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Angiotensin II and the Natriuretic and Blood Pressure Response to Mental Stress in African Americans

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Objectives: To test the hypothesis that Angiotensin II (Ang II) is a contributing factor to the response pattern in African Americans (AAs) who retain rather than excrete sodium during mental stress.

Design/Study Participants: Double-blind, randomized, cross-over trial of 87 healthy AAs aged 18 to 50 years.

Interventions: The study participants received either a placebo or irbesartan, (150 mg PO), an Ang II receptor antagonist, for seven days prior to stress testing. Urinary sodium excretion (UNaV) and systolic blood pressure (SBP) were collected prior to and throughout a mental stress protocol (rest and stress period).

Setting: A southeastern university.

Main Outcome Measures: Ang II, SBP, and sodium retention.

Results: During the placebo condition, 62 participants showed the expected increase in UNaV (excreters) while 25 participants reduced UNaV during stress (retainers). Irbesartan retainers demonstrated a reversal in the direction of their natriuretic response, now increasing UNaV in response to stress (Δ UNaV of -.094 mmol/min with placebo vs .052 mmol/min on irbesartan; P<.001). In excreters, irbesartan reduced SBP levels during both rest (-2.36 mm Hg; P=.03) and stress (-4.59;P<.0001), and an even more pronounced reduction in SBP was demonstrated by retainers on treatment during both rest (-4.29 mm Hg; P=.03) and stress (-6.12; P<.001).

Conclusions: Ang II contributes to sodium retention in retainers. Furthermore, our findings indicate that suppression of Ang II has a beneficial effect on SBP during rest and stress in this population. *Ethn Dis*. 2018;28(4):511-516; doi:10.18865/ed.28.4.511.

Key Words: Angiotensin II; Sodium Retention; Systolic Blood Pressure; Irbesartan; Stress; Retainers, Excreters

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Introduction

Salt intake and stress are two major risk factors for the development of high blood pressure (BP) and BP-related target organ damage. These two risk factors have been shown to have a synergistic effect, and animal studies demonstrated that strains at risk for the development of hypertension retain (rather than excrete) sodium during mental stress.[1-3] Further studies suggest that sodium retention during stress in animals is due, in part, to increased renal sympathetic nerve activity (RSNA) through the regulation of Angiotensin II (Ang II).[4] Studies also demonstrated sodium retention during stress in a significant percentage of humans belonging to high-risk populations, including African Americans (AAs),[5,6] individuals with a positive family history of hypertension,[7] and obese individuals.[8] Although the mechanisms responsible for sodium retention in these individuals have yet to be established, we have associated this pattern to indices of BP-related target organ damage to the heart[6] and kidney.[9]

The primary purpose of this study was to determine if activation of the renin-angiotensin-system (RAS) contributes to sodium retention in AAs. In this study, we performed a randomized clinical trial on AAs, a population with a high prevalence of stress-dependent sodium retention, in which the activation of RAS in regard to sodium retention has not been previously studied, to our knowledge.

Methods

Study Participants

Study participant characteristics are provided in Table 1. Inclusion criteria were: 1) AA; 2) aged 18 to 50 years; 3) not pregnant; 4) healthy and not on medications. Those who demonstrated an impaired natriuretic response to stress (retaining sodium) are defined as retainers. Alternatively, those whose UNaV levels increased in response to stress, an appropriate natriuretic response, are defined as excreters.

Recruitment and Screening

All procedures were approved by the Human Assurance Committee of at the Medical College of Georgia in accordance with the institutional guidelines. Informed consent was obtained prior to any measurements. Study participants were recruited by contacting individuals who had participated in our previous studies and by word-of-mouth. Following consent, we obtained baseline measurements, including height, weight, and resting BP, using a mercury column and stethoscope. Blood samples were collected to exclude individuals with significant kidney, liver, or other chronic illness.

Protocol

The study utilized a randomized, double-blind, placebo-controlled crossover design to determine the effects of an angiotensin receptor antagonist (ARB) (irbesartan) on the natriuretic and BP response to acute mental stress. Irbesartan was chosen for its ability to block the vasoconstrictor and aldosterone-secreting effects of Ang II by selectively blocking, in a noncompetitive manner, the binding of Ang II to the AT1 receptor. The steady-state concentrations are reached within 3 days with a half-life of 11 to 15 hours.

