Neurobiology of Aging 34 (2013) 2889.e11–2889.e13

Contents lists available at SciVerse ScienceDirect

Neurobiology of Aging

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

Brief communication

Assessment of *TREM2* rs75932628 association with Alzheimer's disease in a population-based sample: the Cache County Study

Josue D. Gonzalez Murcia [a,1], Cameron Schmutz [a,1], Caitlin Munger [a], Ammon Perkes [a], Aaron Gustin [a], Michael Peterson [a], Mark T.W. Ebbert [a,b], Maria C. Norton [c,d], JoAnn T. Tschanz [d], Ronald G. Munger [e], Christopher D. Corcoran [f], John S.K. Kauwe [a,*]

[a] *Department of Biology, Brigham Young University, Provo, UT, USA*
[b] *ARUP Institute for Clinical and Experimental Pathology, Salt Lake City, UT, USA*
[c] *Department of Family Consumer and Human Development, Utah State University, Logan, UT, USA*
[d] *Department of Psychology, Utah State University, Logan, UT, USA*
[e] *Department of Nutrition, Dietetics, and Food Sciences, Utah State University, Logan, UT, USA*
[f] *Department of Mathematics and Statistics, Utah State University, Logan, UT, USA*

ARTICLE INFO

Article history:
Received 7 June 2013
Accepted 14 June 2013
Available online 12 July 2013

Keywords:
Alzheimer's disease
Human
TREM2
Rare variant
R47H

ABSTRACT

Recent studies have identified the rs75932628 (R47H) variant in *TREM2* as an Alzheimer's disease risk factor with estimated odds ratio ranging from 2.9 to 5.1. The Cache County Memory Study is a large, population-based sample designed for the study of memory and aging. We genotyped R47H in 2974 samples (427 cases and 2540 control subjects) from the Cache County study using a custom TaqMan assay. We observed 7 heterozygous cases and 12 heterozygous control subjects with an odds ratio of 3.5 (95% confidence interval, 1.3–8.8; $p = 0.0076$). The minor allele frequency and population attributable fraction for R47H were 0.0029 and 0.004, respectively. This study replicates the association between R47H and Alzheimer's disease risk in a large, population-based sample, and estimates the population frequency and attributable risk of this rare variant.

1. Introduction

Alzheimer's disease (AD) is a fatal neurodegenerative disorder and is the leading cause of dementia in the elderly (Jonsson et al., 2013). It is the sixth leading cause of death in the United States, and of the 6, the only one lacking adequate treatment or prevention. AD is characterized by a progressive loss in cognitive function, and strikes memory early in the course. Neuropathological changes include loss of neurons and synapses, extensive accumulation of amyloid plaques, and neurofibrillary tangles. AD has been identified as a proteopathic disease because of its extensive accumulation of amyloid plaques and neurofibrillary tangles (Guerreiro et al., 2013; Jonsson et al., 2013).

Recent research has focused on finding rare genetic variants that increase risk for AD. Specifically, a rare variant known as rs75932628 (R47H) in exon 2 of the *TREM2* gene has been identified as a risk factor with odds ratio (OR) estimates between 2.9 and 5.1 (Guerreiro et al., 2013; Jonsson et al., 2013). Here we have assessed the R47H variant in a true population-based sample to estimate the relative risk and population-attributable fraction of this variant.

2. Methods

2.1. Subjects

The Cache County Study on Memory Health and Aging is a large population-based study initiated in 1994. The 5092 subjects represented approximately 90% of all residents in Cache County, Utah, aged 65 years or older. For the present study, DNA samples were available for 2974 subjects. A more detailed summation of the Cache County Study has been reported previously (Breitner et al., 1999). Briefly, case-control status was determined using a series of cognitive tests and dementia evaluations that were administered triennially for 4 examination waves. In the first stage of screening, cognitive function was measured using the Modified Mini-Mental State Exam-Revised. Individuals who were screened as positive cases and also a randomly selected subset (19%) of individuals were invited to participate in subsequent stages of evaluation. These consisted of an informant interview and a clinical assessment, reviewed by a geropsychiatrist and a neuropsychologist. At this

* Corresponding author at: Brigham Young University, 675 WIDB, Brigham Young University, Provo, UT 84602, USA. Tel.: (801) 422-2993; fax: (801) 422-0090.
E-mail address: kauwe@byu.edu (J.S.K. Kauwe).
[1] These authors contributed equally to this work.

