Neurobiology of Aging 97 (2021) 148.e9–148.e16

Contents lists available at ScienceDirect

Neurobiology of Aging

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

Underlying genetic variation in familial frontotemporal dementia: sequencing of 198 patients

Merel O. Mol[a,*], Jeroen G.J. van Rooij[a,b], Tsz H. Wong[a], Shamiram Melhem[a], Annemieke J.M.H. Verkerk[b], Anneke J.A. Kievit[c], Rick van Minkelen[c], Rosa Rademakers[d], Cyril Pottier[d], Laura Donker Kaat[a,c], Harro Seelaar[a], John C. van Swieten[a], Elise G.P. Dopper[a]

[a] Department of Neurology & Alzheimer Center, Erasmus Medical Center, Rotterdam, the Netherlands
[b] Department of Internal Medicine, Erasmus Medical Center, Rotterdam, the Netherlands
[c] Department of Clinical Genetics, Erasmus Medical Center, Rotterdam, the Netherlands
[d] Neurodegenerative Brain Diseases Group, VIB Center for Molecular Neurology, University of Antwerp, Antwerp, Belgium

ARTICLE INFO

Article history:
Received 17 April 2020
Received in revised form 1 June 2020
Accepted 14 July 2020
Available online 30 July 2020

Keywords:
Frontotemporal dementia
Familial
Whole-exome sequencing
Genetic screen

ABSTRACT

Frontotemporal dementia (FTD) presents with a wide variability in clinical syndromes, genetic etiologies, and underlying pathologies. Despite the discovery of pathogenic variants in several genes, many familial cases remain unsolved. In a large FTD cohort of 198 familial patients, we aimed to determine the types and frequencies of variants in genes related to FTD. Pathogenic or likely pathogenic variants were revealed in 74 (37%) patients, including 4 novel variants. The repeat expansion in *C9orf72* was most common (21%), followed by variants in *MAPT* (6%), *GRN* (4.5%), and *TARDBP* (3.5%). Other pathogenic variants were found in *VCP, TBK1*, *PSEN1,* and a novel homozygous variant in *OPTN*. Furthermore, we identified 15 variants of uncertain significance, including a promising variant in *TUBA4A* and a frameshift in *VCP*, for which additional research is needed to confirm pathogenicity. The patients without identified genetic cause demonstrated a wide clinical and pathological variety. Our study contributes to the clinical characterization of the genetic subtypes and confirms the value of whole-exome sequencing in identifying novel genetic variants.

1. Introduction

Frontotemporal dementia (FTD) is one of the main causes of presenile dementia (Coyle-Gilchrist et al., 2016). FTD constitutes a heterogeneous spectrum with large variability in clinical and pathological features (Mackenzie and Rademakers, 2007; Mann and Snowden, 2017). It has a strong genetic component, and autosomal dominant inheritance is observed in 10%–25% of patients (Convery et al., 2019; Seelaar et al., 2008). Mutations in *C9orf72, GRN,* and *MAPT* account for ~30% of familial cases, with substantial geographical variability in mutation frequencies (Fostinelli et al., 2018; Kim et al., 2018; Moore et al., 2020; Oijerstedt et al., 2019; Seelaar et al., 2008; Tang et al., 2016; Wood et al., 2013). In the past decade, whole-exome sequencing (WES) has emerged as a method to identify novel pathogenic variants not only in these genes, but also likely pathogenic variants or variants of uncertain significance (VUS) in an increasing number of other dementia-associated genes such as *TARDBP, VCP, TBK1,* and *SQSTM1* (Blauwendraat et al., 2018; Dols-Icardo et al., 2018; Ramos et al., 2019, 2020). Nonetheless, around two-thirds of familial cases remain without a known genetic cause, implying yet undiscovered variants (Pottier et al., 2019).

In this study, we systematically assessed a broad set of dementia-related genes in our large cohort of patients with FTD and a positive family history using WES, *C9orf72* repeat-primed PCR, and copy number variation analysis. Our objectives were to investigate the frequencies of pathogenic variants in the Netherlands and to identify potential novel variants, which might ultimately provide new pathophysiological insights.

2. Materials and methods

2.1. Clinical data collection

Patients were selected from our large FTD cohort in the Netherlands (Erasmus Medical Center, Rotterdam) (Seelaar et al.,

* Corresponding author at: Department of Neurology, Erasmus Medical Center, Dr Molewaterplein 40, 3015GD Rotterdam, 3015GD Rotterdam, the Netherlands. Tel.: +31624354255; fax: +31107044721.
E-mail address: m.o.mol@erasmusmc.nl (M.O. Mol).

0197-4580/
https://doi.org/10.1016/j.neurobiolaging.2020.07.014

2008), which currently includes 656 patients with a clinical diagnosis of either the behavioral variant of FTD (bvFTD) or primary progressive aphasia (PPA), classified into 3 different forms (semantic variant [svPPA], nonfluent variant (nfvPPA), and logopenic variant [lvPPA]). We excluded patients and relatives with a pathological diagnosis other than frontotemporal lobar degeneration (FTLD). The family history was considered positive with the presence of at least one first- or second-degree relative affected by an FTLD spectrum disorder (besides bvFTD and PPA, this includes FTD with motor neuron disease, amyotrophic lateral sclerosis [ALS], progressive supranuclear palsy, and corticobasal syndrome [CBS]) or another type of dementia or Parkinson's disease [PD]). Family history was further classified into one of the following adjusted Goldman categories (Goldman et al., 2005). Psychiatric disorders were not considered in this classification as these were not known for all patients.

(1) Autosomal dominant: $\geq$ 2 relatives with either an FTLD spectrum disorder at any age or another type of dementia or PD $<$ 65 years, occurring in at least 2 generations with one person being a first-degree relative of both other 2;
(2) Familial aggregation: $\geq$ 3 relatives (first, second, or third degree) with an FTLD spectrum disorder, another type of dementia or PD at any age, not meeting criteria for autosomal dominant inheritance;
(3) Possible familial: $\geq$ 1 first- or second-degree relative with an FTLD spectrum disorder at any age or another type of dementia or PD $<$ 65 years;
4) Possible familial late-onset: $\geq$ 1 first-degree relative with any type of dementia or PD $>$ 65 years;
(5) Negative family history: none of the above.

From the total cohort ($n = 656$), we selected 198 unrelated patients with a positive family history (Goldman 1–4) and DNA availability (Supplementary Fig. A.1). For 41 familial patients, DNA was not available.

2.2. Sequencing and variant filtering

In 38 patients, targeted Sanger sequencing of *MAPT* or *GRN*, or *C9orf72* repeat-primed PCR had previously revealed a pathogenic variant. WES was performed in 151 patients, and 9 were whole-genome sequenced at the Mayo Clinic Genome Analysis Core as part of another study. As the data were collected from various sources, different capture kits were used (see Appendix A for bioinformatics details). The presence of a *C9orf72* repeat expansion was tested using either repeat-primed PCR or a commercial kit (AmplideX PCR/CE, Asuragen), with a repeat length $\geq$30 considered pathogenic variants (Renton et al., 2011).

We analyzed 26 prespecified genes, based on an extensive literature search of genes associated with FTD, FTD-ALS, and Alzheimer's disease (AD), as AD may clinically resemble FTD (Supplementary Table A.1). Variants were selected based on the following criteria: (1) affecting coding (missense, nonsense, frameshift) or splicing regions; (2) with a minor allele frequency of $<$0.1% in the Genome Aggregation Database (gnomAD); and (3) with a quality by depth score $\geq$5. The untranslated regions (UTRs) of the genes *GRN, MAPT*, and *TARDBP* were investigated for the presence of known pathogenic regulatory variants. Variants reported as pathogenic in the AD&FTD Mutation Database (http://www.molgen.ua.ac.be/ADMutations) were classified accordingly. We classified novel variants as pathogenic, likely pathogenic, or as VUS in a conservative and systematic approach according to the recently refined guidelines by The American College of Medical Genetics and Genomics (ACMG) (Nykamp et al., 2017; Richards et al., 2015). The following criteria were jointly considered to obtain evidence of pathogenicity: (1) bioinformatic *in silico* prediction scores: SIFT, PolyPhen2, MutationTaster, FATHMM, combined annotation dependent depletion; score $\geq$10), Human Splicing Finder, and MaxEnt; (2) presence in other online genetic databases [OMIM, HGMD, ClinVar, AlzGene, Healthy Exomes (HEX) (Guerreiro et al., 2018)]; (3) existing literature on the variant or a different variant in the same position; (4) segregation analysis if available; (5) functional biomarker if available (blood progranulin levels for *GRN*); and (6) pathological confirmation of disease if available. Variants reported in the previously mentioned genetic databases as likely benign were only discarded if these reports were consistent and in concordance with *in silico* prediction tools. Pathogenic and likely pathogenic variants were confirmed by Sanger sequencing.

