View metadata, citation and similar papers at core.ac.uk

brought to you by CORE

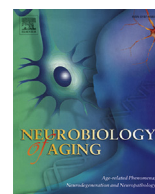
provided by Elsevier - Publisher Connector

Neurobiology of Aging 35 (2014) 2881.e1–2881.e6

Contents lists available at ScienceDirect

Neurobiology of Aging

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



# Investigating the role of rare coding variability in Mendelian dementia genes (*APP*, *PSEN1*, *PSEN2*, *GRN*, *MAPT*, and *PRNP*) in late-onset Alzheimer's disease

Celeste Sassi [a,b,*], Rita Guerreiro [a,b], Raphael Gibbs [a,b], Jinhui Ding [b], Michelle K. Lupton [c], Claire Troakes [c], Safa Al-Sarraj [c], Michael Niblock [c], Jean-Marc Gallo [c], Jihad Adnan [c], Richard Killick [c], Kristelle S. Brown [d], Christopher Medway [d], Jenny Lord [d], James Turton [d], Jose Bras [a], Alzheimer's Research UK Consortium [1], Kevin Morgan [d], John F. Powell [c], Andrew Singleton [b], John Hardy [a]

[a] *Department of Molecular Neuroscience, UCL Institute of Neurology, University College London, London, UK*
[b] *Laboratory of Neurogenetics, National Institute on Aging, National Institutes of Health, Bethesda, MD, USA*
[c] *King's College London Institute of Psychiatry, London, UK*
[d] *Translation Cell Sciences-Human Genetics, School of Life Sciences, Queens Medical Centre, University of Nottingham, Nottingham, UK*

ARTICLE INFO

*Article history:*
Received 20 April 2014
Received in revised form 6 June 2014
Accepted 7 June 2014
Available online 16 June 2014

*Keywords:*
Alzheimer's disease
Neurodegenerative dementia
*APP*
*PSEN1*
*PSEN2*
*MAPT*
*GRN*
*PRNP*
Exome sequencing

ABSTRACT

The overlapping clinical and neuropathologic features between late-onset apparently sporadic Alzheimer's disease (LOAD), familial Alzheimer's disease (FAD), and other neurodegenerative dementias (frontotemporal dementia, corticobasal degeneration, progressive supranuclear palsy, and Creutzfeldt-Jakob disease) raise the question of whether shared genetic risk factors may explain the similar phenotype among these disparate disorders. To investigate this intriguing hypothesis, we analyzed rare coding variability in 6 Mendelian dementia genes (*APP*, *PSEN1*, *PSEN2*, *GRN*, *MAPT*, and *PRNP*), in 141 LOAD patients and 179 elderly controls, neuropathologically proven, from the UK. In our cohort, 14 LOAD cases (10%) and 11 controls (6%) carry at least 1 rare variant in the genes studied. We report a novel variant in *PSEN1* (p.I168T) and a rare variant in *PSEN2* (p.A237V), absent in controls and both likely pathogenic. Our findings support previous studies, suggesting that (1) rare coding variability in *PSEN1* and *PSEN2* may influence the susceptibility for LOAD and (2) *GRN*, *MAPT*, and *PRNP* are not major contributors to LOAD. Thus, genetic screening is pivotal for the clinical differential diagnosis of these neurodegenerative dementias.

© 2014 Elsevier Inc. Open access under CC BY license.

## 1. Introduction

Alzheimer's disease (AD) (OMIM #104310) is the most common cause of progressive dementia in the elderly individuals. Aging and genetic factors play a pivotal role for the disease development. AD incidence increases exponentially from the age of 65 years (1.5% affected) to 80 years and older (30% affected). Twin studies have shown that AD heritability ranges between 60% and 80% (Bergem et al., 1997; Gatz et al., 2006; Raiha et al., 1996). Fully penetrant mutations in amyloid precursor protein (*APP*) and presenilins (*PSEN1* and *PSEN2*) are known to cause familial autosomal dominant AD. The *APOE* ε4 allele is the main risk factor for apparently sporadic AD. In the last 5 years, genome-wide association studies (GWASs) identified several loci, harboring common variants with low risk effect size (OR: 1.2–1.5) (Harold et al., 2009; Hollingworth

* Corresponding author at: Neurogenetics Laboratory, National Institutes of Health, 35 Convent Drive, Bethesda, MD 20892, USA and UCL Institute of Neurology Queen Square London WC1N 3BG, UK. Tel.: +44 (0)20 3456 7890; fax: +44 (0)20 7278 5069.
*E-mail address:* celeste.sassi.10@ucl.ac.uk (C. Sassi).

[1] The Alzheimer's Research UK (ARUK) Consortium: Peter Passmore, David Craig, Janet Johnston, Bernadette McGuinness, Stephen Todd, Queen's University Belfast, UK; Reinhard Heun, Royal Derby Hospital, UK; Heike Kölsch, University of Bonn, Germany; Patrick G. Kehoe, University of Bristol, UK; Nigel M. Hooper, University of Leeds, UK; Emma R.L.C. Vardy, University of Newcastle, UK; David M. Mann, Stuart Pickering-Brown, University of Manchester, UK; Kristelle Brown, James Lowe, Kevin Morgan, University of Nottingham, UK; A. David Smith, Gordon Wilcock, Donald Warden, University of Oxford (OPTIMA), UK; Clive Holmes, University of Southampton, UK.

0197-4580/© 2014 Elsevier Inc. Open access under CC BY license.
http://dx.doi.org/10.1016/j.neurobiolaging.2014.06.002

et al., 2011; Lambert et al., 2009; Lambert et al., 2013; Naj et al., 2011; Seshadri et al., 2010).

Recently, next generation sequencing has led to enormous progress in AD genetics, with the discovery of 2 rare significant risk factors, mapping to *TREM2* (p.R47H) and *PLD3* (p.V232M), and a very rare protective variant in *APP* (p.A637T) (Cruchaga et al., 2013; Guerreiro et al., 2013; Jonsson et al., 2012). In addition, *C9orf72* repeat expansion has been reported in a few patients with clinical AD (Majounie et al., 2012).

