

LETTERS

Mutations in progranulin cause tau-negative frontotemporal dementia linked to chromosome 17

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Frontotemporal dementia (FTD) is the second most common cause of dementia in people under the age of 65 years¹. A large proportion of FTD patients (35–50%) have a family history of dementia, consistent with a strong genetic component to the disease². In 1998, mutations in the gene encoding the microtubule-associated protein tau (*MAPT*) were shown to cause familial FTD with parkinsonism linked to chromosome 17q21 (FTDP-17)³. The neuropathology of patients with defined *MAPT* mutations is characterized by cytoplasmic neurofibrillary inclusions composed of hyperphosphorylated tau^{3,4}. However, in multiple FTD families with significant evidence for linkage to the same region on chromosome 17q21 (D17S1787–D17S806), mutations in *MAPT* have not been found and the patients consistently lack tau-immunoreactive inclusion pathology^{5–12}. In contrast, these patients have ubiquitin (ub)-immunoreactive neuronal cytoplasmic inclusions and characteristic lentiform ub-immunoreactive neuronal intranuclear inclusions^{11–13}. Here we demonstrate that in these families, FTD is caused by mutations in progranulin (*PGRN*) that are likely to create null alleles. *PGRN* is located 1.7 Mb centromeric of *MAPT* on chromosome 17q21.31 and encodes a 68.5-kDa secreted growth factor involved in the regulation of multiple processes including development, wound repair and inflammation¹⁴. *PGRN* has also been strongly linked to tumorigenesis¹⁴. Moreover, *PGRN* expression is increased in activated microglia in many neurodegenerative diseases including Creutzfeldt–Jakob disease, motor neuron disease and Alzheimer's disease^{15,16}. Our results identify mutations in *PGRN* as a cause of neurodegenerative disease and indicate the importance of *PGRN* function for neuronal survival.

FTD is characterized by abnormalities in behaviour, personality and language with relative preservation of perception and memory, and may also be associated with motor dysfunction, including both motor neuron disease (MND) and parkinsonism^{1,17}. The microscopic pathology of FTD varies markedly¹⁸; although some cases (~40%) have tau-positive neuronal inclusions, the majority lack tau-based histopathology and show ub-immunoreactive neurites and neuronal cytoplasmic inclusions (NCI). The ub-immunoreactive inclusions are characteristically found in layer II of the frontal and temporal neocortex and in the dentate fascia of the hippocampus (frontotemporal lobar degeneration with ubiquitin inclusions, FTLD-U)^{1,17}. This same pattern of ub-immunoreactive pathology

is also found in patients with MND and dementia¹⁹. Other than being ubiquitinated, the molecular composition of the NCI is presently unknown.

Previous genetic linkage studies in FTD families revealed a locus on chromosome 17q21²⁰ (Fig. 1a). Subsequently, over 30 mutations in *MAPT* (encoding tau) were identified that account for a proportion of these cases (FTDP-17)^{3,4} (<http://www.molgen.ua.ac.be/FTDmutations>). FTDP-17 patients with *MAPT* mutations inevitably develop tau neurofibrillary inclusion pathology⁴. However, since the identification of *MAPT* mutations in 1998, nine FTD families have been conclusively linked to chr17q21 but lack defined *MAPT* mutations (tau-negative FTD-17)^{5,12,13}. In each family, affected patients lack tau inclusions but develop ub-immunoreactive pathology typical of FTLD-U^{5–9,11–13}. Moreover, several of these families also have ub-immunoreactive lentiform neuronal intranuclear inclusions (NII) with a similar distribution to the NCI (Fig. 2a–c)^{5,6,11–13}. We previously found NII in other families—too small for genetic linkage analysis—with tau-negative FTD however these lesions were uncommon in sporadic FTD²¹. These findings led us to propose that NII are characteristic of FTD associated with this genetic locus^{12,21}.