2.3. SNP array and CNV detection

We performed copy number variant (CNV) analysis of the same 26 genes using single nucleotide polymorphism (SNP) array data to identify deletions or duplications in subjects without a pathogenic variant (including those with a VUS). The SNP array platform used was Illumina GSA BeadChip GSA MD, v2 (Illumina GSA Arrays "Infinium iSelect 24x1 HTS Custom BeadChip Kit"). Samples were processed using the Illumina manufacturer's recommended protocol. CNV calling was performed using Nexus Copy Number software (v.4.1, BioDiscovery, Inc, El Segundo, CA, USA) with default parameters.

2.4. Neuropathology

Neuropathological examination was available in 76 subjects (46 probands and 30 affected relatives). Immunohistochemistry was performed as previously described (Seelaar et al., 2008), and FTLD diagnosis was based on the criteria by Cairns et al. (2007). The pattern of FTLD with TDP-43 or FET pathology was classified into different subtypes according to the morphology and distribution of neuronal inclusions as proposed by Neumann and Mackenzie (2019).

3. Results

3.1. Frequencies of known pathogenic variants

We detected a pathogenic or likely pathogenic genetic variant in 74 of 198 (37%) patients (Table 1). The most common cause was the *C9orf72* repeat expansion identified in 21% (42/198), followed by pathogenic variants in *MAPT* in 6% (11/198; 6 unique variants), *GRN* in 4.5% (9/198; 8 unique variants, 3 of which were not reported previously), and *TARDBP* in 3.5% (7/198, 2 unique variants). Clinical and pathological characteristics of patients carrying genetic variants in these 4 genes are shown in Fig. 1 and Supplementary Table A.2. Furthermore, we identified 2 different pathogenic missense variants in *VCP* (1%), one nonsense variant in *TBK1* (0.5%), one missense variant in *PSEN1* (0.5%), and one novel homozygous variant in *OPTN* (0.5%). Subsequent CNV analysis performed in all remaining cases ($n = 124$) did not reveal any deletions or duplications. No cases were identified with a double pathogenic variant, although this could not be excluded in 38 cases tested for single genes.

3.2. Novel pathogenic and likely pathogenic variants

The novel *OPTN* variant is a homozygous splice-site variant (c.1242+1G>A) in a patient with lvPPA, decreased frontotemporal

Table 1
Pathogenic variants identified in 8 of 26 prespecified genes that were screened associated with FTD, FTD-ALS, and AD

Gene	Nucleotide change	Amino acid change	gnomAD MAF[a]	CADD[b]	#Probands	#Relatives[c]
C9orf72 repeat expansion	NA	NA	NA	NA	42	16
GRN (NM_002087)						
GRN	c.243delC	S82VfsX174	0	NA	1	28
GRN	c.373C>T	Q125X	0	35.0	1	5
GRN	c.1231_1232delGT	V411Sfs*2	0	NA	1	0
GRN	c.945_946delTG	C315X	0	NA	1	0
GRN	c.1160dupG	C388LfsX26	0	NA	1	0
GRN	c.19T>G	W7G	0	26.0	1	0
GRN	c.19T>C	W7R	0	25.9	2	0
GRN	c.1A>C	M1? (p.0)	0	23.9	1	0
MAPT (NM_005910)						
MAPT	c.902C>T	P301L	0	32.0	3	34
MAPT	c.815G>T	G272V	0	29.8	2	6
MAPT	c.944T>G	L315R	0	31.0	1	6
MAPT	c.1216C>T	R406W	1.6e-05	29.8	3	3
MAPT	c.959C>T	S320F	0	32.0	1	0
MAPT	c.841_843delAAG	L281del	2.6−05	NA	1	0
OPTN (NM_001008211)						
OPTN	c.1242+1G>A	NA	4.0e-06	28.6	1	0
PSEN1 (NM_000021)						
PSEN1	c.791C>T	P264L	4.0e-06	32.0	1	0
TARDBP (NM_007375)						
TARDBP	c.1147A>G	I383V	1.9e-05	18.6	6	1
TARDBP	c.787A>G	K263G	0	28.9	1	0
TBK1 (NM_013254)						
TBK1	c.1335G>A	W445X	0	39.0	1	1
VCP (NM_007126)						
VCP	c.785C>G	T262S	0	23.2	1	0
VCP	c.472A>G	M158V	0	23.8	1	0
TOTAL					74	100

Key: AD, Alzheimer's disease; ALS, amyotrophic lateral sclerosis; CADD, combined annotation dependent depletion; FTD, frontotemporal dementia; NA, not available/applicable.
[a] Minor allele frequency of the total population (141,456 exome/genome sequences).
[b] Version CADD score: GRch37-v1.4.
[c] Relatives are all confirmed carriers of the variant.

FDG uptake on positron emission tomography-computed tomography, and a normal profile in cerebrospinal fluid of ptau and amyloid-β, which is incompatible with AD. Family history revealed a sibling diagnosed with nfvPPA and consanguinity between parents (Goldman 3). No other relatives were known to have dementia, PD, or ALS. We considered the variant likely pathogenic

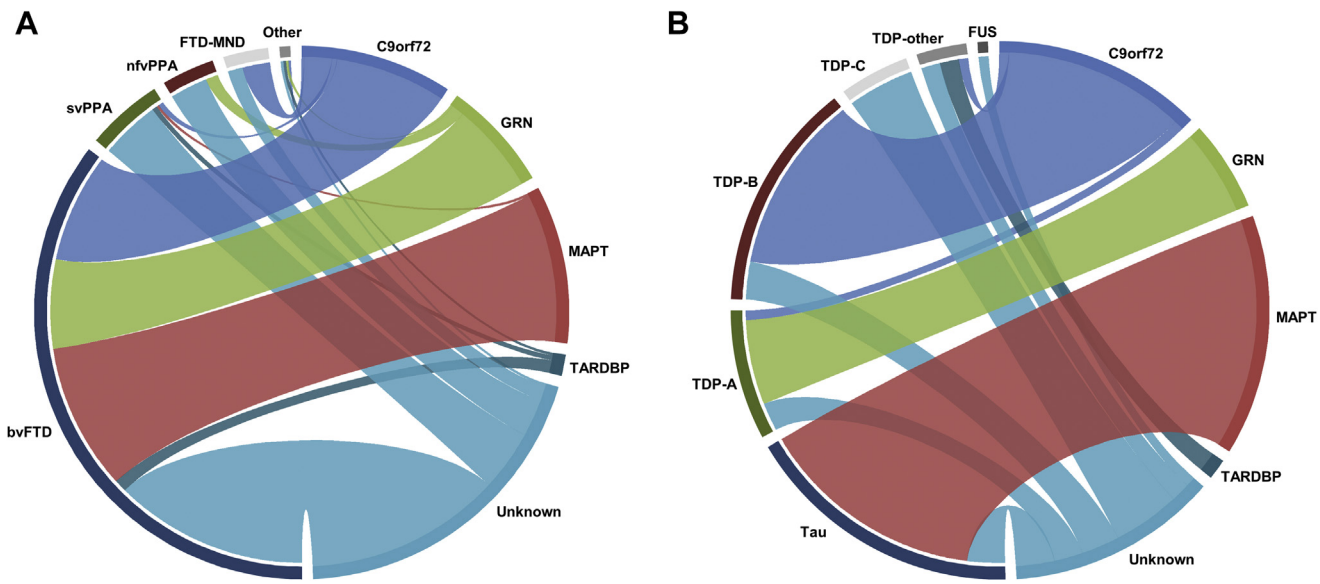

Fig. 1. Circos plots showing correlations between the major genetic subtypes and (A) clinical diagnosis ($n = 292$) and (B) pathological diagnosis ($n = 76$), whereas large heterogeneity is revealed in cases without identified genetic cause. Patients carrying variants in the genes *OPTN, PSEN1, TBK1,* and *VCP* were not included in these figures because of small numbers. The group 'unknown' includes patients with a VUS. Other = other clinical diagnosis (lvPPA, mixed PPA, or benign FTD). TDP-other = type D, type E, or unclassified. Details of all patients can be found in the Supplementary Tables A.2-3. Abbreviations: VUS, variants of uncertain significance; lvPPA, logopenic variant of PPA; PPA, primary progressive aphasia; FTD, frontotemporal dementia.

for the following reasons: (1) it is extremely rare in gnomAD (minor allele frequency, 8.8e−06) and has not been reported in the homozygous state; (2) it is predicted to change the canonical splice donor site resulting in skipping of exon 12 (MaxEnt, NNSplice, HSF), leading to a shift of the open reading frame; (3) the variant segregates with the disease as the sibling with nfvPPA carried the same homozygous variant.