The overlapping clinical and neuropathologic features between AD and other neurodegenerative dementias (frontotemporal dementia [FTD], corticobasal degeneration [CBD], progressive supranuclear palsy [PSP], and Creutzfeldt-Jakob disease [CJD]) lead to a misdiagnosis in 17%–30% of AD cases (Beach et al., 2012). This raises the question of whether genetic risk factors relevant in such dementias may play a role in late-onset Alzheimer's disease (LOAD). GWASs have shown that common noncoding variability in Mendelian dementia genes (*APP*, *PSEN1*, *PSEN2*, *MAPT*, *GRN*, and *PRNP*) does not influence susceptibility to AD. By contrast, a growing body of evidence highlighted the significant role of rare coding variants in *PSEN1* in LOAD (Benitez et al., 2013; Cruchaga et al., 2012). Thus, to test the hypothesis that rare coding variability in genes relevant for familial Alzheimer's disease (FAD) and other types of dementia (*APP*, *PSEN1*, *PSEN2*, *MAPT*, *GRN*, and *PRNP*) may underlie LOAD pathogenesis, we have analyzed exome sequencing data, in a British cohort composed of 141 LOAD cases without any apparent family history and 179 elderly controls autopsy proven.

## 2. Methods

### 2.1. Cases and controls

Our cohort was composed of 141 independent LOAD (age at onset ≥65 years) cases and 179 elderly (>60 years) unrelated controls, neuropathologically confirmed. These patients were referred as apparently sporadic LOAD cases.

All the patients and controls were Caucasian, mostly from the UK (London, Manchester, Nottingham, and Edinburgh) and to a lesser extent from North America. The average age at diagnosis was 76.7 years (range 65–97 years) for the LOAD patients and the mean age of ascertainment was 78 years (range 60–102 years) for the controls (Table 1).

Written informed consent was obtained for each individual and the study was approved by the appropriate institutional review boards.

### 2.2. Exome sequencing

Library preparation for next-generation sequencing was performed according to the NimbleGen (Roche NimbleGen v2) and TruSeq (Illumina) sample-preparation protocols. DNA libraries were then hybridized to exome-capture probes with NimbleGen SeqCap EZ Human Exome Library, version 2.0 (Roche NimbleGen) or TruSeq (Illumina). Each capture method covers the *APP*, *PSEN1*, *PSEN2*, *GRN*, *MAPT*, and *PRNP* loci. Exome-enriched libraries were sequenced on the Illumina HiSeq 2000 using 2 × 100 bp paired end read cycles.

### 2.3. Bioinformatics

Sequence alignment and variant calling were performed against the reference human genome (UCSC hg19). Paired end sequence reads (2 × 100 bp paired end read cycles) were aligned using the Burrows-Wheeler aligner (Li and Durbin, 2009). Format conversion and indexing were performed with Picard (www.picard.sourceforge.net/index.shtml). The Genome Analysis Toolkit was used to recalibrate base quality scores, perform local realignments around indels and to call and filter the variants (McKenna et al., 2010). VCFtools was used to annotate gene information for the remaining novel variants. We used ANNOVAR software to annotate the variants (Wang et al., 2010). Variants were checked against established databases (1000 Genomes Project and dbSNP v.134). The protein coding effects of variants were predicted using SIFT, Polyphen2, and SeattleSeq Annotation (gvs.gs.washington.edu/SeattleSeqAnnotation). All variants within the coding regions of *APP*, *PSEN1*, *PSEN2*, *MAPT*, *GRN*, and *PRNP* were annotated for both cases and controls.

### 2.4. Sanger sequencing

All rare variants identified by whole exome sequencing in the candidate genes were validated by Sanger sequencing.

Primers for exons harboring rare variants were designed in Primer3 (http://bioinfo.ut.ee/primer3-0.4.0/) using UCSC (http://genome.ucsc.edu/) reference sequences NM_000484.3 (*APP*), NM_000021.3 (*PSEN1*), NM_000447.2 (*PSEN2*), NM_001123066.3 (*MAPT*), NM_002087.2 (*GRN*), and NM_000311.3 (*PRNP*).

Purified sequences were analyzed on an ABI 3730 DNA Analyzer (Applied Biosystems, CA, USA) and electropherograms were visualized in Sequencher software (version 4.2 Gene Codes Corporation, MI, USA).

### 2.5. Apoe genotyping

*APOE* genotypes comprising the *APOE* ε2, ε3, and ε4 alleles were assayed using the TaqMan method (Applied Biosystems Inc [ABI], Foster City, CA, USA). SNP-specific primers and probes were designed by ABI (TaqMan genotyping assays).

## 3. Results

We identified 226 variants (nonsynonymous, synonymous, intronic, and UTRs) and 18 indels (coding and intronic) in the genes studied. Of these, we analyzed the 18 rare coding variants (minor allele frequency <1%), 1 splice-site mutation (*MAPT* c.115−2A>T), 1 low frequency and 1 common coding polymorphisms in *PRNP*: a 24 bp deletion (rs138688873) and the p.M129V (rs1799990), respectively. In our cohort, 14 LOAD cases (10%) and 11 controls (6%) carry at least one of these rare variants (Table 2). We detected 5 novel variants: 3 present in cases (*APP* p.Y538H, *PSEN1* p.I168T, and *MAPT* c.115-2A>T) and 2 in controls (*MAPT* p.G200E and *PRNP* p.M134V).

*PRNP* and *APP* harbor an higher relative proportion of rare coding variants in controls (1.3/Kb and 1.2/Kb, respectively), compared to cases (0/Kb and 0.4/Kb, respectively), thus, suggesting that rare coding variability in these genes may be well tolerated (Table 3). On

**Table 1**
Cohort

| Cohort | n | Diagnosis | Sequencing strategy | Age (y) mean ± SD (range) | Male (%) | *APOE* e4+ (%) |
|---|---|---|---|---|---|---|
| LOAD CASES | 141 | Clinical and neuropathologic | Exome sequencing | 76.7 (65–97) | 42 | 62 |
| CONTROLS | 179 | Clinical and neuropathologic | Exome sequencing | 78 (60–102) | 55 | 40.7 |

Key: LOAD, late-onset Alzheimer's disease; SD, standard deviation.

