Serum Uric Acid and Pulse Wave Velocity Among Healthy Adults: Baseline Data From the Brazilian Longitudinal Study of Adult Health (ELSA-Brasil)

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BACKGROUND
We aimed to evaluate a possible association between serum uric acid (SUA) levels and carotid-to-femoral pulse wave velocity (cf-PWV) among healthy participants of the ELSA-Brasil.

METHODS
We excluded subjects using antihypertensive medication, diuretics, allopurinol, binge drinkers, body mass index (BMI) >35 kg/m^2, and those with history of cardiovascular diseases (CVD). In a cross-sectional and sex-specific analysis, linear regression models were built having cf-PWV as dependent variable and SUA as independent variable. Multiple adjustments were subsequently made for age, heart rate and blood pressure, BMI, and fasting glucose levels as covariates. Product interaction terms were built to test interaction between SUA and other covariates.

RESULTS
We analyzed 1,875 men and 1,713 women (mean ages, 48.9±8.4 and 50.2±8.7 years, respectively). SUA was linearly associated with cf-PWV in men (P = 0.01) and in women (P = 0.01). After full adjustment, the association remained significant for men (P = 0.01) and no longer significant for women (P = 0.10). Fully adjusted linear coefficients β (95% CI) were 0.06 (0.015; 0.112) and 0.04 (−0.01; 0.12) in men and women, respectively. Significant interaction between SUA and age (P = 0.02) fasting glucose (P < 0.01) and BMI (P = 0.02) was found only for women.

CONCLUSION
In an apparently healthy population, SUA was significantly associated to cf-PWV in men but not in women.

Keywords: aortic stiffness; blood pressure; gender; hypertension; pulse wave velocity; uric acid.

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Serum uric acid (SUA) is a product of purine metabolism and it has been related to circulatory system disturbance since the XIX Century[1] with contradictory findings concerning to a possible causal association with hypertension, diabetes, and incidence of cardiovascular events.[2–6] There are recent studies that looked at SUA and its possible association to arterial stiffness[7–14] and yet these biological pathways are still not clear. On the other hand, although arterial stiffness has been associated with hypertension and atherosclerosis since the 1930s,[15] pulse wave velocity (PWV) measurement use has been disseminated after the availability of new devices at clinical settings. This new situation leads to the study of the association between high PWV and cardiovascular new events and deaths.[16]

The vast majority of studies addressing uric acid and aortic stiffness is observational. A study in Greece reported SUA levels independently associated with carotid-to-femoral PWV (cf-PWV) in 1,225 never-treated people with high blood pressure.[7] Three independent studies recruiting participants for a periodic health check-up exams in Korea (9,955 women),[8] Japan (982 men and women)[9], and China (940 men and women)[13] revealed a significant association between uric acid and PWV after multivariate adjustment. In the "Cardiometabolic Risk in Chinese Study", a sample of 1,283 women and 2,389 men drawn from a free-living community-based health examination had progressively higher values through uric acid quartiles and resting heart rate and systolic blood pressure played as independent moderator factors between uric acid and PWV.[14] In the VASORISK cohort (366 hypertensive participants), the multiple linear regression analysis yielded a positive association between PWV and uric acid in women after adjusting for classical risk factors.[12] Contrasting with these results, the "Brisighella Heart Study",[10] which was performed in 2,939 participants free of cardiovascular diseases (CVD) and another Korean study[11] performed in 1,276 people with metabolic syndrome

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did not find any association between uric acid and high cf-PWV.

These contrasting results might be an indicative of conflicting data related to different populations, sampling methods, and different PWV measurement devices used in previous studies. Moreover, these studies were carried out on Asian and European populations and aortic stiffness seems to be higher in Blacks living in Angola,[17] South Africa,[18] Brazil[19], and the United States.[20] In order to verify the link "uric acid–pulse wave velocity" in a population with heterogeneous ethnic background encompassing White, Brown, and Black participants and to help clarify the possible association between SUA and cf-PWV, we chose to address this question in the Brazilian Longitudinal Study of Adult Health (ELSA-Brasil).