Three variants in *GRN* have not been reported previously, including 2 truncating (p.C388LfsX26 and p.C315X) and 1 missense variant (p.W7G). The truncating variants were found in 2 patients with bvFTD leading to death within 5 years. Family history revealed an autosomal dominant pattern in the patient with the C388LfsX26 variant (Goldman 1), whereas the patient with the C315X variant only had 2 relatives with dementia at old age (Goldman 4). Segregation analysis could not be performed because of lack of DNA from family members and serum was not available to measure progranulin levels. However, all truncating variants in *GRN* are currently considered as likely pathogenic.

The *GRN* missense variant was identified in a patient who presented with apathy, severe visual hallucinations, fluctuations in cognitive functioning, and a mild asymmetrical hypokinetic rigid syndrome, leading to a differential diagnosis of dementia with Lewy bodies, bvFTD, and CBS. Neuroimaging showed severe left frontal atrophy, suggestive of underlying FTLD. The patient's brother was clinically diagnosed with CBS and also suffered from prominent visual hallucinations. Their father had died at the age of 59 years with severe behavioral and memory disturbances. DNA of these affected relatives was not available for testing. Its pathogenicity is supported by absence in gnomAD, reduced serum progranulin levels (13.4 ng/mL) in the carrier, and the previous report of a different amino acid change in the same codon in 2 other families (p.W7R) (Saracino et al., 2019).

3.3. Variants of uncertain significance

We found 15 different VUS (Table 2 and Supplementary Table A.3). The variant we identified in *TUBA4A* (p.R105C) seems most relevant, as it was found in a proband with an autosomal dominant inheritance pattern, and segregation analysis revealed the same variant in 4 additional affected relatives (2 with bvFTD and 2 with unspecified dementia), whereas it was absent in an unaffected relative (aged>70 years). Its pathogenicity is further supported by its absence in gnomAD, and *in silico* tools predict a deleterious effect. FTLD-TDP pathology was confirmed in the proband, with features fitting subtype A. Based on the ACMG guidelines, without supporting functional data thus far, we interpreted the variant as VUS.

Three other variants (K389Rfs*23 in *VCP*, p.W541C in *GRN*, and p.P1084S in *DCTN1*) in patients with familial aggregation (Goldman 2) are potential candidates, but DNA of family members was not available for segregation analysis. The frameshift variant in *VCP*, due to an insertion resulting in a truncated protein, was found in a patient with bvFTD. Family history was positive for dementia and PD. Its pathogenicity is unknown as frameshift or nonsense variants have not been previously reported in *VCP*. Therefore, this variant was classified as VUS. The missense variant in *GRN* (p.W541C), predicted to be damaging, was found in a patient with nfvPPA, but plasma progranulin levels were not available. The p.P1084S variant in *DCTN1* was found in a patient with bvFTD and additional semantic deficits, without parkinsonism or motor neuron disease.

For the remaining 11 variants, pathogenicity remains questionable either because of benign or contradictory *in silico* predictions or because DNA from other family members was not available for segregation analyses. Of note, the VUS in *SQSTM1* (p.A33V) was detected in 2 unrelated patients. This variant was also found in the Healthy Exomes database (minor allele frequency, 0.004).

3.4. Patients with unknown genetic cause

We did not identify any pathogenic variant, likely pathogenic variant, or VUS in the 26 screened genes in the remaining 108 (55%) patients. Although >75% had Goldman scores 3–4, this group also included 6 (6%) patients with Goldman 1 and 18 (17%) with Goldman 2. The majority (65%) was diagnosed with bvFTD; a relatively large proportion (21%) in this group had svPPA. Other diagnoses included nfvPPA (13%) and lvPPA (1%). Concomitant parkinsonism was present in 14 patients and 6 suffered from ALS. Seventeen patients underwent pathological examination and showed a variety of FTLD pathologies (Supplementary Tables A.3 and A.4).

4. Discussion

In the present study of a large cohort of familial FTD, we revealed pathogenic variants in 8 FTD-related genes, with the

Table 2
Fifteen variants of uncertain significance detected in 16 familial patients

Gene	Transcript	Nucleotide change	Amino acid change	gnomAD MAF[a]	Pat. Tools[b]	CADD score[c]	Goldman score
TUBA4A	NM_006000	exon3:c.313C>T	R105C	0	D/D/D/T	32	1
GRN	NM_002087	exon12:c.1623G>C	W541C	0	D/D/D/T	34	2
DCTN1	NM_004082	exon28:c.3250C>T	P1084S	3.6e-05	T/D/D/T	22.7	2
UNC13A	NM_001080421	exon10:c.1005G>T	E335D	7.6e-05	T/T/T/T	15.7	2
TREM2	NM_018965	exon4:c.514C>T	P172S	2.4e-05	T/T/T/T	14.8	2
VCP	NM_007126	exon10:c.1064_1065ins[d]	K389Rfs*23	0	NA	NA	2
NEK1	NM_001199397	exon32:c.3728A>G	D1243G	0	D/D/D/T	32	3
PRKAR1B	NM_002735	exon3:c.259C>G	P87A	3.9e-05	T/T/D/D	16.3	3
DPP6	NM_130797	exon8:c.805G>A	G269R	3.2e-05	T/D/D/T	24.1	4
SIGMAR1	NM_001282205	exon4:c.463G>C	A155P	1.2e-04	NA	18.3	4
UBQLN2	NM_013444	exon1:c.401C>T	T134I	2.5e-05	T/D/D/D	17.5	4
DPP6	NM_130797	exon17:c.1673G>A	G558D	0	T/T/T/T	16.3	4
NEK1	NM_001199397	exon24:c.2023G>A	V675I	1.27e-05	T/T/D/T	15.9	4
TBK1	NM_013254	exon9:c.1000A>G	I334V	3.2e-05	T/T/T/T	14.3	4
SQSTM1	NM_003900	exon1:c.98C>T	A33V	7.7e-04	T/T/T/D	13.2	4

The variant in *SQSTM1* was detected in 2 patients. Variants are ordered to Goldman score and subsequently to CADD score. Variants with a CADD score <10 were discarded.
Key: NA, not available.
[a] Minor allele frequency of the total population (141,456 exome/genome sequences).
[b] Prediction tools: SIFT/PolyPhen2/MutationTaster/FATHMM, with T = tolerated and D = damaging.
[c] Version CADD score: GRch37-v1.4.
[d] Large insertion of 124 nucleotides leading to frameshift with stopgain.

C9orf72 repeat expansion as most common, followed by variants in *MAPT* and *GRN*. Furthermore, we identified an unexpected high frequency of the p.I383V variant in *TARDBP*, a novel homozygous *OPTN* variant, and 3 novel *GRN* variants. Finally, we found 15 VUS, including a promising variant in *TUBA4A* that cosegregated with the disease. The overall frequency of pathogenic variants sums up to 37%. Confining the analysis to patients with a strong family history (Goldman 1–2; n=70) raises this to 57%. Nonetheless, it indicates that still a substantial proportion of familial cases remains genetically unresolved.

4.1. Frequencies of known pathogenic variants

We found relatively high frequencies of variants in *MAPT* (6%) and *TARDBP* (3.5%) compared with other cohorts (Supplementary Table A.5). Variants in *TARDBP* have been reported in around 4% of familial ALS (Zou et al., 2017) but much less often in FTD (Blauwendraat et al., 2018; Ramos et al., 2019, 2020). Surprisingly, 5 unrelated *TARDBP* carriers harbored the same variant (p.I383V), suggestive of a possible founder effect. The same variant was found in other FTD cohort screens across the world (Caroppo et al., 2016; Ramos et al., 2019, 2020). Family history of our patients was not consistent with autosomal dominant transmission (i.e., high Goldman scores), possibly indicating reduced penetrance of this variant, as also suggested by others (Caroppo et al., 2016).