**Table 2**
Rare variants found in *APP*, *PSEN1*, *PSEN2*, *MAPT*, *GRN*, *PRNP* in 141 LOAD cases and 179 controls

| Variant interpretation | Gene | Position | Nucleotide change | Aa change | Minor allele | status | SIFT/Polyphen | LOAD cases (n = 141) | | | | Comment | CONTROLS (n = 179) | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | | | | | Count (%) | (AAO–AAD) | Genotype | *APOE* | | Count (%) | Genotype | *APOE* |
| *PROBABLE PATHOGENIC** | | | | | | | | | | | | | | | |
| | *PSEN1* | 14:73653583 | c.503T>C | p.I168T | C | novel | possibly-damaging | 1 (0.7) | 86y-94y | T/C | ε2ε4 | p.I168del reported in FAD | 0 | - | - |
| | *PSEN2* | 1:227076673 | c.710 C>T | p.A237V | T | rs200670135 | possibly-damaging | 1 (0.7) | 87y-95y | C/T | ε3ε3 | Homologous residue in *PSEN1* (p.A231) | 0 | - | - |
| *LIKELY RARE BENIGN POLYMORPHISMS* | | | | | | | | | | | | | | | |
| | *APP* | 21:27423376 | c.602 C>T | p.A201V | T | rs149995579 | tolerated | 0 | - | - | - | EXON 5 | 1 (0.5) | C/T | ε3ε4 |
| | | 21:27326979 | c.1612 T>C | p.Y538H | C | novel | possibly-damaging | 1 (0.7) | 69y-77y | T/C | ε3ε4 | EXON 13 | 0 | - | - |
| | | 21:27326907 | c.1684 G>A | p.V562I | A | rs199586073 | tolerated | 0 | - | - | - | EXON 13 | 1 (0.5) | G/A | ε3ε3 |
| | | 21:27284167 | c.1795 G>A | p.E599K | A | rs140304729 | possibly-damaging | 0 | - | - | - | EXON 14 | 1 (0.5) | G/A | ε3ε4 |
| | *PSEN2* | 1:227071448 | c.184 C>T | p.R62C | T | rs150400387 | possibly-damaging | 1 (0.7) | 83y-91y | C/T | ε3ε3 | N-Terminal | 0 | - | - |
| | | 1:227071449 | c.185 G>A | p.R62H | A | rs58973334 | tolerated | 1 (0.7) | 75y-89y | G/A | ε3ε3 | N-Terminal | 0 | - | - |
| | | 1:227073271 | c.389 C>T | p.S130L | T | rs63750197 | possibly-damaging | 1 (0.7) | 69y-77y | C/T | ε3ε3 | | 1 (0.5) | C/T | ε2ε2 |
| | | 1:227083249 | c.1316 A>C | p.D439A | C | rs63750110 | possibly-damaging | 1 (0.7) | 75y-89y | A/C | ε3ε3 | C-Terminal | 1 (0.5) | A/C | ε3ε3 |
| | *GRN* | 17:42428954 | c.970 G>A | p.A324T | A | rs63750541 | tolerated | 0 | - | - | - | | 2 (1.1) | G/A | ε3ε3, ε3ε2 |
| | | 17:42429497 | c.1294 C>T | p.R432C | T | rs63750130 | tolerated | 1 (0.7) | 94y | C/T | ε3e4 | | 0 | - | - |
| | | 17:42429500 | c.1297 C>T | p.R433W | T | rs63750412 | possibly-damaging | 1 (0.7) | 69y-81y | C/T | ε4ε4 | | 0 | - | - |
| | | 17:42430128 | c.1744 G>A | p.A582T | A | rs72824737 | tolerated | 0 | - | - | - | | 1 (0.5) | G/A | ε3ε3 |
| | *MAPT* | 17:44068824 | c.115-2A>T | frameshift | T | novel | possibly-damaging | 1 (0.7) | 81y-89y | A/T | ε4ε4 | | 0 | - | - |
| | | 17:44060841 | c.671 T>G | p.V224G | G | rs141120474 | possibly-damaging | 2 (1.4) | 74y-82y; 88y- | T/G | ε2ε3; ε2ε3 | | 1 (0.5) | T/G | ε3ε2 |
| | | 17:44060807 | c.637 G>A | p.G213R | A | rs76375268 | possibly-damaging | 2 (1.4) | 74y-82y; 75y- | G/A | ε3ε4; ε3ε3 | | 0 | - | - |
| | | 17:44060769 | c.599 G>A | p.G200E | A | novel | possibly-damaging | 0 | - | - | - | | 1 (0.5) | G/A | ε3ε3 |
| | *PRNP* | 20:4680266 | c.400 A>G | p.M134V | G | novel | possibly-damaging | 0 | - | - | - | | 1 (0.5) | A/G | ε3ε2 |
| *LIKELY LOW FREQUENCY AND COMMON BENIGN POLYMORPHISMS* | | | | | | | | | | | | | | | |
| | *PRNP* | 20:4680094-4680118 | | delACAGCC TCATGGTGG TGGCTGGGG | delACAGCC TCATGGTGG TGGCTGGGG | rs138688873 | possibly-damaging | 2 (1.4) | 80y-88y; 76y-83y | delACAG CCTCATGG TGGTGGC TGGGG | ε3ε3; ε3ε3 | | 0 | - | - |
| | | 20:4680251 | c.385 A>G | p.M129V | G | rs1799990 | tolerated | 64 (45) | | A/G | | | 68 (38) | A/G | |

Rare variants in causative genes for the monogenic forms of neurodegenerative dementias (AD, FTD, PSP, CBD, CJD): amyloid precursor protein, *APP* (NM_000484.3); presenilins 1and 2, *PSEN1* (NM_000021.3) and *PSEN2* (NM_000447.2); progranulin, *GRN* (NM_002087.2); microtubule associated protein Tau, *MAPT* (NM_001123066.3); prion protein, *PRNP* (NM_000311.3).
Key: AD, Alzheimer's disease; AAD, age at death; AAO, age at onset; CJD, Creutzfeldt-Jakob disease; FAD, familial Alzheimer's disease; FTD, frontotemporal dementia; PSP, progressive supranuclear palsy; Aa, amino acid.
* Classification based on the algorithm proposed by Guerreiro et al., 2010a.

**Table 3**
Relative frequency of rare variants (rare variants for Kb of coding sequence) in late-onset AD (LOAD) cases and controls (CTRLS) in *APP*, *PSEN1*, *PSEN2*, *MAPT*, *GRN*, *PRNP*

| Gene | LOAD cases (n = 141) | CTRLS (n = 179) |
|---|---|---|
| *APP* | 0.4/Kb | 1.2/Kb |
| *PSEN1* | 0.6/Kb | 0/Kb |
| *PSEN2* | 3.7/Kb | 1.5/Kb |
| *MAPT* | 1.3/Kb | 0.8/Kb |
| *GRN* | 1.1/Kb | 1.1/Kb |
| *PRNP* | 0/Kb | 1.3/Kb |

the other hand, no controls carry any rare variant in *PSEN1*, suggesting that an amino acid change in PS1 is frequently pathogenic.

