Progranulin mutations in FTD - From molecule to phenotype

Michael Hutton Ph.D.
Dept. of Neuroscience
Mayo Clinic Jacksonville

Current address:
Merck Research Laboratories
33 Avenue Louis Pasteur
Boston, MA 02115

Identification of *PGRN* mutations

nature

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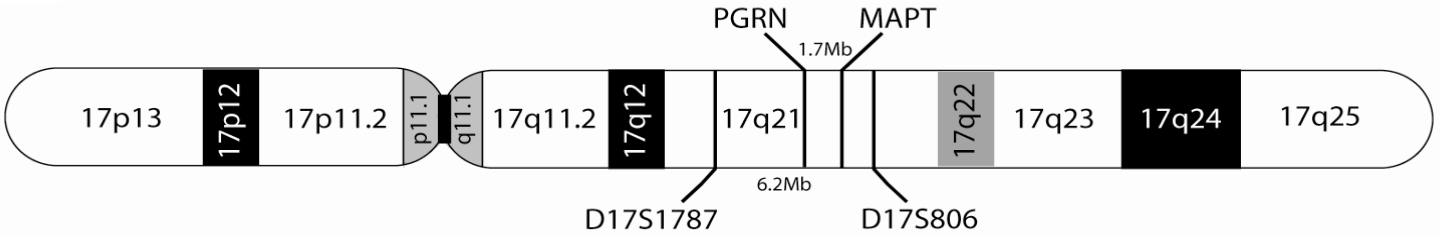
Mutations in progranulin cause tau-negative frontotemporal dementia linked to chromosome 17

Matt Baker[1]*, Ian R. Mackenzie[2]*, Stuart M. Pickering-Brown[5,6]*, Jennifer Gass[1], Rosa Rademakers[1]†, Caroline Lindholm[3], Julie Snowden[6], Jennifer Adamson[1], A. Dessa Sadovnick[3,4], Sara Rollinson[5], Ashley Cannon[1], Emily Dwosh[4], David Neary[6], Stacey Melquist[1], Anna Richardson[6], Dennis Dickson[1], Zdenek Berger[1], Jason Eriksen[1], Todd Robinson[1], Cynthia Zehr[1], Chad A. Dickey[1], Richard Crook[1], Eileen McGowan[1], David Mann[6], Bradley Boeve[7], Howard Feldman[3] & Mike Hutton[1]

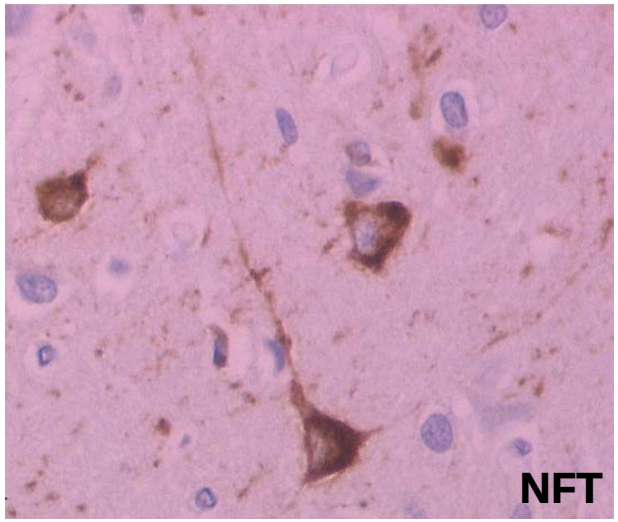
Null mutations in progranulin cause ubiquitin-positive frontotemporal dementia linked to chromosome 17q21

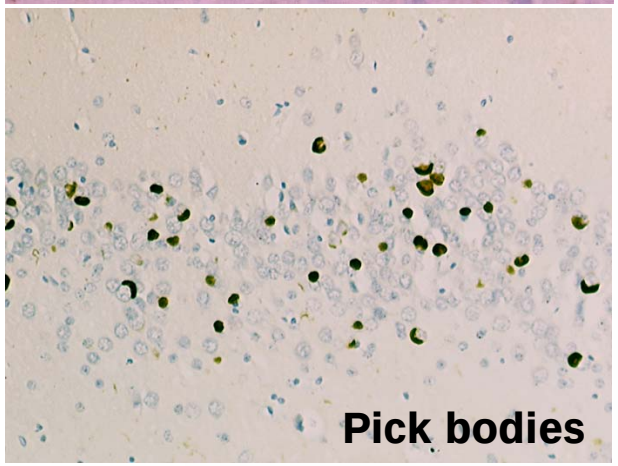
Marc Cruts[1,2,5], Ilse Gijselinck[1,2,5], Julie van der Zee[1,2,5], Sebastiaan Engelborghs[3,5,6], Hans Wils[1,2,5], Daniel Pirici[1,2,5], Rosa Rademakers[1,2,5], Rik Vandenberghe[7], Bart Dermaut[9], Jean-Jacques Martin[4,5], Cornelia van Duijn[10], Karin Peeters[1,2,5], Raf Sciot[8], Patrick Santens[9], Tim De Pooter[1,2,5], Maria Mattheijssens[1,2,5], Marleen Van den Broeck[1,2,5], Ivy Cuijt[1,2,5], Krist'l Vennekens[1,2,5], Peter P. De Deyn[3,5,6], Samir Kumar-Singh[1,2,5] & Christine Van Broeckhoven[1,2,5]



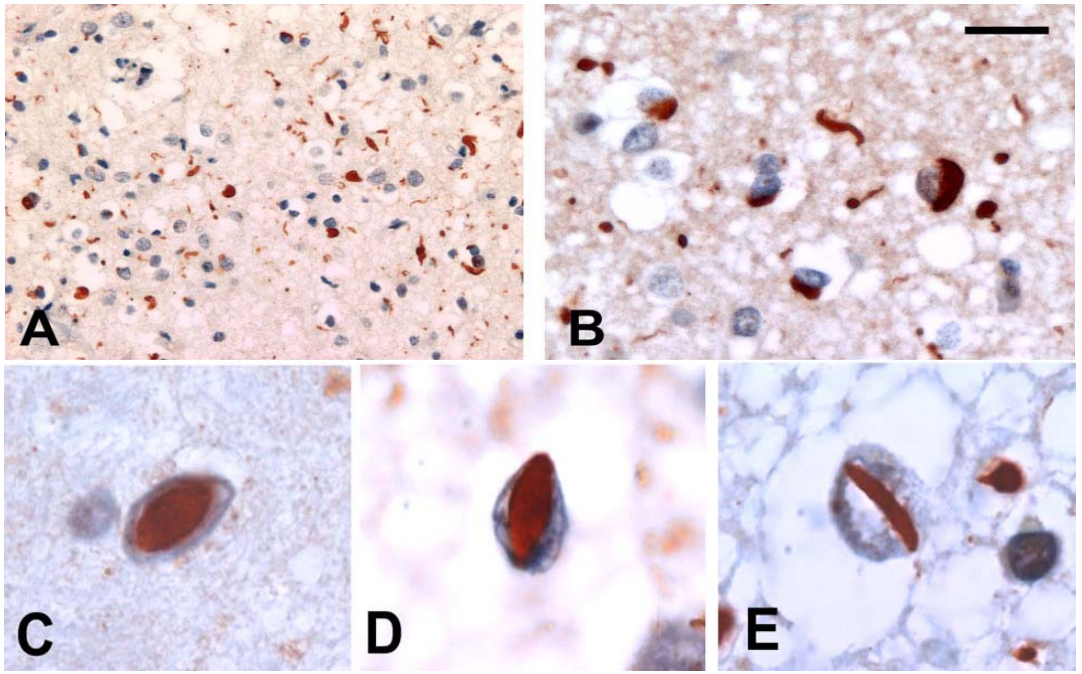
Mutations in *MAPT* (tau) *and Progranulin* explain histopathological subtypes in FTDP-17

p-tau (*MAPT mutations*)