The repeat expansion in *C9orf72* is the most common genetic cause of familial FTD in our cohort, accounting for 21% of cases. This is in line with previous studies revealing it as the major genetic cause of familial and sporadic FTD and ALS (Majounie et al., 2012). However, there is substantial geographical variation with frequencies up to 40% in Scandinavian countries (Fostinelli et al., 2018; Oijerstedt et al., 2019; Ramos et al., 2019), contrasting with its absence in Asian cohorts (Kim et al., 2018; Tang et al., 2016). We found a *GRN* variant in 4.5%, which is less than in other cohorts, especially compared with an Italian study that reported a remarkably high frequency (Fostinelli et al., 2018) (Supplementary Table A.5). The pathogenic variant in *TBK1* (p.W445X) identified in a proband and an affected sibling is the first FTD kindred caused by a variant in *TBK1* in the Netherlands. In contrast, other studies have reported variants in *TBK1* as the fourth most common genetic cause in FTD (Greaves and Rohrer, 2019). Studies of French and Belgian cohorts found frequencies between 1% and 2% in bvFTD and even higher frequencies in FTD-ALS (Gijselinck et al., 2015; Le Ber et al., 2015; van der Zee et al., 2017).

4.2. Novel likely pathogenic variant in OPTN

The presence of a novel homozygous splice-site variant in *OPTN* (c.1242+1G>A) in a patient with lvPPA extends the clinical spectrum of *OPTN* variants, as it has never been associated with this phenotype. *OPTN* variants are extremely rare in FTD; only a few cases have been described with variants in compound heterozygous state or in combination with a *TBK1* variant, which are functionally related genes (Pottier et al., 2015, 2018). Homozygous nonsense/missense *OPTN* variants were first described to cause autosomal recessive ALS (Maruyama et al., 2010). Subsequently, numerous heterozygous variants were reported in ALS as either disease causing or as risk factor (Markovinovic et al., 2017). Thus far, the proband and sibling with nfvPPA are the first FTD cases without motor neuron disease caused by a homozygous *OPTN* variant. The parents of our patient were unaffected. A heterozygous variant in the same position was reported in a patient with familial ALS (c.1242+1delGinsAA) (Belzil et al., 2011). In this case, a second defect—possibly intronic or a copy number variation—in either *OPTN* or *TBK1* cannot be ruled out because the authors performed targeted sequencing of *OPTN* only. Others have also suggested a complex mode of inheritance regarding *OPTN* with an oligogenic basis (Pottier et al., 2015). A recent study on patients with dementia identified heterozygous missense variants in *OPTN*, but functional or segregation analyses were not available (Bartoletti-Stella et al., 2018).

4.3. Variants of uncertain significance

The segregation of a *TUBA4A* variant (p.R105C)—a gene mostly associated with ALS—in several affected family members seems promising. Neuropathologic findings in the proband resembled FTLD-TDP pathology type A. Other groups have also reported likely pathogenic *TUBA4A* variants in clinical ALS and FTD cases, yet without neuropathologic confirmation, suggesting a plausible role for this gene (Perrone et al., 2017; Smith et al., 2014). Functional studies investigating the pathogenicity of the p.R105C variant are currently ongoing.

A novel frameshift variant in *VCP* (p.K389Rfs*23) is also a plausible candidate. This variant was found in a patient with bvFTD and familial aggregation, without any symptoms of motor neuron disease or myopathy. Variants in *VCP* are associated with the classical phenotype of inclusion body myopathy with Paget's disease of bone and frontotemporal dementia (Watts et al., 2004), but cases with pure FTD or ALS have also been described, including 2 other patients in our cohort (Johnson et al., 2010; Wong et al., 2018). Some of the previously reported variants are located in the same D1 domain as this frameshift variant (Abrahao et al., 2016; Watts et al., 2004), which is predicted to lead to a truncated protein. A loss of function mechanism has not been described for *VCP*. Therefore, segregation and/or neuropathological findings consistent with previous *VCP* cases are needed to confirm its pathogenicity.

For the other identified VUS in our cohort (Table 2), genetic screens in additional cohorts, segregation analyses, and functional studies should provide further insight. Of note, the p.A33V variant in *SQSTM1* has been considered as pathogenic despite the lack of functional evidence (Dols-Icardo et al., 2018; Fecto et al., 2011; Le Ber et al., 2013), and it was detected in controls in another study (van der Zee et al., 2014).

4.4. Patients with unknown genetic cause

The wide variety of clinical syndromes and pathologies in the patients with FTD and without an identified genetic cause likely fit various underlying molecular mechanisms. The tau pathology in 4 patients may suggest the presence of unknown causal variants in genes related to *MAPT*, which may have an impact in its transcription or on the physiology of the tau protein. The strong family history in 2 svPPA cases with confirmed TDP type C was remarkable, as svPPA is nearly always sporadic (Convery et al., 2019). In addition, the presence of FUS pathology in a patient with a family history of dementia, PD, and psychiatric disorders contrasts with the sporadic occurrence of FUS cases in the literature (Neumann and Mackenzie, 2019). As FUS is part of the FET protein family, an undefined variant in 1 of the other FET genes, *TAF15* or *EWSR1*, could be considered. Variants in these genes have been reported in a small number of patients, although these were not confirmed to have FUS pathology (Ramos et al., 2019). In our patient, we did not identify any potential causal variants in these genes.

We have not identified variants in patients with bvFTD and concomitant parkinsonism or motor neuron disease, but could not exclude variants in all genes related to these disorders. It might be worthwhile to extend genetic screening to a larger set of genes, as a recent study on sporadic FTD has shown potential variants in genes associated with a variety of disorders (Ciani et al., 2019). Such

genetic pleiotropy alludes to an important issue in all next-generation sequencing studies: the list of genes associated with neurodegeneration continuously grows, and the phenotypical spectra of different subtypes coincide. In this study, we confined to genes associated with FTD, FTD-ALS, and AD to avoid large numbers of VUS and report those that justify further investigation. Fortunately, as the sequencing data permit constant reanalysis of novel genes, we expect that more and more cases will be resolved over time.

4.5. Limitations of the study

As our objective was to give an overview of familial FTD, we focused on cases with a positive family history, representing 43% of our total cohort. Despite our interpretation of a positive family history being rather unconstrained, we might have missed de novo variants or variants with incomplete penetrance in sporadic cases. Several previous studies have revealed *GRN* variants and *C9orf72* repeat expansions in sporadic patients (Blauwendraat et al., 2018; Oijerstedt et al., 2019; Ramos et al., 2019). Nonetheless, we believe that this work reflects clinical practice, where generally familial patients are selected for genetic assessment. As we have not included patients with exclusively psychiatric disorders in the family history, we may have missed the presence of several *GRN* or *C9orf72* carriers (Lanata and Miller, 2016). Finally, as a substantial number of pathogenic variants was identified by targeted single-gene testing, we could not exclude the coexistence of pathogenic variants in other genes (e.g., in *C9orf72* carriers) (Giannoccaro et al., 2017; van Blitterswijk et al., 2013).

5. Conclusions

We present the genetic screen of a large cohort of familial FTD in which we identified a genetic cause in 37% of the patients, including novel pathogenic variants in *OPTN* and *GRN*. A large proportion of carriers of the p.I383V variant in *TARDBP* was found, suggestive of a common founder. We found several VUS, of which the novel variants in *TUBA4A* and *VCP* seem most promising. Future studies are needed to confirm their potential pathogenicity. As a whole, our study contributes to the disentanglement of the wide genetic landscape of FTD.

Disclosure statement

The authors declare no conflict of interest. Several authors of this publication are members of the European Reference Network for Rare Neurological Diseases—Project ID No 739510.

CRediT authorship contribution statement

Merel O. Mol: Data curation, Investigation, Formal analysis, Writing - original draft. **Jeroen G.J. van Rooij:** Conceptualization, Methodology, Writing - review & editing. **Tsz H. Wong:** Investigation, Writing - review & editing. **Shamiram Melhem:** Investigation. **Annemieke J.M.H. Verkerk:** Resources, Writing - review & editing. **Anneke J.A. Kievit:** Writing - review & editing. **Rick van Minkelen:** Resources, Writing - review & editing. **Rosa Rademakers:** Resources, Writing - review & editing. **Cyril Pottier:** Resources, Writing - review & editing. **Laura Donker Kaat:** Conceptualization, Writing - review & editing. **Harro Seelaar:** Conceptualization, Writing - review & editing. **John C. van Swieten:** Conceptualization, Writing - review & editing. **Elise G.P. Dopper:** Conceptualization, Supervision, Writing - review & editing.

Acknowledgements

The authors are indebted to all the patients who made this study possible. The authors also thank Prof. A.J.M. Rozemuller from the Netherlands Brain Bank for the neuropathologic examination of the cases.

This research was funded by Alzheimer Nederland and by The Dutch Research Council (NWO).