In our LOAD cohort, we identified a total of 9 rare coding variants in *APP*, *PSEN1*, *PSEN2*, *MAPT*, *GRN*, and *PRNP*, absent in controls. Of these, 2 are likely to be pathogenic, one in *PSEN1* (p.I168T) and the other in *PSEN2* (p.A237V). In contrast, the variants detected in *APP*, *GRN*, *MAPT*, and *PRNP* are likely tolerated polymorphisms.

Several lines of evidence suggest that p.I168T in *PSEN1* is a deleterious change. First, it clusters in the third transmembrane domain (TM3), on the alpha helix surface, where all the known pathogenic variants have been reported (alpha-helix rule) (Hardy and Crook, 2001). Second, a 4 bp inframe deletion (g.38798_38800delTAT, ΔI167; ΔI168) has already been described in a British family with early-onset Alzheimer's disease (Janssen et al., 2003). The patient carrying this variant (*PSEN1* p.I168T) was diagnosed at 86 years of age, heterozygous for *APOE* ε4 allele (ε2ε4), presented an advanced Alzheimer's disease (Braak V), and did not report any positive family history.

The *PSEN2* p.A237V has been only recently reported by the ClinSeq pilot study (Biesecker et al., 2009) and is likely to be a functional variant with a probable deleterious effect. It clusters on the alpha helix surface of the fifth transmembrane domain (TM5), corresponds to a conserved residue among different species and in *PSEN1* (p.A231), where 2 causative mutations (p.A231V and p.A231T) have been described in a Dutch, French and Canadian family (Campion et al., 1999; Cruts et al., 1998; Rogaeva et al., 2001). The patient carrying the p.A237V variant was diagnosed at 87 years, homozygous for *APOE* ε3 allele, and did not refer any family history of AD.

The other variants detected in our cohort in *PSEN2* (p.R62C, p.R62H, p.S130L, and p.D439A) do not cluster in any TM domain. In addition, the p.S130L and p.D439A have been found also controls.

*GRN* harbors 4 missense mutations (p.A324T, p.R432C, p.R433W, and p.A582T), 2 of which have only been detected in cases (p.R432C and p.R433W). Although the p.R432C variant has been already associated with familial FTD and clinical AD (Brouwers et al., 2008; Cruchaga et al., 2012; Shankaran et al., 2008), its pathogenic role remains unclear. By contrast, the p.R433W has been reported as a nonpathogenic variant (www.molgendatabase).

The variants detected in *MAPT* (c.115-2A>T, p.V224G, p.G213R and p.G200E) cluster outside the microtubule binding domain, where most of the pathogenic mutations have been reported up to date (www.molgendatabase). The c.115-2A>T is predicted to alter Tau exon 7 splicing, introducing a nonsense codon within exon 11. We have not detected any difference in exon 7 and in *MAPT* expression between the splice-site mutation carrier and the other cases and controls. Thus, these findings suggest that c.115-2A>T is not disease related (Supplementary data).

The *MAPT* p.G213R has been found in 2 cases and is absent from the controls. It has been described as possibly damaging by *in silico* predictions and clusters close to a pivotal phosphorylation site (S214) for the serum and glucocorticoid inducible kinase 1 (SGK1). SGK1 controls neurite outgrowth by depolarizing the microtubules through the serine phosphorylation at codon 214 (Yang et al., 2006).

Two cases and no controls carry a 24 bp deletion in the *PRNP* open reading frame (rs138688873), between the repeat 3 (R3) and repeat 4 (R4). This octapeptide deletion (rs138688873) has been described as a risk factor for neurodegenerative diseases with some controversy (Palmer et al., 1993; Perry et al., 1995). Furthermore, although these 2 patients were homozygous for *PRNP* p.M129M, a common polymorphism and risk factor for sporadic Creutzfeldt-Jakob disease (sCJD) (Palmer et al., 1991), p.M129M was not significantly associated with AD in this study ($p$-value = 0.22, OR = 0.75). Thus, suggesting this common variant may be a benign polymorphism.

Finally, we report a significant association between *APOE* ε4 allele and LOAD in our cohort ($p$-value = 0.0002, OR = 2.4). Nevertheless, we have not detected any enrichment for the *APOE* ε4 allele among the patients carrying rare variants in *APP*, *PSEN1*, *PSEN2*, *GRN*, *MAPT*, and *PRNP* (only 6 of 14 LOAD carriers [42%] carry at least one *APOE* ε4 allele). By contrast, *APOE* ε2 allele did not present a significantly higher frequency in controls compared to cases ($p$-value = 0.3, OR = 0.66).

## 4. Discussion

In this study, we tested the hypothesis that the significant phenotypic overlap between sporadic LOAD and other neurodegenerative dementias (FAD, FTD, PSP, CBD, and CJD) may be explained by a common genetic background. Thus, we screened 6 Mendelian dementia genes (*APP*, *PSEN1*, *PSEN2*, *MAPT*, *GRN*, and *PRNP*) aiming to establish whether rare coding variability in these genes is responsible for an appreciable portion of the LOAD risk.

In our LOAD cohort, we found a novel rare variant in *PSEN1* (p.I168T) and a rare variant in *PSEN2* (p.A237V). These variants are likely pathogenic: (1) both cluster in TM domains, on the alpha helix surface; (2) the literature already reported in the same (*PSEN1* p.I168) or homologous residue (*PSEN2* p.A237 and *PSEN1* p.A231) causative mutations for FAD; and (3) the *PSEN1* p.I168T and *PSEN2* p.A237V are classified as possible pathogenic, following the algorithm proposed by Guerreiro et al. (2010a). The other variants detected in our LOAD cases are likely to be tolerated. First, they have already been described as benign polymorphisms (*PSEN2* p.R62H; *PRNP* rs138688873 and *GRN* p.R433W) (Guerreiro et al., 2010a; Palmer et al., 1993; www.molgendatabase). Second, they cluster outside the reported pathogenic domains (*APP* p.A201V, p.Y538H, p.V562I, p.E599K; *PSEN2* p.R62C, p.R62H, p.S130L, p.D439A; *MAPT* p.V224G, p.G213R and p.G200E). Third, they do not alter the gene expression (*MAPT* c.115-2A>T).

Finally, despite the functional consequence of *GRN* p.R432C, the effect of a decreased *GRN* secretion in AD pathogenesis remains controversial. *GRN* pathogenic mutations act through a messenger RNA nonsense-mediate decay, interfering with *GRN* expression and generally are loss of function mutations (LoF) (stop-gain, frameshift mutations, and deletions). The only exception to this rule is represented by the pathogenic missense mutations which cluster in the GRN signal peptide domain (*GRN* p.A9D) (www.molgendatabase). Thus, we suggest, in concert with previous studies (Guerreiro et al., 2010b), that *GRN* missense mutations mapping outside the signal peptide domain are likely to be well tolerated.

