to use the ABPM device by trained technicians to self-measure their daily BP every 20 minutes during the sleep (6:01 AM to 5:59 PM) and awake (6 AM to 6 PM) hours.

STATISTICAL ANALYSES

Statistical power for the analyses was based on the available data for 1,150 participants with APBM in the JHS. The power calculations assumed an additive genetic model with the minor allele frequency varying from 0.05 to 0.45, an alpha of 0.05, 0.005 and 0.0005 and 80% power. This thesis chose the additive genetic model over the recessive or dominant models because it’s working under the assumption that SNPs are neither recessive nor dominant. In addition, the sample size of this study was considerably small. So in an effort to capture all possible associations, individuals who were heterozygous and homozygous for particular SNP alleles were included in the study. This study assumed that each SNP allele contributed differently and that their effect on the outcome was in a dosage-type pattern as they increased from 0, 1, and 2.

The Jackson Heart Study provided data for 1,150 participants who underwent ABPM. However, only 930 participants met the criteria, which were a minimum of 54 of

72 total BP readings, at least one BP reading per hour, and complete diary information of the awake and sleep of hours [35]. Of the 930 participants, two participants were excluded due to extreme BP readings, leaving a starting sample of 928 participants. There were 266 dipper participants with a 10% or more change in blood pressure and 662 non-dippers having less than 10% but greater than 0% change in blood pressure (Fig. 2).

Not all of the participants labeled dipper and non-dippers had their genetic data available. A total of 724 had complete genotyped SNPs. In the linear regression model, 645 to 700 participants were analyzed, while in the logistic regression model, between 645 and 700 participants were observed in the model (Fig. 2).

With an initial sample size of 266 dippers and 662 non-dippers, an odds ratio of 1.4 for a minor allele of 0.35 could be considered statistically significant. As the minor allele frequency (MAF) decreased, the detectable odds ratio increased (for MAF=0.25, the detectable OR=1.5; for MAF=0.15, the detectable OR = 1.6). Power curves for logistic regression were generated taking multiple comparisons into account by varying the alpha from 0.05, 0.005 and 0.0005 (assuming 1, 10 and 100 independent tests, respectively). At a conservative alpha of 0.0005 there was 80% power to detect an odds ratio of 1.5 with a minor allele frequency of 0.35. However, the power was greatly reduced for SNPs with lower minor allele frequencies (MAF<0.15) (Fig. 3a-3c). The power curves for the difference in nighttime mean systolic BP compared to daytime mean systolic BP were also adequately powered. An effect size of 0.04 and a MAF of 0.05 could be detected at 80% power (Fig. 4).

Blood pressure dipping was calculated as a percent difference: mean daytime BP minus the mean nighttime BP divided by the mean nighttime BP times 100. In most studies blood pressure dipping was defined as the difference of at least 10% [35, 44]. However, in the Jackson Heart Study the participants' BP dipping status was divided into four categories: dippers, non-dippers, ex-dippers, and risers. The dippers were defined as having $\geq 20\%$ BP dip; ex-dippers were greater than 10% but less than 20%; non-dippers were greater than 0% but less than 10%; and risers were $\leq 0\%$. For the purposes of this study, the ex-dippers and the risers were grouped along with the non-dippers, making the sample size of non-dippers considerably larger than that of the dippers.

Affymetrix 6.0, a genome-wide human SNP array, was used to determine the SNPs in the *AGT*, *ACE*, and *AGTR1* genes. A total of 112 potential SNPs were identified in the array. Imputed data, a list of polymorphisms in high linkage disequilibrium or linked together in the same gene, were not included in the data analysis because the Jackson Heart Study did not have imputed SNP data readily available. Of the 112 SNPs, only 40 SNPs were identified in JHS population. This list was further pruned to a shorter, non-redundant list of linkage disequilibrium-tagging SNPs ($R^2 < 0.80$) using HaploView, a tool used to analyze and visually interpret haplotypes [45] (Fig. 5a-5c). Linkage-disequilibrium-tagging SNPs are SNPs that have a high correlation ($R^2 < 0.80$) with other SNPs in the same gene. As a result, LD-tagging allows for only one of the SNPs to be represented in the data decreasing multiple comparisons testing and increasing the strength of the p-value in the data. Three of the 40 SNPs were dropped due to their high LD. Thus, an initial 37 SNPs were tested for association with BP dipping (Table 4a-4c).

In addition, the SNP annotation proxy (SNAP) online database was used to find any non-synonymous, functional proxy SNPs with an MAF of 0.05 in LD with the typed SNPs in the JHS population. SNAP uses published SNP data from the International HapMap Project, which is a consortium of published scientific information of genes and SNPs. SNAP has a search engine in which the desirable SNPs in the *AGT*, *AGTRI*, and *ACE* genes to identify proxy SNPs [46]. Since this study was a candidate gene study with a small sample size, the GWAS Catalog was used to determine if there were any genome-wide association studies available out there that had investigated the *AGT*, *AGTRI*, or the *ACE* genes [47]. The GWAS catalog, which contains studies that use an agnostic approach in choosing SNPs to study versus a candidate gene study, which targets specific genes and SNPs based on prior hypotheses, is an online resource of genome-wide association studies available to the public. The studies are found by simply going to web server and typing the gene, SNP, chromosomal region, type of disease, or title of journal article and searching through their database. No studies were found in the GWAS catalog.

Finally, the LocusZoom examined the relationship between the locus, LD, and p-values of SNPs in reference to each other. Lastly, HapMap uses several populations to study polymorphism. Because this analysis focuses on African-Americans, the HapMap black population, the Yoruba of Ibadan (YRI), was used in HaploView, SNAP, and LocusZoom to collect genetic data.