Study participants were randomized by the flip of a coin into two groups by the Augusta University pharmacy, in order to determine the order of intervention in the cross-over study – placebo followed by irbesartan or vice versa. At each phase, the treatment was administered for seven days. Participants took their final dose on the day of testing. The first phase of testing was followed by a 7-14 day washout period after which participants crossed over to the second phase of the study, which was performed under identical conditions. With the cross-over design, each participant acted as his/her own control in the active treatment vs placebo comparison. Therefore, there was not a need to control for variability due to body size or any other confounding factors. This approach has been shown to successfully reduce the variability of sodium intake, as estimated by overnight sodium excretion in free-living individuals.[10]

The primary purpose of this study was to determine if activation of the renin-angiotensin-system (RAS) contributes to sodium retention in African Americans.

Stress Testing Protocol

The mental stress protocol was a modified version of the standard stress testing protocol, originally reported by Light in adults[7] and used by our studies on more than 1,000 pediatric study participants as described previously.[11-13] The stressor was a video game, with opportunity to win a minimal cash reward. On testing day, two study participants were situated in a private room in comfortable chairs. The hemodynamic (BioZ) recording equipment was attached to the participants to obtain measurements at 15-minute intervals throughout the testing period. The participants took their final dose of medication immediately before the testing period. The protocol included 45 minutes of rest, followed by 45 minutes of mild stress. The participants watched a movie during the rest period. Blood and urine samples were obtained at baseline, rest, and stress periods.

Measurements

BPs during the testing protocol were recorded using a Dinamap BP recorder. Sodium excretion was analyzed by the ion selective electrode technique using a NOVA 16 Analyzer (NOVA Biomedical, Waltham, MA). The intra-assay coefficient of variation was <3% and the inter-assay coefficient was 4%.

Classification of Participants

Participants were classified as excreters or retainers, based on a modi-

Table 1. Study participant characteristics

	Retainers (n = 25)			Excreters (n= 62)		
Male	14 (56%)			37 (60%)		
Female	11 (44%)			25 (40%)		
	Mean	STD	95% CI	Mean	STD	95% CI
Height, cm	173.28	10.56	168.92, 177.64	172.50	10.29	169.89, 175.11
Weight, kg	211.80	49.95	191.18, 232.42	180.81	41.69	170.22, 191.39
Age	26.48	10.94	21.96, 31.00	22.92	7.30	21.07, 24.77
BMI	31.86	7.38	28.82, 34.91	27.58	5.98	26.06, 29.10
Resting SBP	115.10	12.27	110.03, 120.17	115.61	10.12	113.044, 118.18

fied version of the technique and criteria we and others used in previous studies.[5,12,14] The classification was performed with data collected during the placebo condition. If patients increased UNaV from the pre-stress to the stress condition, they were designated excreter. In contrast, if UNaV was lower during the stress condition, they were designated retainer.

Statistical Analysis

All statistical analyses were performed using SAS 9.4 version software package. Data are summarized by mean and 95% CI for continuous variables and frequency for categorical variables of retainers and excreters. Because we used a cross-over design, in the first step of our analysis, we tested for carryover effects and period effects using the method outlined by Jones and Kenward.[15] Neither the carryover nor the period effects were significant; therefore, no adjustments were needed in the subsequent analyses. In the next step, we examined the effects of irbesartan on UNaV and systolic BP (SBP). The histogram and Q-Q plot indicated that the normality assumption for the distribution of ΔUNaV was questionable for the placebo and irbesartan arms for both excreters and retainers. Therefore, we performed the Wilcoxon Signed Ranked test to obtain the results for UNaV in Table 2. However, the normality assumption was shown to be reasonable for the SBP study, and hence, we used the paired t-test to obtain the results for SBP in Table 2.