Ethical assurances: Approval of the study was provided by the Medical Ethics Review Board of the Erasmus Medical Center of Rotterdam (MEC-2009-170). Written informed consent was obtained from all participants or their legal representatives. Brain autopsy was performed in accordance with the Legal and Ethical Code of Conduct of the Netherlands Brain Bank.

Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at https://doi.org/10.1016/j.neurobiolaging.2020.07.014.

References

Abrahao, A., Abath Neto, O., Kok, F., Zanoteli, E., Santos, B., Pinto, W.B., Barsottini, O.G., Oliveira, A.S., Pedroso, J.L., 2016. One family, one gene and three phenotypes: a novel VCP (valosin-containing protein) mutation associated with myopathy with rimmed vacuoles, amyotrophic lateral sclerosis and fronto-temporal dementia. J. Neurol. Sci. 368, 352–358.

Bartoletti-Stella, A., Baiardi, S., Stanzani-Maserati, M., Piras, S., Caffarra, P., Raggi, A., Pantieri, R., Baldassari, S., Caporali, L., Abu-Rumeileh, S., Linarello, S., Liguori, R., Parchi, P., Capellari, S., 2018. Identification of rare genetic variants in Italian patients with dementia by targeted gene sequencing. Neurobiol. Aging 66, 180.e23–180.e31.

Belzil, V.V., Daoud, H., Desjarlais, A., Bouchard, J.P., Dupre, N., Camu, W., Dion, P.A., Rouleau, G.A., 2011. Analysis of OPTN as a causative gene for amyotrophic lateral sclerosis. Neurobiol. Aging 32, 555.e13–555.e14.

Blauwendraat, C., Wilke, C., Simon-Sanchez, J., Jansen, I.E., Reifschneider, A., Capell, A., Haass, C., Castillo-Lizardo, M., Biskup, S., Maetzler, W., Rizzu, P., Heutink, P., Synofzik, M., 2018. The wide genetic landscape of clinical frontotemporal dementia: systematic combined sequencing of 121 consecutive subjects. Genet. Med. 20, 240–249.

Cairns, N.J., Bigio, E.H., Mackenzie, I.R., Neumann, M., Lee, V.M., Hatanpaa, K.J., White 3rd, C.L., Schneider, J.A., Grinberg, L.T., Halliday, G., Duyckaerts, C., Lowe, J.S., Holm, I.E., Tolnay, M., Okamoto, K., Yokoo, H., Murayama, S., Woulfe, J., Munoz, D.G., Dickson, D.W., Ince, P.G., Trojanowski, J.Q., Mann, D.M., Consortium for Frontotemporal Lobar Degeneration, 2007. Neuropathologic diagnostic and nosologic criteria for frontotemporal lobar degeneration: consensus of the Consortium for Frontotemporal Lobar Degeneration. Acta Neuropathol. 114, 5–22.

Caroppo, P., Camuzat, A., Guillot-Noel, L., Thomas-Anterion, C., Couratier, P., Wong, T.H., Teichmann, M., Golfier, V., Auriacombe, S., Belliard, S., Laurent, B., Lattante, S., Millecamps, S., Clot, F., Dubois, B., van Swieten, J.C., Brice, A., Le Ber, I., 2016. Defining the spectrum of frontotemporal dementias associated with TARDBP mutations. Neurol. Genet. 2, e80.

Ciani, M., Bonvicini, C., Scassellati, C., Carrara, M., Maj, C., Fostinelli, S., Binetti, G., Ghidoni, R., Benussi, L., 2019. The missing heritability of sporadic frontotemporal dementia: new insights from rare variants in neurodegenerative candidate genes. Int. J. Mol. Sci. 20.

Convery, R., Mead, S., Rohrer, J.D., 2019. Review: clinical, genetic and neuroimaging features of frontotemporal dementia. Neuropathol. Appl. Neurobiol. 45, 6–18.

Coyle-Gilchrist, I.T., Dick, K.M., Patterson, K., Vazquez Rodriquez, P., Wehmann, E., Wilcox, A., Lansdall, C.J., Dawson, K.E., Wiggins, J., Mead, S., Brayne, C., Rowe, J.B., 2016. Prevalence, characteristics, and survival of frontotemporal lobar degeneration syndromes. Neurology 86, 1736–1743.

Dols-Icardo, O., Garcia-Redondo, A., Rojas-Garcia, R., Borrego-Hernandez, D., Illan-Gala, I., Munoz-Blanco, J.L., Rabano, A., Cervera-Carles, L., Juarez-Rufian, A., Spataro, N., De Luna, N., Galan, L., Cortes-Vicente, E., Fortea, J., Blesa, R., Grau-Rivera, O., Lleo, A., Esteban-Perez, J., Gelpi, E., Clarimon, J., 2018. Analysis of known amyotrophic lateral sclerosis and frontotemporal dementia genes reveals a substantial genetic burden in patients manifesting both diseases not carrying the C9orf72 expansion mutation. J. Neurol. Neurosurg. Psychiatry 89, 162–168.

Fecto, F., Yan, J., Vemula, S.P., Liu, E., Yang, Y., Chen, W., Zheng, J.G., Shi, Y., Siddique, N., Arrat, H., Donkervoort, S., Ajroud-Driss, S., Sufit, R.L., Heller, S.L., Deng, H.X., Siddique, T., 2011. SQSTM1 mutations in familial and sporadic amyotrophic lateral sclerosis. Arch. Neurol. 68, 1440–1446.

Fostinelli, S., Ciani, M., Zanardini, R., Zanetti, O., Binetti, G., Ghidoni, R., Benussi, L., 2018. The heritability of frontotemporal lobar degeneration: validation of

