nature publishing group

Watermelon Extract Supplementation Reduces Ankle Blood Pressure and Carotid Augmentation Index in Obese Adults With Prehypertension or Hypertension

Arturo Figueroa[1], Marcos A. Sanchez-Gonzalez[1], Alexei Wong[1] and Bahram H. Arjmandi[1]

BACKGROUND
Ankle-brachial index (ABI) and ankle blood pressure (BP) are associated with increased carotid wave reflection (augmentation index, AIx). Oral L-citrulline and L-arginine from synthetic or watermelon sources have reduced brachial BP, aortic BP, and aortic AIx. A directly measured carotid AIx (cAIx) rather than aortic AIx has been proposed as a better measurement of central AIx. We evaluated the effects of watermelon extract on ankle BP and cAIx in individuals with normal ABI and prehypertension or stage 1 hypertension.

METHODS
Ankle and brachial systolic BP (SBP), diastolic BP (DBP), mean arterial pressure (MAP), cAIx, ABI, and heart rate (HR) were evaluated in the supine position in 14 adults (11 women/3 men, age 58 ± 1 years) with prehypertension or stage 1 hypertension (153 ± 4 mm Hg). Subjects were randomly assigned to 6 weeks of watermelon extract supplementation (L-citrulline/L-arginine, 6 g daily) or placebo followed by a 2-week washout period and then crossover.

RESULTS
Ankle and brachial SBP (−11.5 ± 3.8 and −15.1 ± 2.8 mm Hg), DBP (−7.8 ± 2.3 and −7.6 ± 1.8 mm Hg), and MAP (−9.8 ± 2.6 and −7.3 ± 1.8 mm Hg), and cAIx (−8.8 ± 2.6 %) decreased significantly ($P < 0.05$) after watermelon supplementation compared to placebo. Watermelon supplementation had no significant effect ($P > 0.05$) on ABI and HR.

CONCLUSIONS
This study shows that watermelon extract supplementation reduces ankle BP, brachial BP, and carotid wave reflection in obese middle-aged adults with prehypertension or stage 1 hypertension and normal ABI, which may reflect improved arterial function.

Keywords: *ankle blood pressure; blood pressure; carotid wave reflection; hypertension; L-citrulline; watermelon*

American Journal of Hypertension, advance online publication 8 March 2012; doi:10.1038/ajh.2012.20

Low ankle-brachial index (ABI), the ratio of ankle to brachial systolic blood pressure (SBP), provides information on the presence of lower limb atherosclerosis and increased pressure wave reflection (augmentation index, AIx) from peripheral to central arteries (carotid and aorta).[1,2] Normal ABI is also associated with increased carotid AIx (cAIx) in overweight/obese adults with prehypertension or stage 1 hypertension.[3] However, less attention has been paid to the individual value of ankle SBP as a determinant of arterial function. High ankle SBP is considered as a marker of subclinical atherosclerosis and a predictor of cardiovascular mortality in adults with normal ABI and high brachial BP.[4] It has been reported that ankle SBP and mean arterial pressure (MAP) have a higher influence on cAIx than ABI and brachial SBP in healthy adults.[5]

L-Citrulline from watermelon is efficiently converted to L-arginine, the substrate for endothelial nitric oxide production.[6] L-Citrulline and L-arginine either from synthetic or watermelon sources have shown to decrease brachial BP and aortic BP in adults with high BP via improved endothelial function and aortic AIx.[7–9] However, a directly measured cAIx rather than a synthesized aortic AIx has been recommended for the evaluation of central AIx.[10] To the best of our knowledge, no previous study has evaluated the effect of an antihypertensive intervention on ankle BP. Therefore, the purpose of this study was to examine the hypothesis that watermelon extract would reduce cAIx and ankle BP in obese adults with normal ABI and high brachial BP.

