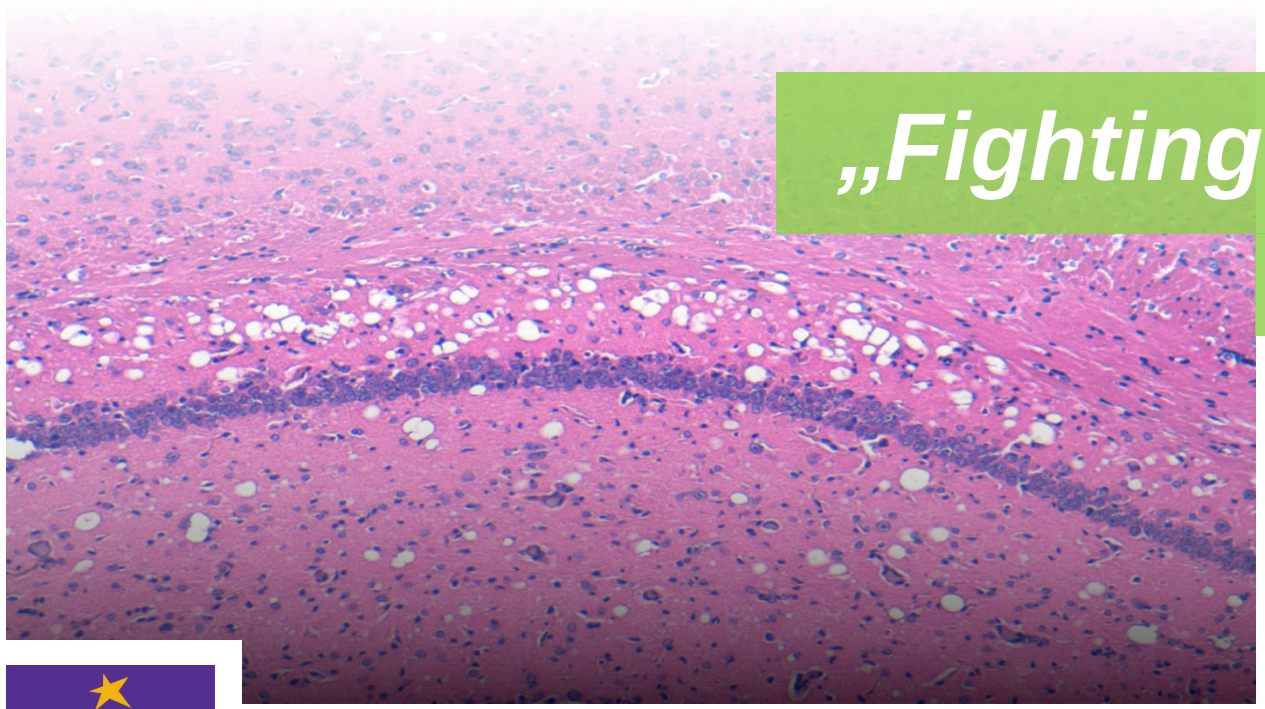
Universitätsklinikum Hamburg-Eppendorf

Investigating the potential of the neuroprotective N1 fragment of the prion protein as a new treatment against prion diseases



„Fighting prions with the prion protein“

Hermann Clemens Altmeppen

Institute of Neuropathology
Junior research group
„Proteolytic Processing of the Prion Protein“
University Medical Center HH-Eppendorf