METHODS

Study subjects

ELSA-Brasil is a multicenter prospective cohort study enrolling 15,105 civil servants aged 35–74 years (54% women) from 6 cities of Brazil (Belo Horizonte, Porto Alegre Rio de Janeiro, Salvador, São Paulo e Vitória)[21–23] which aims to investigate factors associated to the development and progression of CVD and diabetes. We applied a cross-sectional analysis using baseline data of ELSA-Brasil collected from 2008 to 2010. For the purpose of this study, we excluded subjects using antihypertensive medication, diuretics, allopurinol, binge drinkers (men ≥ 210 g alcohol/week; women ≥ 140 g alcohol/week), BMI > 35 kg/m^2, and those with history of CVDs. Hence, this analysis was based on data collected in 3,588 participants (1,875 men and 1,713 women).

Laboratory measurements

After an overnight fasting, blood sample was collected in all participants and uric acid was determined enzymatically by the uricase method. Blood glucose was determined enzymatically by the hexokinase method. Total cholesterol, high-density cholesterol (HDL cholesterol) and triglycerides were determined by the enzymatic colorimetric method. Low-density cholesterol (LDL cholesterol) was calculated using the Friedewald equation and filtration glomerular rate was calculated by CKD-Epi.[24] The strategies for the collection, processing, transportation, and quality control of blood and urine tests in the ELSA-Brasil are described in detail elsewhere.[22]

Cf-PWV measurements

Cf-PWV was measured with an automatic and validated device (Complior, Artech Medicale, France) with the participant lying down in a room with controlled temperature (20 °C–24 °C). Before cf-PWV measurement, blood pressure was measured in the supine position with an automatic device (Omron 705 CP, Japan) at the nondominant arm. The distance from the sternum manubrium to the right femoral pulse was measured with a standardized inelastic tape without correction for abdomen curvature. The pulse sensors were placed on the right carotid and femoral arteries, allowing visualization of the pulse wave on a computer screen. Software registered only the pulse waves with good quality. Cf-PWV is calculated by dividing the distance from the suprasternal notch to the femoral pulse by the time lag between the carotid and femoral pulses. The cf-PWV of each participant was calculated as the arithmetic average obtained in 10 consecutive cardiac cycles in regular heart rhythm. cf-PWV is influenced not only by the stiffness of large arteries (e.g., aorta), but also by the pressure inside the artery in the moment of the exam, which determines recruitment of elastic and collagen fibers. Therefore, cf-PWV obtained in ELSA-Brasil was adjusted by the BP measured immediately before the exam in the logistic models. For the quality control, all cf-PWV tests were recorded in the 6 sites and sent to a centralized Reading Center responsible to validate exams of all ELSA-Brasil participants.[23]

Sociodemographic and anthropometric variables

Other variables included in our analysis were age, sex, education (elementary, high-school, college); smoking status (never, former, and current), age (years), and self-reported ethnicity (White, Brown, Black, Asian, and Native). Height and weight were measured and BMI was calculated dividing body weight to the squared height in meters (kg/m^2). Waist circumference (cm) was measured using standardized procedure. Blood pressure was obtained in the sitting position after a minimum rest period of 5 min. Three consecutive readings were obtained in each participant after 1 min interval each. The mean of the 2 last measurements was defined as the casual blood pressure. Heart rate was measured 3 times in the supine position. The mean of the 2 last measurements was defined as the mean heart rate.

Statistical analysis

Categorical variables are reported as percentages and continuous variables as means and standard deviations unless stated otherwise. Statistical normality was checked with the Kolmogorov–Smirnov test. Age-adjusted correlation was tested between uric acid and cf-PWV.