All the descriptive statistics were obtained using SAS 9.3. Blood pressure dipping was treated both as a continuous and dichotomous variables (linear and logistic regression, respectively). Binary variables (dipper vs. non-dipper) were used in the

logistic regression analyses and the difference between daytime SBP and nighttime SBP was used in the linear model. The models were additive, which tested the association of the SNP on BP dipping based on a dosage-type pattern ranging from the presence of 0, 1, or 2 effect alleles. All the models were formulated on the likelihood that if the odds ratio is greater than one, then the individual is a non-dipper.

Simple (unadjusted) and multivariable (adjusted) models were tested to determine the relationship between the covariates and the outcome. The simple comparison models included only the SNP, while the multivariable models included the SNPs, the statistically significant covariates and ten principal components (PCs). The principal components were previously calculated and provided by the Jackson Heart Study. The PCs are used to decrease correlation between certain variables, such as those pertaining to ancestry and population substructure in African-Americans, which are caused by geographical isolation, genetic drift, or natural selection [48]. Principal components were used in this data to adjust the data and to eliminate any potential confounding due to the genetic variability within different African-American populations.

The other covariates and potential confounders associated with BP dipping were age, gender, hypertension, hypertension medication, smoking, alcohol consumption, and education. Smoking was categorized as non-smoker, previous smoker, and current smoker. Alcohol consumption was given as the percent of drinkers. Education was classified as less than high school (<12yrs), high school graduate (=12yrs) and some college or college graduate (>12yrs). T-tests were used to compare dippers and non-dippers for the continuous variables, while the chi-square (χ^2) was used to compare groups for the categorical variables. The values of the t-tests were expressed in mean $\pm$

SD, while the chi-square tests were in percentages (%). A p-value of 0.05 was the cut-off for statistical significance for all the tests.

The linear and logistic models were adjusted for the covariates that were statistically significant in this population or for covariates, such as age and gender, which were chosen *a priori*. The logistic regression model was adjusted for age, gender, BMI, alcohol consumption, hypertension medication status, and ten principal components. The linear regression was also adjusted for the same risk factors, including mean systolic blood pressure.

RESULTS

Blood pressure data was collected from 1,150 JHS participants. However, 220 participants did not have their BP dipping status available due to missing data. Two other participants, who were extreme outliers, were excluded from the sample, making 928 participants the total starting sample size.

The total female dippers and non-dippers were 181 vs. 460. The total male dippers and non-dippers were 85 vs. 202. Gender was not statistically significant ($p=0.67$). However, the rest of the data was reported based on the percent of females because they were the majority. Thus, seventy-three percent of female dippers were 65 yrs or younger compared to non-dippers at 65% ($p=0.004$). The dippers tended to have a lower mean daytime SBP (dipper: 129.9 ± 13.5 vs. non-dipper: 130.4 ± 13.8 ; $p=0.59$) and mean nighttime SBP (dipper: 120.2 ± 13.1 vs. non-dipper: 127.40 ± 15.2 ; $p<0.0001$).

Non-dippers were more likely to have hypertension (165, 63% vs. 465, 71%; $p=0.02$), have higher a mean BMI (31.40 ± 6.55 vs. 30.40 ± 6.14 ; $p=0.04$), be on hypertension medication (138, 57% vs. 403, 64%; $p=0.04$), and have higher levels of aldosterone ($5.65, \pm 4.7$ vs. 6.2 ± 6.0 ; $p=0.15$) and renin (5.17 ± 34.20 vs. 5.65 ± 23.70 ; $p=0.81$). Non-dippers were also more likely to be non-smokers (173, 65 % vs. 452, 68%; $p=0.42$), and to consume less alcohol (139, 52% vs. 265 40%; $p=0.0007$), and to have less college education (dipper: 181, 68% vs. 412, 62%; $p=0.21$) than dippers (Table 1).

The population displayed a normal distribution of daytime, systolic blood pressures nighttime systolic blood pressure, and the difference between the two (Fig.6a-6c). Because the difference between nighttime and daytime blood pressure was normally distributed, linear regression could be used to model the effect of each SNP on (daytime-nighttime blood pressure). In the linear regression model, the *AGTR1* rs12721234 G allele demonstrated the most negative association ($\beta = -1.52 \pm 1.10$; 95% CI; -3.68-0.64,) between the difference in BP dipping; *AGTR1* rs12721267 G allele demonstrated the most positive association ($\beta = 0.90 \pm 1.12$; 95% CI; -1.29-3.10) (Table 3a-3f). However, the p-values were not statistically significant ($p=0.17$ and $p=0.10$, respectively). Moreover, because none of the results for any of the SNPs were significant, no associations between the SNPs and BP dipping could be established. In fact, none of the SNPs showed any association to BP dipping in either the unadjusted or adjusted linear regression models.

In the logistic regression model, the unadjusted results indicated that the *AGTR1* rs12695891 A allele, observed in 569 participants, had an inverse odd ratios (0.70, 95% CI; 0.40- 1.23) with a p-value of 0.22. In this model, having an odds ratio less than 1.0 means that participants with the *AGTR1* rs12695891 A allele are less likely to be non-dippers than those without the allele. Similar to the linear regression models, none of the SNPs were statistically significantly associated with BP dipping in either the unadjusted and adjusted logistic regression models (Table 2a-2f).