METHODS

Subjects. We studied 14 middle-aged (58 ± 1 years) adults (11 women and 3 men). Inclusion criteria were an ABI between 1.0–1.5 and brachial SBP >120 mm Hg confirmed in two separate days after at least 10 min of rest. Subjects had no apparent cardiovascular or metabolic diseases assessed by medical history. Exclusion criteria included regular consumption of L-citrulline/L-arginine rich foods or supplements, smoking, BP higher than 160/100 mm Hg, and chronic diseases. All women were postmenopausal (>1 year of last menstruation)

[1]Department of Nutrition, Food and Exercise Sciences, College of Human Sciences, Florida State University, Tallahassee, Florida, USA. Correspondence: Arturo Figueroa (afiguero@fsu.edu)

Received 27 October 2011; first decision 3 December 2011; accepted 23 January 2012 .

and were not using exogenous ovarian hormones. None of the subjects was previously treated with antihypertensive medications. Subjects were asked not to modify their diet and exercise habits during the study. The Florida State University Human Subjects committee approved the experimental procedures and written informed consent was obtained from all subjects.

Study design. We used a randomized, two-period, crossover design. On the first visit, subjects received a familiarization with the tests, which were performed in the morning in a quiet temperature-controlled room (23 ± 1 °C) after an overnight fast and avoiding alcohol and caffeinated drinks for at least 24 h before testing. After electrocardiogram, BP, and tonometry instrumentation, subjects rested for at least 20 min before data collection. Brachial BP, ankle BP, and cAIx were collected in the supine position. The tests were conducted at the same time of the day (±1 h) for each subject to reduce possible diurnal variations in vascular parameters at baseline and at the end of each intervention.

Subjects were randomly assigned to watermelon supplementation (6 g of L-citrulline/L-arginine (2/1) per day) or placebo for 6 weeks separated by a 2-week washout period. Watermelon powder from Milne Fruit Products (Prosser, WA) consisted of sieved and freeze dried watermelon solids. The daily watermelon powder would be equivalent to ~2.3 pounds of red watermelon raw. The placebo consisted of sucrose, glucose, and fructose at 2:2:1 to match the sugar composition of the watermelon powder. The last dose of watermelon and placebo was ingested ~24 h before the cardiovascular measurements. Compliance was assessed by powder's bag counts at each visit.

Anthropometrics. Height was measured using a stadiometer to the nearest 0.5 cm and body weight was measured using a seca scale (Sunbeam Products, Boca Raton, FL) to the nearest 0.1 kg. Body mass index was calculated as kg/m^2.

Cardiovascular parameters. Brachial and ankle BP, brachial BP, ABI, heart rate (HR), and cAIx were simultaneously measured in duplicate using an automatic device (VP-2000; Omron Healthcare, Vernon Hills, IL). The BP cuffs were wrapped around both arms (brachial artery) and ankles (posterior tibial artery). BP was measured using the oscillometric method. MAP was estimated from the cuff pressure for maximal oscillations. BPs from the right cuffs is reported in this study. ABI was calculated as ankle SBP/brachial SBP. A tonometer was placed over the right common carotid artery to obtain pressure waveforms. The cAIx was calculated as the difference between the second and first systolic peak multiplied by 100. The average of two measurements was used in the analysis. The validity and reliability of ankle SBP and cAIx measured from VP-2000 have been previously demonstrated.[11] HR was obtained from the electrocardiogram.

Statistical analyses. Student's *t*-test was used to detect possible difference in parameters between interventions at baseline. The effects of watermelon and placebo were evaluated by a two-way ANOVA with repeated measures ((placebo vs. watermelon) × (baseline vs. 6 weeks)). When ANOVA produced a significant interaction, *post-hoc* comparisons were made with paired *t*-tests. A Pearson's correlation was used to determine the relationship between changes in BP and cAIx. Data are shown as means ± s.e. Statistical significance was defined a priori as $P < 0.05$. Statistical analyses were performed using SPSS version 16.0 (SPSS, Chicago, IL).

RESULTS

Watermelon supplementation was well tolerated by all subjects and no adverse effects were reported. Height, weight, body mass index, and waist circumference were 1.66 ± 0.02 m, 102.7 ± 3.6 kg, 37.3 ± 1.8 kg/m^2, and 111.9 ± 4.1 cm, respectively. There were no differences in subject characteristics at baseline and after 6 weeks of both treatments. All subjects were obese by body mass index and waist circumference, and had prehypertension or stage 1 hypertension. There were no significant differences in subject characteristics at baseline and after 6 weeks of both interventions.

