Racial Differences in the Effect of APOE-ε4 Genotypes on Trail Making Test Part B in Alzheimer's disease

Chun Xu (chun.xu@utrgv.edu)
UTRGV: The University of Texas Rio Grande Valley https://orcid.org/0000-0001-7893-6341
Priscila Acevedo
UTRGV: The University of Texas Rio Grande Valley
Yongke Lu
Marshall Academy: Marshall University
Brenda Bin Su
UTRGV: The University of Texas Rio Grande Valley
Victoria Padilla
UTRGV: The University of Texas Rio Grande Valley
Kaysie Ozuna
UTRGV: The University of Texas Rio Grande Valley
Kimberly Moreno
UTRGV: The University of Texas Rio Grande Valley
Annu Karithara
UTRGV: The University of Texas Rio Grande Valley
Chun Xiang Mao
Saint Clair College
Osvaldo Navia
WVU: West Virginia University
Ubolrat Piamjariyakul
WVU: West Virginia University
Kesheng Wang (kesheng.wang@hsc.wvu.edu)
WVU: West Virginia University

Research Article

Keywords: Alzheimer's disease, TMT-B, APOE-ε4 allele, Mixed model, Racial differences

Posted Date: February 18th, 2021

DOI: https://doi.org/10.21203/rs.3.rs-194964/v1

Abstract

The trail making test part B (TMT-B) evaluates "executive" functions, memory, and sensorimotor functions. No previous studies were found to examine the longitudinal effect of apolipoprotein E epsilon 4 (APOE-ε4) genotypes on the TMT-B scores in Alzheimer's disease (AD), and/or mild cognitive impairment (MCI) participants. This study used data from the Alzheimer's Disease Neuroimaging Initiative (ADNI): 482 participants with AD, 503 with cognitive normal (CN), 1,293 with MCI at baseline and follow-up of four years. The multivariable linear mixed model was used to determine the longitudinal changes in the TMT-B scores. The individuals with 1 or 2 *APOE-ε4* alleles revealed significantly higher TMT-B scores (poor cognitive function) compared with individuals without *APOE-ε4* allele at baseline and four follow-up visits (all p values < 0.0001). Compared with Whites, non-Hispanic African American and Hispanic populations had higher TMT-B scores (p = 0.0007 and 0.0044, respectively). Furthermore, stratified by diagnosis, the African American CN group had the highest TMT-B scores (p < 0.0001) and the Hispanic AD group had higher TMT-B scores (p = 0.0123) compared with Whites, while both African American and Hispanic MCI groups had higher TMT-B scores compared with Whites (p = 0.162 and 0.0274, respectively). In addition, stratified by racial groups, the subjects with *APOE-ε4*-homozgous and *APOE-ε4*-heterozgous showed higher TMT-B scores compared with subjects who carry zero *APOE-ε4* allele just in Whites (p = 0.0023 and 0.0003, respectively); whereas there was no difference among *APOE-ε4* genotypes in AA and Hispanics. In conclusion, the *APOE-ε4* allele was associated with increased TMT-B scores but such associations varied by racial groups.

Introduction

Alzheimer's disease (AD), the most common cause of major neurocognitive disorder (dementia), is a chronic neurodegenerative disease with progressive memory problems that are centered around their episodic memory (Lane et al. 2018). AD is recognized for its cognitive decline in two or more areas in the brain, affecting personality, memory, language and behavior (Weller and Budson 2018). Mild Cognitive Impairment (MCI) is the middle stage between normal aging and dementia (Sanford 2017). As the disease progresses and cognitive problems increase, the patients become more debilitated and die within the span of 8.5 years after the disease first presented itself (Lane et al. 2018). It is estimated that 5.8 million Americans will have Alzheimer's disease in 2020 and it is projected to grow exponentially, exceeding 13 million by 2050 (Hebert et al. 2013). The high prevalence is found among older adults from ages of 65 and above (Masters et al. 2015). Recent studies have demonstrated the prevalence of AD in various ethnic populations in the United States. For example, inequalities in dementia incidence between six racial and ethnic groups were identified, with the highest for African Americans and American Indians, intermediate for Latinos, Pacific Islanders, and whites, and the lowest among Asian-Americans (Mayeda et al. 2016). A meta-study showed that AD rates were higher in African Americans than in Caucasian Whites (Steenland et al. 2016).

AD is a multifactorial disease that has a multitude of risk factors that range from environmental to genetic. Non-genetic risk factors associated with AD include age, cardiovascular and lifestyle habits,

diabetes, hypertension, obesity, diet, smoking, and physical activity (Crous-Bou et al. 2017). Genetic factors associated with the development of AD include mutations in the Apolipoprotein E (APOE) gene, amyloid precursor protein (APP) gene, presenilin 1 (PSEN1) gene, and presenilin 2 (PSEN2) gene; while PSEN1 and PSEN2 have been found to cause familial AD, which leads to the development of AD at an earlier age (2016 Alzheimer's disease facts and figures - – 2016 - Alzheimer's & Dementia - Wiley Online Library ; Watanabe and Shen 2017).