According to SNAP, in the *AGT* gene, nonsynonymous SNP rs4762 (MAF=0.178) was in weak LD ($R^2=0.3$ in YRI) with SNP rs11122577, genotyped in this study. The weak link between these two SNPs suggested that there is a small possibility that neither

of these SNP affect the BP dipping. In the *ACE* gene, nonsynonymous SNP rs4303 (MAF=0.069) was in perfect LD ($R^2=1.0$) with rs4297 (genotyped in this study). The SNP rs4303 and was also in strong LD ($R^2=0.8$) with rs1270926 (genotyped in this study), so SNP rs4303 s most probably has no association with BP dipping.

Nonsynonymous SNP rs4364 (MAF=0.077) was in weak LD ($R^2=0.125$) with rs4329 (genotyped in this study), so the affect of rs4364 on the outcome is inconclusive. There were no nonsynonymous functional SNPs found in the *AGTR1* genes, so SNAP was not utilized to find any proxy SNPs. In addition, the LocusZoom regional plots did not offer any addition insight to the association between SNPs and the outcome (Fig. 7a-7c).

DISCUSSION

To our knowledge, this study is the first to examine genes in the renin-angiotensin-aldosterone system and the association of genetic polymorphisms in BP dipping in African-Americans. None of the SNPs tested was significantly associated with BP dipping in this population of African-Americans. Although aldosterone ($p=0.15$) and renin ($p=0.81$) play a vital role in RAAS regulation in blood pressure, neither of them, was associated with BP dipping in our data (Table 1).

This population was well educated with more than two thirds of dippers and non-dippers having at least some college education. Both groups were overwhelmingly non-smokers or previous smokers. However, gender, smoking status and education were not significantly associated with BP dipping. Nonetheless, as prior studies have noted, other traditional risk factors for hypertension were significantly associated with BP dipping (age, BMI, and alcohol). Individuals who were dippers tended to be younger, have a lower BMI, and consume more alcohol.

This study was able to survey several SNPs across the three candidate genes, allowing for the identification of several types of SNPs (non-synonymous, functional, intronic, or near 5' of the gene) (Table 4). Consistent with the literature, this study found that blood pressure dipping is highly correlated with both SBP and DBP. However, blood pressure dipping was only correlated with nighttime SBP and DBP, while daytime blood pressures had no association with the outcome.

The variance inflation factors for all adjusted models indicated that multicollinearity among the covariates was not a problem. Furthermore, using the ABPM technique to collect multiple BP readings of daytime and nighttime systolic and diastolic BP allowed for accuracy in the determination of BP dipping status in this population. Additionally, having principal components available to adjust for in the data may have decreased potential confounders associated with ancestry and admixture within this African-American population.

This study did not correct for multiple SNPs tested. Since no association was found between SNPs and BP dipping in this population, methods to correct for multiple comparisons like Bonferroni correction were not necessary. However, even if the results had shown any associations between the predictors and the outcome, correcting for multiple corrections may have still been inappropriate. Though multiple corrections may prevent Type 1 errors, given the high *a priori* evidence for these genes and their dependence among SNPs other more lenient methods would have had to be explored.

Although none of the SNPs displayed any significant associations with BP dipping there are some limitations to this study that should be taken into consideration. There was sufficient power to detect an effect size but the sample size was small. From the small sample size available, there were participants that had missing data, decreasing the sample sometimes to half its size. Also, not all the possible SNPs were analyzed because the imputed data were not available for this analysis. In addition, most of the SNPs analyzed were not functional.

This study was also limited in the ability to generalize its results. As previously stated, African-Americans are particularly affected by hypertension compared to whites or Hispanics. Although, only white Americans and different SNPs were examined, the most similar study to this investigation, the Wisconsin Sleep Study, evaluated normotensive participants to determine SNP involvement with BP dipping [16]. However, the majority of this population, whether they were labeled dipper or non-dipper, were hypertensive or on antihypertensive medication. Observing a population of African-Americans with neither hypertension nor taking hypertension medication might a better way of measuring the genetic effects on BP dipping. In any case, the results of this thesis could mainly benefit African-Americans living in other communities similar to the health profile of Jackson, Mississippi or populations who have or are at great risk of hypertension.

CONCLUSION

To our knowledge, this is the first analysis done on the effect of SNPs in the *AGT*, *AGTR1*, and *ACE* genes in a population of only African-Americans. Although no associations were captured in this analysis, one cannot rule out the possibility of other factors, beyond this study, interacting with genes and BP dipping. Only common variants were analyzed in this study. In future studies, rare variants should also be included because it allows for a better survey of genetic variants across the gene. In addition, gene-environment interactions and other types of genetic variations, such as deletions, insertions, and copy number variants should be taken into consideration. These variants might also affect blood pressure dipping. Moreover, a larger sample size, of perhaps different races and ethnicities should be included in further studies to increase the ability to generalize results across different populations. In conclusion, hypertension is a major health problem in African-American communities. Further investigation in this population will be needed to determine if genetic factors are involved in the lack of BP dipping in African-Americans.

Figure 1. The Renin- Angiotensin-Aldosterone System

Figure 2. Breakdown of sample size in the Jackson Heart Study.

Figure 3a. Power to Detect Effect Size in logistic regression analysis: : 1 tests, $\alpha=0.05$

Figure 3b. Power to Detect Effect Size in logistic regression analysis: : 10 tests, $\alpha=0.005$

Figure 3c. Power to Detect Effect Size in logistic regression analysis: 100 tests, $\alpha=0.0005$

Figure 4. Power: Difference in daytime-nighttime mean systolic Blood Pressure

Figure 5a. Haplotype Map displaying the varying linkage-disequilibrium of SNPs in *AGT* gene.

Figure 5b. Haplotype Map displaying the varying linkage-disequilibrium of SNPs in *AGTR1* gene.