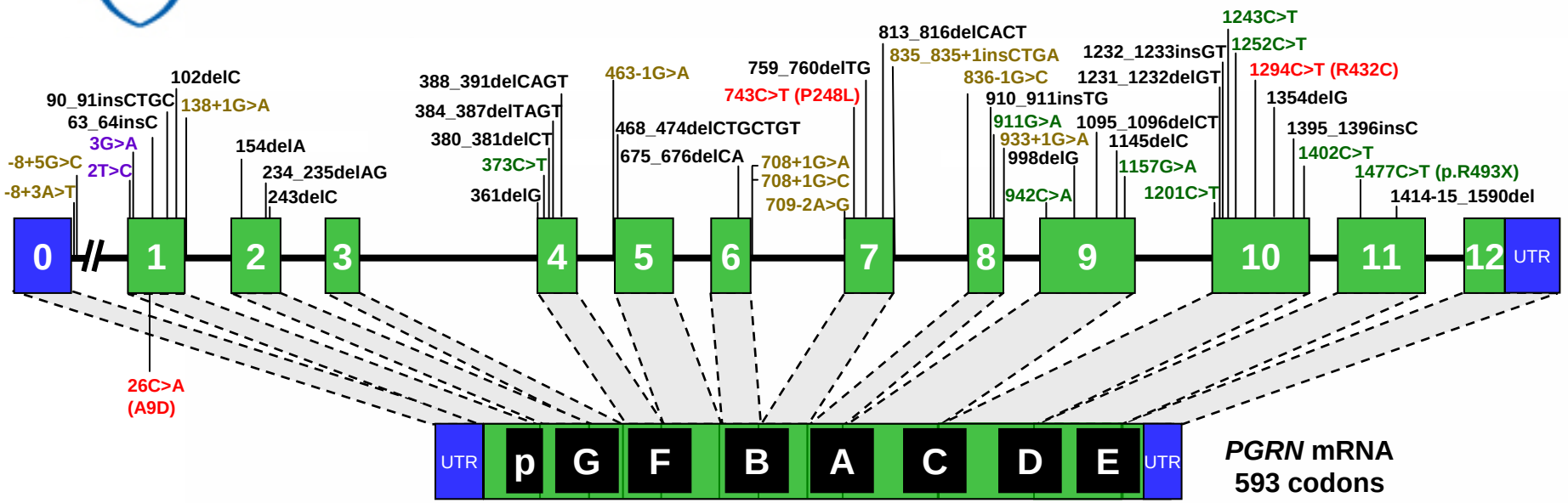
Ubiquitin/TDP-43 (*PGRN* mutations)



A, B – intracytoplasmic inclusions
C,D,E – intranuclear inclusions

NO *MAPT* mutations in families with ubiquitin-positive inclusion pathology

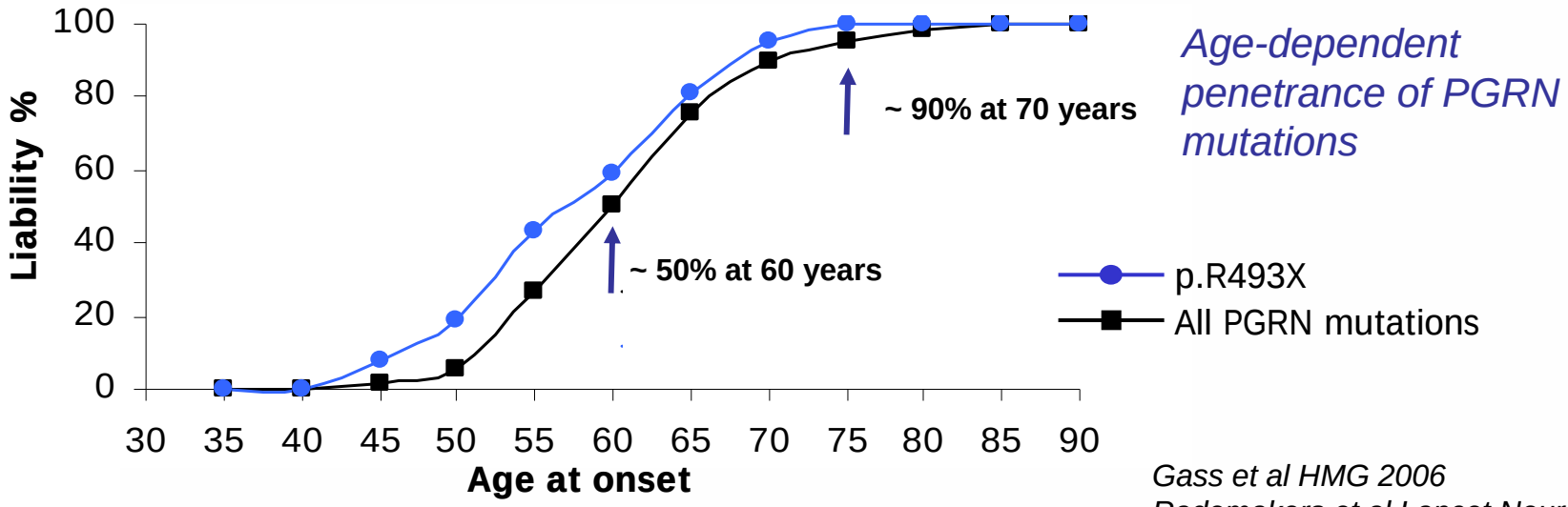
PGRN mutations in FTDP-17



- All mutations cause partial loss of functional PGRN (haploinsufficiency) - 47 mutations reported to date
  - 2 Kozak sequence mutations (p.M1?)
  - 3 Missense changes – (loss of protein function)
  - 10 Splice site mutations
  - 23 Small in/del mutations that cause frame shifts
  - 9 Nonsense mutations (most common mutation is p.493X)

Penetrance and clinicopathologic features associated with *PGRN* mutations

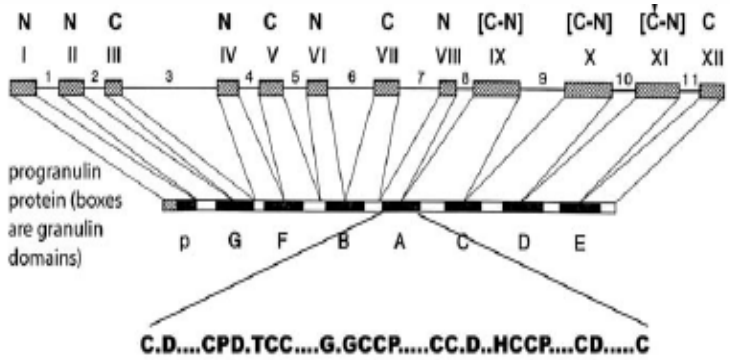
- Frequency 5.4% (10/185) in Mayo Clinic ADRC referral FTD series
  - 13.1% in cases with positive family history
- Mean age at onset 59 ± 7 years (range 48-83)
- FTD and Primary Progressive Aphasia most frequent initial clinical diagnoses
  - ~80% of carriers develop progressive language impairment during course
  - ~70% of *PGRN* mutation carriers develop Parkinsonism during disease course
  - Motor neuron disease features are very rare
- Ubiquitin/TDP-43 cytoplasmic and intranuclear inclusions in all with autopsy



Age-dependent penetrance of PGRN mutations

Gass et al HMG 2006
Rademakers et al Lancet Neurol 2007

PGRN is a pluripotent mitogenic factor



- PGRN is reported to be a 68.5 kDa secreted growth/mitogenic factor composed of 7.5 tandem repeats of a 12 cysteine motif (granulins, GRNs)
- PGRN can be processed to 6KDa GRNs peptides by extracellular protease (elastase)
- Regulates cell cycle progression and cell motility in multiple tissue remodeling processes including development and wound healing
  - **PGRN and GRNs have opposing actions in regulation of wound repair**
- PGRN overexpression associated with tumorigenesis (growth and invasion)
- PGRN is upregulated in activated microglia in neurodegenerative disease