The most well-known documented gene associated with late AD is APOE. There are three types of APOE alleles: ε2, ε3, and ε4, which can make different combinations: ε2/ε2, ε2/ε3, ε2/ε4, ε3/ε3, ε3/ε4, and ε4/ε4. The most common APOE allele among all ethnic groups is ε3 (Abondio et al. 2019). In the USA, about 61% of the population carry ε3/ε3, 23% of the population have ε3/ε4, and 11% have ε2/ε3 (2016 Alzheimer's disease facts and figures - – 2016 - Alzheimer's & Dementia - Wiley Online Library). APOE-ε3 helps in maintaining the structural integrity of cholesterol-rich lipoproteins, enhances their solubilization in blood plasma and regulates lipid homeostasis. However, the *APOE* genotype that accounts for most AD risk factors, and AD pathology is APOE-ε4. About 26% of Americans carry APOE-ε4, including ε2/ε4, ε3/ε4, and ε4/ε4 (Breitve et al. 2016). With one copy of APOE-ε4 (ε2/ε4 and ε3/ε4) and two copies of APOE-ε4 (ε4/ε4), risk of developing the disease rises 4-fold and 12-fold, respectively (Spinney 2014). The APOE-ε4 allele is more dominant in African American (AA) and Hispanic populations than in White populations (Abondio et al. 2019), which is consistent with the notion that AA and Hispanic populations have a higher prevalence of AD (Mayeda et al. 2016). APOE-ε4 is a special allele that has been demonstrated to link with neurologic abnormalities, vascular diseases, and aging. There is an acceleration in brain aging when the APOE-ε4 is present. It has been demonstrated that there can be a decline in cognitive thinking if people carry this allele (Müller-Gerards et al. 2019). APOE-ε4 has a greater risk of increasing amyloid beta plaques that build up in the brain and causes the death of neurons that are present in advanced stages of AD (Flowers and Rebeck 2020).

The Trail Making Test (TMT) is a cognitive function test that has been commonly used in both clinical and research settings to assess the progression of AD (Hagenaars et al. 2018). The TMT test is administered in two parts. Part A (TMT-A) consists of a patient connecting alternating numbers that reside in circles (Salthouse 2012). Part B (TMT-B) involves the patient connecting alternating numbers and letters within circles (Llinàs-Reglà et al. 2017). Different studies using the TMT have been able to use genetic analysis to find genes associated with cognitive abilities in diseases such as attention deficit hyperactivity disorder (Zhang et al. 2019). However, no previous studies were found to examine the longitudinal effect of apolipoprotein E epsilon 4 (APOE-ε4) genotypes on the TMT-B scores in Alzheimer’s disease (AD), and/or mild cognitive impairment (MCI) participants. In this study, we examined the longitudinal effect of APOE-ε4 genotype on the TMT-B scores in AD and mild cognitive impairment (MCI) participants with different ethnicities to detect racial differences in the effect.

Materials And Methods

Study subjects

Data used in this article's preparation was obtained from the Alzheimer's Disease Neuroimaging Initiative (ADNI) database (adni.loni.usc.edu). The ADNI was launched in 2003 as a public-private partnership, led by Principal Investigator Michael W. Weiner, MD. The primary goal of ADNI has been to test whether serial magnetic resonance imaging (MRI), positron emission tomography (PET), other biological markers, and clinical and neuropsychological assessment can be combined to measure the progression of MCI and early AD. The ADNI is an ongoing, longitudinal, multicenter study designed to develop clinical, imaging, genetic, and biochemical biomarkers for the early detection and tracking of AD. There was an Institutional Review Board exemption for the current study due to secondary data analysis.

The baseline data included 482 individuals with AD, 1,293 with MCI, and 503 with cognitive normal. Social-demographic factors included gender, age, race, and educational level. Gender was self-reported as either male or female. Age was classified into three groups: ≤ 65 years, 66-75 years, and 76+ years. Race consisted of three subgroups: non-Hispanic White, non-Hispanic African American, and Hispanic. Years of education was classified into ≤ 12 years, 13-16 years, and 17+ years.

Trail Making Test (TMT)-B

The TMT-B provides cognitive flexibility and measures psychomotor processing speed, visual scanning, and attentional set-shifting. An array of numbers and letters were presented to the subjects and they were asked to draw connecting lines while alternating between numbers and letters in sequential order. The results for TMT-B were reported as the number of seconds required to complete the task. The higher scores reveal greater impairment (Reitan and Wolfson 1985; Reitan and Wolfson 1995). The TMT-B were measured longitudinally at 0 (designated as baseline), 12, 24, 36, and 48 months, respectively.

APOE-ε4 genotyping

APOE genotyping was performed on DNA samples obtained from the blood samples from studied subjects using an APOE genotyping kit, as described in http://www.adni-info.org/Scientists/Pdfs/adniproceduresmanual12.pdf (also see http://www.adni-info.org for detailed information on blood sample collection, DNA preparation, and genotyping methods). APOE-ε4 carriers were defined as individuals with at least one ε4 allele (ε4/ ε4 designated as APOE-ε4-2, ε4/ ε3 or ε4/ ε2 as APOE-ε4-1), while non-carriers were defined as individuals with no ε4 allele (APOE-ε4-0) (Table 1).

Statistical Methods

The categorical variables were presented in their raw values along with the proportions for categorical variables, and continuous variables were presented in the form of mean ± SD. The skewness and kurtosis of TMT-B score were 1.06 and -0.18, respectively (Figure 1). For the analysis of baseline data, the Chi-square test was used to examine the associations of categorical variables with racial groups, while one-way ANOVA was performed to determine the differences in continuous variables among racial groups. The linear mixed models (LLMs) were conducted to examine the longitudinal changes in TMT-B as a continuous trait. The APOE-ε4 was entered as fixed effect and time point (baseline and up to 4 years of

follow-up) as random effect, adjusted for age, gender, education, and baseline AD diagnosis. Furthermore, the multivariable LMM analyses were conducted stratified by diagnosis and racial groups. All analyses were performed with SAS version 9.4 (Cary, NC. U.S.A.).