0197-4580/$ – see front matter © 2013 Elsevier Inc. All rights reserved.
http://dx.doi.org/10.1016/j.neurobiolaging.2013.06.004

time a preliminary diagnosis was assigned as dementia or another cognitive disorder. Those who were diagnosed with dementia were invited to participate in standard laboratory tests for dementia, a magnetic resonance imaging scan, and a geropsychiatrist examination. At the conclusion of these tests, the final cognitive status of participants was determined by an expert panel consisting of neuropsychologists, geropsychiatrists, a neurologist, and a cognitive neuroscientist. By consensus, diagnoses of AD were assigned by National Institute of Neurological and Communicative Disorders and Stroke and the Alzheimer's Disease and Related Disorders Association criteria (McKhann et al., 1984) and included probable AD cases. Cases for this study were those diagnosed with AD only and no comorbid types of dementia. The control subjects were identified as individuals who had negative test results in all examinations and who showed no signs of dementia.

2.2. Genotyping

We performed single-nucleotide polymorphism genotyping for 2974 DNA samples from participants in the Cache County Study for R47H using a custom TaqMan assay. Included in the 2974 samples were 427 clinically ascertained prevalent and incident AD cases and 2540 cognitively normal participants.

2.3. Analysis

Association between R47H and case control status was tested using logistic regression models with adjustment for age, sex, and *APOE* ε4 genotype. We calculated population-attributable fractions using Equation 1 (Ebbert et al., 2013; International Parkinson Disease Genomics Consortium, 2011; Naj et al., 2011), where p equals the allele frequency and OR represents the OR:

$$PAF = \frac{p(OR-1)}{p(OR-1)+1} \quad \text{Eq. 1}$$

3. Results

Our results replicated the significant association between R47H and AD. Of our 2974 samples we observed 7 heterozygous cases and 12 heterozygous controls with an OR of 3.5 (95% confidence interval, 1.3–8.8; $p = 7.6E{-}03$). The minor allele frequency and population-attributable fraction for R47H were 0.0029 and 0.004, respectively.

4. Discussion

Recent studies (Guerreiro et al., 2013; Jonsson et al., 2013) identified an R47H variant in *TREM2* as a risk factor for late-onset AD. Several studies have replicated this work using different ethnic cohorts, study designs, and sample sizes. One study (Pottier et al., 2013) examined the association between the R47H variant and early-onset AD in Caucasian subjects of French origin, consisting of 726 AD cases and 783 control subjects. The study concluded that the variant increases risk for AD with onset earlier than 65 years (OR, 4.07; $p = 0.009$). Another study (Benitez et al., 2013) researched a Spanish population and directly genotyped the R47H variant in 504 AD cases and 550 control subjects. R47H was found in 1.4% (7/504) of AD cases and in 0% (0/550) of control subjects ($p < 0.009$). A familial study (Giraldo et al., 2013) tested a consanguineous Colombian family, that included 3 cases and 5 control subjects, reporting a missense mutation in exon 4 of *TREM2* causing frontotemporal dementia. They also found the R47H variant to increase risk for late- and early-onset AD in 185 AD cases and 183 control subjects (OR, 3.3). Despite the differences in demographics and size, each study concludes that the R47H variant increases AD risk.

Our work further supports that the R47H variant increases risk for late-onset AD. Unlike previous studies, however, the OR and population-attributable fractions estimated here are from a large population-based sample. As such, they better estimate the effects of this allele in the general population. The estimated OR of 3.5 in our study suggests that the increased AD risk for R47H is comparable with the *APOE* ε4 allele. However, because of the low frequency of this variant in the general population, the population-attributable fraction of the R47H variant is very low (0.004 vs. 0.20 for the *APOE* ε4 allele in this sample). Therefore, the population effects are much lower than those of the *APOE* ε4 allele, lessening the potential clinical and diagnostic effect of this discovery. In conclusion, although the population-level effects of the R47H variant do not approach those of the *APOE* ε4 allele, *TREM2* has a clear and replicable effect on AD risk. The loss-of-function caused by the R47H variant contributes to the disruption of an immune response that triggers an inflammatory response leading to neuronal cell death and might contribute to the degeneration of phagocytic pathways that aid in the clearance of neuronal cell debris (Neumann and Daly, 2013). These functions clearly play a role in AD and future preventative and therapeutic efforts can leverage this important new information.