In conclusion, our findings support recent studies, suggesting that rare coding variability in *PSEN1* and *PSEN2* contributes to susceptibility for apparently sporadic LOAD. Therefore, sporadic LOAD and FAD may be influenced by the same genes and thus pathogenic mechanisms. On the contrary, rare coding variants in *MAPT*, *GRN*, and *PRNP* are not major players in the development of

LOAD. Thus, genetic screening is fundamental for the differential diagnosis of these disparate neurodegenerative dementias.

## Acknowledgements

This study was supported by the Alzheimer's Research UK, the Medical Research Council (MRC), and the Wellcome Trust/MRC Joint Call in Neurodegeneration Award (WT089698) to the UK Parkinson's Disease Consortium (whose members are from the University College London, the National Institute for Health Research Biomedical Research Centre for Mental Health and Biomedical Research Unit for Dementia at the South London, and Maudsley NHS Foundation Trust and Kings College London; Institute of Neurology, the University of Sheffield, and the MRC Protein Phosphorylation Unit at the University of Dundee), grants (P50 AG016574, U01 AG006786, and R01 AG18023), the National Institute for Health Research Biomedical Research Unit in Dementia at University College London Hospitals, University College London; an anonymous donor, the Big Lottery (to Dr Morgan); a fellowship from Alzheimer's Research UK (to Dr Guerreiro); and the Intramural Research Programs of the National Institute on Aging and the National Institute of Neurological Disease and Stroke, National Institutes of Health (Department of Health and Human Services Project number, ZO1 AG000950-10). The MRC London Neurodegenerative Diseases Brain Bank and the Manchester Brain Bank from Brains for Dementia Research are jointly funded from Alzheimer's Research UK and Alzheimer's Association. The views expressed are those of the authors and not necessarily those of the NHS, the NIHR, or the Department of Health.

## Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.neurobiolaging.2014.06.002.

## References

Beach, T.G., Monsell, S.E., Phillips, L.E., Kukull, W., 2012. Accuracy of the clinical diagnosis of Alzheimer disease at National Institute on Aging Alzheimer Disease Centers, 2005-2010. J. Neuropathol. Exp. Neurol. 71, 266–273.

Benitez, B.A., Karch, C.M., Cai, Y., Jin, S.C., Cooper, B., Carrell, D., Bertelsen, S., Chibnik, L., Schneider, J.A., Bennett, D.A., Alzheimer's Disease Neuroimaging Initiative, Genetic and Environmental Risk for Alzheimer's Disease Consortium GERAD, Fagan, A.M., Holtzman, D., Morris, J.C., Goate, A.M., Cruchaga, C., 2013. The PSEN1, p.E318G variant increases the risk of Alzheimer's disease in APOE-ε4 carriers. PLoS Genet. 9, e1003685.

Bergem, A.L., Engedal, K., Kringlen, E., 1997. The role of heredity in late-onset Alzheimer disease and vascular dementia. A twin study. Arch. Gen. Psychiatry 54, 264–270.

Biesecker, L.G., Mullikin, J.C., Facio, F.M., Turner, C., Cherukuri, P.F., Blakesley, R.W., Bouffard, G.G., Chines, P.S., Cruz, P., Hansen, N.F., Teer, J.K., Maskeri, B., Young, A.C., NISC Comparative Sequencing Program, Manolio, T.A., Wilson, A.F., Finkel, T., Hwang, P., Arai, A., Remaley, A.T., Sachdev, V., Shamburek, R., Cannon, R.O., Green, E.D., 2009. The ClinSeq Project: piloting large-scale genome sequencing for research in genomic medicine. Genome Res. 19, 1665–1674.

Brouwers, N., Sleegers, K., Engelborghs, S., Maurer-Stroh, S., Gijselinck, I., van der Zee, J., Pickut, B.A., Van den Broeck, M., Mattheijssens, M., Peeters, K., Schymkowitz, J., Rousseau, F., Martin, J.-J., Cruts, M., De Deyn, P.P., Van Broeckhoven, C., 2008. Genetic variability in progranulin contributes to risk for clinically diagnosed Alzheimer disease. Neurology 71, 656–664.

Campion, D., Dumanchin, C., Hannequin, D., Dubois, B., Belliard, S., Puel, M., Thomas-Anterion, C., Michon, A., Martin, C., Charbonnier, F., Raux, G., Camuzat, A., Penet, C., Mesnage, V., Martinez, M., Clerget-Darpoux, F., Brice, A., Frebourg, T., 1999. Early-onset autosomal dominant Alzheimer disease: prevalence, genetic heterogeneity, and mutation spectrum. Am. J. Hum. Genet. 65, 664–670.

Cruchaga, C., Haller, G., Chakraverty, S., Mayo, K., Vallania, F.L.M., Mitra, R.D., Faber, K., Williamson, J., Bird, T., Diaz-Arrastia, R., Foroud, T.M., Boeve, B.F., Graff-Radford, N.R., St Jean, P., Lawson, M., Ehm, M.G., Mayeux, R., Goate, A.M., NIA-LOAD/NCRAD Family Study Consortium, 2012. Rare variants in APP, PSEN1 and PSEN2 increase risk for AD in late-onset Alzheimer's disease families. PloS One 7, e31039.

Cruchaga, C., Karch, C.M., Jin, S.C., Benitez, B.A., Cai, Y., Guerreiro, R., Harari, O., Norton, J., Budde, J., Bertelsen, S., Jeng, A.T., Cooper, B., Skorupa, T., Carrell, D., Levitch, D., Hsu, S., Choi, J., Ryten, M., UK Brain Expression Consortium (UKBEC), Hardy, J., Ryten, M., Trabzuni, D., Weale, M.E., Ramasamy, A., Smith, C., Sassi, C., Bras, J., Gibbs, J.R., Hernandez, D.G., Lupton, M.K., Powell, J., Forabosco, P., Ridge, P.G., Corcoran, C.D., Tschanz, J.T., Norton, M.C., Munger, R.G., Schmutz, C., Leary, M., Demirci, F.Y., Bamne, M.N., Wang, X., Lopez, O.L., Ganguli, M., Medway, C., Turton, J., Lord, J., Braae, A., Barber, I., Brown, K., The Alzheimer's Research UK (ARUK) Consortium, Passmore, P., Craig, D., Johnston, J., McGuinness, B., Todd, S., Heun, R., Kölsch, H., Kehoe, P.G., Hooper, N.M., Vardy, E.R.L.C., Mann, D.M., Pickering-Brown, S., Brown, K., Kalsheker, N., Lowe, J., Morgan, K., David Smith, A., Wilcock, G., Warden, D., Holmes, C., Pastor, P., Lorenzo-Betancor, O., Brkanac, Z., Scott, E., Topol, E., Morgan, K., Rogaeva, E., Singleton, A.B., Hardy, J., Kamboh, M.I., St George-Hyslop, P., Cairns, N., Morris, J.C., Kauwe, J.S.K., Goate, A.M., 2013. Rare coding variants in the phospholipase D3 gene confer risk for Alzheimer's disease. Nature 505, 550–554.