H_2N-MWTLVSWVALTAGLVAGTR-

CPDGQFCPVACCLDPGGASYSCCRP – paragranulin (P)

-LLDKWPTTLSRHLGGP-

CQVDAHCSAGHSCIFTVSGTSSCCPFPEAVACGDGHHCCPRGFHCSADGRSC – granulin 7 (G)

-FQRSGNNSVGAIQ-

CPDSQFECPDFSTCCVMVDGSWGCCPMPQASCCEDRVHCCPHGAFCDLVHTRC – granulin 6 (F)

-ITPTGTHPLAKKLPAQRTNRAVALSSSVM-

CPDARSRCPDGSTCCELPSGKYGCCPMPNATCCSDHLHCCPQDTVCDLIQSKC – granulin 2 (B)

-LSKENATTDLLTKLPAHTVGDVK-

CDMEVSCPDGYTCCRLQSGAWGCCPFTQAVCCEDHIHCCPAGFTCDTQKGTC – granulin 1 (A)

-EQGPHQVPWMEKAPAHLSLPDPQALKRDVP-

CDNVSSCPSSDTCCQLTSGEWGCCPIPEAVCCSDHQHCCPQGYTCVAEGQC – granulin 3 (C)

-QRGSEIVAGLEKMPARRASLSHPRDIG-

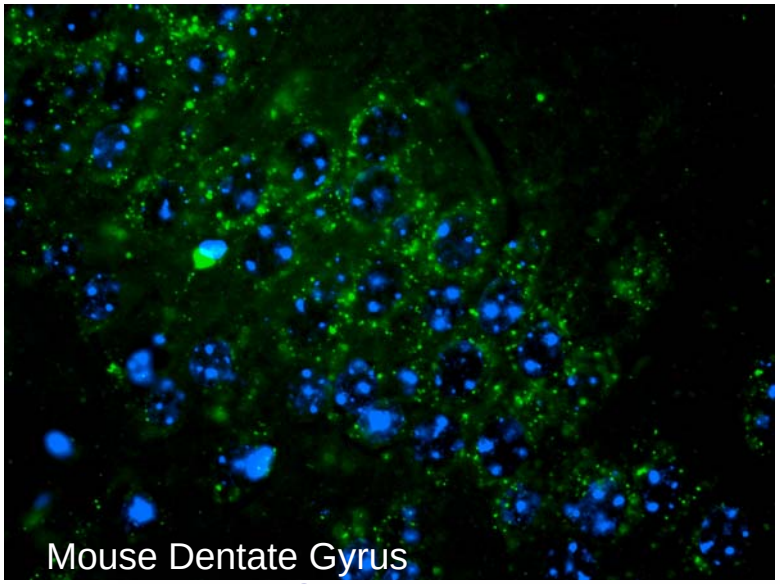
CDQHTSCPVGQTCCPSLGGSWACCQLPHAVCCEDRQHCCPAGYTCNVKARSC – granulin 4 (D)

-EKEVVSAQPATFLARSPHVGVKDVE-

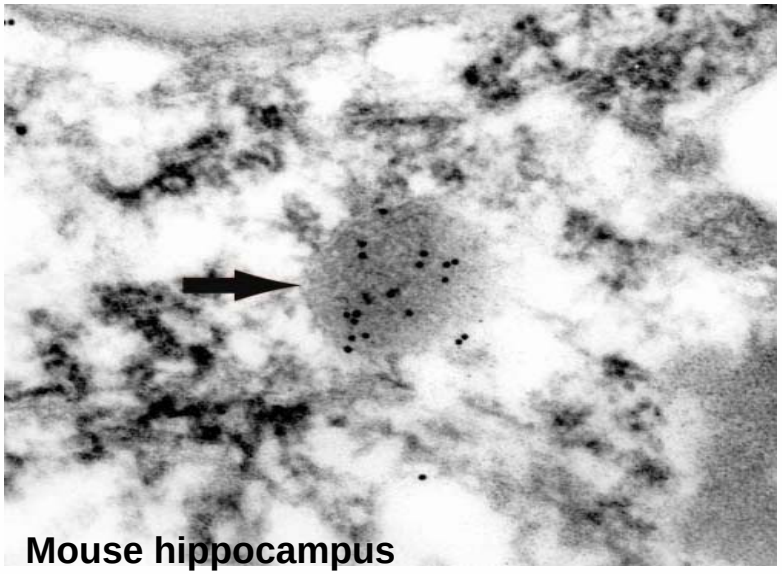
CGEGHFCHDNQTCCRDNRQGWACCPYRQGVCCADRRHCCPAGFRCAARGTKC – granulin 5 (E)

-LRREAPRWDAPLRDPALRQLL^{593}-COOH

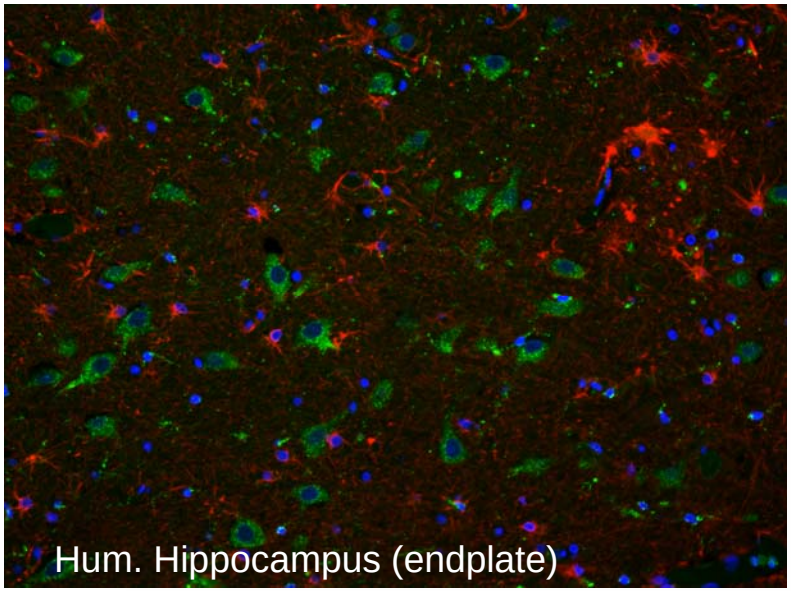
PGRN is expressed in neurons and microglia but not in astrocytes or oligodendrocytes (Dennis Dickson / Eileen McGowan)