Disclosure statement

The authors report no conflicts of interest.

All data collection and procedures were conducted under IRB approval from Brigham Young University and Utah State University.

Acknowledgements

This work was supported by grants from NIH (R01AG11380, R01AG21136, R01AG31272, R01AG042611), the Alzheimer's Association (MNIRG-11-205368), the Charleston Conference on Alzheimer's Disease and the Brigham Young University Gerontology Program. The authors thank the participants and staff of the Dementia Progression Study, the Utah Population Database, and the Cache County Study on Memory Health and Aging for their important contributions to this work. The funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.

References

Benitez, B.A., Cooper, B., Pastor, P., Jin, S.C., Lorenzo, E., Cervantes, S., Cruchaga, C., 2013. TREM2 is associated with the risk of Alzheimer's disease in Spanish population. Neurobiol. Aging 34, 1711.e15–1711.e17.

Breitner, J.C., Wyse, B.W., Anthony, J.C., Welsh-Bohmer, K.A., Steffens, D.C., Norton, M.C., Tschanz, J.T., Plassman, B.L., Meyer, M.R., Skoog, I., Khachaturian, A., 1999. APOE-epsilon4 count predicts age when prevalence of AD increases, then declines: the Cache County Study. Neurology 53, 321–331.

Ebbert, M., Ridge, P.G., Wilson, A., Sharp, A., Bailey, M., Norton, M., Tschanz, J., Munger, R., Corcoran, C., Kauwe, J.S., 2013. Population-based analysis of Alzheimer's disease risk alleles implicates genetic interactions. Biol. Psychiatry, In press.

Giraldo, M., Lopera, F., Siniard, A.L., Corneveaux, J.J., Schrauwen, I., Carvajal, J., Munoz, C., Ramirez-Restrepo, M., Gaiteri, C., Myers, A.J., Caselli, R.J., Kosik, K.S., Reiman, E.M., Huentelman, M.J., 2013. Variants in triggering receptor expressed on myeloid cells 2 are associated with both behavioral variant frontotemporal lobar degeneration and Alzheimer's disease. Neurobiol. Aging 34, 2077.e11–2077.e18.

Guerreiro, R., Wojtas, A., Bras, J., Carrasquillo, M., Rogaeva, E., Majounie, E., Cruchaga, C., Sassi, C., Kauwe, J.S., Younkin, S., Hazrati, L., Collinge, J., Pocock, J., Lashley, T., Williams, J., Lambert, J.C., Amouyel, P., Goate, A., Rademakers, R., Morgan, K., Powell, J., St George-Hyslop, P., Singleton, A., Hardy, J., Alzheimer

Genetic Analysis Group, 2013. TREM2 variants in Alzheimer's disease. N. Engl. J. Med. 368, 117–127.

International Parkinson Disease Genomics Consortium, Nalls, M.A., Plagnol, V., Hernandez, D.G., Sharma, M., Sheerin, U.M., Saad, M., Simon-Sanchez, J., Schulte, C., Lesage, S., Sveinbjornsdottir, S., Stefansson, K., Martinez, M., Hardy, J., Heutink, P., Brice, A., Gasser, T., Singleton, A.B., Wood, N.W., 2011. Imputation of sequence variants for identification of genetic risks for Parkinson's disease: a meta-analysis of genome-wide association studies. Lancet 377, 641–649.

Jonsson, T., Stefansson, H., Steinberg, S., Jonsdottir, I., Jonsson, P.V., Snaedal, J., Bjornsson, S., Huttenlocher, J., Levey, A.I., Lah, J.J., Rujescu, D., Hampel, H., Giegling, I., Andreassen, O.A., Engedal, K., Ulstein, I., Djurovic, S., Ibrahim-Verbaas, C., Hofman, A., Ikram, M.A., van Duijn, C.M., Thorsteinsdottir, U., Kong, A., Stefansson, K., 2013. Variant of TREM2 associated with the risk of Alzheimer's disease. N. Engl. J. Med. 368, 107–116.