Figure 5c. Haplotype Map displaying the varying linkage-disequilibrium of SNPs in *ACE* gene.

Figure 6a. Distribution of mean daytime systolic blood pressure.

Figure 6b. Distributions of mean nighttime systolic blood pressure.

Figure 6c. Distribution of difference in mean daytime-nighttime systolic blood pressure.

Figure 7a. The association between linkage disequilibrium, recombination rate, and statistical significance of SNPs in *AGT*.

Figure 7b. The association between linkage disequilibrium, recombination rate, and statistical significance of SNPs in *AGTR1*.

Figure 7c. The association between linkage disequilibrium, recombination rate, and statistical significance of SNPs in *ACE*.

Table 1. Characteristics of blood pressure dippers and nondippers in the Jackson Heart Study (N=930)

	Dipper (N=266)	Nondipper (N=662)	P-value ^a
Age			
1 (21-44)	41 (15%)	70 (11%)	0.004
2 (45-54)	66 (25 %)	134 (20%)	
3 (55-64)	87 (33%)	226 (34%)	
4(65-74)	65 (24%)	176 (27%)	
5 (75-84)	7 (3%)	56 (8%)	
Gender (% female)	181 (68%)	460 (69%)	0.67
Aldosterone (mean, SD)	5.65 (4.7)	6.2 (6.0)	0.15
Renin (mean, SD)	5.17 (34.2)	5.65 (23.7)	0.81
Mean daytime SBP (mean, SD)	129.9 (13.5)	130.4 (13.8)	0.59
Mean nighttime SBP (mean, SD)	120.2 (13.1)	127.2 (15.2)	<.0001
Mean daytime DBP (mean, SD)	79.9 (9.2)	79.0 (9.4)	0.17
Mean nighttime DBP (mean, SD)	70.1 (8.9)	73.1 (9.7)	<.0001
BMI (mean, SD)	30.4 (6.1)	31.4 (6.6)	0.04
Hypertension (%)	165 (63%)	465 (71%)	0.02
Hypertension medication (%)	138 (57%)	403 (64%)	0.04
Smoking Status (%)			0.42
Non smoker	173 (65%)	450 (68%)	
Previous smoker	61 (23%)	150 (23%)	
Current smoker	32 (12%)	61 (9%)	
Alcohol consumption (% drinkers)	139 (52%)	265 (40%)	0.0007
Education (%)			0.21
Less than high school (<12yrs)	47 (18%)	141 (21%)	
High school graduate (=12yrs)	38 (14%)	111 (17%)	
Some college or college graduate(>12yrs)	181 (68%)	410 (62%)	

^aFor categorical variables, a Chi-square test was used; for continuous, a t-test was used.

Table 2-a. Associations of *AGT* polymorphisms with odds of blood pressure dipping.

SNP	Effect Allele	N	Odds ratio	95% CI		P-value
rs1926723	C	593	0.88	0.40	1.90	0.74
rs2493129	A	593	0.98	0.60	1.63	0.93
rs11122576	C	593	1.15	0.57	2.34	0.70
rs11568026	G	593	1.20	0.82	1.75	0.35
rs11122577	A	592	1.36	0.90	2.04	0.15
rs11122578	A	593	1.15	0.57	2.34	0.70

Table 2-b. Associations of *ACE* polymorphisms with odds of blood pressure dipping.

SNP	Effect Allele	N	Odds ratio	95% CI		P-value
rs4297	C	591	1.00	0.64	1.57	0.99
rs12709426	G	588	1.07	0.63	1.79	0.81
rs4324	G	590	0.97	0.75	1.25	0.80
rs4325	A	593	0.87	0.61	1.24	0.44
rs4329	G	593	0.97	0.75	1.25	0.81
rs4351	G	586	1.01	0.79	1.31	0.92

Table 2-c. Associations of *AGTR1* polymorphisms with odds of blood pressure dipping.

SNP	Effect Allele	N	Odds ratio	95% CI		P-value
rs275651	A	579	1.10	0.82	1.48	0.52
rs275652	G	592	1.18	0.87	1.60	0.28
rs12721267	G	593	0.86	0.48	1.54	0.61
rs2933249	A	593	0.87	0.50	1.50	0.60
rs12721286	T	591	1.26	0.74	2.12	0.40
rs12721221	C	585	0.97	0.71	1.33	0.84
rs2131127	G	592	1.00	0.76	1.31	0.99
rs12695877	T	592	0.98	0.75	1.27	0.87
rs4681440	C	593	1.06	0.80	1.41	0.68
rs12721255	T	593	0.97	0.72	1.31	0.85
rs4681443	G	592	1.04	0.80	1.34	0.78
rs4681444	A	593	1.12	0.84	1.49	0.44
rs1492103	G	593	0.92	0.69	1.22	0.55
rs1492100	T	592	0.97	0.74	1.26	0.80
rs3772616	C	592	1.03	0.80	1.33	0.80
rs4524238	G	592	0.93	0.70	1.23	0.60
rs12695891	A	586	0.77	0.45	1.31	0.33
rs12721328	G	593	0.88	0.50	1.57	0.68
rs1826361	C	593	0.86	0.66	1.12	0.26
rs12695902	G	593	1.58	0.95	2.62	0.08
rs12721234	T	583	1.06	0.55	2.06	0.87
rs12695918	C	593	0.97	0.68	1.38	0.85
rs1800766	C	593	0.80	0.54	1.18	0.26
rs12695923	C	592	1.00	0.75	1.33	1.00

Table 2-d. Adjusted associations of *AGT* polymorphisms with odds of blood pressure dipping.