Cardiovascular parameters

Cardiovascular parameters before and after the interventions are presented in **Table 1**. There were no significant differences between the interventions in baseline values. Changes in ankle SBP and cAIx are shown in **Figure 1**. There were significant decreases in ankle SBP ($P < 0.01$), diastolic BP (DBP) ($P < 0.05$) and MAP ($P < 0.05$); brachial SBP ($P < 0.01$), DBP ($P < 0.01$) and MAP ($P < 0.01$); and cAIx ($P < 0.05$) after watermelon compared to placebo. There was no change in ABI and HR after both interventions. There was no significant correlation between ankle and brachial BP with cAIx.

DISCUSSION

The novel findings of the present study are that 6 weeks watermelon supplementation containing 6 g daily of L-citrulline/L-arginine

Table 1 | Hemodynamic parameters before and after the interventions (*n* = 14)

	Watermelon		Placebo	
Variables	Before	After	Before	After
Ankle SBP (mm Hg)	185 ± 5	173 ± 6**,***	183 ± 5	181 ± 5
Ankle DBP (mm Hg)	99 ± 4	91 ± 5*,***	96 ± 4	97 ± 4
Ankle MAP (mm Hg)	131 ± 4	121 ± 5*,***	129 ± 4	129 ± 4
Brachial SBP (mm Hg)	152 ± 5	137 ± 5**,†	151 ± 5	147 ± 6
Brachial DBP (mm Hg)	89 ± 4	82 ± 3**,†	88 ± 4	86 ± 4
Brachial MAP (mm Hg)	115 ± 4	106 ± 3**,†	113 ± 5	112 ± 4
Carotid AIx (%)	31 ± 4	23 ± 5*,***	29 ± 4	30 ± 4
ABI (ratio)	1.25 ± 0.1	1.26 ± 0.1	1.26 ± 0.1	1.24 ± 0.1
Heart rate (beats/min)	68 ± 4	67 ± 3	67 ± 4	67 ± 4

Data are mean ± s.e.
ABI, ankle-brachial index; AIx, augmentation index; DBP, diastolic blood pressure; MAP, mean arterial pressure; SBP, systolic blood pressure.
*$P < 0.05$ and **$P < 0.01$ different from before; ***$P < 0.05$ and †$P < 0.01$ different from placebo.

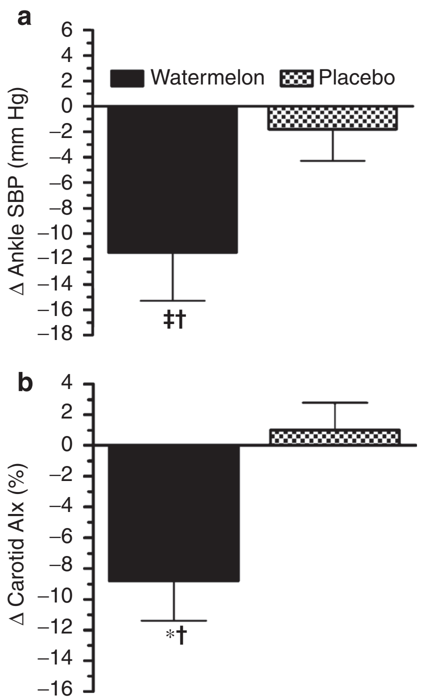

Figure 1 | Changes in (**a**) ankle systolic blood pressure (SBP) and (**b**) carotid augmentation index (AIx) after 6 weeks of watermelon supplementation and placebo. *$P < 0.05$ different from before. ‡$P < 0.01$ different from before. †$P < 0.05$ different from placebo. Data are mean ± s.e.

reduced ankle BP, brachial BP, and cAIx in obese middle-aged adults with normal ABI and prehypertension or stage 1 hypertension.

The hypotensive effect of L-citrulline and L-arginine from synthetic or watermelon sources on brachial and aortic BP has been previously demonstrated in adults with prehypertension and hypertension.[7,8] We previously reported that 4 g daily of L-citrulline/L-arginine from watermelon supplementation was effective to reduce aortic SBP but not aortic DBP or brachial SBP and DBP.[7] In the present study, brachial SBP, DBP, and MAP decreased by 15.1, 6.9, and 8.7 mm Hg after an increased dose of the amino acids from watermelon extract compared with placebo. Since a low dose of watermelon extract did not significantly affect brachial BP in our previous study,[7] the present findings indicate that the hypotensive effect of watermelon is dose-dependent. Moreover, ankle SBP, DBP, and MAP decreased by 11.5, 7.8, and 9.8 mm Hg with watermelon extract compared with no changes after placebo. Thus the lack of effect on ABI is explained by the concurrent decreases in ankle and brachial SBP. Interestingly, watermelon extract reduced ankle SBP from above 175 mm Hg to below this cut-point in 50% of the subjects. An ankle SBP >175 mm Hg is associated with prehypertension and hypertension and subclinical arterial damage in overweight/obese adults.[12,13] A normal ankle SBP is clinical important as cardiovascular events were lower by ~2.0-fold in those with normal ankle SBP than in those with high ankle SBP during a 15-year follow-up.[4,12]