The absence of obvious *MAPT* mutations in tau-negative FTD-17 families suggested that this form of FTD is caused either by novel *MAPT* mutations that have eluded detection or by mutations in a different gene. However, extensive sequencing of *MAPT* intronic and regulatory regions in two linked FTD-17 families (UBC17 and 1083), and analysis of soluble tau from patient brain extracts (UBC17), previously failed to find evidence of pathogenic *MAPT* mutations^{12,22}. Therefore, we examined other candidate genes within the 3.53-centimorgan (6.19-Mb) critical region defined by haplotype analysis in reported families (D17S1787–D17S806)¹². The coding exons of candidate genes were first sequenced in affected and unaffected members of UBC17 (Table 1), a large Canadian tau-negative FTD family with highly significant evidence for linkage to chromosome 17q21 (two-point LOD score of 3.65)¹². Analysis of over 80 genes (out of ~165 in the region) failed to identify a pathogenic mutation. However, when progranulin (*PGRN*) was sequenced, an insertion mutation of 4 base pairs (bp) was detected in exon 1 (c.90_91insCTGC) causing a frameshift at codon 31 that introduces a premature termination codon after a read through of 34 residues (C31LfsX34). *PGRN* is a 593-amino-acid (68.5-kDa) multifunctional growth factor that is composed of seven-and-a-half tandem

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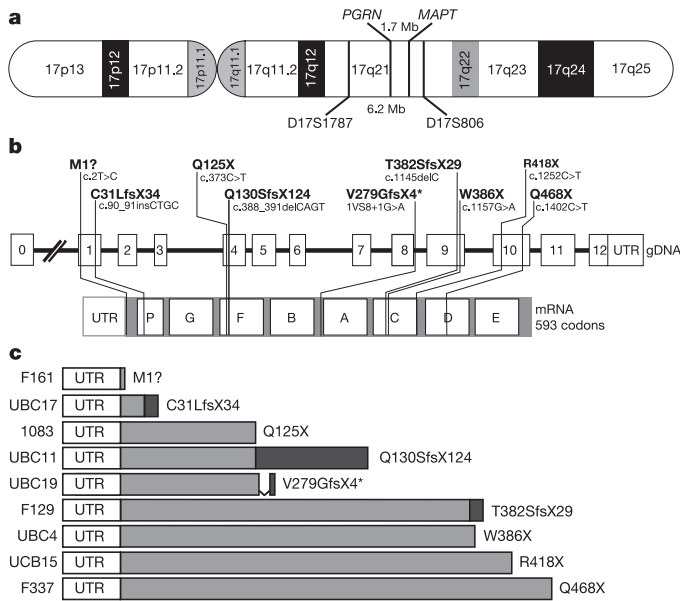


Figure 1 | Null mutations in *PGRN* cause tau-negative FTD linked to chromosome 17. **a**, Schematic representation of chromosome 17. *PGRN* is located 1.7 Mb from *MAPT*, which is mutated in FTDP-17. **b**, Schematic representation of the *PGRN* gene and mRNA encoding the PGRN protein, with positions of tau-negative FTD-17 mutations indicated. Lettered boxes refer to individual granulin repeats. **c**, Location of the mutation altering the Met 1 codon, and the premature termination codons created by the truncating mutations, in *PGRN* mRNA. The mutant mRNAs are subject to nonsense-mediated decay resulting in null alleles. The IVS8+1G>A mutation (indicated by an asterisk) is predicted to cause skipping of exon 8 (V279GfsX4).

repeats of a 12-cysteine granulin motif¹⁴. The mutant (C31LfsX34) *PGRN* protein is predicted to be only 65 residues in length, including the signal peptide, and does not contain a single intact cysteinyl repeat (Fig. 1). The C31LfsX34 mutation segregated with disease in UBC17 (Table 1, Supplementary Fig. 1), and was absent in 550 North American control individuals.