SNP	Effect Allele	N	Odds ratio	95% CI		P-value ^a
rs1926723	C	575	1.03	0.46	2.34	0.93
rs2493129	A	575	1.00	0.60	1.70	1.00
rs11122576	C	575	1.15	0.54	2.47	0.71
rs11568026	G	575	1.18	0.80	1.75	0.41
rs11122577	A	574	1.48	0.95	2.30	0.08
rs11122578	A	575	1.15	0.54	2.47	0.71

^aAdjusted for age, gender, BMI, alcohol consumption, hypertension, hypertension medication and principal components.

Table 2-e. Adjusted associations of *ACE* polymorphisms with odds of blood pressure dipping.

SNP	Effect Allele	N	Odds ratio	95% CI		P-value ^a
rs4297	C	573	0.97	0.62	1.59	0.96
rs12709426	G	572	1.01	0.58	1.76	0.96
rs4324	G	572	0.93	0.71	1.21	0.57
rs4325	A	575	0.79	0.53	1.17	0.24
rs4329	G	575	0.93	0.71	1.03	0.59
rs4351	G	568	1.07	0.82	1.40	0.63

^aAdjusted for age, gender, BMI, alcohol consumption, hypertension, hypertension medication and principal components.

Table 2-f. Adjusted associations of *AGTR1* polymorphisms with odds of blood pressure dipping.

SNP	Effect Allele	N	Odds ratio	95% CI		P-value
rs275651	A	562	1.12	0.82	1.53	0.48
rs275652	G	574	1.17	0.85	1.61	0.33
rs12721267	G	575	0.86	0.47	1.57	0.62
rs2933249	A	575	0.87	0.48	1.60	0.65
rs12721286	T	573	1.24	0.72	2.13	0.44
rs12721221	C	567	0.92	0.66	1.32	0.68
rs2131127	G	574	1.03	0.77	1.38	0.85
rs12695877	T	575	1.02	0.76	1.36	0.90
rs4681440	C	575	1.16	0.85	1.58	0.36
rs12721255	T	575	0.96	0.67	1.36	0.80
rs4681443	G	574	1.11	0.83	1.46	0.48
rs4681444	A	575	1.53	0.90	1.72	0.18
rs1492103	G	575	0.94	0.66	1.33	0.71
rs1492100	T	574	1.00	0.75	1.33	0.98
rs3772616	C	574	1.20	0.85	1.48	0.42
rs4524238	G	574	0.99	0.72	1.36	0.94
rs12695891	A	569	0.70	0.400	1.23	0.22
rs12721328	G	575	0.82	0.45	1.48	0.51
rs1826361	C	575	0.88	0.67	1.16	0.36
rs12695902	G	575	1.61	0.92	2.80	0.09
rs12721234	T	565	1.25	0.61	2.56	0.54
rs12695918	C	575	1.03	0.71	1.49	0.88
rs1800766	C	575	0.85	0.56	1.29	0.44
rs12695923	C	574	0.96	0.71	1.30	0.80

^aAdjusted for age, gender, BMI, alcohol consumption, hypertension, hypertension medication and principal components.

Table 3-a. Unadjusted associations of *AGT* polymorphisms with difference in blood pressure (mean daytime - mean nighttime blood pressure).

SNP	Effect Allele	N	Beta (β)	Standard error	95% CI		P-value
rs1926723	C	703	0.64	1.17	-1.65	2.93	0.58
rs2493129	A	703	1.06	0.85	-0.61	2.74	0.21
rs11122576	C	703	0.64	1.08	-1.47	2.76	0.60
rs11568026	G	702	-0.54	0.61	-1.74	0.66	0.38
rs11122577	A	702	-0.40	0.66	-1.70	0.89	0.54
rs11122578	A	703	0.64	1.07	-1.47	2.76	0.55

Table 3-b. Unadjusted associations of *ACE* polymorphisms with difference in blood pressure (mean daytime - mean nighttime blood pressure).

SNP	Effect Allele	N	Beta (β)	Standard error	95% CI		P-value
rs4297	C	701	-1.14	0.76	-2.62	0.35	0.14
rs12709426	G	697	-0.47	0.89	-2.22	1.26	0.59
rs4329	G	703	-0.17	0.43	-1.02	0.68	0.69
rs4324	G	700	-0.14	0.44	-1.01	0.70	0.72
rs4325	A	703	-0.21	0.62	-1.43	1.01	0.73
rs4351	G	696	0.21	0.43	-0.64	1.05	0.63

Table 3-c. Unadjusted associations of *AGTRI* polymorphisms with difference in blood pressure (mean daytime - mean nighttime blood pressure).

SNP	Effect Allele	N	Beta (β)	Standard error	95% CI		P-value
rs275651	A	688	-0.66	0.49	-1.62	0.30	0.18
rs275652	G	702	-0.77	0.50	-1.74	0.20	0.12
rs12721267	G	703	0.82	1.04	-1.24	2.87	0.43
rs2933249	A	703	0.25	0.95	-1.61	2.12	0.79
rs12721286	T	700	0.76	0.84	-0.89	2.41	0.37
rs12721221	C	695	-0.45	0.53	-1.50	0.60	0.40
rs2131127	G	702	-0.51	0.46	-1.41	0.39	0.27
rs12695877	T	702	-0.47	0.45	-1.36	0.41	0.29
rs4681440	C	703	-0.42	0.47	-1.34	0.51	0.38
rs12721255	T	703	0.06	0.51	-1.06	0.94	0.91
rs4681443	G	702	-0.63	0.44	-1.5	0.24	0.16
rs4681444	A	703	-0.44	0.47	-1.37	0.48	0.35
rs1492103	G	703	0.21	0.52	-0.80	1.22	0.69
rs1492100	T	702	-0.25	0.46	-1.16	0.64	0.57
rs3772616	C	702	0.11	0.44	-0.97	0.74	0.78
rs4524238	G	702	0.09	0.49	-0.87	1.04	0.86
rs12695891	A	696	-0.79	0.93	-2.62	1.04	0.40
rs12721328	G	703	0.13	1.02	-1.87	2.14	0.90
rs1826361	C	703	-0.71	0.43	-1.56	0.14	0.10
rs12695902	G	703	0.30	0.80	-1.27	1.87	0.70
rs12721234	T	691	-1.17	1.04	-3.20	0.87	0.26
rs12695918	C	703	0.21	0.60	-0.97	1.38	0.73
rs1800766	C	703	0.02	0.70	-1.38	1.34	0.98
rs12695923	C	702	-0.37	0.47	-1.29	0.55	0.43