Results

Descriptive statistics by racial groups

Baseline features of the studied subjects are summarized in Table 1. There are gender differences among three ethnic groups. More male subjects were observed in the White population, but more females were noted in AA and Hispanic. In terms of age of the participants, there were more White seniors (75+) as compared to other 2 ethnic groups, i.e. AA and Hispanic (p=0.0007). There were differences in years of education (p=0.0431) among racial groups. There was no difference in diagnosis and APOE-ε4 genotype among the three ethnic groups. TMT-B scores were significantly higher in AA and Hispanic participants than in White participants (p=0.0088).

APOE-ε4with TMT-B by year

The TMT-B evaluations were performed at five time points which include baseline, 12 months, 24 months, 36 months, and 48 months. Detailed data regarding TMT-B scores by five time points and APOE-ε4 alleles are presented in Table 2. The subjects with two APOE-ε4 alleles showed significantly higher TMT-B scores, than subjects with 0 or 1 copies of APOE-ε4 allele, in all time points (all p values <0.0001). Furthermore, Table 2 reveals that the TMT-B scores increased as the APOE-ε4 allele increased from baseline to the 48 months of visits.

Longitudinal analysis of APOE-ε4with TMT-B

The longitudinal changes in TMT-B were examined in the multivariable LMM adjust for baseline characteristics (Table 3). The individuals with 1 or 2 *APOE*-ε4 alleles revealed significantly higher TMT-B scores (β=10.70, p=0.0002; β=15.81, p=0.001, respectively) compared with individuals without the APOE-ε4 allele. Being elder (75+), non-Hispanic African Americans and Hispanic participants were associated with increased TMT-B scores (β=27.03, p<0.0001; β=20.10, p=0.0007, and β= 20.61, p=0.0044, respectively). Individuals with AD and MCI were associated with increased TMT-B scores (both p values <0.0001) compared with cognitive normal (CN). There was no difference in TMT-B scores between males and females. Furthermore, there was a significant time effect: compared with baseline, all follow-up time points showed significantly higher TMT-B scores (all p values <0.0001).

LMM analysis of APOE4 with TMT-B by diagnosis

Multivariable LMM analyses of APOE4-ε4 with TMT-B by diagnosis are presented in Table 4. The APOE-ε4 allele was associated with TMT-B scores in MCI and CN groups. In the MCI group, the subjects with APOE-ε4-2 and APOE-ε4-1 showed higher TMT-B scores (β= 20.79, p=0.0006; β= 14.83, p<0.0001, respectively) compared with subjects who carry zero APOE-ε4 alleles; while in CN group, the subjects with

APOE-ε4-2 showed higher TMT-B scores (β= 27.19, p=0.0051) compared with subjects who carry zero APOE-ε4 alleles. Compared with White, Hispanics but not AA had higher TMT-B scores in AD group, yet AA but not Hispanics had higher TMT-B scores in CN group, while both AA and Hispanics had the higher TMT-B scores in MCI group. Furthermore, being elder (75+) in CN and MCI groups (β= 35.26, p<0.0001; β= 36.86, p<0.0001, respectively) was associated with increased TMT-B scores compared with the younger group (≤65 years old). Higher education was associated with decreased TMT-B scores in CN and MCI groups. Additionally, there was a significant time-response effect in AD and MCI groups: all follow-up time points showed significantly higher TMT-B scores (all *p* values <0.0001).

Racial differences in multivariable LMM analysis of APOE4 with TMT-B

The multivariable LMM analysis of APOE-ε4 stratified by racial groups can be found in Table 5. In Whites, the subjects with APOE-ε4-2 and APOE-ε4-1 showed higher TMT-B scores (β= 15.08, p=0.0023; β= 10.71, p=0.0003, respectively) compared with subjects who carry zero APOE-ε4 allele; whereas, in AA and Hispanics, there was no difference among APOE-ε4 genotypes. Compared with CN, AD was associated with higher TMT-B scores in all racial groups, while MCI was associated with higher TMT-B scores just in Whites and AA. Females were associated with lower TMT-B scores in AA and Hispanics, while elder (75+) was associated with higher TMT-B scores in Whites and AA. Higher education was associated with lower TMT-B scores in all racial groups. Additionally, there was a significant time effect in Whites (all *p* values <0.0001), while time effect was found only at 36 months in AA (p=0.0179) and at 48 months in Hispanics (p=0.0011).

Discussion

TMT-B provides cognitive flexibility measures, such as psychomotor processing speed, visual scanning, and attentional set shifting. Higher TMT-B scores indicate poor cognitive function. TMT-B tests had been used for capturing subclinical dysfunction responsible for gait abnormality in patients with AD and associations between physical performance and executive function in older adults with mild cognitive impairment (McGough et al. 2011). In the current study, using a multivariable linear mixed model analysis we found that higher TMT-B scores (poor visual attention/executive functioning) were associated with lower levels of education, inheriting APOE ε4 allele(s), old age, and ethnical minority among most diagnostic groups. TMT-B scores were higher in AA than in Hispanics and Whites. In term of the different populations with AD, Hispanics had higher TMT-B scores compared with Whites and AA, while in MCI groups, both AA and Hispanics had higher TMT-B scores compared with Whites. Our results are consistent with the report that the White subjects (N = 63) had better TMT-B/A scores as compared with the African American subjects (N = 135) (Gupta et al. 2016). However, no difference was found in cognition between Hispanics and Whites (Díaz-Venegas et al. 2016). The discrepancies between the studies may be due to differences in cognitive measurements (e.g., sampling, sub-populations, and statistical analysis). The present study further added that non-Hispanic AA and Hispanic participants were associated with increased TMT-B scores in the multivariable LMM. Specifically, our results showed that the AA and Hispanics in the MCI group had the higher TMT-B scores, while the Hispanics in the AD

group had higher TMT-B scores compared with Whites; while in CN group, AA had higher TMT-B scores compared with Whites.