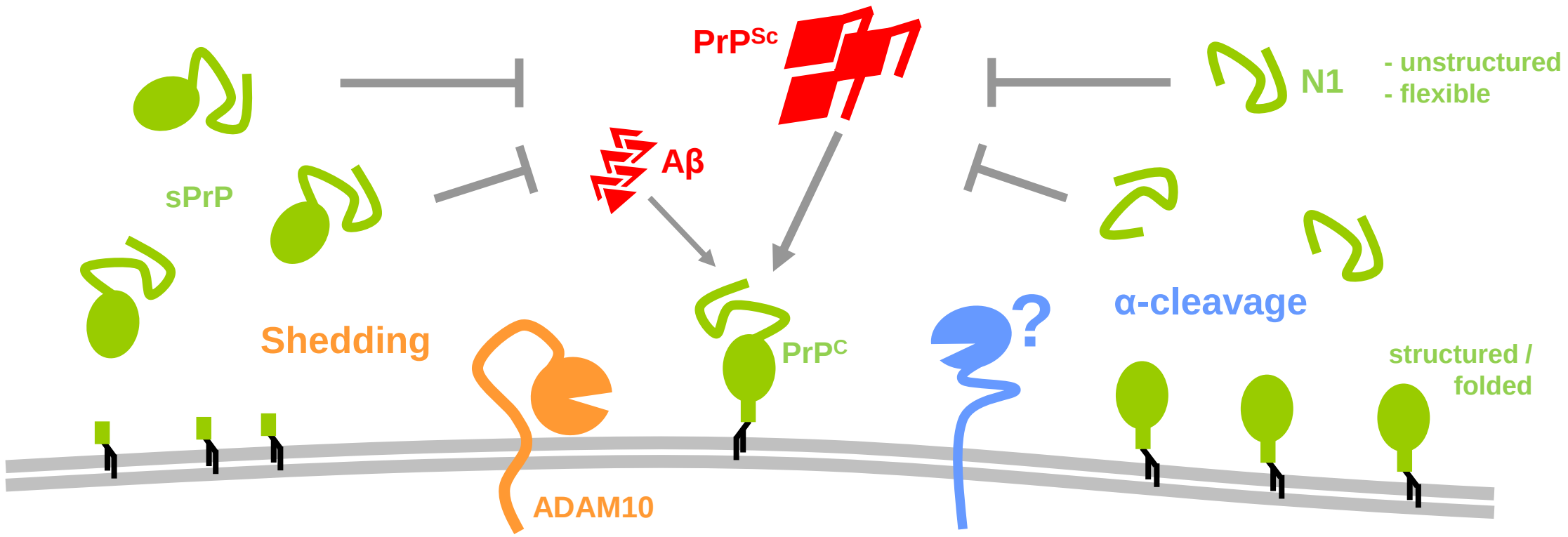
CREUTZFELDT-JAKOB DISEASE FOUNDATION, INC.

Supporting Families Affected by Prion Disease

Source: www.dfb.de

„Fighting prions with the prion protein“

The protective role of molecular cleavage events



2016 grant (work ongoing)

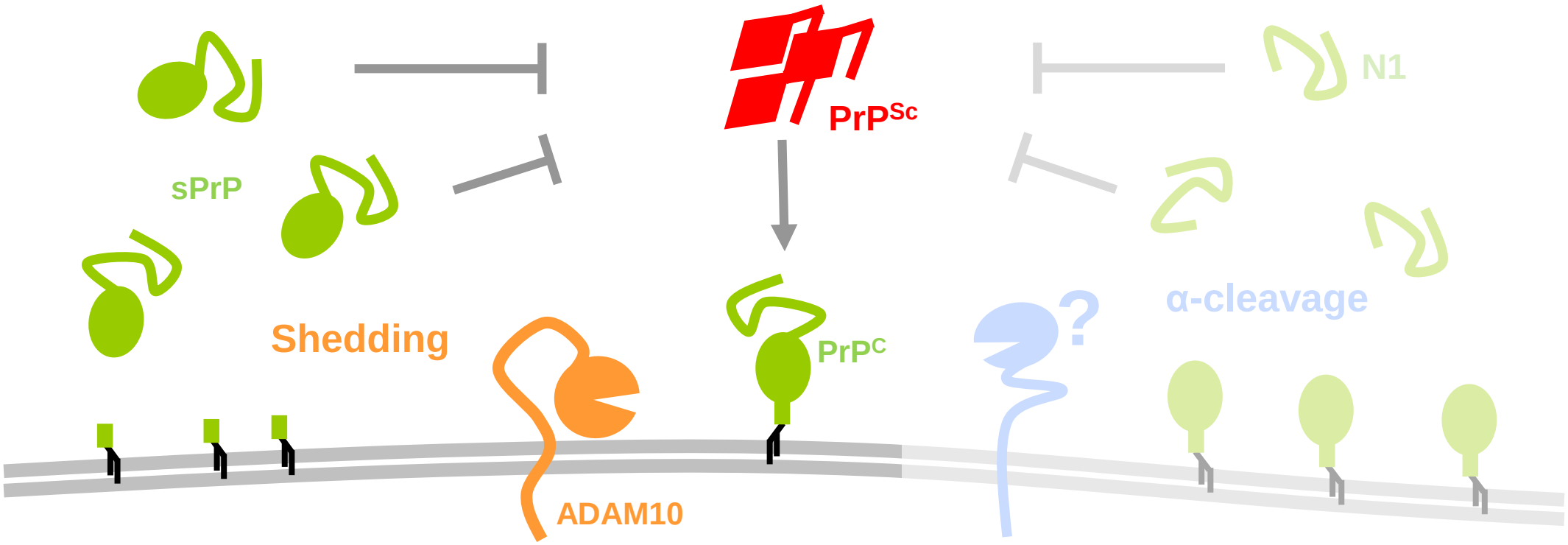
The Mary Sue Riffl Memorial Grant
The Neil W. Foster Memorial Grant
The Mary S. Friel and Mary T. Friel Memorial Grant
The CJD Foundation Memorial Grant