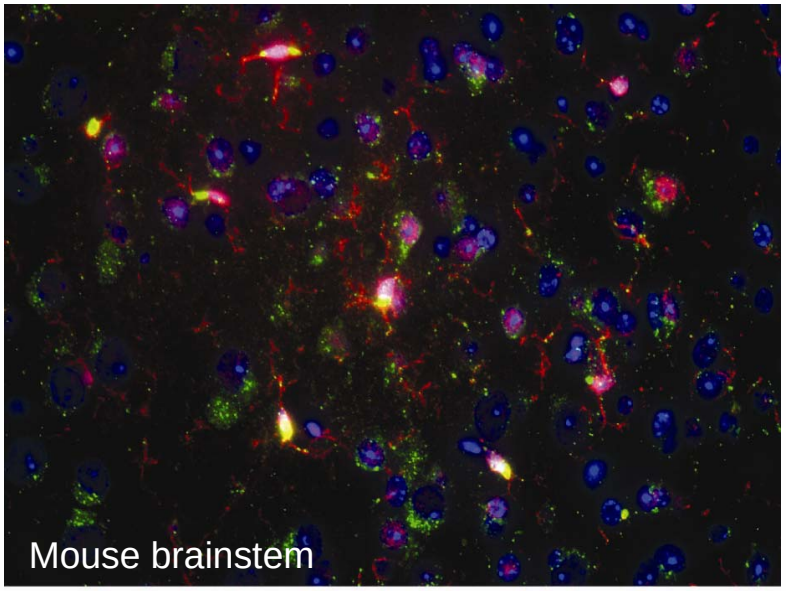
PGRN - green



PGRN – immunogold labelling

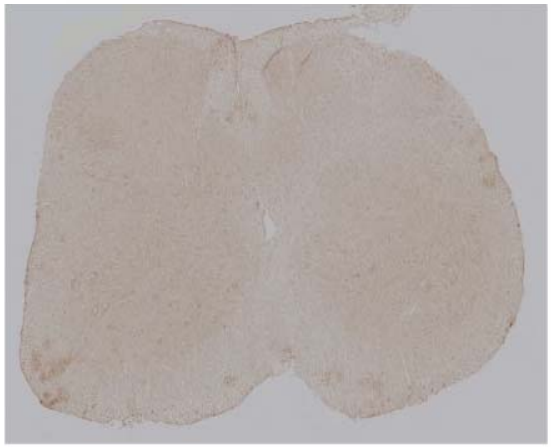
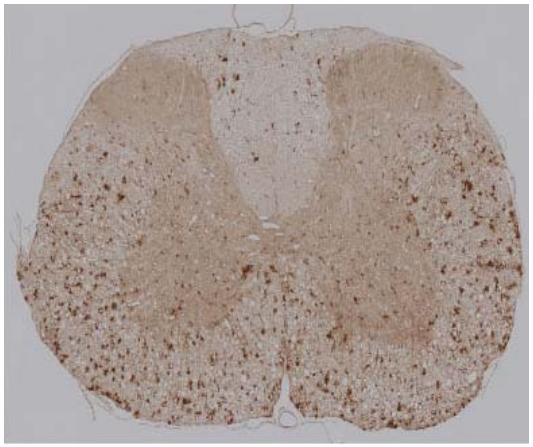
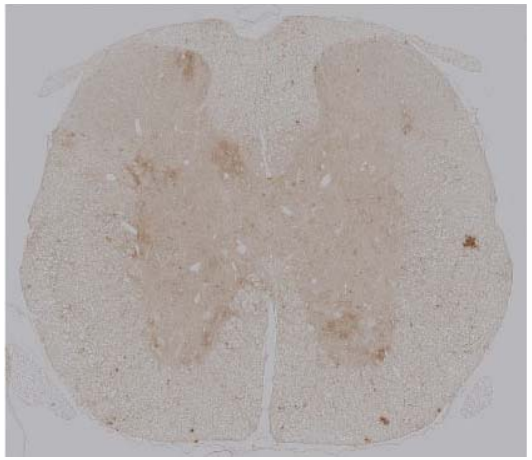


PGRN – green, GFAP - red



PGRN – green, IBA1 (microglia) - red

PGRN in spinal cord in tau (P301L) transgenic mice (JNPL3)

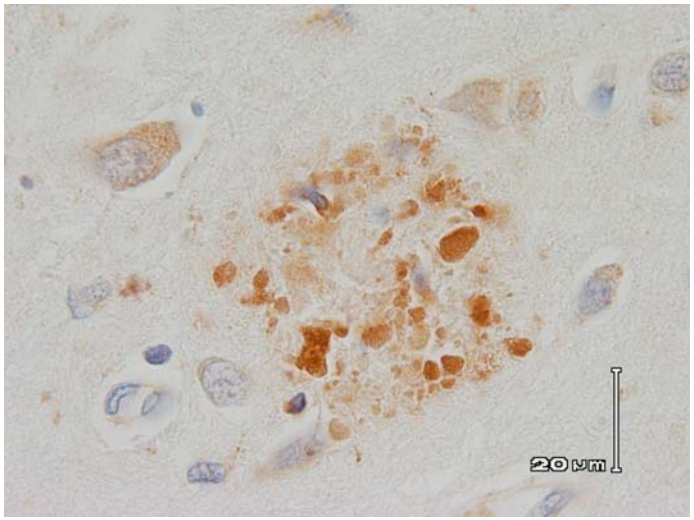
	2 mo (unaffected)	12 mo (severe)	
JNPL3 (P301L)			Massive upregulation of PGRN in microglia observed with development of tau pathology and neurodegeneration in JNPL3 mice (12months)
Non-Tg mice			Increase in neuronal PGRN expression with aging in Non-Tg mice (note darker staining in gray matter)

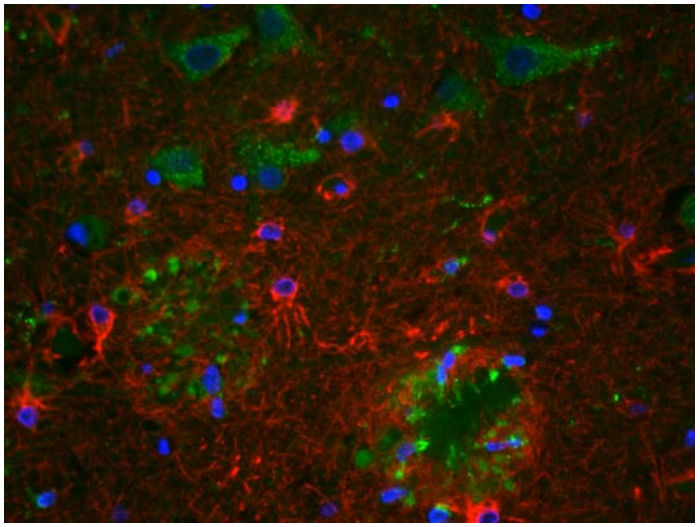
Jason Eriksen

PGRN accumulates around amyloid plaques in Alzheimer's disease and transgenic mice

PGRN IHC

Human AD





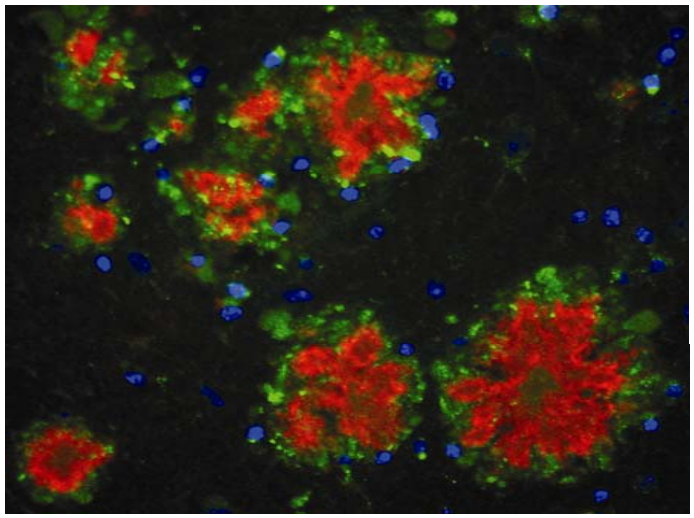
Astrocytes (Red)

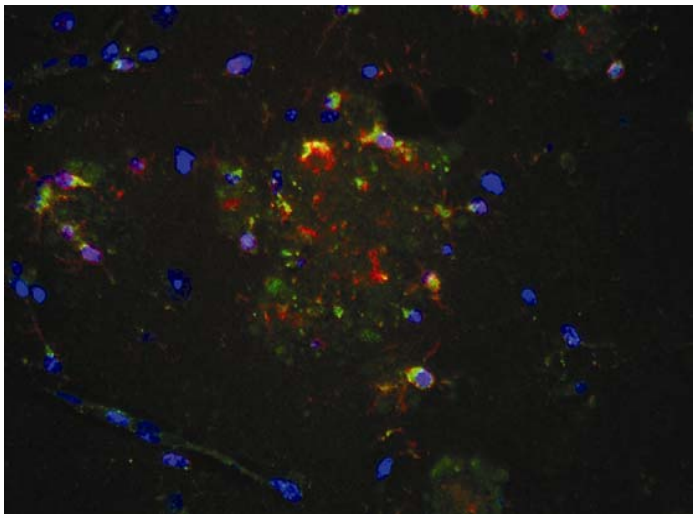
APP Tg mouse

PGRN (Green)

