Ronald L. Walton, BSc*

Alexandra I. Soto-Ortolaza, BSc*

Melissa E. Murray, PhD

Oswaldo Lorenzo-Betancor, MD, PhD

Kotaro Ogaki, MD, PhD

Michael G. Heckman, MS

Sruti Rayaprolu, BSc

Rosa Rademakers, PhD

Nilüfer Ertekin-Taner, MD, PhD

Ryan J. Uitti, MD

Jay A. van Gerpen, MD

Zbigniew K. Wszolek, MD

Glenn E. Smith, MD

Kejal Kantarci, MD

Val J. Lowe, MD

Joseph E. Parisi, MD

David T. Jones, MD

Rodolfo Savica, MD, PhD

Jonathan Graff-Radford, MD

David S. Knopman, MD

Ronald C. Petersen, MD, PhD

Neill R. Graff-Radford, MD

Tanis J. Ferman, PhD

Dennis W. Dickson, MD

Bradley F. Boeve, MD

Owen A. Ross, PhD

Catherine Labbé, PhD

TREM2 p.R47H SUBSTITUTION IS NOT ASSOCIATED WITH DEMENTIA WITH LEWY BODIES

OPEN

Dementia with Lewy bodies (DLB) is the second leading cause of neurodegenerative dementia in the elderly and is clinically characterized by the presence of cognitive decline, parkinsonism, REM sleep behavior disorder, and visual hallucinations.[1,2] At autopsy, α-synuclein–positive Lewy-related pathology is observed throughout the brain. Concomitant Alzheimer disease–related pathology including amyloid plaques and, to a lesser degree, neurofibrillary tangles are often present.[2] The clinical characteristics of DLB share overlapping features with Alzheimer disease dementia (AD) and Parkinson disease (PD). A recent genetic association study examining known hits from PD and AD identified variants at both the α-synuclein (*SNCA*) and *APOE* loci as influencing the individual risk to DLB.[3] These findings would suggest that DLB may be a distinct disease with shared genetic risk factors with PD and AD.

The p.R47H substitution in the triggering receptor expressed on myeloid cells 2 (TREM2) protein is now a well-established risk factor for AD and has been suggested to play a more general role in other neurodegenerative disorders,[4,5] although these latter findings were not confirmed in a large follow-up study.[6] TREM2 p.R47H has not been evaluated in relation to risk of DLB to date. Therefore, we assessed the frequency of TREM2 p.R47H in a clinical series of patients with DLB and a pathologically defined series of Lewy body disease (LBD) cases defined by the McKeith criteria for likelihood of DLB.[2]

A total of 442 clinical DLB cases, 829 pathologically diagnosed LBD cases, and 1,154 clinical controls were included. Using the consensus criteria,[7] pathologic samples were categorized as high (349), intermediate (254), and low clinical DLB likelihood (226). The clinical cases were enrolled as part of the Mayo Alzheimer's Disease Research Center and the Udall Center for Parkinson's Research, and pathologically diagnosed LBD cases included those from the brain bank for neurodegenerative disorders at Mayo Clinic Jacksonville. Seventy-three clinical DLB cases were also in the pathologic LBD group. Our controls were individuals free of dementia or movement disorder at the time of examination and were included in our previous *TREM2* study.[5] All patients were unrelated Caucasians. Median age at study in controls was 70 years (range: 45–95 years), and 44.6% were male. In patients with clinical DLB, median age at onset was 79 years (range: 49–100 years), and male sex was most common (74.4%). Median age at death in pathologic LBD cases was 80 years (range: 50–100 years), and 56.8% were male. The Mayo Clinic Institutional Review Board approved the study, and all patients or legal next of kin provided written informed consent. Screening of the *TREM2* c.140G>A (rs75932628; p.R47H) was performed using a custom Taqman genotyping assay (Life Technologies, Carlsbad, CA), with a call rate of 100%. Carrier status was confirmed using standard Sanger sequencing. There was no evidence of a departure from Hardy-Weinberg equilibrium in controls (χ^2 $p \geq 0.99$).

Frequencies of TREM2 p.R47H were compared between controls and the different disease groups using unadjusted logistic regression models (unless no p.R47H carriers were observed in which case the Fisher exact test was used). We did not adjust logistic regression models for age or sex because this additional complexity would not be appropriate for this rare variant. The threshold for statistical significance was set at $p \leq 0.05$. PLINK (v1.07) (http://pngu.mgh.harvard.edu/purcell/plink/) was used to perform all statistical analyses.

The frequency of the p.R47H substitution was 0.17% in our controls, which is very similar to what is detected in the ExAC sample population (0.21%) (Cambridge, MA [http://exac.broadinstitute.org]). The age range of the control carriers was 58–78 (table e-1 at Neurology.org/ng), and thus we cannot exclude that some will convert to AD later on. The variant was not observed in the clinical DLB series ($p = 0.58$ vs controls) and was only seen in 1 pathologic LBD case of high-likelihood DLB with a similar frequency (0.14%, $p = 0.86$) to that of healthy controls (table 1). We detected an increased frequency in the pathologically defined LBD cases with intermediate (0.78%, odds ratio [OR] = 4.6, $p = 0.032$) and low (0.88%, OR = 5.18, $p = 0.021$) likelihood DLB in comparison with controls (table 1).