Normality, linearity, and homoscedasticity assumptions of the linear model were checked. We tested the linear association between SUA and PWV by building sex-specific multilinear regression models using PWV continuously as a dependent variable and SUA as independent variable. Adjustments were subsequently made by age, blood pressure measured at the time of cf-PWV measurement, body mass index, and fasting glucose.

As cf-PWV is known to increase with age and blood pressure,[25] our first model was adjusted for age and blood pressure measured immediately before the cf-PWV measure. Further adjustments were made including fasting glucose and heart rate as covariates. Product interaction terms were built to test interaction between SUA and other covariates. A 2-tailed level of significance was set at 5%. Analyses were carried out using the SPSS 20.0 statistical package.

RESULTS

We analyzed 1,875 men and 1,713 women (mean ages, 48.9 ± 8.4; 50.2 ± 8.7 years respectively). As predicted, SUA values were higher in men than in women (6.2 ± 1.2 mg/dl vs. 4.53 ± 0.9 mg/dl). Mean PWV also differed among genders (9.06 ± 1.41 m/s vs. 8.4 ± 1.30 m/s for men and women, respectively) justifying a sex-specific analysis. Age-adjusted Pearson correlation between SUA and cf-PWV showed coefficients of $r = 0.089$ for men ($P = 0.01$) and $r = 0.073$ for women ($P = 0.01$).

Pulse wave velocity measurements (m/s) by SUA units (mg/dl) are shown in at Table 1. Univariate analysis showed significant linear association between SUA and PWV for men and women ($P = 0.01$ for both genders). After adjustment for age, blood pressure, and heart rate the linear association lost significance for men ($P = 0.06$) and remained significant in women ($P = 0.01$). After further adjustments for BMI and glucose levels the association was positive and significant for men ($P = 0.01$). Linear coefficient showed that there is a difference of 0.06 (0.015; 0.112) m/s in men cf-PWV by unit in SUA (mg/dl). Nonsignificant interaction between SUA and age, blood pressure, BMI, or fasting glucose was found for men. For women, we found significant interaction between SUA and age ($P = 0.02$) fasting glucose ($P = 0.01$) and BMI ($P = 0.02$).

DISCUSSION

We found linear association between SUA and cf-PWV beyond the natural required adjustments for age and blood pressure at the time of CF-PWV measurement in men on a healthy multiethnic population. Our main findings suggest a different association of SUA and cf-PWV between genders.

Some previous studies have addressed the association between uric acid and cf-PWV. Vlachopoulos *et al.*[7] evaluated 1,225 newly diagnosed hypertensive patients treatment, not using antidiabetic drugs and acid lowering medication, with mean age of 53.3 years and found no association between cf-PWV and SUA levels. After adjustment for confounders, a positive association was found for men and women separately. Gomez-Marcos evaluated 366 hypertensive men and women. They found a positive association between cf-PWV and uric acid tertiles only for women after adjustment for sociodemographic and classical risk factors. However, this association lost significance after further adjustment for glomerular filtration rate, waist measurement, us-C-reactive protein, and use of diuretics. No association was found for men.[12] Again, in a sample of 1,276 Korean individuals with a frequency of almost 25% of metabolic syndrome, no association was found between quartiles of uric acid and heart-femoral and brachial-ankle cf-PWV for females and males separately.[11]

Possible reasons for the discrepancy between our data and the literature could be due to PWV adjustment by sex and not a sex-specific analysis.[14] Potential bias may also be secondary to the inclusion of patients taking medications that could introduce potential bias due to inclusion of participants taking medications that could affect both the dependent and the independent variables.[9] Moreover differently from our studied population, some studies recruited participants within clinical relevant conditions.[8]

Results from studies in apparently healthy populations are not clear either. Ishizaka *et al.*[9] evaluated brachial-ankle cf-PWV in 982 men and women and found a positive association for both genders analyzing uric acid quartiles and after multivariate adjustment for age, total and HDL cholesterol, systolic blood pressure, triglycerides, fasting glucose, and smoking status. Liang *et al.*[14] evaluated a population of 1,283 women and 2,389 men drawn from a community based health examination with an exclusion criteria of vascular disease, diabetes mellitus, or hyperlipidemia treated with medication, and renal failure. After adjustment for age, sex, BMI, total cholesterol, triglyceride, HDL-C, LDL-C, blood pressure, heart rate, and fasting glucose, they found SUA was associated with elevated aortic arterial stiffness in Chinese adults, independent of traditional cardiovascular risk factors.