Cruts, M., van Duijn, C.M., Backhovens, H., Van den Broeck, M., Wehnert, A., Serneels, S., Sherrington, R., Hutton, M., Hardy, J., St George-Hyslop, P.H., Hofman, A., Van Broeckhoven, C., 1998. Estimation of the genetic contribution of presenilin-1 and -2 mutations in a population-based study of presenile Alzheimer disease. Hum. Mol. Genet. 7, 43–51.

Gatz, M., Reynolds, C.A., Fratiglioni, L., Johansson, B., Mortimer, J.A., Berg, S., Fiske, A., Pedersen, N.L., 2006. Role of genes and environments for explaining Alzheimer disease. Arch. Gen. Psychiatry 63, 168–174.

Guerreiro, R.J., Baquero, M., Blesa, R., Boada, M., Brás, J.M., Bullido, M.J., Calado, A., Crook, R., Ferreira, C., Frank, A., Gómez-Isla, T., Hernández, I., Lleó, A., Machado, A., Martínez-Lage, P., Masdeu, J., Molina-Porcel, L., Molinuevo, J.L., Pastor, P., Pérez-Tur, J., Relvas, R., Oliveira, C.R., Ribeiro, M.H., Rogaeva, E., Sa, A., Samaranch, L., Sánchez-Valle, R., Santana, I., Tàrraga, L., Valdivieso, F., Singleton, A., Hardy, J., Clarimón, J., 2010a. Genetic screening of Alzheimer's disease genes in Iberian and African samples yields novel mutations in presenilins and APP. Neurobiol. Aging 31, 725–731.

Guerreiro, R.J., Washecka, N., Hardy, J., Singleton, A., 2010b. A thorough assessment of benign genetic variability in GRN and MAPT. Hum. Mutat. 31, E1126–E1140.

Guerreiro, R., Wojtas, A., Bras, J., Carrasquillo, M., Rogaeva, E., Majounie, E., Cruchaga, C., Sassi, C., Kauwe, J.S.K., 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.

Hardy, J., Crook, R., 2001. Presenilin mutations line up along transmembrane alpha-helices. Neurosci. Lett. 306, 203–205.

Harold, D., Abraham, R., Hollingworth, P., Sims, R., Gerrish, A., Hamshere, M.L., Pahwa, J.S., Moskvina, V., Dowzell, K., Williams, A., Jones, N., Thomas, C., Stretton, A., Morgan, A.R., Lovestone, S., Powell, J., Proitsi, P., Lupton, M.K., Brayne, C., Rubinsztein, D.C., Gill, M., Lawlor, B., Lynch, A., Morgan, K., Brown, K.S., Passmore, P.A., Craig, D., McGuinness, B., Todd, S., Holmes, C., Mann, D., Smith, A.D., Love, S., Kehoe, P.G., Hardy, J., Mead, S., Fox, N., Rossor, M., Collinge, J., Maier, W., Jessen, F., Schürmann, B., Heun, R., van den Bussche, H., Heuser, I., Kornhuber, J., Wiltfang, J., Dichgans, M., Frölich, L., Hampel, H., Hüll, M., Rujescu, D., Goate, A.M., Kauwe, J.S.K., Cruchaga, C., Nowotny, P., Morris, J.C., Mayo, K., Sleegers, K., Bettens, K., Engelborghs, S., De Deyn, P.P., Van Broeckhoven, C., Livingston, G., Bass, N.J., Gurling, H., McQuillin, A., Gwilliam, R., Deloukas, P., Al-Chalabi, A., Shaw, C.E., Tsolaki, M., Singleton, A.B., Guerreiro, R., Mühleisen, T.W., Nöthen, M.M., Moebus, S., Jöckel, K.H., Klopp, N., Wichmann, H.E., Carrasquillo, M.M., Pankratz, V.S., Younkin, S.G., Holmans, P.A., O'Donovan, M., Owen, M.J., Williams, J., 2009. Genome-wide association study identifies variants at CLU and PICALM associated with Alzheimer's disease. Nat. Genet. 41, 1088–1093.

Hollingworth, P., Harold, D., Sims, R., Gerrish, A., Lambert, J.C., Carrasquillo, M.M., Abraham, R., Hamshere, M.L., Pahwa, J.S., Moskvina, V., Dowzell, K., Jones, N., Stretton, A., Thomas, C., Richards, A., Ivanov, D., Widdowson, C., Chapman, J., Lovestone, S., Powell, J., Proitsi, P., Lupton, M.K., Brayne, C., Rubinsztein, D.C., Gill, M., Lawlor, B., Lynch, A., Brown, K.S., Passmore, P.A., Craig, D., McGuinness, B., Todd, S., Holmes, C., Mann, D., Smith, A.D., Beaumont, H., Warden, D., Wilcock, G., Love, S., Kehoe, P.G., Hooper, N.M., Vardy, E.R.L.C., Hardy, J., Mead, S., Fox, N.C., Rossor, M., Collinge, J., Maier, W., Jessen, F., Rüther, E., Schürmann, B., Heun, R., Kölsch, H., van den Bussche, H., Heuser, I., Kornhuber, J., Wiltfang, J., Dichgans, M., Frölich, L., Hampel, H., Gallacher, J., Hüll, M., Rujescu, D., Giegling, I., Goate, A.M., Kauwe, J.S.K., Cruchaga, C., Nowotny, P., Morris, J.C., Mayo, K., Sleegers, K., Bettens, K., Engelborghs, S., De Deyn, P.P., Van Broeckhoven, C., Livingston, G., Bass, N.J., Gurling, H., McQuillin, A., Gwilliam, R., Deloukas, P., Al-Chalabi, A., Shaw, C.E., Tsolaki, M., Singleton, A.B., Guerreiro, R., Mühleisen, T.W., Nöthen, M.M., Moebus, S., Jöckel, K.-H., Klopp, N., Wichmann, H.-E., Pankratz, V.S., Sando, S.B., Aasly, J.O., Barcikowska, M., Wszolek, Z.K., Dickson, D.W., Graff-Radford, N.R., Petersen, R.C., Alzheimer's Disease Neuroimaging Initiative, van Duijn, C.M., Breteler, M.M.B., Ikram, M.A., DeStefano, A.L., Fitzpatrick, A.L., Lopez, O., Launer, L.J., Seshadri, S., CHARGE consortium, Berr, C., Campion, D., Epelbaum, J., Dartigues, J.-F., Tzourio, C., Alpérovitch, A., Lathrop, M., EADI1 consortium, Feulner, T.M., Friedrich, P., Riehle, C., Krawczak, M., Schreiber, S., Mayhaus, M., Nicolhaus, S., Wagenpfeil, S., Steinberg, S., Stefansson, H.,