APOE-ε4 is the most significant genetic risk factor for AD. Human and animal studies have indicated that APOE-ε4 significantly affects several biological pathways in the brain, and the individuals carrying one or more APOE-ε4 alleles are prone or predisposed to AD development. Indeed, individuals with two copies of APOE-ε4 alleles (homozygous subjects) are at even greater risk, and the odds ratio for developing AD is five times greater compared to individuals with one APOE-ε4 allele (as heterozygous subjects) (Farrer et al. 1997). In addition to the coding region of APOE, non-coding variability in this locus also has independent contribution to the AD development (Zhou et al. 2019). Interestingly, the present study showed that the subjects with one or two APOE-ε4 alleles had significantly higher TMT-B scores compared with subjects without APOE-ε4 allele. More interestingly, this study found that the APOE-ε4 genotype was associated with TMT-B in Whites, however, no difference was observed between APOE-ε4 allele and TMT-B score among the AA and Hispanic populations. Findings of APOE-ε4 allele status associated with TMT-B scores could have a major impact on the understanding, treatment, and prevention of neurodegenerative disorders, such as AD and MCI. Furthermore, we found that the APOE-ε4 allele was associated with TMT-B scores in MCI and CN groups but had no association in the AD group. Those subjects in the AD group may be at an early stage of AD, or they may carry the APOE ε2 allele (as a protective allele). Further studies are needed to explore the association of APOE-ε4 with TMT-B scores under different neurodegenerative diseases.

Our results revealed that as age increased (from baseline to 4 years follow-up), the TMT-B scores increased; while being elder (75+) was associated with increased TMT-B scores in the whole sample, especially in CN and MCI groups, which supports previous findings (Roe et al. 2018). In addition, elder (75+) was associated with increased TMT-B scores in Whites and AA but not in Hispanics. Identification of conditions by the TMT-B test at an early age, such as 50, 60, or 65 years of age has implications for clinical management and preservation of dementia related disorders. Initial management may begin with health counseling and lifestyle modifications such as continuing education (e.g., go to Retirement Learning Center), since higher educated subjects have improved TMT-B scores. Better TMT-B scores increased with greater educational attainment for all ethnic groups and most diagnostic groups, except patients with AD. Our results are consistent with recent reports that better TMT-B scores are observed in those higher levels of education (Gupta et al. 2016; Lipnicki et al. 2019). Thus, the multidisciplinary intervention to prevent or slow the progress of cognitive decline and AD is warranted.

The TMT-B measurements (cognitive flexibility) are used not only for neuro-degenerative disorders (e.g., AD), but also have been reported to assess improvements of other treatments for many diseases and clinical phenotypes, such as acute lymphoblastic leukemia, who have the longest follow-up of survivors (median of 30 years) (Mulrooney et al. 2019), and the cognitive profile in migraine patients (Baschi et al. 2019). A previous study found that the interictal epileptiform discharges (IEDs) in non-rapid eye movement (NREM) were associated with visuospatial and memory impairment and IED led to poorer performance in TMT-B among patients with adult epilepsy; while IEDs were associated with only TMT-B

but not associated with any other cognitive tests (Liu et al. 2016). Our current study, together with the other previous studies, highlights that TMT-B is a highly sensitive, reliable, and effective tool for measurement of cognitive function. The current findings are consistent with one previous study where the authors demonstrated that the TMT-B is highly sensitive in revealing differences in reaction time among not only AD, MCI, and CN groups, but also among the patients with a very mild form of AD (Lin et al. 2016); therefore, TMT-B can reveal obvious impairment of executive functioning. One recent study found that the TMT-B test independently contributed to the prediction of the Clinical Dementia Rating; while individuals who were resilient to an underlying AD pathology tended to be younger and have better performance on the TMT-A and B tests (Roe et al. 2018).

The study has major strengths. First, the linear mixed model addressed time series effects and was adjusted for all covariates. To date, this is the first study to use this statistical method for TMT-B, APOE-ε4, and socio-demographic variables from ADNI data. Second, the findings in such a moderate sample size using the national database generally supports the trajectory of TMT-B and may help to provide specific time points as to when TMT-B scores change to early detection of cognitive decline. So far, there is no cure for patients with AD, thus, early diagnosis of AD, such as MCI or any types of cognitive impairment, is key for early clinical intervention using sensitive tools or measurements, such as the TMT-B tests.

We acknowledge some limitations. First, small sample size for non-White populations (African American and Hispanic), particularly once we further divided into sub-groups (e.g., APOE-ε4 allele status or diagnostic groups), the sample size becomes even smaller, limiting the generalizability of the findings. Therefore, using TMT-B with a large sample size of African American and Hispanic samples for subgroup analysis may be worthwhile for future research. Second, we did not include other modifiable risk factors (such as dietary, lifestyles, and exercise), which has been demonstrated to contribute to cognitive function, in our current study (Lehert et al. 2015).