Dichotomizing ABI into groups above and below the median showed a significantly higher cAIx in middle-aged individuals with a lower ABI (>1.0–1.5) than those with an ABI higher than 1.5.[3] Consistent with the previous study,[3] we found increased cAIx and brachial SBP in adults with an ABI between 1.0 and 1.5, suggesting an increased left ventricular afterload.[14] A strong association has been reported between ankle BP and cAIx in middle-aged adults with normal ABI and brachial BP.[5] We recently reported that watermelon supplementation (4 g daily of L-citrulline/L-arginine) decreased aortic AIx (−6%) in adults with prehypertension. However, the use of the generalized transfer function to estimate aortic AIx from radial tonometry has been criticized.[10,15] Alternatively, a directly measured cAIx has been suggested for the measurement of central wave reflection.[10] It has been shown that vasoactive drug treatment decrease central AIx by attenuating the magnitude of the reflected wave.[16–19] A decrease in cAIx (~6%) has been observed with reductions in brachial SBP and DBP after 3 months of calcium channel blocker therapy in untreated hypertensives.[19] Our present findings showed that cAIx decreased by ~9% after watermelon supplementation. Although ankle and brachial SBP, DBP, and MAP decreased in the present study, the changes in cAIx and peripheral BP were not significantly correlated. Consistent with our findings, previous studies have shown that vasodilating drugs reduce central AIx independently of brachial BP reduction.[18,19] Our study adds to the existing knowledge that the decrease in cAIx is independent of the fall in ankle BP. This dissociation may be explained by a greater influence of central BP on central AIx than peripheral BP.[14] It has been shown that the reduction in central AIx produced by vasodilating drugs is attributed to the greater decrease in central than in brachial SBP.[20] The effect of watermelon extract on cAIx may be important as reduction in central AIx after antihypertensive treatment lead to reduced left ventricular afterload and improved ventricular-arterial coupling.[17]

A possible mechanism underlying the effect of watermelon supplementation on AIx is through a decrease in wave reflection amplitude independently of aortic stiffness and brachial BP.[7] A reduced vascular tone by vasodilating drugs can cause a decrease in reflected wave amplitude and AIx.[14,17,19] This effect is achieved by affecting peripheral muscular arteries.[14,17,19] Previous studies suggest that L-citrulline and L-arginine supplementation can improve peripheral artery endothelial function by increasing L-arginine bioavailability and nitric oxide production.[8,9]

Potential limitations of the present study include a relatively small simple size, a mildly elevated BP, and a population composed predominantly of obese postmenopausal women. Therefore, our findings may not be generalized to other populations.

In conclusion, watermelon supplementation reduced ankle BP, brachial BP, and carotid wave reflection in obese middle-aged adults with prehypertension or stage 1 hypertension and normal ABI. This study suggests that watermelon extract improves arterial function independently of the reduction in peripheral BP.

Acknowledgment: We thank Milne Fruit Products for donation of the watermelon and placebo powders.

Disclosure: The authors declared no conflict of interest.