Results

Effect of Irbesartan on Sodium Excretion

The effects of treatment on levels of UNaV during mental stress for ex-

Table 2. Analysis of UNaV and SBP in retainers and excreters

	Retainers, n=25				
	Placebo	Irbesartan	Irbesartan-Placebo	P	95% CI
Rest UNaV, mmol/min	.339	.284	–.055	.216	–.143, .034
Stress UNaV, mmol/min	.245	.336	.091	.045	.032, .181
Δ UNaV, mmol/min	–.094	.052	.146	<.001	.076, .215
Rest SBP, mm Hg	115.10	110.81	–4.29 (–3.7%)	.032	–8.170, –.410
Stress SBP, mm Hg	118.52	112.40	–6.12 (–5.1%)	<.001	.402
ΔSBP, mm Hg	3.42	1.59	–1.83	.402	–6.242, –2.589
	Excreters, n=62				
	Placebo	Irbesartan	Irbesartan-Placebo	P	95% CI
Rest UNaV, mmol/min	.191	.251	.024	.303	–.022, .070
Stress UNaV, mmol/min	.298	.319	.021	.408	–.029, .070
Δ UNaV, mmol/min	.107	.104	–.003	.860	–.052, .057
Rest SBP, mm Hg	115.61	113.25	–2.36 (–2.0%)	.033	– 4.526, –.192
Stress SBP, mm Hg	119.63	115.04	–4.59 (–3.8%)	<.0001	– 6.089, –3.102
ΔSBP, mm Hg	4.02	1.79	–2.23	.051	– 4.484, .011

Table 3. Average SBP in combined sample

	Placebo	Irbesartan	Irbesartan-Placebo	P	95% CI
Rest SBP, mm Hg	115.46	112.54	– 2.92 (– 2.5%)	.003	–4.785, –1.043
Stress SBP, mm Hg	119.31	114.28	– 5.03 (– 4.2%)	<.0001	–6.422, –3.643
ΔSBP, mm Hg	3.85	1.74	– 2.11	.037	–4.112, –.125

creters and retainers are provided in Table 2. Within the retainer group, the UNaV levels decreased in response to mental stress during the placebo phase, by definition. The effect was reversed after irbesartan treatment. The difference in the change was statistically significant (P<.001), showing a beneficial effect of irbesartan among retainers. In contrast, among excreters, the UNaV levels increased (by definition) at the same rate in both phases during stress, showing no significant effect of irbesartan on sodium excretion in this group (P=.86).

Effect of Irbesartan on BP

Table 2 shows the effects of treatment on BP levels for excreters and retainers, with results indicating that absolute SBP levels were ameliorated in both retainers and excreters when treated with irbesartan. Both groups saw a significant drop in SBP levels during the rest and stress periods. Although not statistically significant, the extent of BP increase from rest to stress lessened under irbesartan treatment, as compared with the placebo phase for both retainers and excreters. We also analyzed the combined data from both retainers and excreters (N=87). Results indicated, again, that SBP levels during rest and stress were significantly lowered by treatment with an ARB. In addition, the analysis revealed a significant impact of irbesartan on the change in SBP in response to stress (ΔSBP of –2.11 mm Hg, P=.037). (Table 3)

Discussion

The most significant finding in this study was that treatment with an ARB increased UNaV during stress in AA study participants who otherwise retained sodium. As such, these findings provide evidence that this response pattern is mediated, at least in part, by Ang II. To our knowledge, this is the first clinical trial to demonstrate the benefits of an RAS inhibitor on the stress-induced rise in BP in AAs. Taken together, these findings provide evidence for the potential effectiveness of an ARB in reducing the negative impact of stress in this select population of AAs.

The findings from this study using an ARB are consistent with those observed in previous studies[16] on Caucasians using angiotensin converting enzyme inhibitors (ACEi). For example, Fauvel[17] measured stress-induced changes in UNaV in 20 patients with hypertension, 10 on a placebo and 10 on an ACEi (Lisinopril), for one month. Consistent with our findings, there were no changes in UNaV following treatment in the placebo group. In contrast with the placebo group, the study participants on the ACEi demonstrated lower rest and stress BPs as well as an increase in UNaV during stress. Rollnik[14] reported on a group of 48 Caucasian individuals, including 27 normotensives and 21 with mild hypertension. As with our previous studies, Rollnik[14] found that approximately one in three retained sodium during mental stress, based on changes within the individual. Captopril improved the change in UNaV from –20mmol/hr (placebo) to +2.8 mmol/hr (treatment). Schneider[18] reported the effects of Captopril on UNaV during stress in 33 normotensive patients with no family history of hypertension, 33 normotensives with a positive family history of hypertension, and 36 patients with mild hypertension. All three groups increased UNaV during stress, with hypertensive patients demonstrating the smallest increase, suggesting a blunted natriuretic response to stress in the early phase of hypertension. In

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addition, patients with hypertension demonstrated the greatest decrease in BP but the smallest increase in UNaV when treated with an ACEi, revealing their likely impaired sodium handling and suggesting that RAS plays a significant role in the response pattern.