This study has several clinical implications. It is better to use similar analyzing strategies to examine if the TMT-B scores based on the APOE-ε2 allele dose, may have an opposite trend for different diagnostic groups, levels of education, and ages as compared to APOE-ε4 allele status. As a most recent study demonstrated that the APOE-ε2 allele is associated with an exceptionally low likelihood of AD/dementia (Reiman et al. 2020). In the future, the possibility of combining genetic markers (e.g., APOE alleles) with the TMT-B scores, might offer early diagnosis of AD, therefore leading to early interventional treatment. Since the findings of these combined results complement existing hypothesized models, in which molecular biomarker changes occur prior to structural or cognitive markers in the AD’s pathological process (Jack et al. 2013). A breakthrough study showed a total of nine patients (who carried APOE-ε4 allele) had significant cognitive function improvement after completion of metabolic enhancement for neurodegeneration (MEND) treatment for 5-24 months (Bredesen et al. 2016). Thus, it is promising that with the advance of the science of molecular genetic study and clinical intervention, we might be able to conquer the battle of AD.

In conclusion, APOE-ε4 allele is associated with TMT-B scores and there are racial differences in TMT-B scores, however, the associations of APOE-ε4 with TMT-B scores differed by racial groups. Our findings suggest that TMT-B is a sensitive, reliable, and one of the best predictors for cognitive flexibility measures. The results of our current study show that it is more desirable to use the TMT-B package to assess the level of cognitive impairment for CN, MCI, and AD subjects as well as it being used as a screening tool for detecting AD at an early stage and improve treatment for other cognitive related diseases/phenotypes. Together with previous findings, our results with a moderate sample size again suggest that TMT-B is one of the best indicators of executive function. Further research, particularly on a larger sample size in African American and Hispanic samples seems especially warranted.

Declarations

Note: [a] Data used in preparation of this article were obtained from the Alzheimer's Disease Neuroimaging Initiative (ADNI) database (adni.loni.usc.edu). As such, the investigators within the ADNI contributed to the design and implementation of ADNI and/or provided data but did not participate in analysis or writing of this report. A complete listing of ADNI investigators can be found at: http://adni.loni.usc.edu/wp-content/uploads/how_to_apply/ADNI_Acknowledgement_List.pdf.

Role of the funding sources

No funding source is given for the present paper.

Disclosure

All authors have reported no financial interests or potential conflicts of interest.

Acknowledgement

The present study is a secondary data analysis. The original study and ADNI was funded by the Alzheimer's Disease Neuroimaging Initiative (ADNI) (National Institutes of Health Grant U01 AG024904) and DOD ADNI (Department of Defense award number W81XWH-12-2-0012). ADNI is funded by the National Institute on Aging, the National Institute of Biomedical Imaging and Bioengineering, and through generous contributions from the following: AbbVie, Alzheimer's Association; Alzheimer's Drug Discovery Foundation; Araclon Biotech; BioClinica, Inc.; Biogen; Bristol-Myers Squibb Company; CereSpir, Inc.; Cogstate; Eisai Inc.; Elan Pharmaceuticals, Inc.; Eli Lilly and Company; EuroImmun; F. Hoffmann-La Roche Ltd and its affiliated company Genentech, Inc.; Fujirebio; GE Healthcare; IXICO Ltd.; Janssen Alzheimer Immunotherapy Research & Development, LLC.; Johnson & Johnson Pharmaceutical Research & Development LLC.; Lumosity; Lundbeck; Merck & Co., Inc.; Meso Scale Diagnostics, LLC.; NeuroRx Research; Neurotrack Technologies; Novartis Pharmaceuticals Corporation; Pfizer Inc.; Piramal Imaging; Servier; Takeda Pharmaceutical Company; and Transition Therapeutics. The Canadian Institutes of Health Research is providing funds to support ADNI clinical sites in Canada. Private sector contributions are facilitated by the Foundation for the National Institutes of Health (www.fnih.org). The grantee

organization is the Northern California Institute for Research and Education, and the study is coordinated by the Alzheimer's Therapeutic Research Institute at the University of Southern California. ADNI data are disseminated by the Laboratory for Neuro Imaging at the University of Southern California.

References

2016 Alzheimer's disease facts and figures - - 2016 - Alzheimer's & Dementia - Wiley Online Library

Abondio P et al. (2019) The Genetic Variability of APOE in Different Human Populations and Its Implications for Longevity Genes (Basel) 10 doi:10.3390/genes10030222

Baschi R, Monastero R, Cosentino G, Costa V, Giglia G, Fierro B, Brighina F (2019) Visuospatial learning is fostered in migraine: evidence by a neuropsychological study Neurol Sci 40:2343-2348 doi:10.1007/s10072-019-03973-6

Bredesen DE, Amos EC, Canick J, Ackerley M, Raji C, Fiala M, Ahdidan J (2016) Reversal of cognitive decline in Alzheimer's disease Aging (Albany NY) 8:1250-1258 doi:10.18632/aging.100981

Breitve MH, Hynninen MJ, Brønnick K, Chwiszczuk LJ, Auestad BH, Aarsland D, Rongve A (2016) A longitudinal study of anxiety and cognitive decline in dementia with Lewy bodies and Alzheimer's disease Alzheimer's Research & Therapy 8:3 doi:10.1186/s13195-016-0171-4

Crous-Bou M, Minguillón C, Gramunt N, Molinuevo JL (2017) Alzheimer's disease prevention: from risk factors to early intervention Alzheimer's Research & Therapy 9 doi:10.1186/s13195-017-0297-z