Contrarily, in the 619 men and women participants of the Brisighella Heart Study, who were not taking antihypertensive, antidiabetic, lipid-lowering and uric acid-lowering drugs with mean age of 53.3 years, no significant association between SUA and cf-PWV was found after adjustment for age.[10]

Potential mechanisms for the effect of aortic stiffness to cardiovascular risk factors are accredited to breaks in elastin fibers, accumulation of collagen, fibrosis, inflammation, medial smooth muscle necrosis, and medial calcifications.[26] On the other hand, experimental studies have shown that uric acid may favor microvascular disease stimulating vascular smooth muscle cell proliferation *in vitro*, with the production of proinflammatory, prooxidative, and vasoconstrictive substances.[2,3,27] Therefore, possible explanation for

Table 1. Linear association between serum uric acid with pulse wave velocity by genders

	Men (n = 1,865)			Women (n = 1,713)		
	β	CI (95%)	P	β	CI (95%)	P
Univariate	0.07	(0.03, 0.13)	<0.01	0.14	(0.13, 0.26)	<0.01
Adjusted for age	0.36	(0.05, 0.67)	<0.01	0.38	(0.05, 0.07)	<0.01
Plus blood pressure and heart rate	0.04	(−0.003, 0.089)	0.06	0.07	(0.013, 0.125)	0.01
Fully adjusted	0.06	(0.015, 0.112)	0.01	0.04	(−0.01, 0.12)	0.10

Fully adjusted model: age, blood pressure, and heart rate measured before pulse wave velocity measurement, body mass index, and fasting glucose. Results are presented as difference in cf-PWV unit by each serum uric acid unit (mg/dl).

the association found in some studies is that uric acid could regulate critical proinflammatory pathways in smooth muscle cell suggesting that it might have a role in the vascular changes.[28]

As cf-PWV increases with age and it is influenced by sympathetic and parasympathetic activity,[25] we adjusted our model to age, blood pressure, and heart rate measured in supine position immediately before the cf-PWV recording. Moreover, as SUA has been associated with fasting blood glucose and incident type 2 diabetes, we also adjusted our multivariate model for fasting blood glucose. As previous studies looked at the association between SUA and blood pressure found positive results that could be confounded by BMI,[6] we therefore included BMI in our adjustments without material changes in results. Although some authors have adjusted their analysis for other covariables, we chose to exclude those participants from the sample as possible confounders.

Interestingly, most of the studies reporting positive cross-sectional associations between SUA and arterial stiffness were conducted among Asian populations.[8,9,13,14] This highlights the importance of our study among a large healthy non-Asian population.

Our study has some strengths and limitations that merit consideration. We present data of CF-PWV of a well-defined population. Our study was powered to analyze SUA levels and cf-PWV in a sex-specific analysis even after exclusion of potential confounders to the association studied as use of antihypertensive, allopurinol, higher BMI, and binge drinkers. Nevertheless, due to the cross-sectional nature of the design we were not able to clear the possible pathway between SUA and cf-PWV.

In conclusion, our findings support that in an apparently healthy and multiethnic population SUA was significantly associated with PWV in men independently of age, blood pressure, heart rate, BMI, and fasting blood glucose. Future prospective studies using data from ELSA-Brasil would clarify about a causal relationship and bring more information about possible mechanisms that can explain the association between SUA and gender-related differences.