McKhann, G., Drachman, D., Folstein, M., Katzman, R., Price, D., Stadlan, E.M., 1984. Clinical diagnosis of Alzheimer's disease: report of the National Institute of Neurological and Communicative Disorders and Stroke and the Alzheimer's Disease and Related Disorders Association Work Group under the auspices of Department of Health and Human Services Task Force on Alzheimer's Disease. Neurology 34, 939–944.

Naj, A.C., Jun, G., Beecham, G.W., Wang, L.S., Vardarajan, B.N., Buros, J., Gallins, P.J., Buxbaum, J.D., Jarvik, G.P., Crane, P.K., Larson, E.B., Bird, T.D., Boeve, B.F., Graff-Radford, N.R., De Jager, P.L., Evans, D., Schneider, J.A., Carrasquillo, M.M., Ertekin-Taner, N., Younkin, S.G., Cruchaga, C., Kauwe, J.S.K., Nowotny, P., Kramer, P., Hardy, J., Huentelman, M.J., Myers, A.J., Barmada, M.M., Demirci, F.Y., Baldwin, C.T., Green, R.C., Rogaeva, E., St George-Hyslop, P., Arnold, S.E., Barber, R., Beach, T., Bigio, E.H., Bowen, J.D., Boxer, A., Burke, J.R., Cairns, N.J., Carlson, C.S., Carney, R.M., Carroll, S.L., Chui, H.C., Clark, D.G., Corneveaux, J., Cotman, C.W., Cummings, J.L., DeCarli, C., DeKosky, S.T., Diaz-Arrastia, R., Dick, M., Dickson, D.W., Ellis, W.G., Faber, K.M., Fallon, K.B., Farlow, M.R., Ferris, S., Frosch, M.P., Galasko, D.R., Ganguli, M., Gearing, M., Geschwind, D.H., Ghetti, B., Gilbert, J.R., Gilman, S., Giordani, B., Glass, J.D., Growdon, J.H., Hamilton, R.L., Harrell, L.E., Head, E., Honig, L.S., Hulette, C.M., Hyman, B.T., Jicha, G.A., Jin, L.W., Johnson, N., Karlawish, J., Karydas, A., Kaye, J.A., Kim, R., Koo, E.H., Kowall, N.W., Lah, J.J., Levey, A.I., Lieberman, A.P., Lopez, O.L., Mack, W.J., Marson, D.C., Martiniuk, F., Mash, D.C., Masliah, E., McCormick, W.C., McCurry, S.M., McDavid, A.N., Mckee, A.C., Mesulam, M., Miller, B.L., Miller, C.A., Miller, J.W., Parisi, J.E., Perl, D.P., Peskind, E., Petersen, R.C., Poon, W.W., Quinn, J.F., Rajbhandary, R.A., Raskind, M., Reisberg, B., Ringman, J.M., Roberson, E.D., Rosenberg, R.N., Sano, M., Schneider, L.S., Seeley, W., Shelanski, M.L., Slifer, M.A., Smith, C.D., Sonnen, J.A., Spina, S., Stern, R.A., Tanzi, R.E., Trojanowski, J.Q., Troncoso, J.C., Van Deerlin, V.M., Vinters, H.V., Vonsattel, J.P., Weintraub, S., Welsh-Bohmer, K.A., Williamson, J., Woltjer, R.L., Cantwell, L.B., Dombroski, B.A., Beekly, D., Lunetta, K.L., Martin, E.R., Kamboh, M.I., Saykin, A.J., Reiman, E.M., Bennett, D.A., Morris, J.C., Montine, T.J., Goate, A.M., Blacker, D., Tsuang, D.W., Hakonarson, H., Kukull, W.A., Foroud, T.M., Haines, J.L., Mayeux, R., Pericak-Vance, M.A., Farrer, L.A., Schellenberg, G.D., 2011. Common variants at MS4A4/MS4A6E, CD2AP, CD33 and EPHA1 are associated with late-onset Alzheimer's disease. Nat. Genet. 43, 436–441.

Neumann, H., Daly, M.J., 2013. Variant TREM2 as risk factor for Alzheimer's disease. N. Engl. J. Med. 368, 182–184.

Pottier, C., Wallon, D., Rousseau, S., Rovelet-Lecrux, A., Richard, A.C., Rollin-Sillaire, A., Frebourg, T., Campion, D., Hannequin, D., 2013. TREM2 R47H variant as a risk factor for early-onset Alzheimer's disease. J. Alzheimers Dis. 35, 45–49.