Díaz-Venegas C, Downer B, Langa KM, Wong R (2016) Racial and ethnic differences in cognitive function among older adults in the USA International journal of geriatric psychiatry 31:1004-1012 doi:10.1002/gps.4410

Farrer LA et al. (1997) Effects of Age, Sex, and Ethnicity on the Association Between Apolipoprotein E Genotype and Alzheimer Disease: A Meta-analysis JAMA 278:1349-1356 doi:10.1001/jama.1997.03550160069041

Flowers SA, Rebeck GW (2020) APOE in the normal brain Neurobiology of Disease 136:104724 doi:10.1016/j.nbd.2019.104724

Gupta VK et al. (2016) Disparities in Age-Associated Cognitive Decline Between African-American and Caucasian Populations: The Roles of Health Literacy and Education Journal of the American Geriatrics Society 64:1716-1723 doi:10.1111/jgs.14257

Hagenaars SP et al. (2018) Genetic contributions to Trail Making Test performance in UK Biobank Molecular Psychiatry 23:1575-1583 doi:10.1038/mp.2017.189

Hebert LE, Weuve J, Scherr PA, Evans DA (2013) Alzheimer disease in the United States (2010-2050) estimated using the 2010 census Neurology 80:1778-1783 doi:10.1212/WNL.0b013e31828726f5

Jack CR et al. (2013) Update on hypothetical model of Alzheimer's disease biomarkers Lancet Neurol 12:207-216 doi:10.1016/S1474-4422(12)70291-0

Lane CA, Hardy J, Schott JM (2018) Alzheimer's disease European Journal of Neurology 25:59-70 doi:10.1111/ene.13439

Lehert P, Villaseca P, Hogervorst E, Maki PM, Henderson VW (2015) Individually modifiable risk factors to ameliorate cognitive aging: a systematic review and meta-analysis Climacteric 18:678-689 doi:10.3109/13697137.2015.1078106

Lin Y-C, Hsu W-C, Wu C-K, Chang W-H, Wu KP-H, Wong AMK (2016) Comparison of motor performance of upper and lower extremities in dual-task tests in patients with mild Alzheimer's dementia Aging Clin Exp Res 28:491-496 doi:10.1007/s40520-015-0441-1

Lipnicki DM et al. (2019) Determinants of cognitive performance and decline in 20 diverse ethno-regional groups: A COSMIC collaboration cohort study PLOS Medicine 16:e1002853 doi:10.1371/journal.pmed.1002853

Liu X-Y, Shi T, Yin W-N, Ren Z-Y, Deng Y-L, Chen S-D (2016) Interictal epileptiform discharges were associated with poorer cognitive performance in adult epileptic patients Epilepsy Research 128:1-5 doi:10.1016/j.eplepsyres.2016.09.022

Llinàs-Reglà J, Vilalta-Franch J, López-Pousa S, Calvó-Perxas L, Torrents Rodas D, Garre-Olmo J (2017) The Trail Making Test: Association With Other Neuropsychological Measures and Normative Values for Adults Aged 55 Years and Older From a Spanish-Speaking Population-Based Sample Assessment 24:183-196 doi:10.1177/1073191115602552

Masters CL, Bateman R, Blennow K, Rowe CC, Sperling RA, Cummings JL (2015) Alzheimer's disease Nature Reviews Disease Primers 1:1-18 doi:10.1038/nrdp.2015.56

Mayeda ER, Glymour MM, Quesenberry CP, Whitmer RA (2016) Inequalities in dementia incidence between six racial and ethnic groups over 14 years Alzheimer's & dementia : the journal of the Alzheimer's Association 12:216-224 doi:10.1016/j.jalz.2015.12.007

McGough EL, Kelly VE, Logsdon RG, McCurry SM, Cochrane BB, Engel JM, Teri L (2011) Associations Between Physical Performance and Executive Function in Older Adults With Mild Cognitive Impairment: Gait Speed and the Timed "Up & Go" Test Phys Ther 91:1198-1207 doi:10.2522/ptj.20100372

Müller-Gerards D et al. (2019) Subjective cognitive decline, APOE ε4, and incident mild cognitive impairment in men and women Alzheimers Dement (Amst) 11:221-230 doi:10.1016/j.dadm.2019.01.007

Mulrooney DA et al. (2019) The changing burden of long-term health outcomes in survivors of childhood acute lymphoblastic leukaemia: a retrospective analysis of the St Jude Lifetime Cohort Study The Lancet Haematology 6:e306-e316 doi:10.1016/S2352-3026(19)30050-X

Reiman EM et al. (2020) Exceptionally low likelihood of Alzheimer's dementia in APOE2 homozygotes from a 5,000-person neuropathological study Nature Communications 11:667 doi:10.1038/s41467-019-14279-8

Reitan R, Wolfson D (1985) The Halstead-Reitan Neuropsychological Test Battery: Theory and Interpretattion. The Halstead-Reitan Neuropsychological Test Battery,

Reitan R, Wolfson D (1995) Category test and trail making test as measures of frontal lobe functions 9:50-56

Roe CM et al. (2018) Incident cognitive impairment: longitudinal changes in molecular, structural and cognitive biomarkers Brain 141:3233-3248 doi:10.1093/brain/awy244

Salthouse T (2012) Consequences of Age-Related Cognitive Declines Annu Rev Psychol 63:201-226 doi:10.1146/annurev-psych-120710-100328

Sanford AM (2017) Mild Cognitive Impairment Clinics in Geriatric Medicine 33:325-337 doi:10.1016/j.cger.2017.02.005

Spinney L (2014) Alzheimer's disease: The forgetting gene Nature 510:26 doi:10.1038/510026a