Stefansson, K., Snaedal, J., Björnsson, S., Jonsson, P.V., Chouraki, V., Genier-Boley, B., Hiltunen, M., Soininen, H., Combarros, O., Zelenika, D., Delepine, M., Bullido, M.J., Pasquier, F., Mateo, I., Frank-Garcia, A., Porcellini, E., Hanon, O., Coto, E., Alvarez, V., Bosco, P., Siciliano, G., Mancuso, M., Panza, F., Solfrizzi, V., Nacmias, B., Sorbi, S., Bossù, P., Piccardi, P., Arosio, B., Annoni, G., Seripa, D., Pilotto, A., Scarpini, E., Galimberti, D., Brice, A., Hannequin, D., Licastro, F., Jones, L., Holmans, P.A., Jonsson, T., Riemenschneider, M., Morgan, K., Younkin, S.G., Owen, M.J., O'Donovan, M., Amouyel, P., Williams, J., 2011. Common variants at ABCA7, MS4A6A/MS4A4E, EPHA1, CD33 and CD2AP are associated with Alzheimer's disease. Nat. Genet. 43, 429–435.

Janssen, J.C., Beck, J.A., Campbell, T.A., Dickinson, A., Fox, N.C., Harvey, R.J., Houlden, H., Rossor, M.N., Collinge, J., 2003. Early onset familial Alzheimer's disease: mutation frequency in 31 families. Neurology 60, 235–239.

Jonsson, T., Atwal, J.K., Steinberg, S., Snaedal, J., Jonsson, P.V., Bjornsson, S., Stefansson, H., Sulem, P., Gudbjartsson, D., Maloney, J., Hoyte, K., Gustafson, A., Liu, Y., Lu, Y., Bhangale, T., Graham, R.R., Huttenlocher, J., Bjornsdottir, G., Andreassen, O.A., Jönsson, E.G., Palotie, A., Behrens, T.W., Magnusson, O.T., Kong, A., Thorsteinsdottir, U., Watts, R.J., Stefansson, K., 2012. A mutation in APP protects against Alzheimer's disease and age-related cognitive decline. Nature 488, 96–99.

Lambert, J.C., Heath, S., Even, G., Campion, D., Sleegers, K., Hiltunen, M., Combarros, O., Zelenika, D., Bullido, M.J., Tavernier, B., Letenneur, L., Bettens, K., Berr, C., Pasquier, F., Fiévet, N., Barberger-Gateau, P., Engelborghs, S., De Deyn, P., Mateo, I., Franck, A., Helisalmi, S., Porcellini, E., Hanon, O., EuropeanAlzheimer's Disease Initiative Investigators, de Pancorbo, M.M., Lendon, C., Dufouil, C., Jaillard, C., Leveillard, T., Alvarez, V., Bosco, P., Mancuso, M., Panza, F., Nacmias, B., Bossù, P., Piccardi, P., Annoni, G., Seripa, D., Galimberti, D., Hannequin, D., Licastro, F., Soininen, H., Ritchie, K., Blanché, H., Dartigues, J.-F., Tzourio, C., Gut, I., Van Broeckhoven, C., Alpérovitch, A., Lathrop, M., Amouyel, P., 2009. Genome-wide association study identifies variants at CLU and CR1 associatedCLU and PICALM associated with Alzheimer's disease. with Alzheimer's disease. Nat. Genet. 41, 1094–1099.

Lambert, J.C., Ibrahim-Verbaas, C.A., Harold, D., Naj, A.C., Sims, R., Bellenguez, C., Jun, G., Destefano, A.L., Bis, J.C., Beecham, G.W., Grenier-Boley, B., Russo, G., Thornton-Wells, T.A., Jones, N., Smith, A.V., Chouraki, V., Thomas, C., Ikram, M.A., Zelenika, D., Vardarajan, B.N., Kamatani, Y., Lin, C.-F., Gerrish, A., Schmidt, H., Kunkle, B., Dunstan, M.L., Ruiz, A., Bihoreau, M.T., Choi, S.H., Reitz, C., Pasquier, F., Hollingworth, P., Ramirez, A., Hanon, O., Fitzpatrick, A.L., Buxbaum, J.D., Campion, D., Crane, P.K., Baldwin, C., Becker, T., Gudnason, V., Cruchaga, C., Craig, D., Amin, N., Berr, C., Lopez, O.L., De Jager, P.L., Deramecourt, V., Johnston, J.A., Evans, D., Lovestone, S., Letenneur, L., Morón, F.J., Rubinsztein, D.C., Eiriksdottir, G., Sleegers, K., Goate, A.M., Fiévet, N., Huentelman, M.J., Gill, M., Brown, K., Kamboh, M.I., Keller, L., Barberger-Gateau, P., McGuinness, B., Larson, E.B., Green, R., Myers, A.J., Dufouil, C., Todd, S., Wallon, D., Love, S., Rogaeva, E., Gallacher, J., St George-Hyslop, P., Clarimon, J., Lleo, A., Bayer, A., Tsuang, D.W., Yu, L., Tsolaki, M., Bossù, P., Spalletta, G., Proitsi, P., Collinge, J., Sorbi, S., Sanchez-Garcia, F., Fox, N.C., Hardy, J., Naranjo, M.C.D., Bosco, P., Clarke, R., Brayne, C., Galimberti, D., Mancuso, M., Matthews, F., European Alzheimer's Disease Initiative (EADI), Genetic and Environmental Risk in Alzheimer's Disease, Alzheimer's Disease Genetic Consortium, Cohorts for Heart and Aging Research in Genomic Epidemiology, Moebus, S., Mecocci, P., Del Zompo, M., Maier, W., Hampel, H., Pilotto, A., Bullido, M., Panza, F., Caffarra, P., Nacmias, B., Gilbert, J.R., Mayhaus, M., Lannfelt, L., Hakonarson, H., Pichler, S., Carrasquillo, M.M., Ingelsson, M., Beekly, D., Alvarez, V., Zou, F., Valladares, O., Younkin, S.G., Coto, E., Hamilton-Nelson, K.L., Gu, W., Razquin, C., Pastor, P., Mateo, I., Owen, M.J., Faber, K.M., Jonsson, P.V., Combarros, O., O'Donovan, M.C., Cantwell, L.B., Soininen, H., Blacker, D., Mead, S., Mosley Jr., T.H., Bennett, D.A., Harris, T.B., Fratiglioni, L., Holmes, C., de Bruijn, R.F.A.G., Passmore, P., Montine, T.J., Bettens, K., Rotter, J.I., Brice, A., Morgan, K., Foroud, T.M., Kukull, W.A., Hannequin, D., Powell, J.F., Nalls, M.A., Ritchie, K., Lunetta, K.L., Kauwe, J.S.K., Boerwinkle, E., Riemenschneider, M., Boada, M., Hiltunen, M., Martin, E.R., Schmidt, R., Rujescu, D., Wang, L.S., Dartigues, J.-F., Mayeux, R., Tzourio, C., Hofman, A., Nöthen, M.M., Graff, C., Psaty, B.M., Jones, L., Haines, J.L., Holmans, P.A., Lathrop, M., Pericak-Vance, M.A., Launer, L.J., Farrer, L.A., van Duijn, C.M., Van Broeckhoven, C., Moskvina, V., Seshadri, S., Williams, J., Schellenberg, G.D., Amouyel, P., 2013. Meta-analysis of 74,046 individuals identifies 11 new susceptibility loci for Alzheimer's disease. Nat. Genet. 45, 1452–1458.