PGRN was then sequenced in affected individuals from an additional 41 families with clinical and pathological features consistent with tau-negative FTD. Families were identified in Canada (7 families), the USA (8 families), the UK (17 families), the Netherlands (1 family) and Scandinavia (8 families). This analysis identified an additional seven *PGRN* mutations in eight families, each predicted to cause premature termination of the coding sequence (Table 1, Fig. 1c). The mutations include four nonsense mutations (Q125X, W386X, R418X

and Q468X), two frameshift mutations (Q130SfsX124, T382SfsX29) and a mutation in the 5' splice site of exon 8 (IVS8+1G>A; in UBC19). The latter is likely to lead to skipping of exon 8 from the *PGRN* messenger RNA, resulting in a frameshift (V279GfsX4); however, this could not be confirmed as a source of RNA was not available. The Q130SfsX124 mutation was found in two FTD families ascertained independently in Canada (UBC11) and the UK (F53). All seven mutations segregated with disease in the relevant families (Table 1, Supplementary Fig. 1) and were absent in 200 North American and 95 UK controls.

The only two FTD families with significant evidence for linkage to 17q21 (UBC17¹² and 1083^{5,23}) were both found to have mutations in *PGRN* (Fig. 1, Supplementary Fig. 2). Notably, patients from all nine families with *PGRN* mutations met clinical criteria for FTD¹, without signs of MND, and all had neuropathological findings that included ub-immunoreactive NII, as predicted by our hypothesis that these lesions are a characteristic feature of FTD linked to this locus^{12,21}. Analysis of a further series of FTD families from Belgium also identified mutations in *PGRN*, including another family (DR8) with published evidence for linkage to chromosome 17q21^{11,23}.

To investigate the pathogenic mechanism of these truncating mutations, we determined whether premature termination of the *PGRN* coding sequence resulted in nonsense-mediated decay (NMD) of the mutant mRNAs²⁴. Quantitative PCR with reverse transcription (qRT-PCR) on RNA extracted from patient lymphoblasts carrying the R418X (UBC15) and C31LfsX34 (UBC17) mutations showed that both were associated with a ~50% reduction in total *PGRN* mRNA relative to lymphoblasts from unaffected individuals (Fig. 3a). In addition, *PGRN* mRNA from both families consisted almost entirely of wild-type mRNA with little of the mutant mRNAs detected (Fig. 3c, Supplementary Fig. 3b). Treatment of patient lymphoblasts with cycloheximide—a known inhibitor of NMD²⁴—resulted in an increase in levels of total *PGRN* mRNA (Fig. 3b) that was associated with a selective increase in the C31LfsX34 and R418X mutant mRNAs (Fig. 3c, Supplementary Fig. 3c). Furthermore, western blot analysis showed that wild-type *PGRN* protein was reduced in extracts from R418X and C31LfsX34 lymphoblasts (mean reduction 34%, $P = 0.01$, t -test) relative to extracts from unaffected relatives (Fig. 3d). We were also unable to detect the predicted mutant proteins in extracts from patient brain tissue (not shown) and lymphoblastoid cells (Fig. 3d). These data suggest that the observed premature termination mutations in *PGRN* cause disease by creating null alleles, with the mutant mRNAs being degraded by NMD. This results in loss of functional *PGRN* (haploinsufficiency) and presumably explains why there is no relationship between the location of each mutation and clinical phenotype, as each mutation has the same effect—creation of a null allele.

