Behavioural Brain Research 296 (2016) 85–93

Contents lists available at ScienceDirect

Behavioural Brain Research

journal homepage: www.elsevier.com/locate/bbr

Research report

Neuromarkers of the common angiotensinogen polymorphism in healthy older adults: A comprehensive assessment of white matter integrity and cognition

Lauren E. Salminen[a,*], Peter R. Schofield[b,c], Kerrie D. Pierce[b], Yi Zhao[d], Xi Luo[d], Youdan Wang[d], David H. Laidlaw[e], Ryan P. Cabeen[e], Thomas E. Conturo[f], David F. Tate[g], Erbil Akbudak[f], Elizabeth M. Lane[h], Jodi M. Heaps[g], Jacob D. Bolzenius[a], Laurie M. Baker[a], Lee M. Cagle[a], Robert H. Paul[a,g]

[a] University of Missouri-St. Louis, Department of Psychological Sciences, 1 University Blvd., Stadler Hall 442A, St. Louis, MO 63121, USA
[b] Neuroscience Research Australia, Barker Street Randwick, Sydney, NSW 2031, Australia
[c] School of Medical Sciences, University of New South Wales, Sydney, NSW 2052, Australia
[d] Brown University, Department of Biostatistics and Center for Statistical Sciences, Providence, RI 02912, USA
[e] Brown University, Computer Science Department, Providence, RI 02912, USA
[f] Washington University School of Medicine, Mallinckrodt Institute of Radiology, 510 S. Kingshighway, St. Louis, MO 63110, USA
[g] Missouri Institute of Mental Health,4633 World Parkway Circle, Berkeley, MO 63134-3115, USA
[h] Vanderbilt University Medical Center, 1211 Medical Center Drive, Nashville, TN 37232, USA

HIGHLIGHTS

- The *AGT* M268T SNP is associated with cardiovascular abnormalities.
- The 268T variant is a risk factor for reduced white matter in healthy adults.
- The superior longitudinal fasciculus and cingulum are vulnerable to 268T.
- Attention/processing speed and language are compromised among TT genotypes.
- White matter and cognition are impacted independent of hyperintensity burden.

ARTICLE INFO

Article history:
Received 5 May 2015
Received in revised form 18 August 2015
Accepted 20 August 2015
Available online 28 August 2015

Keywords:
AGT
M235T
M268T
rs699
White matter
Cognition

ABSTRACT

The common angiotensinogen (*AGT*) M268T polymorphism (rs699; historically referred to as M235T) has been identified as a significant risk factor for cerebrovascular pathologies, yet it is unclear if healthy older adults carrying the threonine amino acid variant have a greater risk for white matter damage in specific fiber tracts. Further, the impact of the threonine variant on cognitive function remains unknown. The present study utilized multiple indices of diffusion tensor imaging (DTI) and neuropsychological assessment to examine the integrity of specific white matter tracts and cognition between individuals with homozygous genotypes of M268T (MetMet $n = 27$, ThrThr $n = 27$). Differences in subcortical hyperintensity (SH) volume were also examined between groups. Results indicated that the threonine variant was associated with significantly reduced integrity in the superior longitudinal fasciculus (SLF) and the cingulate gyrus segment of the cingulum bundle (cingulum CG) compared to those with the methionine variant, and poorer cognitive performance on tests of attention/processing speed and language. Despite these associations, integrity of these tracts did not significantly mediate relationships between cognition and genetic status, and SH did not differ significantly between groups. Collectively our results suggest that the threonine variant of M268T is a significant risk factor for abnormalities in specific white matter tracts and cognitive domains in healthy older adults, independent of SH burden.

1. Introduction

Advanced age is the most common predictor of cerebrovascular disease (CVD) and is evident among the majority of individuals

* Corresponding author. Fax: +1 314 516 5392.
E-mail address: LSalminen@mail.umsl.edu (L.E. Salminen).

http://dx.doi.org/10.1016/j.bbr.2015.08.028
0166-4328/

over the age of 65 [1,2]. CVD is a result of ischemic damage that results in lacunar infarcts and lesions that often aggregate in subcortical white matter of the vascular bed [3,4]. These lesions can be visualized *in vivo* as areas of high signal intensity on T2-weighted MRI [5] or hypointense signals on computed tomography (CT). The most common form of CVD is subcortical ischemic vascular disease, which represents a significant risk factor for vascular dementia [6] and [7]. While the etiological mechanisms of CVD are numerous and diverse, abnormalities in blood pressure are common antecedents of cerebral ischemia [8–11].

Blood pressure is endogenously regulated by localized renin-angiotensin system (RAS) activity across multiple organ tissues [12,13]. This biosynthetic pathway is initiated by an enzyme-substrate reaction between renin and angiotensinogen (AGT) that determines the synthesis rate of the main effector peptide, angiotensin II (AngII) [14]. When bound to the type 1 receptor, AngII causes constriction of the artery wall and a subsequent increase in blood pressure. Chronic elevations in circulating AngII have been associated with vascular factors such as hypertension and atherosclerosis, resulting in upregulation of the RAS and further damage to vessel structure [15–18]. Evidence suggests that genetic expression of the AngII precursor molecule, AGT, may explain a degree of inter-individual variation in AngII concentrations [19,20].