pedigree classification criteria in a northern Italy cohort. J. Alzheimers Dis. 61, 753–760.
Giannoccaro, M.P., Bartoletti-Stella, A., Piras, S., Pession, A., De Massis, P., Oppi, F., Stanzani-Maserati, M., Pasini, E., Baiardi, S., Avoni, P., Parchi, P., Liguori, R., Capellari, S., 2017. Multiple variants in families with amyotrophic lateral sclerosis and frontotemporal dementia related to C9orf72 repeat expansion: further observations on their oligogenic nature. J. Neurol. 264, 1426–1433.
Gijselinck, I., Van Mossevelde, S., van der Zee, J., Sieben, A., Philtjens, S., Heeman, B., Engelborghs, S., Vandenbulcke, M., De Baets, G., Baumer, V., Cuijt, I., Van den Broeck, M., Peeters, K., Mattheijssens, M., Rousseau, F., Vandenberghe, R., De Jonghe, P., Cras, P., De Deyn, P.P., Martin, J.J., Cruts, M., Van Broeckhoven, C., BELNEU Consortium, 2015. Loss of TBK1 is a frequent cause of frontotemporal dementia in a Belgian cohort. Neurology 85, 2116–2125.
Goldman, J.S., Farmer, J.M., Wood, E.M., Johnson, J.K., Boxer, A., Neuhaus, J., Lomen-Hoerth, C., Wilhelmsen, K.C., Lee, V.M., Grossman, M., Miller, B.L., 2005. Comparison of family histories in FTLD subtypes and related tauopathies. Neurology 65, 1817–1819.
Greaves, C.V., Rohrer, J.D., 2019. An update on genetic frontotemporal dementia. J. Neurol. 266, 2075–2086.
Guerreiro, R., Sassi, C., Raphael Gibbs, J., Edsall, C., Hernandez, D., Brown, K., Lupton, M.K., Parkinnen, L., Ansorge, O., Hodges, A., Ryten, M., Tienari, P.J., Van Deerlin, V.M., Trojanowski, J.Q., Morgan, K., Powell, J., Singleton, A., Hardy, J., Bras, J., 2018. A comprehensive assessment of benign genetic variability for neurodegenerative disorders. bioRxiv 270686.
Johnson, J.O., Mandrioli, J., Benatar, M., Abramzon, Y., Van Deerlin, V.M., Trojanowski, J.Q., Gibbs, J.R., Brunetti, M., Gronka, S., Wuu, J., Ding, J., McCluskey, L., Martinez-Lage, M., Falcone, D., Hernandez, D.G., Arepalli, S., Chong, S., Schymick, J.C., Rothstein, J., Landi, F., Wang, Y.D., Calvo, A., Mora, G., Sabatelli, M., Monsurro, M.R., Battistini, S., Salvi, F., Spataro, R., Sola, P., Borghero, G., , ITALSGEN Consortium, Galassi, G., Scholz, S.W., Taylor, J.P., Restagno, G., Chio, A., Traynor, B.J., 2010. Exome sequencing reveals VCP mutations as a cause of familial ALS. Neuron 68, 857–864.
Kim, E.J., Kim, Y.E., Jang, J.H., Cho, E.H., Na, D.L., Seo, S.W., Jung, N.Y., Jeong, J.H., Kwon, J.C., Park, K.H., Park, K.W., Lee, J.H., Roh, J.H., Kim, H.J., Yoon, S.J., Choi, S.H., Jang, J.W., Ki, C.S., Kim, S.H., 2018. Analysis of frontotemporal dementia, amyotrophic lateral sclerosis, and other dementia-related genes in 107 Korean patients with frontotemporal dementia. Neurobiol. Aging 72, 186.e1–186.e7.
Lanata, S.C., Miller, B.L., 2016. The behavioural variant frontotemporal dementia (bvFTD) syndrome in psychiatry. J. Neurol. Neurosurg. Psychiatry 87, 501–511.
Le Ber, I., Camuzat, A., Guerreiro, R., Bouya-Ahmed, K., Bras, J., Nicolas, G., Gabelle, A., Didic, M., De Septenville, A., Millecamps, S., Lenglet, T., Latouche, M., Kabashi, E., Campion, D., Hannequin, D., Hardy, J., Brice, A., French Clinical and Genetic Research Network on FTD/FTD-ALS, 2013. SQSTM1 mutations in French patients with frontotemporal dementia or frontotemporal dementia with amyotrophic lateral sclerosis. JAMA Neurol. 70, 1403–1410.
Le Ber, I., De Septenville, A., Millecamps, S., Camuzat, A., Caroppo, P., Couratier, P., Blanc, F., Lacomblez, L., Sellal, F., Fleury, M.C., Meininger, V., Cazeneuve, C., Clot, F., Flabeau, O., LeGuern, E., Brice, A., French, C., French Clinical and Genetic Research Network on FTLD/FTLD-ALS, 2015. TBK1 mutation frequencies in French frontotemporal dementia and amyotrophic lateral sclerosis cohorts. Neurobiol. Aging 36, 3116.e5–3116.e8.
Mackenzie, I.R., Rademakers, R., 2007. The molecular genetics and neuropathology of frontotemporal lobar degeneration: recent developments. Neurogenetics 8, 237–248.
Majounie, E., Renton, A.E., Mok, K., Dopper, E.G., Waite, A., Rollinson, S., Chio, A., Restagno, G., Nicolaou, N., Simon-Sanchez, J., van Swieten, J.C., Abramzon, Y., Johnson, J.O., Sendtner, M., Pamphlett, R., Orrell, R.W., Mead, S., Sidle, K.C., Houlden, H., Rohrer, J.D., Morrison, K.E., Pall, H., Talbot, K., Ansorge, O., , Chromosome 9-ALS/FTD Consortium, French research network on FTLD/FTLD/ALS, ITALSGEN Consortium, Hernandez, D.G., Arepalli, S., Sabatelli, M., Mora, G., Corbo, M., Giannini, F., Calvo, A., Englund, E., Borghero, G., Floris, G.L., Remes, A.M., Laaksovirta, H., McCluskey, L., Trojanowski, J.Q., Van Deerlin, V.M., Schellenberg, G.D., Nalls, M.A., Drory, V.E., Lu, C.S., Yeh, T.H., Ishiura, H., Takahashi, Y., Tsuji, S., Le Ber, I., Brice, A., Drepper, C., Williams, N., Kirby, J., Shaw, P., Hardy, J., Tienari, P.J., Heutink, P., Morris, H.R., Pickering-Brown, S., Traynor, B.J., 2012. Frequency of the C9orf72 hexanucleotide repeat expansion in patients with amyotrophic lateral sclerosis and frontotemporal dementia: a cross-sectional study. Lancet Neurol. 11, 323–330.
Mann, D.M.A., Snowden, J.S., 2017. Frontotemporal lobar degeneration: Pathogenesis, pathology and pathways to phenotype. Brain Pathol. 27, 723–736.
Markovinovic, A., Cimbro, R., Ljutic, T., Kriz, J., Rogelj, B., Munitic, I., 2017. Optineurin in amyotrophic lateral sclerosis: multifunctional adaptor protein at the crossroads of different neuroprotective mechanisms. Prog. Neurobiol. 154, 1–20.
Maruyama, H., Morino, H., Ito, H., Izumi, Y., Kato, H., Watanabe, Y., Kinoshita, Y., Kamada, M., Nodera, H., Suzuki, H., Komure, O., Matsuura, S., Kobatake, K., Morimoto, N., Abe, K., Suzuki, N., Aoki, M., Kawata, A., Hirai, T., Kato, T., Ogasawara, K., Hirano, A., Takumi, T., Kusaka, H., Hagiwara, K., Kaji, R., Kawakami, H., 2010. Mutations of optineurin in amyotrophic lateral sclerosis. Nature 465, 223–226.
Moore, K.M., Nicholas, J., Grossman, M., McMillan, C.T., Irwin, D.J., Massimo, L., Van Deerlin, V.M., Warren, J.D., Fox, N.C., Rossor, M.N., Mead, S., Bocchetta, M., Boeve, B.F., Knopman, D.S., Graff-Radford, N.R., Forsberg, L.K., Rademakers, R., Wszolek, Z.K., van Swieten, J.C., Jiskoot, L.C., Meeter, L.H., Dopper, E.G., Papma, J.M., Snowden, J.S., Saxon, J., Jones, M., Pickering-Brown, S., Le Ber, I., Camuzat, A., Brice, A., Caroppo, P., Ghidoni, R., Pievani, M., Benussi, L., Binetti, G., Dickerson, B.C., Lucente, D., Krivensky, S., Graff, C., Öijerstedt, L., Fallström, M., Thonberg, H., Ghoshal, N., Morris, J.C., Borroni, B., Benussi, A., Padovani, A., Galimberti, D., Scarpini, E., Fumagalli, G.G., Mackenzie, I.R., Hsiung, G.R., Sengdy, P., Boxer, A.L., Rosen, H., Taylor, J.B., Synofzik, M., Wilke, C., Sulzer, P., Hodges, J.R., Halliday, G., Kwok, J., Sanchez-Valle, R., Lladó, A., Borrego-Ecija, S., Santana, I., Almeida, M.R., Tábuas-Pereira, M., Moreno, F., Barandiaran, M., Indakoetxea, B., Levin, J., Danek, A., Rowe, J.B., Cope, T.E., Otto, M., Anderl-Straub, S., de Mendonça, A., Maruta, C., Masellis, M., Black, S.E., Couratier, P., Lautrette, G., Huey, E.D., Sorbi, S., Nacmias, B., Laforce Jr., R., Tremblay, M.L., Vandenberghe, R., Damme, P.V., Rogalski, E.J., Weintraub, S., Gerhard, A., Onyike, C.U., Ducharme, S., Papageorgiou, S.G., Ng, A.S.L., Brodtmann, A., Finger, E., Guerreiro, R., Bras, J., Rohrer, J.D., FTD Prevention Initiative, 2020. Age at symptom onset and death and disease duration in genetic frontotemporal dementia: an international retrospective cohort study. Lancet Neurol. 19, 145–156.
Neumann, M., Mackenzie, I.R.A., 2019. Review: neuropathology of non-tau frontotemporal lobar degeneration. Neuropathol. Appl. Neurobiol. 45, 19–40.
Nykamp, K., Anderson, M., Powers, M., Garcia, J., Herrera, B., Ho, Y.Y., Kobayashi, Y., Patil, N., Thusberg, J., Westbrook, M., Invitae Clinical Genomics Group; Scott Topper, 2017. Sherloc: a comprehensive refinement of the ACMG-AMP variant classification criteria. Genet. Med. 19, 1105–1117.
Oijerstedt, L., Chiang, H.H., Bjorkstrom, J., Forsell, C., Lilius, L., Lindstrom, A.K., Thonberg, H., Graff, C., 2019. Confirmation of high frequency of C9orf72 mutations in patients with frontotemporal dementia from Sweden. Neurobiol. Aging.
Perrone, F., Nguyen, H.P., Van Mossevelde, S., Moisse, M., Sieben, A., Santens, P., De Bleecker, J., Vandenbulcke, M., Engelborghs, S., Baets, J., Cras, P., Vandenberghe, R., De Jonghe, P., De Deyn, P.P., Martin, J.J., Van Damme, P., Van Broeckhoven, C., van der Zee, J., Belgian Neurology Consortium, 2017. Investigating the role of ALS genes CHCHD10 and TUBA4A in Belgian FTD-ALS spectrum patients. Neurobiol. Aging 51, 177.e9–177.e16.
Pottier, C., Bieniek, K.F., Finch, N., van de Vorst, M., Baker, M., Perkersen, R., Brown, P., Ravenscroft, T., van Blitterswijk, M., Nicholson, A.M., DeTure, M., Knopman, D.S., Josephs, K.A., Parisi, J.E., Petersen, R.C., Boylan, K.B., Boeve, B.F., Graff-Radford, N.R., Veltman, J.A., Gilissen, C., Murray, M.E., Dickson, D.W., Rademakers, R., 2015. Whole-genome sequencing reveals important role for TBK1 and OPTN mutations in frontotemporal lobar degeneration without motor neuron disease. Acta Neuropathol. 130, 77–92.
Pottier, C., Rampersaud, E., Baker, M., Wu, G., Wuu, J., McCauley, J.L., Zuchner, S., Schule, R., Bermudez, C., Hussain, S., Cooley, A., Wallace, M., Zhang, J., Taylor, J.P., Benatar, M., Rademakers, R., 2018. Identification of compound heterozygous variants in OPTN in an ALS-FTD patient from the CReATe consortium: a case report. Amyotroph. Lateral Scler. Frontotemporal. Degener. 19, 469–471.
Pottier, C., Ren, Y., Perkerson 3rd, R.B., Baker, M., Jenkins, G.D., van Blitterswijk, M., DeJesus-Hernandez, M., van Rooij, J.G.J., Murray, M.E., Christopher, E., McDonnell, S.K., Fogarty, Z., Batzler, A., Tian, S., Vicente, C.T., Matchett, B., Karydas, A.M., Hsiung, G.R., Seelaar, H., Mol, M.O., Finger, E.C., Graff, C., Oijerstedt, L., Neumann, M., Heutink, P., Synofzik, M., Wilke, C., Prudlo, J., Rizzu, P., Simon-Sanchez, J., Edbauer, D., Roeber, S., Diehl-Schmid, J., Evers, B.M., King, A., Mesulam, M.M., Weintraub, S., Geula, C., Bieniek, K.F., Petrucelli, L., Ahern, G.L., Reiman, E.M., Woodruff, B.K., Caselli, R.J., Huey, E.D., Farlow, M.R., Grafman, J., Mead, S., Grinberg, L.T., Spina, S., Grossman, M., Irwin, D.J., Lee, E.B., Suh, E., Snowden, J., Mann, D., Ertekin-Taner, N., Uitti, R.J., Wszolek, Z.K., Josephs, K.A., Parisi, J.E., Knopman, D.S., Petersen, R.C., Hodges, J.R., Piguet, O., Geier, E.G., Yokoyama, J.S., Rissman, R.A., Rogaeva, E., Keith, J., Zinman, L., Tartaglia, M.C., Cairns, N.J., Cruchaga, C., Ghetti, B., Kofler, J., Lopez, O.L., Beach, T.G., Arzberger, T., Herms, J., Honig, L.S., Vonsattel, J.P., Halliday, G.M., Kwok, J.B., White 3rd, C.L., Gearing, M., Glass, J., Rollinson, S., Pickering-Brown, S., Rohrer, J.D., Trojanowski, J.Q., Van Deerlin, V., Bigio, E.H., Troakes, C., Al-Sarraj, S., Asmann, Y., Miller, B.L., Graff-Radford, N.R., Boeve, B.F., Seeley, W.W., Mackenzie, I.R.A., van Swieten, J.C., Dickson, D.W., Biernacka, J.M., Rademakers, R., 2019. Genome-wide analyses as part of the international FTLD-TDP whole-genome sequencing consortium reveals novel disease risk factors and increases support for immune dysfunction in FTLD. Acta Neuropathol. 137, 879–899.
Ramos, E.M., Dokuru, D.R., Van Berlo, V., Wojta, K., Wang, Q., Huang, A.Y., Deverasetty, S., Qin, Y., van Blitterswijk, M., Jackson, J., Appleby, B., Bordelon, Y., Brannelly, P., Brushaber, D.E., Dickerson, B., Dickinson, S., Domoto-Reilly, K., Faber, K., Fields, J., Fong, J., Foroud, T., Forsberg, L.K., Gavrilova, R., Ghoshal, N., Goldman, J., Graff-Radford, J., Graff-Radford, N., Grant, I., Grossman, M., Heuer, H.W., Hsiung, G.R., Huey, E., Irwin, D., Kantarci, K., Karydas, A., Kaufer, D., Kerwin, D., Knopman, D., Kornak, J., Kramer, J.H., Kremers, W., Kukull, W., Litvan, I., Ljubenkov, P., Lungu, C., Mackenzie, I., Mendez, M.F., Miller, B.L., Onyike, C., Pantelyat, A., Pearlman, R., Petrucelli, L., Potter, M., Rankin, K.P., Rascovsky, K., Roberson, E.D., Rogalski, E., Shaw, L., Syrjanen, J., Tartaglia, M.C., Tatton, N., Taylor, J., Toga, A., Trojanowski, J.Q., Weintraub, S., Wong, B., Wszolek, Z., Rademakers, R., Boeve, B.F., Rosen, H.J., Boxer, A.L., consortium, A.L., Coppola, G., 2020. Genetic screening of a large series of North American sporadic and familial frontotemporal dementia cases. Alzheimers Dement. 16, 118–130.
Ramos, E.M., Koros, C., Dokuru, D.R., Van Berlo, V., Kroupis, C., Wojta, K., Wang, Q., Andronas, N., Matsi, S., Beratis, I.N., Huang, A.Y., Lee, S.E., Bonakis, A., Florou-Hatziyiannidou, C., Fragkiadaki, S., Kontaxopoulou, D., Agiomyrgiannakis, D., Kamtsadeli, V., Tsinia, N., Papastefanopoulou, V., Stamelou, M., Miller, B.L., Stefanis, L., Papatriantafyllou, J.D., Papageorgiou, S.G., Coppola, G., 2019. Frontotemporal dementia spectrum: first genetic screen in a Greek cohort. Neurobiol. Aging 75, 224.e1–224.e8.
Renton, A.E., Majounie, E., Waite, A., Simon-Sanchez, J., Rollinson, S., Gibbs, J.R., Schymick, J.C., Laaksovirta, H., van Swieten, J.C., Myllykangas, L., Kalimo, H.,