The proposed haploinsufficiency mechanism is also consistent

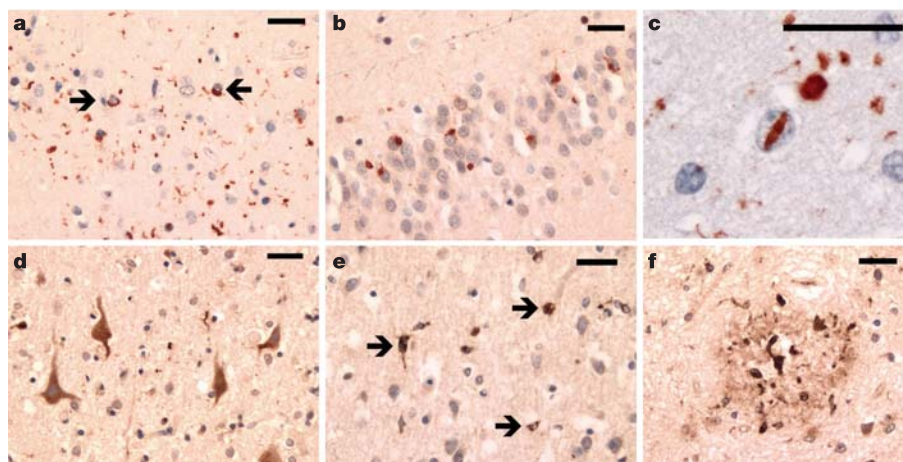


Figure 2 | Immunohistochemistry in FTD with *PGRN* mutations. **a–c**, Ubiquitin immunohistochemistry demonstrates neurites and neuronal cytoplasmic inclusions (NCI) (arrows) in layer II of the frontal neocortex (**a**), NCI in hippocampal dentate granule cells (**b**), and neuronal intranuclear inclusions (NII) in the superficial neocortex (**c**). **d–f**, *PGRN* immunohistochemistry was positive in some neocortical neurons (**d**), but did not stain NCI or NII in layer II cortex (**e**). Activated microglia (arrows) showed strong *PGRN* expression in affected areas of FTD (**e**) and around senile plaques in Alzheimer's disease (**f**). Scale bars, 30 μ m.

Table 1 | Families with premature termination mutations in PGRN

Family	Origin	Affecteds*	Mean age onset	Mutation (nucleotide)	Mutation (protein)
F161	USA	1 (1)	Early 50s	c.2T>C	p.M1?
UBC17†	Canada	17 (4)	58	c.90_91insCTGC	p.C31LfsX34
1083‡	Netherlands	19 (1)	65	c.373C>T	p.Q125X
UBC11	Canada	6 (1)	68	c.388_391delCAGT	p.Q130SfsX124
F53	UK	3 (1)	60	c.388_391delCAGT	p.Q130SfsX124
UBC19	Canada	9 (1)	61	c.IV58+1G>A	p.V279GfsX4‡
F129	USA	3 (2)	54	c.1145delC	p.T382SfsX29
UBC4	Canada	7 (1)	65	c.1157G>A	p.W386X
UBC15	Canada	10 (4)	60	c.1252C>T	p.R418X
F337	UK	5 (1)	59	c.1402C>T	p.Q468X

*Number of affecteds with NII pathology confirmed is in parentheses.

†Families with previously published linkage to chr17.

‡Predicted effect of IV58+1G>A mutation.

with our subsequent identification of a *PGRN* mutation that destroys the normal Kozak sequence by altering the Met 1 codon (c.2T>C, M1?) in another tau-negative FTD case (F161) with ub-immunoreactive pathology and NII (Table 1, Fig. 1, Supplementary Fig. 4). This mutation was absent in 200 North American controls and sequence analysis of brain complementary DNA showed a substantial reduction in mutant mRNA, providing evidence of a null allele (Supplementary Fig. 4). A second mutation that alters the Met 1 codon (c.3G>A) was also observed in a Belgian FTD patient series²³.

Next, immunohistochemistry was performed on post-mortem brain tissue using a panel of antibodies that recognize all regions of the *PGRN* protein. Consistent with previously published data on the distribution of *PGRN*^{15,16,25}, immunoreactivity was observed in a subset of cortical neurons and intensely in activated microglial cells, both in patients from FTD families with *PGRN* mutations (UBC17, UBC15 and F129) and in normal aged and Alzheimer's disease subjects (Fig. 2d–f). However, the ub-immunoreactive NCI and NII in the FTD cases with mutations showed no staining for *PGRN* (Fig. 2e). Although this finding indicates that the disease mechanism does not cause the accumulation of *PGRN* in these lesions, it leaves the identity of the ubiquitinated protein species in NCI and NII unknown.