The M268T polymorphism, historically referred to as M235T, (rs699; p.Met268Thr) of the *AGT* gene is a missense mutation whereby a single nucleotide polymorphism in exon 2 (c.803T>C) is predicted to encode threonine instead of methionine at residue 268 of the peptide sequence [21,22]. The threonine variant of M268T has been associated with arterial hypertension, carotid atherosclerosis, coronary heart disease, ischemic heart disease, and ischemic stroke [20,22,23]. In addition to peripheral vascular damage, the 268T allele has been associated with progression of deep subcortical white matter lesions among community-dwelling elderly individuals [24] and increased lesion severity independent of arterial hypertension [25]. Not all studies have identified these relationships, however, as Paternoster et al. [26] revealed no significant relationship between 268T and white matter lesions in a meta-analysis of six studies. Differences in imaging methods may explain the variability in results, as most studies that did not report significant relationships utilized brain imaging techniques with lower sensitivity to white matter lesions compared to studies that reported significant results [27].

While previous investigations of M268T and brain integrity have focused on the development and progression of white matter lesions, evidence suggests that microstructural damage to myelinated axon fibers precedes lesion development [28]. Thus, tract-specific measures of fiber integrity may be more useful for assessing the impact of M268T in healthy older adults. Diffusion tensor imaging (DTI) is a non-invasive imaging technique that provides information about the integrity of white matter fibers by measuring directional properties of water diffusion on two-dimensional (2D) grayscale maps [29]. Scalar metrics such as fractional anisotropy (FA) and mean diffusivity (MD) can be calculated from the diffusion tensor to evaluate the directional restriction and rate of water movement along an axon fiber. Decreased FA and increased MD are indicative of white matter degeneration, likely due to demyelination and/or axonal loss [30].

In addition to scalar metrics, recent studies have shown that length-based diffusion tractography MRI (dtMRI) is sensitive to aging processes [31–36]. This technique combines DTI scalar metrics with traditional tractography methods [37] to computationally reconstruct fiber tract lines [34]. As a result, dtMRI captures subtle changes in white matter microstructure along the entire length of a fiber tract that may not reach a threshold to impact traditional scalar metrics [34,36]. Multimodal utilization of DTI scalar metrics and dtMRI may reveal independent patterns of white matter aging that are influenced by cerebrovascular dysfunction. Further, these methods can provide region-specific insight into the biological consequences of M268T on microstructural white matter integrity.

The purpose of this study was to examine the impact of the *AGT* M268T polymorphism on white matter integrity using DTI in two groups of 27 healthy older adults representing the MetMet (MM) and ThrThr (TT) genotypes. Given the known relationships between cardiovascular health and cognitive status [1,38–40], we additionally examined the impact of M268T on neuropsychological performance. In order to provide a comprehensive assessment of microstructural integrity, white matter was measured using dtMRI and DTI scalar metrics of FA and MD. As a secondary aim, genetic differences in subcortical hyperintensity (SH) volume were also investigated to allow for comparisons to previous research. Participants were grouped according to genotype so that only homozygous individuals with either the MM or TT genotypes were studied in order to preserve power and facilitate interpretation of study outcomes. We hypothesized that individuals with the threonine variant would exhibit poorer cognitive performance compared to those with the methionine variant, and that poor performance would be mediated by abnormalities in white matter microstructure.

2. Methods

2.1. Participants

Data were obtained from 54 healthy, mixed race, older adults (males $n = 22$, females $n = 32$) involved in larger study of cognitive aging. Participants were recruited from the local community using radio and print advertisements that advertised “healthy aging.” A small subset of participants was recruited from the Research Participant Registry of the Washington University Institute of Clinical and Translational Sciences (ICTS).

2.1.1. Inclusion criteria

English-speaking, between the ages of 50–85, and able to complete basic and instrumental activities of daily living (ADLs) according to the Lawton and Brody activities of daily living scale [41].

2.1.2. Exclusion criteria

History of medical or neurological disorder capable of influencing cognition (e.g., multiple sclerosis, thyroid disease, etc.), all Axis I and II psychological disorders with the exception of treated depression, history of significant head injury defined as a loss of consciousness >5 min, past or current substance abuse, treatment-dependent diabetes, a score <24 on the mini mental state examination (MMSE) [42], and contraindications for MRI (e.g., claustrophobia). Blood pressure was recorded as the average of three separate time points during the neuropsychological evaluation. Although we did not exclude individuals who met criteria for hypertension (systolic ≥ 140; diastolic ≥ 90), the frequency distribution of hypertension was examined between groups. A physician visually scanned all images to rule out gross radiological abnormalities (e.g., hydrocephalus) and those with abnormal scans were excluded from the study. Participants provided informed consent prior to completion of study procedures and were financially compensated for their time. All study procedures were approved by the institutional review board (IRB) of the corresponding institutions.

2.2. Genotyping

Saliva samples were collected during the initial neuropsychological evaluation using the Oragene DNA collection kit (DNA