Aβ (Red)

APP Tg mouse

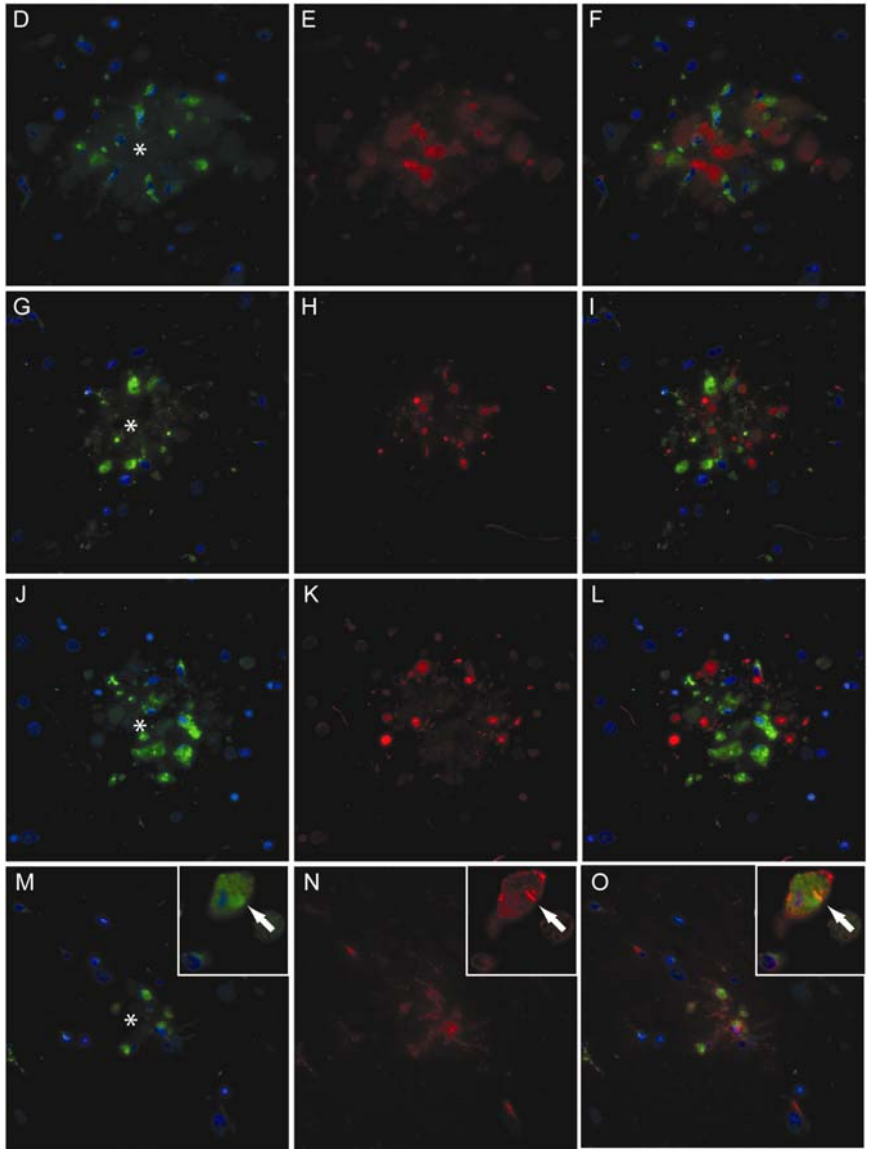




microglia (Red)

Human AD

PGRN IR is predominantly localized to microglia around amyloid plaques in human AD



APP
(dystrophic neurites)

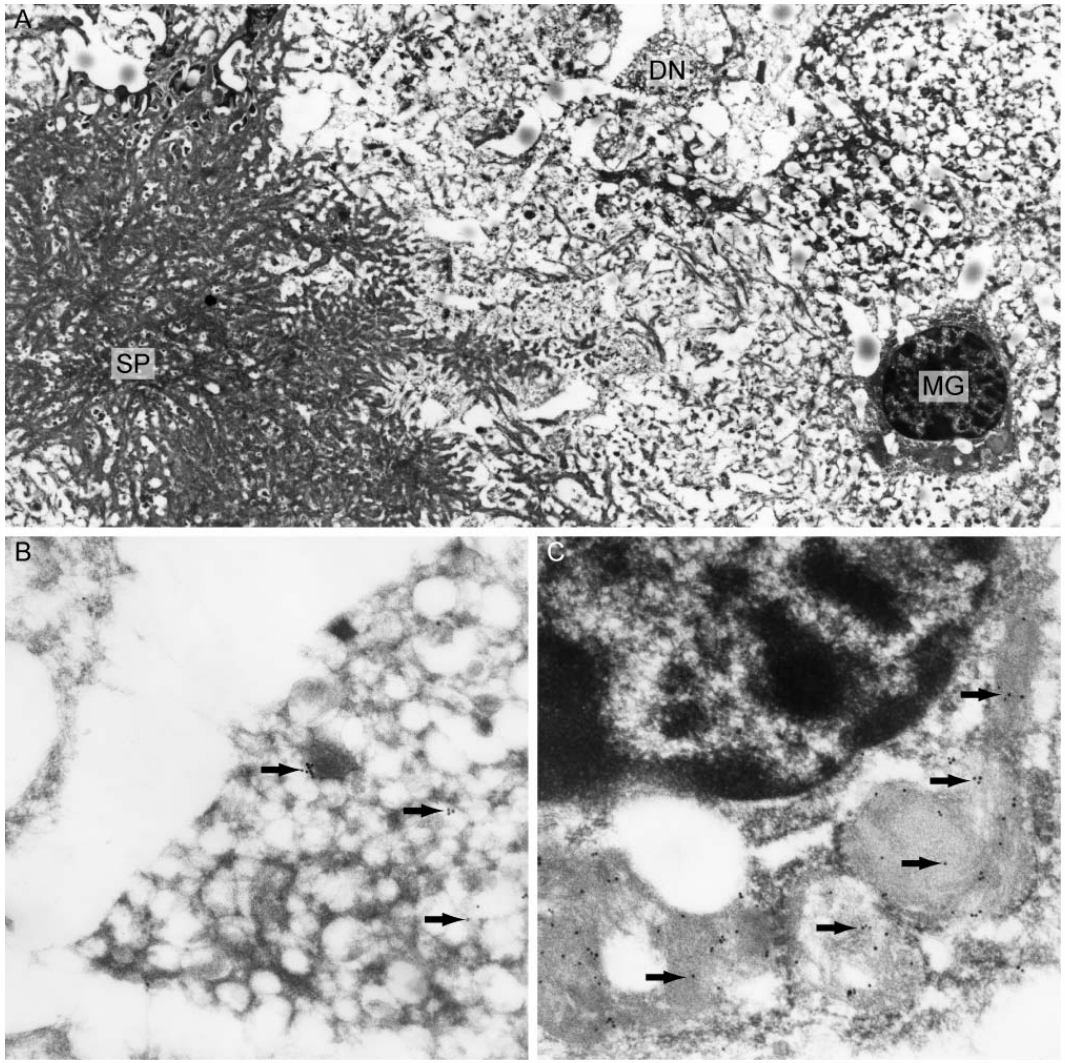
Tau
(dystrophic neurites)

Ubiquitin
(dystrophic neurites)

HLA-DR
(microglia)