Although the function of *PGRN* in neurons has not yet been determined¹⁴, our findings imply that *PGRN* is essential for neuronal survival and even partial loss of *PGRN* eventually leads to neurodegeneration. This supports the more general hypothesis that loss of growth factor support can cause neurodegenerative disease^{26,27}. *PGRN* is expressed in many tissues and mediates its role in development, wound repair and inflammation by activating signalling cascades that control cell-cycle progression and cell motility¹⁴. These include the mitogen-activated protein kinase (MAPK) and phosphatidylinositol-3-OH kinase (PI(3)K) cascades^{14,28}, both of which regulate crucial functions in neurons. *PGRN* also stimulates the induction of other growth factors including vascular endothelial growth factor (VEGF)²⁹. It is also of interest, that although partial loss of *PGRN* apparently results in adult-onset neurodegenerative disease, increased expression of *PGRN* has consistently been linked to tumorigenesis^{14,28}.

The identification of mutations in *PGRN* fully resolves a ten-year-old conundrum, namely the genetic basis for FTD linked to chromosome 17²⁰, and explains why multiple families linked to this region lack *MAPT* mutations. The fact that *PGRN* is located within 2 Mb of *MAPT* and mutations in both genes independently yield indistinguishable clinical phenotypes is presumably an extraordinary coincidence. Our findings are also highly reminiscent of the recent identification of probable loss-of-function mutations affecting another secreted factor—angiogenin (ANG)—in patients with MND³⁰. Tau-negative FTD and MND share overlapping clinical spectrums and have similar ub-immunoreactive pathology¹⁹. Furthermore, *PGRN* and ANG are both potent inducers of angiogenesis and are linked with tumorigenesis^{14,30,31}. Although their precise role