Steenland K, Goldstein FC, Levey A, Wharton W (2016) A Meta-Analysis of Alzheimer's Disease Incidence and Prevalence Comparing African-Americans and Caucasians Journal of Alzheimer's disease : JAD 50:71-76 doi:10.3233/JAD-150778

Watanabe H, Shen J (2017) Dominant negative mechanism of Presenilin-1 mutations in FAD Proc Natl Acad Sci USA 114:12635-12637 doi:10.1073/pnas.1717180114

Weller J, Budson A (2018) Current understanding of Alzheimer's disease diagnosis and treatment F1000Res 7 doi:10.12688/f1000research.14506.1

Zhang K, Fan Z, Wang Y, Faraone SV, Yang L, Chang S (2019) Genetic analysis for cognitive flexibility in the trail-making test in attention deficit hyperactivity disorder patients from single nucleotide polymorphism, gene to pathway level The World Journal of Biological Psychiatry 20:476-485 doi:10.1080/15622975.2017.1386324

Zhou X et al. (2019) Non-coding variability at the APOE locus contributes to the Alzheimer's risk Nature Communications 10 doi:10.1038/s41467-019-10945-z

Tables

Table 1 Descriptive statistics at baseline.

Variable	White	AA	Hispanic	χ^2/F	*P-values*
Gender					
Male	1091	41	36	20.4988	**<0.0001**
Female	898	72	52		
APOE-ε4					
0	1029	49	40	0.5979	0.9633
1	701	32	26		
2	183	8	5		
Age (year)					
≤ 65	231	21	18	19.4095	**0.0007**
65-75	905	58	46		
76+	853	34	24		
Education (year)					
≤ 12	289	24	17	9.8448	**0.0431**
13-16	851	53	42		
17+	849	36	29		
Diagnosis					
CN	449	32	22	2.6999	0.1057
AD	350	18	14		
MCI	1181	60	52		
TMT-B					
Mean ±SD	113.74±72.18	129.84±79.36	135.33±86.63	4.74	**0.0088**

Abbreviations: AA: non-Hispanic African American; CN: Cognitive normal; MCI: Mild Cognitive Impairment; AD: Alzheimer Disease; TMT-B: Trail Making Test Part B; SD: Standard deviation. p value is based on Chi-square test or F test in ANOVA.

Table 2 One-Way ANOVA of TMT-B scores by APOE-ε4 genotype.

Visit	N	APOE-ε4-0 Mean ± SD	APOE-ε4-1 Mean ± SD	APOE-ε4-2 Mean ± SD	F-values	*P* values
Baseline	2190	103.85±66.04	126.10±77.54	138.41±82.87	31.71	**<0.0001**
12 months	1768	109.19±71.98	137.38±86.27	147.09±92.71	28.33	**<0.0001**
24 months	1370	102.43±69.39	131.65±87.27	145.50±93.15	26.86	**<0.0001**
36 months	827	103.94±66.77	127.40±92.78	146.74±92.81	12.73	**<0.0001**
48 months	695	95.75±59.18	133.56±74.48	150.50±91.98	15.03	**<0.0001**

Abbreviations: TMT-B: Trail Making Test Part B; SD: standard deviation. p value is based on F test in one-way ANOVA.

Table 3 Linear mixed model analysis of APOE-ε4 genotype with the TMT-B.

Variable	β ± SE	T-values	*P* values
Gender (ref=male)			
Female	-3.69±2.67	-1.38	0.1669
APOE-ε4 (ref=0)			
1	10.70±2.86	3.74	**0.0002**
2	15.81±4.80	3.29	**0.0010**
Age (ref=<65)			
65-75	3.49±4.31	0.81	0.4181
75+	27.03±4.38	6.17	<**0.000**1
Education (ref =<12)			
13-16	-23.18±3.94	-5.88	<**0.0001**
17+	-31.48±4.01	-7.86	<**0.0001**
Race (ref=White)			
AA	22.10±6.48	3.41	**0.0007**
Hispanic	20.61±7.23	2.85	**0.0044**
Diagnosis (ref=CN)			
AD	110.70±4.47	24.77	<**0.0001**
MCI	25.74±3.23	7.96	<**0.0001**
Visit (ref=baseline)			
12 months	7.17±1.30	5.50	<**0.0001**
24 months	11.43±1.60	7.13	<**0.0001**
36 months	20.96±2.18	9.60	<**0.0001**
48 months	24.54±2.29	10.71	<**0.0001**

Abbreviations: AA: non-Hispanic African American; CN: Cognitive normal; MCI: Mild Cognitive Impairment; AD: Alzheimer Disease; TMT-B: Trail Making Test Part B; β is adjusted regression coefficient, SE is standard error. p value is based on t test in LMM.

Table 4 Linear mixed model analysis of APOE- ε4 genotype with the TMT-B by diagnosis.