Amy Innes
Zeshan Ahmed

PGRN is localized to lysosomal structures in activated microglia



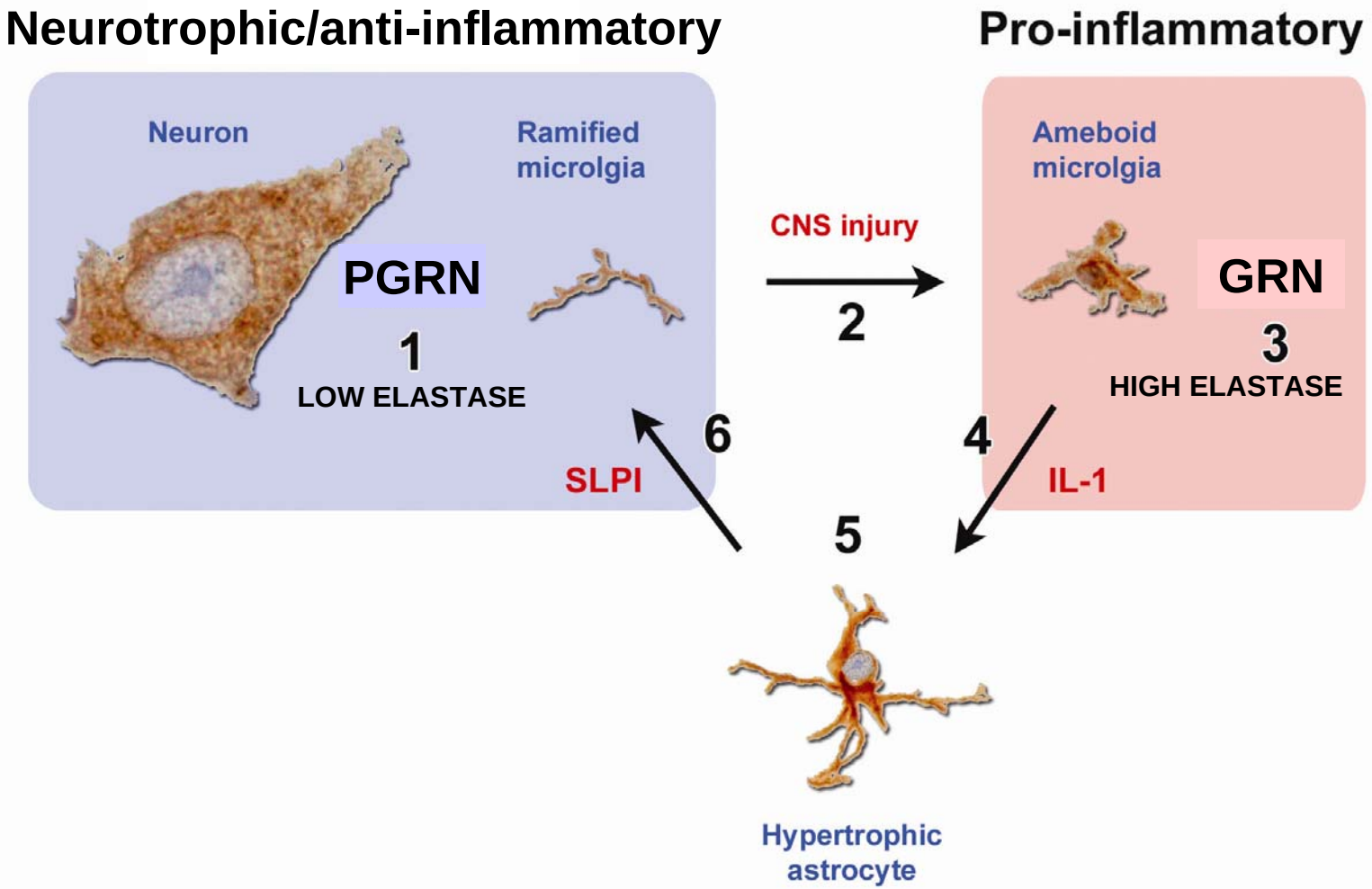
A. Low power of Senile plaque (SP)

B. Dystrophic neurite (DN)
   - Diffuse cytoplasmic staining

C. Activated microglia (MG)
   - Gold particles over lysosomal endosomal structures

Wen-Lang Lin

PGRN may regulate repair/inflammatory response to brain injury – as in periphery



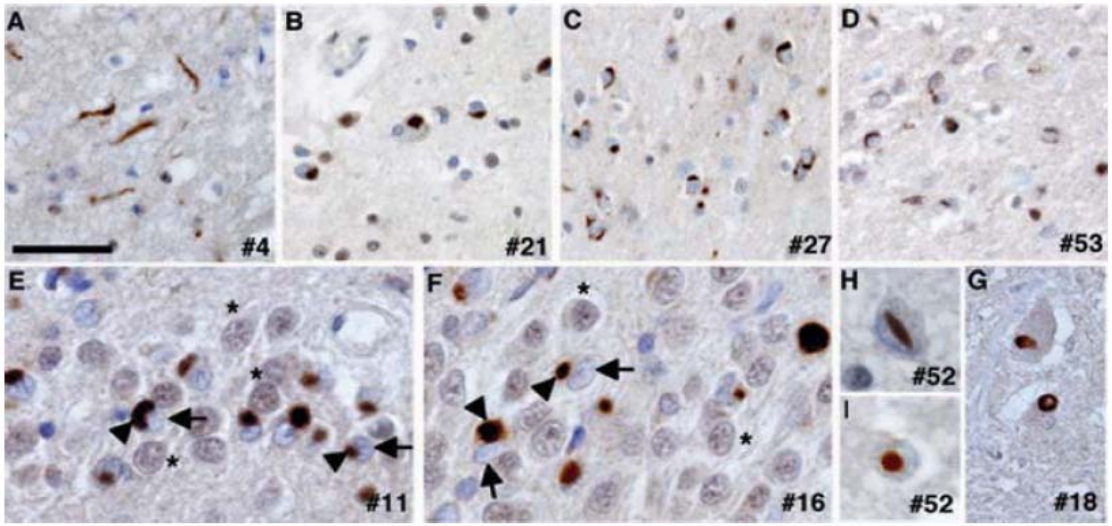
Ahmed et al. J. Neuroinflamm. 2007

Ubiquitinated TDP-43 in Frontotemporal Lobar Degeneration and Amyotrophic Lateral Sclerosis

Science

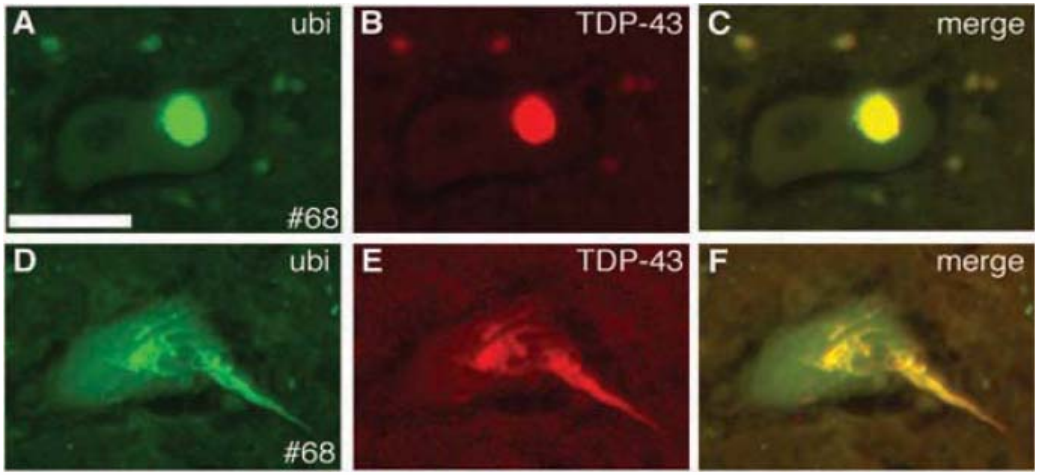
(2006) 314, 130 - 133

Manuela Neumann,[1,11*] Deepak M. Sampathu,[1*] Linda K. Kwong,[1*] Adam C. Truax,[1]
Matthew C. Micsenyi,[1] Thomas T. Chou,[2] Jennifer Bruce,[1] Theresa Schuck,[1] Murray Grossman,[3,4]
Christopher M. Clark,[3,4] Leo F. McCluskey,[3] Bruce L. Miller,[6] Eliezer Masliah,[7]
Ian R. Mackenzie,[8] Howard Feldman,[9] Wolfgang Feiden,[10] Hans A. Kretzschmar,[11]
John Q. Trojanowski,[1,4,5] Virginia M.-Y. Lee[1,4,5]†