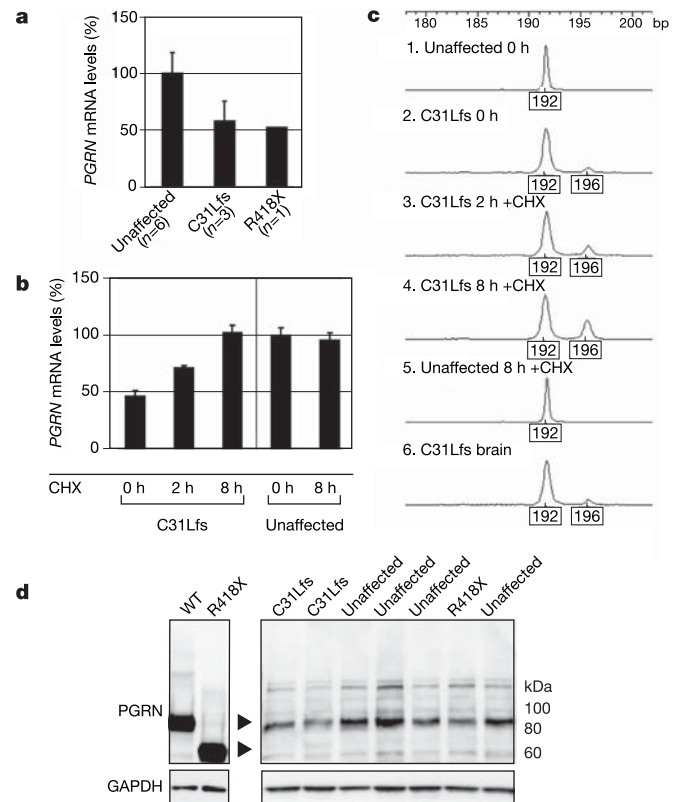


Figure 3 | Mutant *PGRN* mRNAs with premature termination codons are degraded by nonsense-mediated decay. **a**, qRT-PCR analysis shows a ~50% reduction in *PGRN* mRNA in lymphoblastoid cells from individuals carrying the C31LfsX34 (UBC17, $n = 3$) and R418X (UBC15, $n = 1$) mutations. *PGRN* mRNA levels are shown as a percentage of levels in cells from unaffected individuals ($n = 6$). Error bars indicate s.e.m. **b**, Treatment of a C31LfsX34 cell line ($n = 1$) with the NMD inhibitor cycloheximide (CHX; 500 μ M) for 2 h and 8 h results in a progressive increase in total *PGRN* mRNA. *PGRN* mRNA levels are mean values from three replicates. Error bars denote s.d. **c**, RT-PCR fragment analysis in lymphoblastoid cells (trace 2) and brain (trace 6) from patients with the C31LfsX34 mutation shows that the mutant mRNA (196 bp) is virtually absent. Treatment with CHX (traces 3–5) results in the selective increase in C31LfsX34 mutant mRNA. **d**, Western blot analysis of lymphoblastoid extracts shows that wild-type (WT) *PGRN* protein is reduced in lymphoblasts from mutation carriers (C31LfsX34, $n = 2$ and R418X, $n = 1$) relative to unaffected relatives ($n = 4$) (right panel; mean reduction 34%, $P = 0.01$, t -test). In addition, the C31LfsX34 (UBC17) and R418X (UBC15) mutant proteins are not detected. Arrows indicate wild-type *PGRN* and the expected position of the R418X mutant protein. R418X *PGRN* generated from an intronless cDNA construct (not subject to NMD) expressed in HeLa cells (left panel) is included to demonstrate that the mutant protein (if made) is stable.

in neurons has yet to be fully established, it appears that reduced levels of these functionally related factors represents a common mechanism of neurodegeneration in these two diseases. Moreover, these findings imply that replacement of these factors may represent a novel therapeutic strategy in both conditions.

METHODS

See Supplementary Information for further details on the methods used in this study.

PGRN genetic analysis. All *PGRN* exons were amplified from genomic DNA by PCR using the primers listed in Supplementary Table 1, and then sequenced using Applied Biosystems protocol. Following initial identification, the C31LfsX34 mutation was verified with a fluorescent fragment analysis assay, and then checked for both the segregation with disease (in family UBC17) and its presence in 550 North American controls. Sequence analysis of all *PGRN* exons was used to screen 200 aged North American and 95 UK control individuals for all remaining mutations. Segregation of all other mutations in relevant families was checked by sequencing of relevant exons.

Immunohistochemical methods. Immunohistochemistry was performed on post-mortem tissue sections of frontal cortex and hippocampus from two cases of familial FTD with *PGRN* mutations (UBC17 and UBC15), one case of Alzheimer's disease, and one age-matched control individual. The primary antibodies used recognize ubiquitin and all regions of the *PGRN* protein including the amino terminus, carboxy terminus, and the full-length recombinant human *PGRN* protein.

Analysis of *PGRN* protein and *PGRN* RNA. *PGRN* protein levels were quantified by western blot analysis in extracts from lymphoblastoid cells from patients and unaffected relatives using primary antibodies to the N terminus of human *PGRN*, and to *GAPDH* for normalization. *PGRN* mRNA levels were analysed in patients and unaffected relatives by qRT-PCR using SYBR green on RNA isolated from lymphoblastoid cells and from brain samples. Mass values for *PGRN* mRNA were normalized to 28S ribosomal RNA and divided by *GAPDH* mRNA to determine fold-change in expression. The relative levels of mutant and wild-type *PGRN* mRNA in C31LfsX34 lymphoblasts and brain tissue were analysed by RT-PCR fragment analysis using primers spanning exons 1 and 2. Sequence analysis of RT-PCR products was further used to analyse levels of C31LfsX34 and R418X mutant mRNAs compared with wild-type in brain tissue and lymphoblasts. To study the effect of NMD, lymphoblast cells were treated with cycloheximide for 2–8 h.

Received 12 May; accepted 29 June 2006.

Published online 16 July 2006.