Paetau, A., Abramzon, Y., Remes, A.M., Kaganovich, A., Scholz, S.W., Duckworth, J., Ding, J., Harmer, D.W., Hernandez, D.G., Johnson, J.O., Mok, K., Ryten, M., Trabzuni, D., Guerreiro, R.J., Orrell, R.W., Neal, J., Murray, A., Pearson, J., Jansen, I.E., Sondervan, D., Seelaar, H., Blake, D., Young, K., Halliwell, N., Callister, J.B., Toulson, G., Richardson, A., Gerhard, A., Snowden, J., Mann, D., Neary, D., Nalls, M.A., Peuralinna, T., Jansson, L., Isoviita, V.M., Kaivorinne, A.L., Holtta-Vuori, M., Ikonen, E., Sulkava, R., Benatar, M., Wuu, J., Chio, A., Restagno, G., Borghero, G., Sabatelli, M., Consortium, I., Heckerman, D., Rogaeva, E., Zinman, L., Rothstein, J.D., Sendtner, M., Drepper, C., Eichler, E.E., Alkan, C., Abdullaev, Z., Pack, S.D., Dutra, A., Pak, E., Hardy, J., Singleton, A., Williams, N.M., Heutink, P., Pickering-Brown, S., Morris, H.R., Tienari, P.J., Traynor, B.J., 2011. A hexanucleotide repeat expansion in C9ORF72 is the cause of chromosome 9p21-linked ALS-FTD. Neuron 72, 257–268.

Richards, S., Aziz, N., Bale, S., Bick, D., Das, S., Gastier-Foster, J., Grody, W.W., Hegde, M., Lyon, E., Spector, E., Voelkerding, K., Rehm, H.L., ACMG Laboratory Quality Assurance Committee, 2015. Standards and guidelines for the interpretation of sequence variants: a joint consensus recommendation of the American College of medical genetics and Genomics and the association for molecular pathology. Genet. Med. 17, 405–424.

Saracino, D., Sellami, L., Clot, F., Camuzat, A., Lamari, F., Rucheton, B., Benyounes, I., Roue-Jagot, C., Lagarde, J., Sarazin, M., Jornea, L., Forlani, S., LeGuern, E., Dubois, B., Brice, A., Le Ber, I., 2019. The missense p.Trp7Arg mutation in GRN gene leads to progranulin haploinsufficiency. Neurobiol. Aging.