Table 3-d. Adjusted model on the associations of *AGT* polymorphisms with difference in blood pressure (mean daytime - mean nighttime blood pressure).

SNP	Effect Allele	N	Beta (β)	Standard error	95% CI		P-value ^a
rs1926723	C	645	-0.36	1.14	-2.60	1.80	0.75
rs2493129	A	645	0.89	0.89	-0.85	2.65	0.32
rs11122576	C	645	0.18	1.16	-2.09	2.45	0.89
rs11568026	G	645	-0.56	0.65	-1.84	0.72	0.39
rs11122577	A	644	-0.05	0.72	-1.45	1.36	0.95
rs11122578	A	645	-0.18	1.16	-2.09	2.45	0.63

^aAdjusted for age, gender, BMI, alcohol consumption, mean systolic blood pressure, hypertension, hypertension medication and principal components.

Table 3-e. Adjusted model on the associations of *ACE* polymorphisms with difference in blood pressure (mean daytime - mean nighttime blood pressure).

SNP	Effect Allele	N	Beta (β)	Standard error	95% CI		P-value ^a
rs4297	C	644	-0.80	0.81	-2.39	0.79	0.32
rs12709426	G	645	-0.17	0.95	-2.02	1.69	0.86
rs4329	G	644	.015	0.46	-1.05	0.75	0.75
rs4324	G	643	-0.14	0.46	-1.05	0.76	0.75
rs4325	A	644	-0.30	0.67	-1.62	1.03	0.66
rs4351	G	638	0.04	0.46	-0.85	0.94	0.92

^aAdjusted for age, gender, BMI, alcohol consumption, mean systolic blood pressure, hypertension, hypertension medication and principal components.

Table 3-f. Adjusted model on the associations of *AGTR1* polymorphisms with difference in blood pressure (mean daytime - mean nighttime blood pressure).

SNP	Effect Allele	N	Beta (β)	Standard error	95% CI		P-value ^a
rs275651	A	631	-0.76	0.52	-1.78	0.26	0.14
rs275652	G	645	-0.86	0.52	-1.88	0.17	0.10
rs12721267	G	645	0.90	1.12	-1.29	3.10	0.42
rs2933249	A	645	0.44	1.04	-1.60	2.48	0.67
rs12721286	T	642	0.68	0.87	-1.04	2.29	0.44
rs12721221	C	638	-0.71	0.60	-1.89	0.48	0.24
rs2131127	G	644	-0.54	0.50	-1.51	0.43	0.28
rs12695877	T	644	-0.64	0.49	-1.61	0.32	0.19
rs4681440	C	645	0.27	0.53	-1.31	0.77	0.61
rs12721255	T	645	-0.41	0.60	-1.61	0.78	0.50
rs4681443	G	644	0.80	0.48	-1.75	0.14	0.10
rs4681444	A	645	-0.45	0.53	-1.49	0.59	0.40
rs1492103	G	645	-0.30	0.61	-1.50	0.92	0.63
rs1492100	T	644	-0.30	0.49	-1.26	0.66	0.54
rs3772616	C	644	-0.27	0.47	-1.20	0.66	0.57
rs4524238	G	644	-0.03	0.54	-1.09	1.03	0.96
rs12695891	A	639	0.90	0.98	-2.82	1.03	0.36
rs12721328	G	645	0.86	1.14	-1.38	3.10	0.45
rs1826361	C	645	-0.94	0.46	-1.85	-0.03	0.04
rs12695902	G	645	0.30	0.87	0.73	-1.42	2.02
rs12721234	T	633	-1.52	1.10	-3.68	0.64	0.17
rs12695918	C	645	-0.02	0.64	1.27	1.24	0.98
rs1800766	C	645	0.32	0.75	-1.16	1.79	0.67
rs12695923	C	644	-0.44	0.50	-1.43	0.55	0.70

^aAdjusted for age, gender, BMI, alcohol consumption, mean systolic blood pressure, hypertension, hypertension medication and principal components.

Table 4-a. Linkage disequilibrium-tagging SNPs in *AGT*.

SNP	Effect Allele	Position	EAF ^a	Function
rs1926723	C	228906719	0.034	Intron
rs2493129	A	228907565	0.073	Intron
rs11122576	C	228913302	0.039	Intron
rs11568026	G	228914146	0.132	Intron
rs11122577	A	228914179	0.118	Intron
rs11122578	A	228914412	0.041	Intron

*EAF = Effect Allele Frequency. Effect allele= minor allele.

Table 4-b. Linkage disequilibrium-tagging SNPs in *ACE*.