1. Resnick HE, Lindsay RS, McDermott MM, Devereux RB, Jones KL, Fabsitz RR, Howard BV. Relationship of high and low ankle brachial index to all-cause and cardiovascular disease mortality: the Strong Heart Study. *Circulation* 2004; 109:733–739.
2. Khaleghi M, Kullo IJ. Aortic augmentation index is associated with the ankle-brachial index: a community-based study. *Atherosclerosis* 2007; 195:248–253.
3. Rabkin SW, Chan SH, Sweeney C. Ankle-brachial index as an indicator of arterial stiffness in patients without peripheral artery disease. *Angiology* 2012; 63:150–154.
4. Hietanen H, Pääkkönen R, Salomaa V. Ankle blood pressure as a predictor of total and cardiovascular mortality. *BMC Cardiovasc Disord* 2008; 8:3.
5. Tarumi T, Sugawara J, Tanaka H. Association between ankle blood pressure and central arterial wave reflection. *J Hum Hypertens* 2011; 25:539–544.
6. Collins JK, Wu G, Perkins-Veazie P, Spears K, Claypool PL, Baker RA, Clevidence BA. Watermelon consumption increases plasma arginine concentrations in adults. *Nutrition* 2007; 23:261–266.
7. Figueroa A, Sanchez-Gonzalez MA, Perkins-Veazie PM, Arjmandi BH. Effects of watermelon supplementation on aortic blood pressure and wave reflection in individuals with prehypertension: a pilot study. *Am J Hypertens* 2011; 24:40–44.
8. Orea-Tejeda A, Orozco-Gutiérrez JJ, Castillo-Martínez L, Keirns-Davies C, Montano-Hernández P, Vázquez-Díaz O, Valdespino-Trejo A, Infante O, Martínez-Memije R. The effect of L-arginine and citrulline on endothelial function in patients in heart failure with preserved ejection fraction. *Cardiol J* 2010; 17:464–470.
9. Ochiai M, Hayashi T, Morita M, Ina K, Maeda M, Watanabe F, Morishita K. Short-term effects of l-citrulline supplementation on arterial stiffness in middle-aged men. *Int J Cardiol* 2010.
10. Segers P, Rietzschel E, Heireman S, De Buyzere M, Gillebert T, Verdonck P, Van Bortel L. Carotid tonometry versus synthesized aorta pressure waves for the estimation of central systolic blood pressure and augmentation index. *Am J Hypertens* 2005; 18:1168–1173.
11. Cortez-Cooper MY, Supak JA, Tanaka H. A new device for automatic measurements of arterial stiffness and ankle-brachial index. *Am J Cardiol* 2003; 91:1519–22, A9.
12. Hietanen H, Pääkkönen R, Salomaa V. Ankle and exercise blood pressures as predictors of coronary morbidity and mortality in a prospective follow-up study. *J Hum Hypertens* 2010; 24:577–584.
13. Hietanen HJ, Pääkkönen R, Salomaa V. Ankle blood pressure and pulse pressure as predictors of cerebrovascular morbidity and mortality in a prospective follow-up study. *Stroke Res Treat* 2011; 2010:729391.
14. Nichols WW, Denardo SJ, Wilkinson IB, McEniery CM, Cockcroft J, O'Rourke MF. Effects of arterial stiffness, pulse wave velocity, and wave reflections on the central aortic pressure waveform. *J Clin Hypertens (Greenwich)* 2008; 10:295–303.
15. Hope SA, Meredith IT, Tay D, Cameron JD. 'Generalizability' of a radial-aortic transfer function for the derivation of central aortic waveform parameters. *J Hypertens* 2007; 25:1812–1820.
16. Manisty C, Mayet J, Tapp RJ, Sever PS, Poulter N, McG Thom SA, Hughes AD; ASCOT Investigators. Atorvastatin treatment is associated with less augmentation of the carotid pressure waveform in hypertension: a substudy of the Anglo-Scandinavian Cardiac Outcome Trial (ASCOT). *Hypertension* 2009; 54:1009–1013.
17. Hashimoto J, Westerhof BE, Westerhof N, Imai Y, O'Rourke MF. Different role of wave reflection magnitude and timing on left ventricular mass reduction during antihypertensive treatment. *J Hypertens* 2008; 26:1017–1024.
18. Mahmud A, Feely J. Beta-blockers reduce aortic stiffness in hypertension but nebivolol, not atenolol, reduces wave reflection. *Am J Hypertens* 2008; 21:663–667.
19. Palombo C, Malshi E, Morizzo C, Rakebrandt F, Corretti V, Santini F, Fraser AG, Kozakova M. Arterial wave reflection during antihypertensive therapy with barnidipine: a 6-month, open-label study using an integrated cardiovascular ultrasound approach in patients with newly diagnosed hypertension. *Clin Ther* 2009; 31:2873–2885.
20. Hirata K, Vlachopoulos C, Adji A, O'Rourke MF. Benefits from angiotensin-converting enzyme inhibitor 'beyond blood pressure lowering': beyond blood pressure or beyond the brachial artery? *J Hypertens* 2005; 23:551–556.