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DISCLOSURE

The authors declared no conflict of interest.

REFERENCES

1. Mahomed F. On chronic Bright's disease, and its essential symptoms. *The Lancet* 1879; 113:399–401.
2. Hayden MR, Tyagi SC. Uric acid: a new look at an old risk marker for cardiovascular disease, metabolic syndrome, and type 2 diabetes mellitus: the urate redox shuttle. *Nutr Metab (Lond)* 2004; 1:10.
3. Johnson RJ, Kanbay M, Sánchez-Lozada LG. The rediscovery of uric acid in cardiorenal disease: introduction. *Semin Nephrol* 2011; 31:391–393.
4. Johnson RJ, Lanaspa MA, Gaucher EA. Uric acid: a danger signal from the RNA world that may have a role in the epidemic of obesity, metabolic syndrome, and cardiorenal disease: evolutionary considerations. *Semin Nephrol* 2011; 31:394–399.
5. Kanbay M, Segal M, Afsar B, Kang DH, Rodriguez-Iturbe B, Johnson RJ. The role of uric acid in the pathogenesis of human cardiovascular disease. *Heart* 2013; 99:759–766.
6. Palmer TM, Nordestgaard BG, Benn M, Tybjærg-Hansen A, Davey Smith G, Lawlor DA, Timpson NJ. Association of plasma uric acid with ischaemic heart disease and blood pressure: mendelian randomisation analysis of two large cohorts. *BMJ* 2013; 347:f4262.
7. Vlachopoulos C, Xaplanteris P, Vyssoulis G, Bratsas A, Baou K, Tzamou V, Aznaouridis K, Dima I, Lazaros G, Stefanadis C. Association of serum uric acid level with aortic stiffness and arterial wave reflections in newly diagnosed, never-treated hypertension. *Am J Hypertens* 2011; 24:33–39.
8. Park JS, Kang S, Ahn CW, Cha BS, Kim KR, Lee HC. Relationships between serum uric acid, adiponectin and arterial stiffness in postmenopausal women. *Maturitas* 2012; 73:344–348.
9. Ishizaka N, Ishizaka Y, Toda E, Hashimoto H, Nagai R, Yamakado M. Higher serum uric acid is associated with increased arterial stiffness in Japanese individuals. *Atherosclerosis* 2007; 192:131–137.
10. Cicero AF, Salvi P, D'Addato S, Rosticci M, Borghi C, group BHS. Association between serum uric acid, hypertension, vascular stiffness and subclinical atherosclerosis: data from the Brisighella Heart Study. *J Hypertens* 2014, 32:57–64.
11. Lim JH, Kim YK, Kim YS, Na SH, Rhee MY, Lee MM. Relationship between serum uric Acid levels, metabolic syndrome, and arterial stiffness in korean. *Korean Circ J* 2010; 40:314–320.
12. Gomez-Marcos MA, Recio-Rodriguez JI, Patino-Alonso MC, Agudo-Conde C, Rodriguez-Sanchez E, Gomez-Sanchez L, Gomez-Sanchez M, Garcia-Ortiz L: Relationship between uric acid and vascular structure and function in hypertensive patients and sex-related differences. *Am J Hypertens* 2013, 26:599–607.
13. Chen X, Li Y, Sheng CS, Huang QF, Zheng Y, Wang JG. Association of serum uric acid with aortic stiffness and pressure in a Chinese workplace setting. *Am J Hypertens* 2010; 23:387–392.
14. Liang J, Li Y, Zhou N, Teng F, Zhao J, Zou C, Qi L. Synergistic effects of serum uric acid and cardiometabolic risk factors on early stage atherosclerosis: the cardiometabolic risk in Chinese study. *PLoS One* 2012; 7:e51101.
15. Haynes FW, Ellis LB, Weiss S: Pulse wave velocity and arterial elasticity in arterial hypertension, arteriosclerosis, and related conditions. *Am Heart J* 1936, 11:385–401.
16. Vlachopoulos C, Aznaouridis K, Stefanadis C. Prediction of cardiovascular events and all-cause mortality with arterial stiffness: a systematic review and meta-analysis. *J Am Coll Cardiol* 2010; 55:1318–1327.
17. Magalhães P, Capingana DP, Silva AB, Ferreira AV, de Sá Cunha R, Rodrigues SL, Mill JG. Age- and gender-specific reference values of pulse wave velocity for African adults: preliminary results. *Age (Dordr)* 2013; 35:2345–2355.
18. Schutte AE, Huisman HW, Schutte R, Van Rooyen JM, Malan L, Malan NT, Reimann M. Arterial stiffness profiles: investigating various sections of the arterial tree of African and Caucasian people. *Clin Exp Hypertens* 2011; 33:511–517.