SNP	Effect Allele	Position	EAF ^a	Function
rs4297	C	58910286	0.088	Intron
rs12709426	G	58915488	0.063	Missense
rs4324	G	58916903	0.422	Intron
rs4325	A	58916932	0.137	Intron
rs4329	G	58917190	0.423	Intron
rs4351	G	58923464	0.464	Intron

*EAF = Effect Allele Frequency. Effect allele= minor allele.

Table 4-c. Linkage disequilibrium-tagging SNPs in *AGTR1*.

SNP	Effect Allele	Position	EAF ^a	Function
rs409742	G	149895055	0.234	5' near gene
rs275651	A	149897577	0.15	5' near gene
rs275652	G	149897674	0.269	5' near gene
rs12721267	G	149899017	0.246	Intron
rs2933249	A	149899210	0.046	Intron
rs12721286	T	149901605	0.072	Intron
rs12721221	C	149902287	0.187	Intron
rs2131127	G	149906833	0.316	Intron
rs12695877	T	149909724	0.342	Intron
rs4681440	C	149914722	0.238	Intron
rs12721255	T	149914903	0.189	Intron
rs4681443	G	149915159	0.375	Intron
rs4681444	A	149915264	0.224	Intron
rs1492103	G	149915654	0.16	Intron
rs1492100	T	149920117	0.298	Intron
rs3772616	C	149920881	0.328	Intron
rs4524238	G	149922478	0.224	Intron
rs12695891	A	149923336	0.058	Intron
rs12721328	G	149925644	0.046	Intron
rs1826361	C	149927418	0.441	Intron
rs12695902	G	149931108	0.082	Intron
rs12721234	T	149938624	0.048	Intron
rs12695918	C	149939317	0.153	Intron
rs1800766	C	149940332	0.107	Intron
rs12695923	C	149940600	0.268	Intron

^aEAF = Effect Allele Frequency. Effect allele= minor allele.

REFERENCES

1. Mathews, T.J., et al., *Annual summary of vital statistics: 2008*. Pediatrics, 2011. **127**(1): p. 146-57.
2. Jolly, S., et al., *Higher Cardiovascular Disease Prevalence and Mortality among Younger Blacks Compared to Whites*. The American journal of medicine, 2010. **123**(9): p. 811-818.
3. Elliott, W.J., *The economic impact of hypertension*. Journal of clinical hypertension, 2003. **5**(3 Suppl 2): p. 3-13.
4. Miller, G., P. Hughes-Cromwick, and C. Roehrig, *National spending on cardiovascular disease, 1996-2008*. Journal of the American College of Cardiology, 2011. **58**(19): p. 2017-9.
5. Heidenreich, P.A., et al., *Forecasting the Future of Cardiovascular Disease in the United States*. Circulation, 2011.
6. Pleis, J.R., B.W. Ward, and J.W. Lucas, *Summary health statistics for U.S. adults: National Health Interview Survey, 2009*. Vital and health statistics. Series 10, Data from the National Health Survey, 2010(249): p. 1-207.
7. Ong, K.L., et al., *Prevalence, awareness, treatment, and control of hypertension among United States adults 1999-2004*. Hypertension, 2007. **49**(1): p. 69-75.
8. Onusko, E., *Diagnosing secondary hypertension*. American family physician, 2003. **67**(1): p. 67-74.
9. Keenan, N.L. and K.A. Rosendorf, *Prevalence of hypertension and controlled hypertension - United States, 2005-2008*. MMWR. Surveillance summaries : Morbidity and mortality weekly report. Surveillance summaries / CDC, 2011. **60** Suppl: p. 94-7.
10. Schiller, J.S., et al., *Summary health statistics for U.S. adults: National Health Interview Survey, 2010*. Vital and health statistics. Series 10, Data from the National Health Survey, 2012(252): p. 1-207.
11. Sherwood, A., et al., *Nighttime blood pressure dipping: the role of the sympathetic nervous system*. American journal of hypertension, 2002. **15**(2 Pt 1): p. 111-8.
12. Paul, M., A. Poyan Mehr, and R. Kreutz, *Physiology of local renin-angiotensin systems*. Physiological reviews, 2006. **86**(3): p. 747-803.
13. Dickson, M.E. and C.D. Sigmund, *Genetic Basis of Hypertension*. Hypertension, 2006. **48**(1): p. 14-20.
14. Atlas, S.A., *The renin-angiotensin aldosterone system: pathophysiological role and pharmacologic inhibition*. Journal of managed care pharmacy : JMCP, 2007. **13**(8 Suppl B): p. 9-20.
15. Rossier, B.C., *Hypertension finds a new rhythm*. Nature medicine, 2010. **16**(1): p. 27-8.
16. Vardeny, O., et al., *beta2 adrenergic receptor polymorphisms and nocturnal blood pressure dipping status in the Wisconsin Sleep Cohort Study*. Journal of the American Society of Hypertension : JASH, 2011. **5**(2): p. 114-22.