- Brun, A. *et al.* Clinical and neuropathological criteria for frontotemporal dementia. The Lund and Manchester groups. *J. Neurol. Neurosurg. Psychiatry* **57**, 416–418 (1994).
- Chow, T. W., Miller, B. L., Hayashi, V. N. & Geschwind, D. H. Inheritance of frontotemporal dementia. *Arch. Neurol.* **56**, 817–822 (1999).
- Hutton, M. *et al.* Association of missense and 5' splice-site mutations in *tau* with the inherited dementia FTDP-17. *Nature* **393**, 702–705 (1998).
- Ingram, E. M. & Spillantini, M. G. *Tau* gene mutations: dissecting the pathogenesis of FTDP-17. *Trends Mol. Med.* **8**, 555–562 (2002).
- Rademakers, R. *et al.* Tau negative frontal lobe dementia at 17q21: significant finemapping of the candidate region to a 4.8 cM interval. *Mol. Psychiatry* **7**, 1064–1074 (2002).
- Rosso, S. M. *et al.* Familial frontotemporal dementia with ubiquitin-positive inclusions is linked to chromosome 17q21–22. *Brain* **124**, 1948–1957 (2001).
- Lendon, C. L. *et al.* Hereditary dysphasic disinhibition dementia: a frontotemporal dementia linked to 17q21–22. *Neurology* **50**, 1546–1555 (1998).
- Kertesz, A. *et al.* Familial frontotemporal dementia with ubiquitin-positive, tau-negative inclusions. *Neurology* **54**, 818–827 (2000).
- Froelich, S. *et al.* Mapping of a disease locus for familial rapidly progressive frontotemporal dementia to chromosome 17q12–21. *Am. J. Med. Genet.* **74**, 380–385 (1997).
- Bird, T. D. *et al.* Chromosome 17 and hereditary dementia: linkage studies in three non-Alzheimer families and kindreds with late-onset FAD. *Neurology* **48**, 949–954 (1997).
- van der Zee, J. *et al.* A Belgian ancestral haplotype harbours a highly prevalent mutation for 17q21-linked tau-negative FTD. *Brain* **129**, 841–852 (2006).
- Mackenzie, I. R. *et al.* A family with tau-negative frontotemporal dementia and neuronal intranuclear inclusions linked to chromosome 17. *Brain* **129**, 853–867 (2006).
- Rademakers, R. *et al.* In *IPSEN Meeting Research and Perspectives in Alzheimer's Disease: Genotype-Proteotype-Phenotype Relationships in Neurodegenerative Diseases* (ed. Cummings, J.) 117–137 (Springer, Paris, 2005).
- He, Z. & Bateman, A. Progranulin (granulin-epithelin precursor, PC-cell-derived growth factor, acrogranin) mediates tissue repair and tumorigenesis. *J. Mol. Med.* **81**, 600–612 (2003).
- Malaspina, A., Kaushik, N. & de Belleruche, J. Differential expression of 14 genes in amyotrophic lateral sclerosis spinal cord detected using gridded cDNA arrays. *J. Neurochem.* **77**, 132–145 (2001).
- Baker, C. A. & Manuelidis, L. Unique inflammatory RNA profiles of microglia in Creutzfeldt–Jakob disease. *Proc. Natl Acad. Sci. USA* **100**, 675–679 (2003).
- Neary, D., Snowden, J. S. & Mann, D. M. Classification and description of frontotemporal dementias. *Ann. NY Acad. Sci.* **920**, 46–51 (2000).
- Trojanowski, J. Q. & Dickson, D. Update on the neuropathological diagnosis of frontotemporal dementias. *J. Neuropathol. Exp. Neurol.* **60**, 1123–1126 (2001).
- Mackenzie, I. R. & Feldman, H. H. Ubiquitin immunohistochemistry suggests classic motor neuron disease, motor neuron disease with dementia, and frontotemporal dementia of the motor neuron disease type represent a clinicopathologic spectrum. *J. Neuropathol. Exp. Neurol.* **64**, 730–739 (2005).
- Foster, N. L. *et al.* Frontotemporal dementia and parkinsonism linked to chromosome 17: a consensus conference. *Ann. Neurol.* **41**, 706–715 (1997).
- Mackenzie, I. R. & Feldman, H. Neuronal intranuclear inclusions distinguish familial FTD–MND type from sporadic cases. *Acta Neuropathol. (Berl.)* **105**, 543–548 (2003).
- Cruts, M. *et al.* Genomic architecture of human 17q21 linked to frontotemporal dementia uncovers a highly homologous family of low-copy repeats in the *tau* region. *Hum. Mol. Genet.* **14**, 1753–1762 (2005).
- Cruts, M. *et al.* Null mutations in progranulin cause ubiquitin-positive frontotemporal dementia linked to chromosome 17q21. *Nature* advance online publication, doi:10.1038/nature05017 (16 July 2006).
- Maquat, L. E. Nonsense-mediated mRNA decay: splicing, translation and mRNP dynamics. *Nature Rev. Mol. Cell Biol.* **5**, 89–99 (2004).
- Daniel, R., He, Z., Carmichael, K. P., Halper, J. & Bateman, A. Cellular localization of gene expression for progranulin. *J. Histochem. Cytochem.* **48**, 999–1009 (2000).
- Capsoni, S. *et al.* Alzheimer-like neurodegeneration in aged antinerve growth factor transgenic mice. *Proc. Natl Acad. Sci. USA* **97**, 6826–6831 (2000).
- Salehi, A., Delcroix, J. D. & Swaab, D. F. Alzheimer's disease and NGF signaling. *J. Neural Transm.* **111**, 323–345 (2004).
- Lu, R. & Serrero, G. Mediation of estrogen mitogenic effect in human breast cancer MCF-7 cells by PC-cell-derived growth factor (PCDGF/granulin precursor). *Proc. Natl Acad. Sci. USA* **98**, 142–147 (2001).
- Tangkeangsirisin, W. & Serrero, G. PC cell-derived growth factor (PCDGF/GP88, progranulin) stimulates migration, invasiveness and VEGF expression in breast cancer cells. *Carcinogenesis* **25**, 1587–1592 (2004).
- Greenway, M. J. *et al.* ANG mutations segregate with familial and 'sporadic' amyotrophic lateral sclerosis. *Nature Genet.* **38**, 411–413 (2006).
- He, Z., Ong, C. H., Halper, J. & Bateman, A. Progranulin is a mediator of the wound response. *Nature Med.* **9**, 225–229 (2003).