19. de Lima Santos PCJ, de Oliveira Alvim R, Ferreira NE, de Sá Cunha R, Krieger JE, Mill JG, Pereira AC: Ethnicity and arterial stiffness in Brazil. *Am J Hypertens* 2011; 24:278–284.
20. Shah AS, Dolan LM, Gao Z, Kimball TR, Urbina EM. Racial differences in arterial stiffness among adolescents and young adults with type 2 diabetes. *Pediatr Diabetes* 2012; 13:170–175.
21. Schmidt MI, Duncan BB, Mill JG, Lotufo PA, Chor D, Barreto SM, Aquino EM, Passos VM, Matos SM, Molina MD, Carvalho MS, Bensenor IM. Cohort Profile: Longitudinal Study of Adult Health (ELSA-Brasil). *Int J Epidemiol* 2014; 1–8. doi:10.1093/ije/dyu027.
22. Bensenor IM, Griep RH, Pinto KA, Faria CPd, Felisbino-Mendes M, Caetano EI, Albuquerque LdS, Schmidt MI: Routines of organization of clinical tests and interviews in the ELSA-Brasil investigation center. *Revista de Saúde Pública* 2013; 47:37–47.
23. Mill JG, Pinto K, Griep RH, Goulart A, Foppa M, Lotufo PA, Maestri MK, Ribeiro AL, Andreao RV, Dantas EM. Medical assessments and measurements in ELSA-Brasil. *Revista de Saúde Pública* 2013; 47:54–62.
24. Levey AS, Stevens LA. Estimating GFR using the CKD epidemiology collaboration (CKD-EPI) creatinine equation: more accurate GFR estimates, lower CKD prevalence estimates, and better risk predictions. *Am J Kidney Dis* 2010; 55:622.
25. Collaboration RVfAS: Determinants of pulse wave velocity in healthy people and in the presence of cardiovascular risk factors:'establishing normal and reference values'. *Eur Heart J* 2010; 31: 2338–2350.
26. Laurent S, Cockcroft J, Van Bortel L, Boutouyrie P, Giannattasio C, Hayoz D, Pannier B, Vlachopoulos C, Wilkinson I, Struijker-Boudier H: Abridged version of the expert consensus document on arterial stiffness. *Artery Res* 2007; 1:2–12.
27. Johnson RJ, Lanaspa MA, Gaucher EA. Uric acid: a danger signal from the RNA world that may have a role in the epidemic of obesity, metabolic syndrome, and cardiorenal disease: evolutionary considerations. In *Seminars in Nephrology: 2011*. *Elsevier*, 2011, pp. 394–399.
28. Kanellis J, Watanabe S, Li JH, Kang DH, Li P, Nakagawa T, Wamsley A, Sheikh-Hamad D, Lan HY, Feng L, Johnson RJ. Uric acid stimulates monocyte chemoattractant protein-1 production in vascular smooth muscle cells via mitogen-activated protein kinase and cyclooxygenase-2. *Hypertension* 2003; 41:1287–1293.