FTLD-U
(including PGRN cases)

Idiopathic ALS
(NOT SOD1 cases)

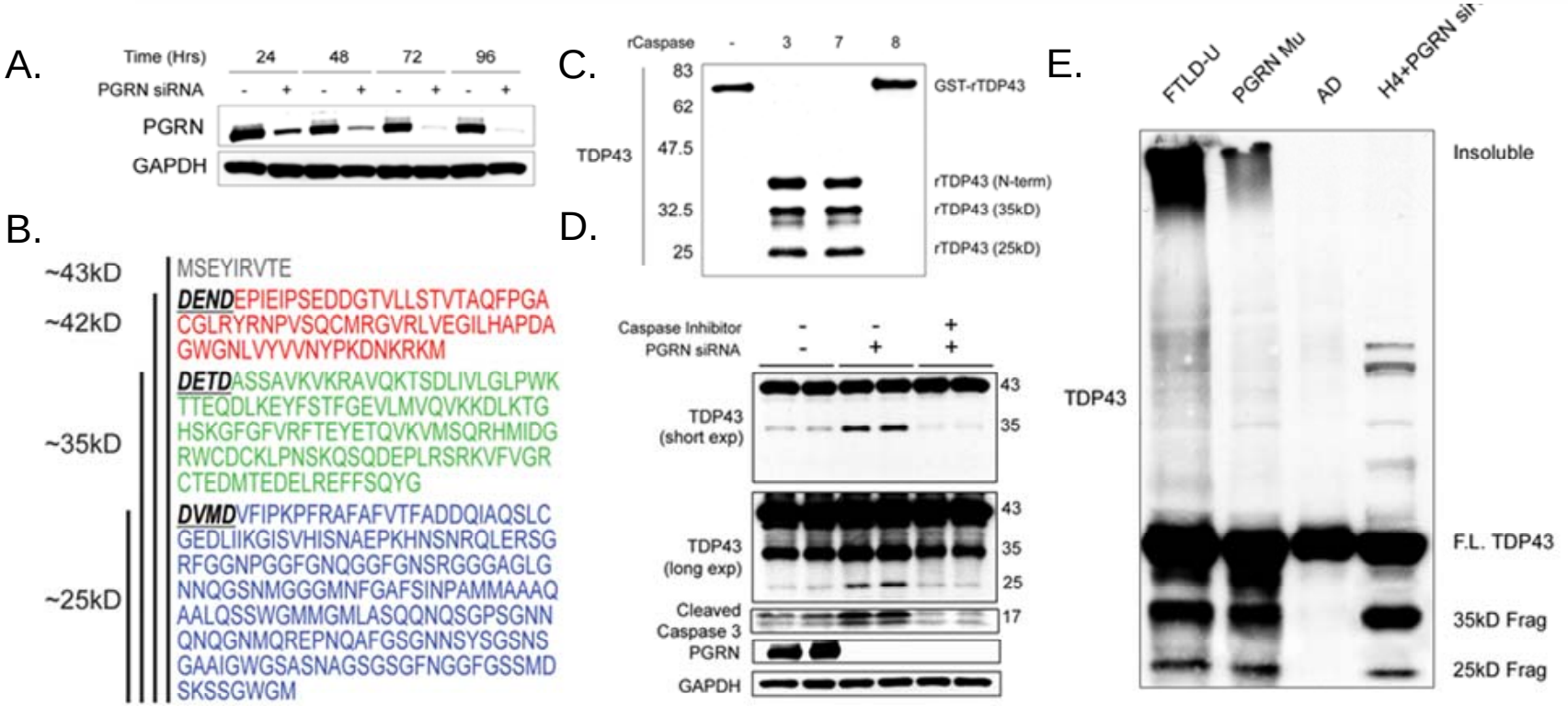


Indicates that FTLD-U (including PGRN cases) and ALS belong to a clinicopathologic spectrum of neurodegenerative diseases.

Progranulin Mediates Caspase-Dependent Cleavage of TAR DNA Binding Protein-43

Yong-Jie Zhang,[1]* Ya-fei Xu,[1]* Chad A. Dickey,[2] Emanuele Buratti,[3] Francisco Baralle,[3] Rachel Bailey,[1] Stuart Pickering-Brown,[4] Dennis Dickson,[1] and Leonard Petrucelli[1]

[1]Department of Neuroscience, Mayo Clinic College of Medicine, Jacksonville, Florida 32224, [2]Department of Molecular Pharmacology and Physiology, University of South Florida, Tampa, Florida 33612, [3]International Centre for Genetic Engineering and Biotechnology, 34012 Trieste, Italy, and [4]Division of Regenerative Medicine, University of Manchester, Manchester M13 9PT, United Kingdom



The Journal of Neuroscience, September 26, 2007 • 27(39):10530–10534

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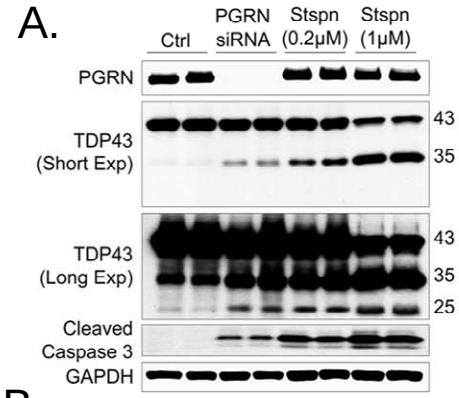
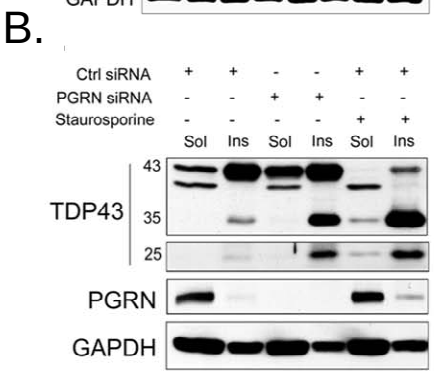
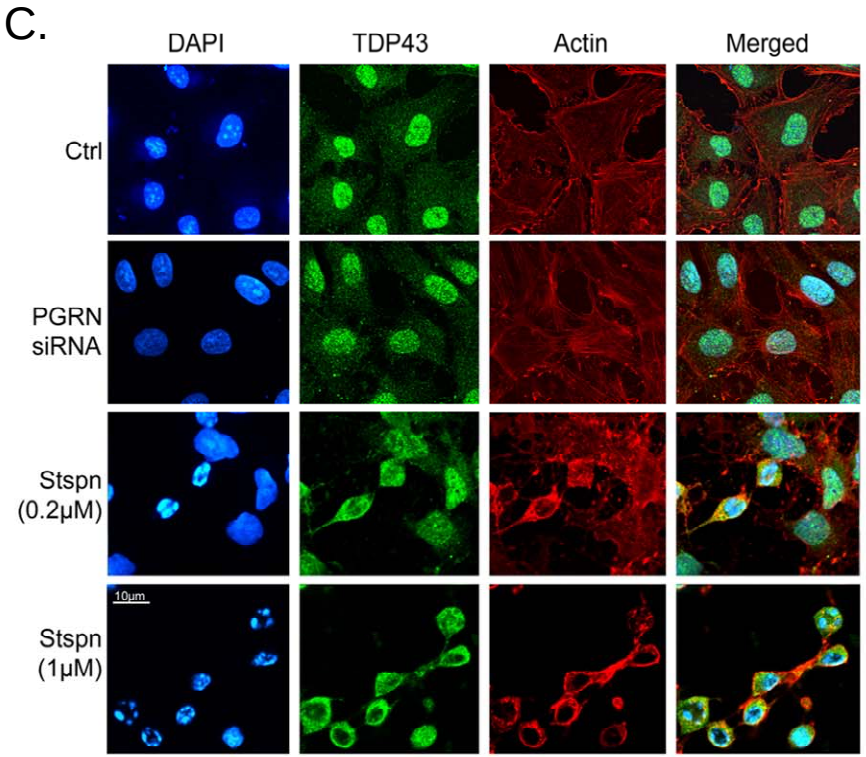
[1]Department of Neuroscience, Mayo Clinic College of Medicine, Jacksonville, Florida 32224, [2]Department of Molecular Pharmacology and Physiology, University of South Florida, Tampa, Florida 33612, [3]International Centre for Genetic Engineering and Biotechnology, 34012 Trieste, Italy, and [4]Division of Regenerative Medicine, University of Manchester, Manchester M13 9PT, United Kingdom