Seelaar, H., Kamphorst, W., Rosso, S.M., Azmani, A., Masdjedi, R., de Koning, I., Maat-Kievit, J.A., Anar, B., Donker Kaat, L., Breedveld, G.J., Dooijes, D., Rozemuller, J.M., Bronner, I.F., Rizzu, P., van Swieten, J.C., 2008. Distinct genetic forms of frontotemporal dementia. Neurology 71, 1220–1226.

Smith, B.N., Ticozzi, N., Fallini, C., Gkazi, A.S., Topp, S., Kenna, K.P., Scotter, E.L., Kost, J., Keagle, P., Miller, J.W., Calini, D., Vance, C., Danielson, E.W., Troakes, C., Tiloca, C., Al-Sarraj, S., Lewis, E.A., King, A., Colombrita, C., Pensato, V., Castellotti, B., de Belleroche, J., Baas, F., ten Asbroek, A.L., Sapp, P.C., McKenna-Yasek, D., McLaughlin, R.L., Polak, M., Asress, S., Esteban-Perez, J., Munoz-Blanco, J.L., Simpson, M., , SLAGEN Consortium, van Rheenen, W., Diekstra, F.P., Lauria, G., Duga, S., Corti, S., Cereda, C., Corrado, L., Soraru, G., Morrison, K.E., Williams, K.L., Nicholson, G.A., Blair, I.P., Dion, P.A., Leblond, C.S., Rouleau, G.A., Hardiman, O., Veldink, J.H., van den Berg, L.H., Al-Chalabi, A., Pall, H., Shaw, P.J., Turner, M.R., Talbot, K., Taroni, F., Garcia-Redondo, A., Wu, Z., Glass, J.D., Gellera, C., Ratti, A., Brown Jr., R.H., Silani, V., Shaw, C.E., Landers, J.E., 2014. Exome-wide rare variant analysis identifies TUBA4A mutations associated with familial ALS. Neuron 84, 324–331.

Tang, M., Gu, X., Wei, J., Jiao, B., Zhou, L., Zhou, Y., Weng, L., Yan, X., Tang, B., Xu, J., Shen, L., 2016. Analyses MAPT, GRN, and C9orf72 mutations in Chinese patients with frontotemporal dementia. Neurobiol. Aging 46, 235.e11–235.e15.

van Blitterswijk, M., Baker, M.C., DeJesus-Hernandez, M., Ghidoni, R., Benussi, L., Finger, E., Hsiung, G.Y., Kelley, B.J., Murray, M.E., Rutherford, N.J., Brown, P.E., Ravenscroft, T., Mullen, B., Ash, P.E., Bieniek, K.F., Hatanpaa, K.J., Karydas, A., Wood, E.M., Coppola, G., Bigio, E.H., Lippa, C., Strong, M.J., Beach, T.G., Knopman, D.S., Huey, E.D., Mesulam, M., Bird, T., White 3rd, C.L., Kertesz, A., Geschwind, D.H., Van Deerlin, V.M., Petersen, R.C., Binetti, G., Miller, B.L., Petrucelli, L., Wszolek, Z.K., Boylan, K.B., Graff-Radford, N.R., Mackenzie, I.R., Boeve, B.F., Dickson, D.W., Rademakers, R., 2013. C9ORF72 repeat expansions in cases with previously identified pathogenic mutations. Neurology 81, 1332–1341.

van der Zee, J., Gijselinck, I., Van Mossevelde, S., Perrone, F., Dillen, L., Heeman, B., Baumer, V., Engelborghs, S., De Bleecker, J., Baets, J., Gelpi, E., Rojas-Garcia, R., Clarimon, J., Lleo, A., Diehl-Schmid, J., Alexopoulos, P., Perneczky, R., Synofzik, M., Just, J., Schols, L., Graff, C., Thonberg, H., Borroni, B., Padovani, A., Jordanova, A., Sarafov, S., Tournev, I., de Mendonca, A., Miltenberger-Miltenyi, G., Simoes do Couto, F., Ramirez, A., Jessen, F., Heneka, M.T., Gomez-Tortosa, E., Danek, A., Cras, P., Vandenberghe, R., De Jonghe, P., De Deyn, P.P., Sleegers, K., Cruts, M., Van Broeckhoven, C., Goeman, J., Nuytten, D., Smets, K., Robberecht, W., Damme, P.V., Bleecker, J., Santens, P., Dermaut, B., Versijpt, J., Michotte, A., Ivanoiu, A., Deryck, O., Bergmans, B., Delbeck, J., Bruyland, M., Willems, C., Salmon, E., Pastor, P., Ortega-Cubero, S., Benussi, L., Ghidoni, R., Binetti, G., Hernandez, I., Boada, M., Ruiz, A., Sorbi, S., Nacmias, B., Bagnoli, S., Sorbi, S., Sanchez-Valle, R., Llado, A., Santana, I., Rosario Almeida, M., Frisoni, G.B., Maetzler, W., Matej, R., Fraidakis, M.J., Kovacs, G.G., Fabrizi, G.M., Testi, S., 2017. TBK1 mutation spectrum in an extended European patient cohort with frontotemporal dementia and amyotrophic lateral sclerosis. Hum. Mutat. 38, 297–309.

van der Zee, J., Van Langenhove, T., Kovacs, G.G., Dillen, L., Deschamps, W., Engelborghs, S., Matej, R., Vandenbulcke, M., Sieben, A., Dermaut, B., Smets, K., Van Damme, P., Merlin, C., Laureys, A., Van Den Broeck, M., Mattheijssens, M., Peeters, K., Benussi, L., Binetti, G., Ghidoni, R., Borroni, B., Padovani, A., Archetti, S., Pastor, P., Razquin, C., Ortega-Cubero, S., Hernandez, I., Boada, M., Ruiz, A., de Mendonca, A., Miltenberger-Miltenyi, G., do Couto, F.S., Sorbi, S., Nacmias, B., Bagnoli, S., Graff, C., Chiang, H.H., Thonberg, H., Perneczky, R., Diehl-Schmid, J., Alexopoulos, P., Frisoni, G.B., Bonvicini, C., Synofzik, M., Maetzler, W., vom Hagen, J.M., Schols, L., Haack, T.B., Strom, T.M., Prokisch, H., Dols-Icardo, O., Clarimon, J., Lleo, A., Santana, I., Almeida, M.R., Santiago, B., Heneka, M.T., Jessen, F., Ramirez, A., Sanchez-Valle, R., Llado, A., Gelpi, E., Sarafov, S., Tournev, I., Jordanova, A., Parobkova, E., Fabrizi, G.M., Testi, S., Salmon, E., Strobel, T., Santens, P., Robberecht, W., De Jonghe, P., Martin, J.J., Cras, P., Vandenberghe, R., De Deyn, P.P., Cruts, M., Sleegers, K., Van Broeckhoven, C., 2014. Rare mutations in SQSTM1 modify susceptibility to frontotemporal lobar degeneration. Acta Neuropathol. 128, 397–410.

Watts, G.D., Wymer, J., Kovach, M.J., Mehta, S.G., Mumm, S., Darvish, D., Pestronk, A., Whyte, M.P., Kimonis, V.E., 2004. Inclusion body myopathy associated with Paget disease of bone and frontotemporal dementia is caused by mutant valosin-containing protein. Nat. Genet. 36, 377–381.

Wong, T.H., Pottier, C., Hondius, D.C., Meeter, L.H.H., van Rooij, J.G.J., Melhem, S., Netherlands Brain, b., van Minkelen, R., van Duijn, C.M., Rozemuller, A.J.M., Seelaar, H., Rademakers, R., van Swieten, J.C., 2018. Three VCP mutations in patients with frontotemporal dementia. J. Alzheimers Dis. 65, 1139–1146.

Wood, E.M., Falcone, D., Suh, E., Irwin, D.J., Chen-Plotkin, A.S., Lee, E.B., Xie, S.X., Van Deerlin, V.M., Grossman, M., 2013. Development and validation of pedigree classification criteria for frontotemporal lobar degeneration. JAMA Neurol. 70, 1411–1417.

Zou, Z.Y., Zhou, Z.R., Che, C.H., Liu, C.Y., He, R.L., Huang, H.P., 2017. Genetic epidemiology of amyotrophic lateral sclerosis: a systematic review and meta-analysis. J. Neurol. Neurosurg. Psychiatry 88, 540–549.