A third goal of our study was to determine if an ARB reduced the BP stress response to mental stress. We observed that the ARB reduced the SBP response to stress independent of group status (excreter/retainer). The combined data demonstrated that the effect of the ARB on SBP levels during stress was almost twice that of the effect during rest (2.5% during rest vs 4.2% during stress). Further analysis of the combined data also revealed that ARB treatment significantly lessened the stress-induced increase in SBP (Table 3).

This research was based on observational studies by our group and others, which reported that AAs demonstrate a higher frequency of sodium retention during stress. Light[5] examined 28 adults, 14 AA and 14 Caucasian patients, and reported that the AA patients "tended" to show lower UNaV during a 1-hour stress period, with a reduction in UNaV for 6 of the 14 AAs and 2 of the 14 Caucasians. A study by our group reported race differences in the natriuretic response to stress across a series of tasks (playing video games and ice to forehead) in AA and Caucasian youths with a positive family history of hypertension.[12] Averaged across tasks, the AA study participants had a greater increase in BP coupled with a smaller increase in UNaV. We also reported findings on a study of 118 AAs using a video game stressor.[12] Of these, 38 (32%) retained sodium during stress. The increase in sodium retention was associated with an increase in cardiac output. These findings support the role of sodium loading and volume increases in generating higher BPs during stress in those who retained sodium. In contrast, vasoconstriction was the source of elevated BP in those who demonstrated a normal natriuretic response during stress.

Study Limitations

Our study design minimized many of the potential limitations. However, remaining limitations to the study may impact the interpretation of the findings. Among these, the study participants were all normotensive, and the findings may not generalize to AAs with hypertension. A Caucasian comparison group was not tested due to financial considerations, which limited comparisons to Caucasians and studies in the literature using a different RAS blocker. Findings with the stress protocol we used (video game) may not be generalized to other forms of stress. With respect to this limitation, the same response patterns have been reported in response to a variety of stressors and protocols.[5,12,14] The study participants may have experienced different levels of stress during the placebo and ARB arms, which could potentially affect the changes in UNAV observed and the grouping of participants into retainers and excreters. The effects of the ARB on BP are modest. However, this is consistent with previous studies that reported minimal effects of an ARB on BP in normotensive AAs.[16] It is well described that ARBs have a small effect on reductions of BP in normotensive individuals.[16] Also, it should be noted that the dosage[19] of ARB utilized in this study was the lowest recommended (for safety purposes) and a higher dose may be needed to have an effect on blood pressure. Lastly, in regard to BP, the lack of statistical evidence may be due to an inadequate sample size to have sufficient statistical power at the 5% level.

Summary and Conclusion

Our study supports the hypothesis that, as in Caucasians, activation of the RAS contributes to sodium retention during mental stress in a significant percentage of AAs. The sodium retention, in turn, contributes to a greater pressure load in these individuals. Our findings indicate that stress may increase RSNA activity, inducing sodium retention through activation of Ang II. An important aspect of these findings is that ARBs are effective in controlling sodium reabsorption during stress in AAs.

Conflict of Interest

No conflicts of interest to report.

Author Contributions

Research concept and design: Harshfield, Hanevold, J Pollock, D Pollock, Treiber, Dong; Acquisition of data: Harshfield; Data analysis and interpretation: Hanevold, Jasti, Ghosh, George; Manuscript draft: Harshfield, Hanevold, Jasti, Ghosh, J Pollock, D Pollock, Dong, George; Statistical expertise: Ghosh, George; Acquisition of funding: Harshfield, J Pollock, D Pollock, Treiber, Dong; Administrative: Harshfield, Hanevold, Jasti, George; Supervision: Harshfield

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