17. Sherwood, A., et al., *Blood pressure dipping: ethnicity, sleep quality, and sympathetic nervous system activity*. American journal of hypertension, 2011. **24**(9): p. 982-8.
18. Rotimi, C., et al., *Polymorphisms of Renin-Angiotensin Genes Among Nigerians, Jamaicans, and African Americans*. Hypertension, 1996. **27**(3): p. 558-563.
19. Goldenberg, I., et al., *Polymorphism in the angiotensinogen gene, hypertension, and ethnic differences in the risk of recurrent coronary events*. Hypertension, 2006. **48**(4): p. 693-9.
20. Giner, V., et al., *Renin-angiotensin system genetic polymorphisms and salt sensitivity in essential hypertension*. Hypertension, 2000. **35**(1 Pt 2): p. 512-7.
21. Jeunemaitre, X., et al., *Genetic polymorphisms of the renin-angiotensin system and angiographic extent and severity of coronary artery disease: the CORGENE study*. Human genetics, 1997. **99**(1): p. 66-73.
22. Goldenberg, I., et al., *Polymorphism in the Angiotensinogen Gene, Hypertension, and Ethnic Differences in the Risk of Recurrent Coronary Events*. Hypertension, 2006. **48**(4): p. 693-699.
23. Kunz, R., et al., *Association Between the Angiotensinogen 235T-Variant and Essential Hypertension in Whites : A Systematic Review and Methodological Appraisal*. Hypertension, 1997. **30**(6): p. 1331-1337.
24. Martinez-Rodriguez, N., et al., *Association of angiotensin II type 1-receptor gene polymorphisms with the risk of developing hypertension in Mexican individuals*. Journal of the renin-angiotensin-aldosterone system : JRAAS, 2011.
25. Niu, W. and Y. Qi, *Association of the angiotensin II type I receptor gene +1166 A>C polymorphism with hypertension risk: evidence from a meta-analysis of 16474 subjects*. Hypertension research : official journal of the Japanese Society of Hypertension, 2010. **33**(11): p. 1137-43.
26. O'Donnell, C.J., et al., *Evidence for association and genetic linkage of the angiotensin-converting enzyme locus with hypertension and blood pressure in men but not women in the Framingham Heart Study*. Circulation, 1998. **97**(18): p. 1766-72.
27. Cooper, R., et al., *The prevalence of hypertension in seven populations of west African origin*. American journal of public health, 1997. **87**(2): p. 160-8.
28. Adeyemo, A., et al., *A genome-wide association study of hypertension and blood pressure in African Americans*. PLoS genetics, 2009. **5**(7): p. e1000564.
29. Profant, J. and J.E. Dimsdale, *Race and diurnal blood pressure patterns. A review and meta-analysis*. Hypertension, 1999. **33**(5): p. 1099-104.
30. Wang, X., et al., *Genetic influences on heart rate variability at rest and during stress*. Psychophysiology, 2009. **46**(3): p. 458-65.
31. Bristow, M.R., *β -Adrenergic Receptor Blockade in Chronic Heart Failure*. Circulation, 2000. **101**(5): p. 558-569.
32. Julve, R., et al., *Polymorphism insertion/deletion of the ACE gene and ambulatory blood pressure circadian variability in essential hypertension*. Blood pressure monitoring, 2001. **6**(1): p. 27-32.

33. Jian, M., et al., *Polymorphism of angiotensin I converting enzyme gene in the older Chinese: linked to ambulatory blood pressure levels and circadian blood pressure rhythm*. International Journal of Cardiology, 1996. **55**(1): p. 33-40.
34. Kario, K., et al., *Angiotensinogen and angiotensin-converting enzyme genotypes, and day and night blood pressures in elderly Japanese hypertensives*. Hypertension research : official journal of the Japanese Society of Hypertension, 1999. **22**(2): p. 95-103.
35. Hickson, D.A., et al., *Socioeconomic position is positively associated with blood pressure dipping among African-American adults: the Jackson Heart Study*. American journal of hypertension, 2011. **24**(9): p. 1015-21.
36. Barksdale, D.J., C. Woods-Giscombé, and J.G. Logan, *Stress, Cortisol, and Nighttime Blood Pressure Dipping in Nonhypertensive Black American Women*. Biological Research For Nursing, 2012.
37. Tomfohr, L., et al., *Everyday discrimination and nocturnal blood pressure dipping in black and white americans*. Psychosomatic medicine, 2010. **72**(3): p. 266-72.
38. Mellman, T.A., et al., *Posttraumatic stress disorder and nocturnal blood pressure dipping in young adult African Americans*. Psychosomatic medicine, 2009. **71**(6): p. 627-30.
39. Bureau, U.S.C., *The American Community- Blacks: 2004*. 2007.
40. *The 2005 Mississippi State of the Heart Report*, M.D.o. Health, O.o.P.H.a.O.o.H.S.D.U.a.t.A. Heart, and Association, Editors. 2005.
41. Wyatt, S.B., et al., *Prevalence, awareness, treatment, and control of hypertension in the Jackson Heart Study*. Hypertension, 2008. **51**(3): p. 650-6.
42. Fuqua, S.R., et al., *Recruiting African-American research participation in the Jackson Heart Study: methods, response rates, and sample description*. Ethnicity & disease, 2005. **15**(4 Suppl 6): p. S6-18-29.
43. Hickson, D.A., et al., *Geographic Representation of the Jackson Heart Study Cohort to the African-American Population in Jackson, Mississippi*. American journal of epidemiology, 2010.
44. Sachdeva, A. and A.B. Weder, *Nocturnal sodium excretion, blood pressure dipping, and sodium sensitivity*. Hypertension, 2006. **48**(4): p. 527-33.
45. Barrett, J.C., et al., *Haploview: analysis and visualization of LD and haplotype maps*. Bioinformatics, 2005. **21**(2): p. 263-265.
46. Johnson, A.D., et al., *SNAP: a web-based tool for identification and annotation of proxy SNPs using HapMap*. Bioinformatics, 2008. **24**(24): p. 2938-2939.
47. Hindorff, L.A., et al., *Potential etiologic and functional implications of genome-wide association loci for human diseases and traits*. Proceedings of the National Academy of Sciences, 2009. **106**(23): p. 9362-9367.
48. Lewis, C.M. and J. Knight, *Introduction to Genetic Association Studies*. Cold Spring Harbor Protocols, 2012. **2012**(3): p. pdb.top068163.

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