Variable	CN β ± SE	*P* values	AD β ± SE	*P* values	MCI β ± SE	*P* values
Gender (ref=male)						
Female	-2.27±3.16	0.4733	2.97±9.35	0.7513	-5.93±3.41	0.0827
APOE-ε4 (ref=0)						
1	3.97±3.55	0.4034	-3.99±10.40	0.7016	14.83±3.56	**<0.0001**
2	27.19±9.67	**0.0051**	-12.10±13.164	0.3583	20.79±6.06	**0.0006**
Age (ref =<65)						
65-75	14.81±8.78	0.0923	-19.80±15.47	0.2015	5.96±4.95	0.2281
75+	35.26±8.92	<**0.0001**	-20.88±14.95	0.1635	36.86±5.09	**<0.0001**
Education (ref=<12)						
13-16	-17.07±5.41	**0.0017**	-10.96±11.14	0.3262	-29.51±5.12	**<0.0001**
17+	-25.56±5.44	**<0.0001**	-17.08±12.48	0.1721	-37.92±5.12	**<0.0001**
Race (ref=White)						
AA	35.26±6.26	<**0.0001**	8.57±23.61	0.7169	21.89±9.09	**0.0162**
Hispanic	2.09±8.63	0.8089	62.41±24.78	**0.0123**	20.62±9.33	**0.0274**
Visit (ref=baseline)						
12 months	-2.46±1.81	0.1740	23.65±5.09	**<0.0001**	6.80±1.56	**<0.0001**
24 months	-0.28±2.18	0.8965	41.05±7.46	**<0.0001**	11.06±1.99	**<0.0001**
36 months	2.36±2.42	0.3307	83.10±16.10	**<0.0001**	23.90±2.86	**<0.0001**
48 months	4.72±2.71	0.0820	92.40±12.54	**<0.0001**	30.59±3.15	**<0.0001**

Abbreviations: AA: non-Hispanic African American; CN: Cognitive normal; MCI: Mild Cognitive Impairment; AD: Alzheimer Disease; TMT-B: Trail Making Test Part B; β is adjusted regression coefficient, SE is standard error. p value is based on t test in LMM

Figures

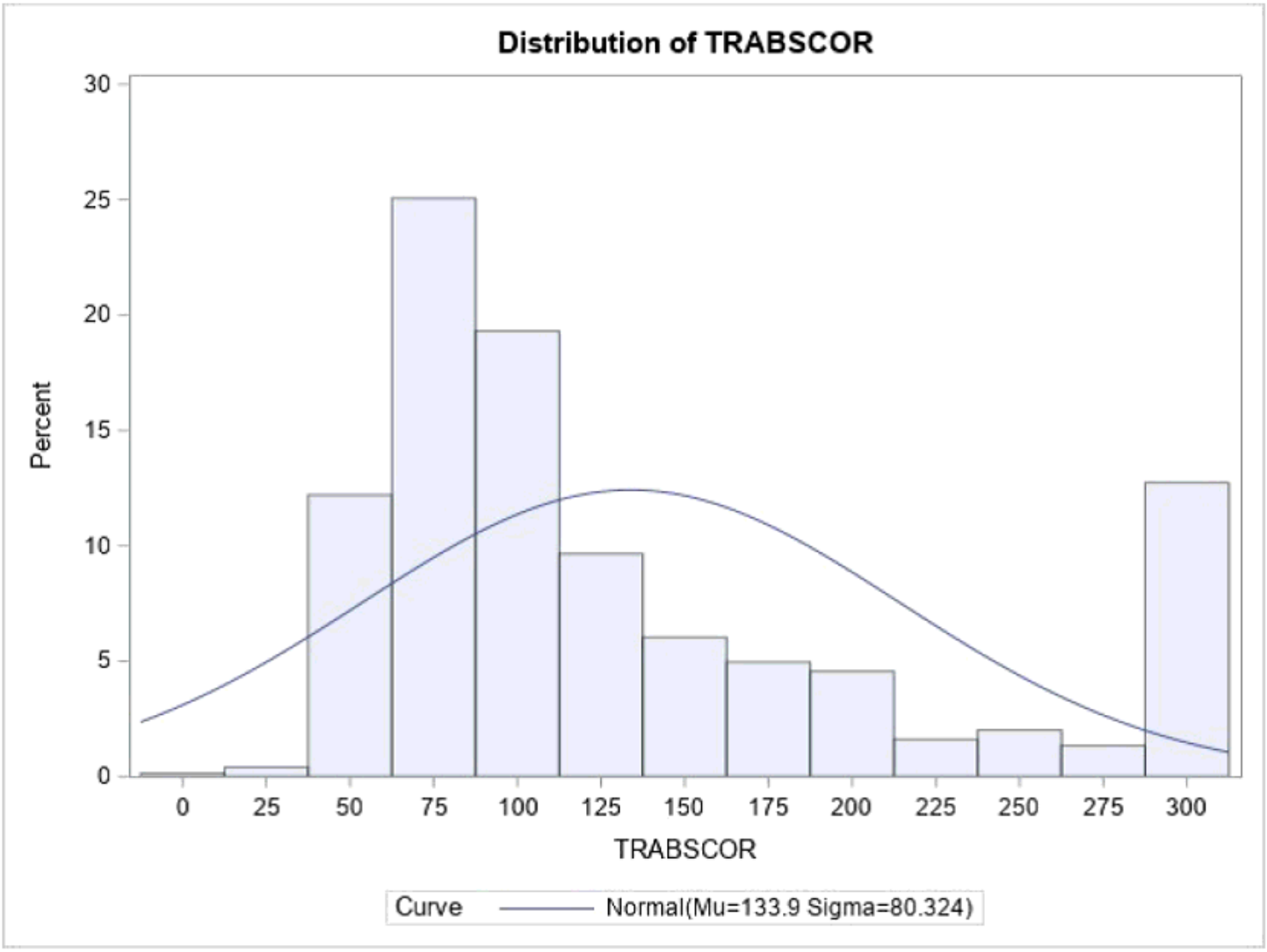

Figure 1

Histogram of transformed Trail Making Test (TMT) Part B. TRABSCOR refers to TMT-B.