Table 1 Association analysis of TREM2 variant p.R47H in dementia with Lewy bodies and Lewy body disease

Disease group	N	Minor allele frequency, n (%)	OR (95% CI)	p Value
Controls	1,154	4 (0.17)	1.00 (reference)	NA
Clinical DLB	442	0	NA	0.58
Pathologic LBD[2]	829	9 (0.54)	3.16 (0.97-10.31)	0.056
High CDLB likelihood	349	1 (0.14)	0.83 (0.09-7.41)	0.86
Intermediate CDLB likelihood	254	4 (0.78)	4.60 (1.14-18.52)	0.032
Low CDLB likelihood	226	4 (0.88)	5.18 (1.29-20.87)	0.021

Abbreviations: OR = odds ratio; CI = confidence interval; DLB = dementia with Lewy bodies; LBD = Lewy body disease; CDLB = based on McKeith criteria for likelihood of clinical DLB.[2]
ORs, 95% CIs, and p values result from unadjusted logistic regression models. For the clinical DLB vs controls comparison, logistic regression analysis was not possible owing to the lack of patients with clinical DLB who were p.R47H carriers. Therefore, an OR and 95% CI were not given for this comparison and the p value results from the Fisher exact test.

Power to detect associations with p.R47H for each disease group is displayed in table e-2.

We identified only 1 carrier of TREM2 p.R47H in our pathologic high-likelihood DLB series and none in our clinical DLB series. The increased frequency in the cases of intermediate and low likelihood of DLB may well reflect the fact that 5 of these cases are clinical AD for which TREM2 p.R47H is an established risk.[6] In addition, 7 of 9 carriers have predominant AD pathology that categorizes them as having low or intermediate likelihood of clinical DLB. The other 2 carriers have brainstem LBD and diffuse LBD (table e-1). The findings of this study would suggest that the TREM2 p.R47H substitution is not a risk for DLB, leaving AD as currently the only neurodegenerative condition showing association with this TREM2 variant.

*These authors contributed equally to the manuscript.

From the Department of Neuroscience (R.L.W., A.I.S.-O., M.E.M., O.L.-B., K.O., S.R., R.R., N.E.-T., D.W.D., O.A.R., C.L.), Division of Biomedical Statistics and Informatics (M.G.H.), Department of Neurology (N.E.-T., R.J.U., J.A.v.G., Z.K.W., N.R.G.-R.), Department of Psychiatry and Psychology (T.J.F., O.A.R.), Mayo Clinic, Jacksonville, FL; Department of Biology (A.I.S.-O., O.A.R.), University of North Florida, Jacksonville; Department of Psychiatry and Psychology (G.E.S.), Department of Radiology (K.K., V.J.L., D.T.J.), Department of Laboratory Medicine and Pathology (J.E.P., B.F.B.), Department of Neurology (J.E.P., D.T.J., R.S., J.G.-R., D.S.K., R.C.P.), Mayo Clinic, Rochester, MN; Department of Clinical and Health Psychology (G.E.S.), University of Florida, Gainesville; and Mayo Graduate School (T.J.F., O.A.R.), Neurobiology of Disease, Jacksonville, FL.

Author contributions: Ronald L. Walton, Alexandra I. Soto-Ortolaza, Melissa E. Murray, Michael G. Heckman, and Sruti Rayaprolu: revising the manuscript for content, analysis and interpretation of data. Oswaldo Lorenzo-Betancor, Kotaro Ogaki, and Nilüfer Ertekin-Taner: revising the manuscript for content. Rosa Rademakers, Ryan J. Uitti, Jay A. van Gerpen, Zbigniew K. Wszolek, Glenn E. Smith, Kejal Kantarci, Val J. Lowe, Joseph E. Parisi, David T. Jones, Rodolfo Savica, Jonathan Graff-Radford, David S. Knopman, Ronald C. Petersen, Neill R. Graff-Radford, Tanis J. Ferman, and Bradley F. Boeve: revising the manuscript for content, acquisition of data. Dennis W. Dickson: revising the manuscript for content, analysis or interpretation of data, acquisition of data. Owen A. Ross and Catherine Labbé: drafting/revising the manuscript for content, study concept and design, analysis and interpretation of data.

1. Ferman TJ, Boeve BF, Smith GE, et al. Inclusion of RBD improves the diagnostic classification of dementia with Lewy bodies. Neurology 2011;77:875–882.

2. McKeith IG, Galasko D, Kosaka K, et al. Consensus guidelines for the clinical and pathologic diagnosis of dementia with Lewy bodies (DLB): report of the consortium on DLB international workshop. Neurology 1996;47:1113–1124.

3. Bras J, Guerreiro R, Darwent L, et al. Genetic analysis implicates APOE, SNCA and suggests lysosomal dysfunction in the etiology of dementia with Lewy bodies. Hum Mol Genet 2014;23:6139–6146.

4. Benitez BA, Cruchaga C. TREM2 and neurodegenerative disease. N Engl J Med 2013;369:1567–1568.

5. Rayaprolu S, Mullen B, Baker M, et al. TREM2 in neurodegeneration: evidence for association of the p.R47H variant with frontotemporal dementia and Parkinson's disease. Mol Neurodegener 2013;8:19.

6. Lill CM, Rengmark A, Pihlstrom L, et al. The role of TREM2 R47H as a risk factor for Alzheimer's disease, frontotemporal lobar degeneration, amyotrophic lateral sclerosis, and Parkinson's disease. Alzheimers Dement 2015;11:1407–1416.

7. McKeith IG, Dickson DW, Lowe J, et al. Diagnosis and management of dementia with Lewy bodies: third report of the DLB Consortium. Neurology 2005;65:1863–1872.