Supplementary Information is linked to the online version of the paper at www.nature.com/nature.

Acknowledgements We thank the FTD research team at Vancouver Coastal Health and the University of British Columbia, and particularly G. Y. R. Hsiung, for identification and follow-up of FTD families; D. Warden, P. Whitbread and E. King (OPTIMA project, Oxford, UK) for assisting with collection of UBC17 family samples; J. Chow (Department of Pathology, University of British Columbia) for help in performing the *PGRN* immunohistochemistry; and M. Yue, J. Gonzales (Mayo Clinic), T. de Pooter and M. Van den Broeck (University of Antwerp) for technical support. This research was funded as part of the Mayo Clinic ADRC grant from the National Institute on Aging (to M.H.), the Mayo Foundation (M.H.), and the Robert and Clarice Smith Fellowship program (to S.M.). I.R.M. and H.F. were funded by the Canadian Institutes of Health research operating grant. S.M.P.-B. received grants from the Medical Research Council (UK) and the Motor Neuron Disease Association. R.R. is a postdoctoral fellow of the Fund for Scientific Research Flanders and a visiting scientist from the Neurodegenerative Brain Diseases Group of the Department of Molecular Genetics, VIB, University of Antwerp, Belgium. Finally, we acknowledge and thank the families who contributed samples, as without them this study would not have been possible.

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