PGRN mut (haploins)

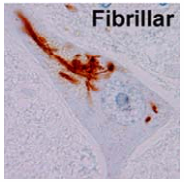
Caspase activation

TDP-43 cleavage

TDP-43 mislocalization

Cleaved TDP-43 Nucleates aggregation

TDP-43 sequestration

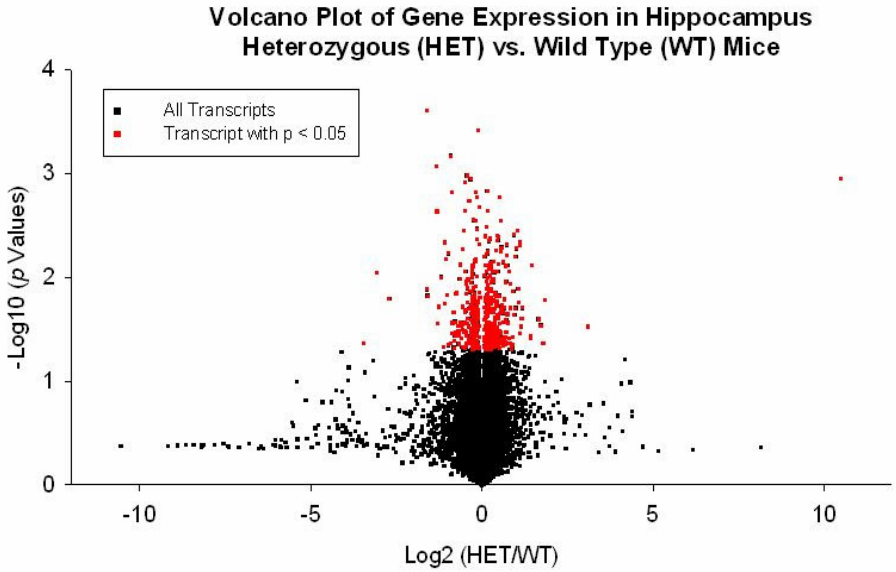


Nuclear TDP-43 Depletion/toxoicity

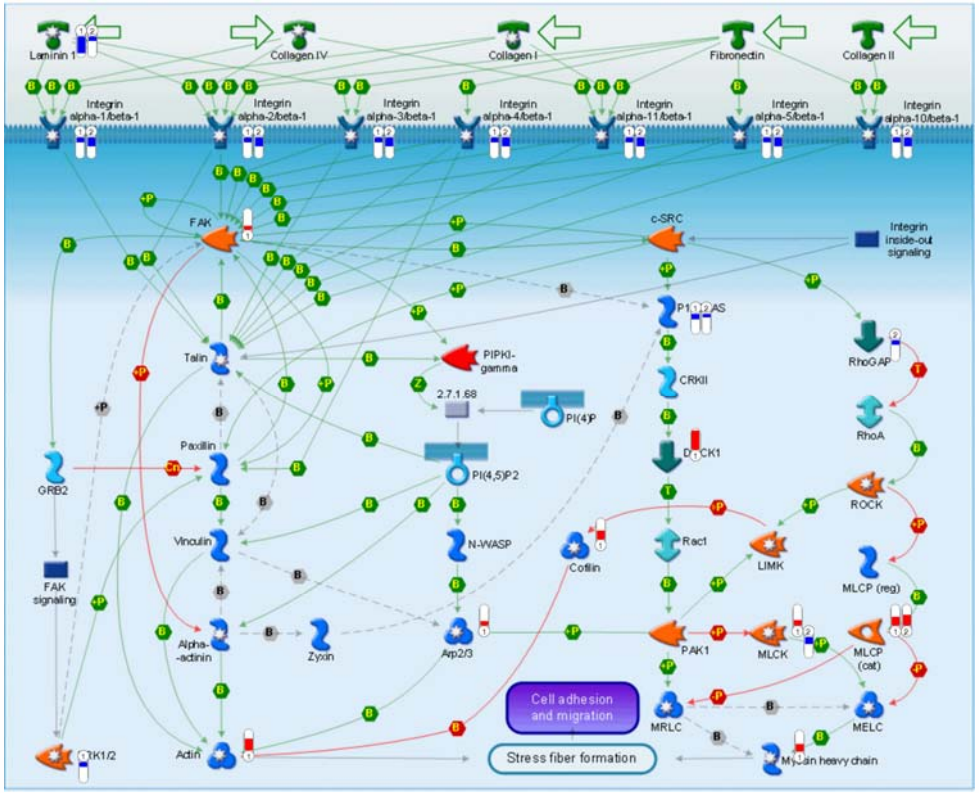
The Journal of Neuroscience, September 26, 2007 • 27(39):10530–10534

Microarray analysis in Progranulin null mice (Masugi Nishihara University of Tokyo)

- PGRN -/- mice display abnormalities in gender-specific brain function and behavior (Kayasuga et al., Behav. Brain Res. epub July 2007)
- No obvious progressive neuropathological changes up to 8 months of age in -/- mice
- Identifying progressive gene expression changes linked to PGRN haploinsufficiency (+/-), can then test if treatments that increase PGRN reverse this shift



3 month old Het mice



Mutations in *PGRN* cause tau-negative FTDP-17

- Loss of function (mostly null) mutations in *PGRN* account for **all** tau-negative FTDP-17 families
  - *PGRN* lies within 2Mb of *MAPT* (1.7Mb)
  - Clinical phenotype similar to idiopathic FTLD but language disorders are prominent
  - Frequency is 5% in unselected ADRC FTD patient series
  - Penetrance is high (90% by 70yrs) but with wide age at onset (40s-80s)
- PGRN localized to neurons and microglia in normal brain
  - PGRN IR increased in activated microglia in neurodegenerative disease
  - Localized to microglia/dystrophic neurites around amyloid plaques
  - Localized to lysosomes in activated microglia – role in phagocytosis?
  - Consistent with role modulating brain repair/inflammation (similar to chemokines, lipoxins)
- PGRN null mutations may deplete normal neurotrophic support leading to neurodegeneration and/or cause defective response to initial neuronal injury
- TDP-43 is a major component of Ubiquitin-ir inclusions found in FTLD-U
  - Progranulin haploinsufficiency leads to caspase-like cleavage of TDP-43 which may cause mislocalization and seed aggregation (Petrucelli and colleagues)
- PGRN -/- mice are viable. -/- and +/- mice show no obvious neuropathological changes to 8 months
  - Microarray analysis reveals altered gene expression patterns at 3months
  - May provide pharmacodynamic model to test compounds that increase PGRN expression

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Mayo Clinic ADRC staff

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  - Howard Feldman
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  - Charles L. White III
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  - Marsel Mesulam (Northwestern U)
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