Li, H., Durbin, R., 2009. Fast and accurate short read alignment with Burrows-Wheeler transform. Bioinformatics 25, 1754–1760.

Majounie, E., Abramzon, Y., Renton, A.E., Perry, R., Bassett, S.S., Pletnikova, O., Troncoso, J.C., Hardy, J., Singleton, A.B., Traynor, B.J., 2012. Repeat expansion in C9ORF72 in Alzheimer's disease. N. Engl. J. Med. 366, 283–284.

McKenna, A., Hanna, M., Banks, E., Sivachenko, A., Cibulskis, K., Kernytsky, A., Garimella, K., Altshuler, D., Gabriel, S., Daly, M., DePristo, M.A., 2010. The Genome Analysis Toolkit: a MapReduce framework for analyzing next-generation DNA sequencing data. Genome Res. 20, 1297–1303.

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.

Palmer, M.S., Dryden, A.J., Hughes, J.T., Collinge, J., 1991. Homozygous prion protein genotype predisposes to sporadic Creutzfeldt-Jakob disease. Nature 352, 340–342.

Palmer, M.S., Mahal, S.P., Campbell, T.A., Hill, A.F., Sidle, K.C., Laplanche, J.L., Collinge, J., 1993. Deletions in the prion protein gene are not associated with CJD. Hum. Mol. Genet. 2, 541–544.

Perry, R.T., Go, R.C., Harrell, L.E., Acton, R.T., 1995. SSCP analysis and sequencing of the human prion protein gene (PRNP) detects two different 24 bp deletions in an atypical Alzheimer's disease family. Am. J. Med. Genet. 60, 12–18.

Raiha, I., Kaprio, J., Koskenvuo, M., Rajala, T., Sourander, L., 1996. Alzheimer's disease in Finnish twins. Lancet 347, 573–578.

Rogaeva, E.A., Fafel, K.C., Song, Y.Q., Medeiros, H., Sato, C., Liang, Y., Richard, E., Rogaev, E.I., Frommelt, P., Sadovnick, A.D., Meschino, W., Rockwood, K., Boss, M.A., Mayeux, R., St George-Hyslop, P., 2001. Screening for PS1 mutations in a referral-based series of AD cases: 21 novel mutations. Neurology 57, 621–625.

Seshadri, S., Fitzpatrick, A.L., Ikram, M.A., DeStefano, A.L., Gudnason, V., Boada, M., Bis, J.C., Smith, A.V., Carassquillo, M.M., Lambert, J.C., Harold, D., Schrijvers, E.M.C., Ramirez-Lorca, R., Debette, S., Longstreth Jr., W.T., Janssens, A.C.J.W., Pankratz, V.S., Dartigues, J.F., Hollingworth, P., Aspelund, T., Hernandez, I., Beiser, A., Kuller, L.H., Koudstaal, P.J., Dickson, D.W., Tzourio, C., Abraham, R., Antunez, C., Du, Y., Rotter, J.I., Aulchenko, Y.S., Harris, T.B., Petersen, R.C., Berr, C., Owen, M.J., Lopez-Arrieta, J., Varadarajan, B.N., Becker, J.T., Rivadeneira, F., Nalls, M.A., Graff-Radford, N.R., Campion, D., Auerbach, S., Rice, K., Hofman, A., Jonsson, P.V., Schmidt, H., Lathrop, M., Mosley, T.H., Au, R., Psaty, B.M., Uitterlinden, A.G., Farrer, L.A., Lumley, T., Ruiz, A., Williams, J., Amouyel, P., Younkin, S.G., Wolf, P.A., Launer, L.J., Lopez, O.L., van Duijn, C.M., Breteler, M.M.B., CHARGE Consortium, GERAD1 Consortium, EADI1 Consortium, 2010. Genome-wide analysis of genetic loci associated with Alzheimer disease. JAMA 303, 1832–1840.

Shankaran, S.S., Capell, A., Hruscha, A.T., Fellerer, K., Neumann, M., Schmid, B., Haass, C., 2008. Missense mutations in the progranulin gene linked to frontotemporal lobar degeneration with ubiquitin-immunoreactive inclusions reduce progranulin production and secretion. J. Biol. Chem. 283, 1744–1753.

Wang, K., Li, M., Hakonarson, H., 2010. ANNOVAR: functional annotation of genetic variants from high-throughput sequencing data. Nucleic Acids Res. 38, e164.

Yang, Y.C., Lin, C.H., Lee, E.H.Y., 2006. Serum- and glucocorticoid-inducible kinase 1 (SGK1) increases neurite formation through microtubule depolymerization by SGK1 and by SGK1 phosphorylation of tau. Mol. Cell. Biol. 26, 8357–8370.