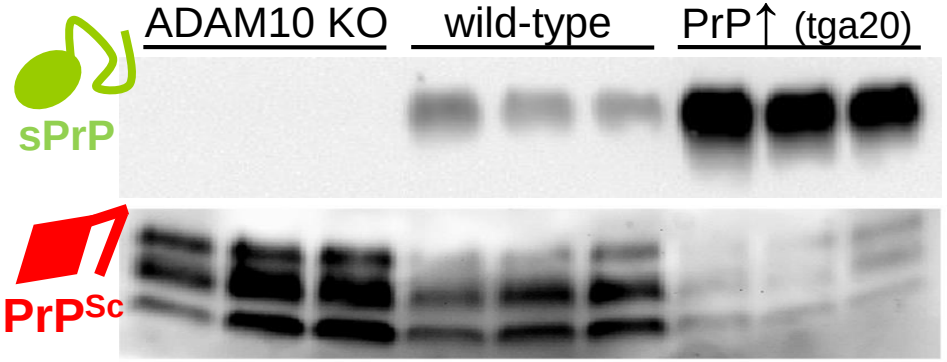
2018 grant

The Garry Buttermann IV Memorial Grant
The Davey Kock Memorial Grant
The Jeffrey A. Smith Memorial Grant
The Strides for CJD Grant

„Fighting prions with the prion protein“

The protective role of molecular cleavage events





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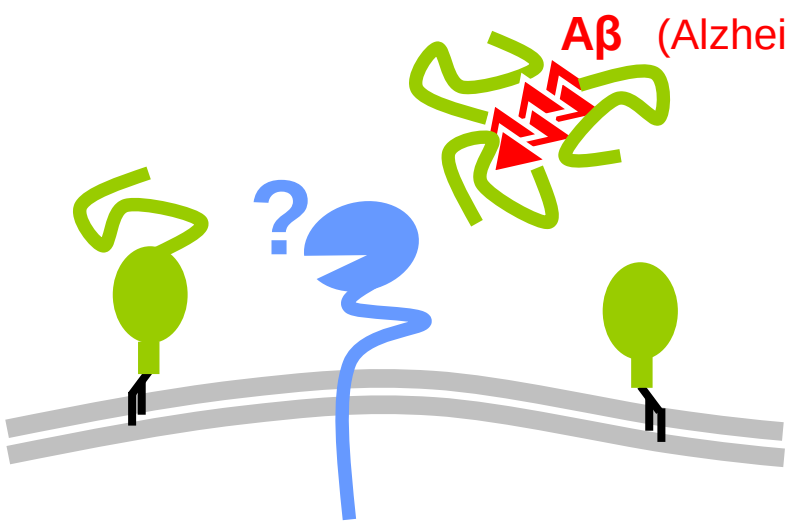
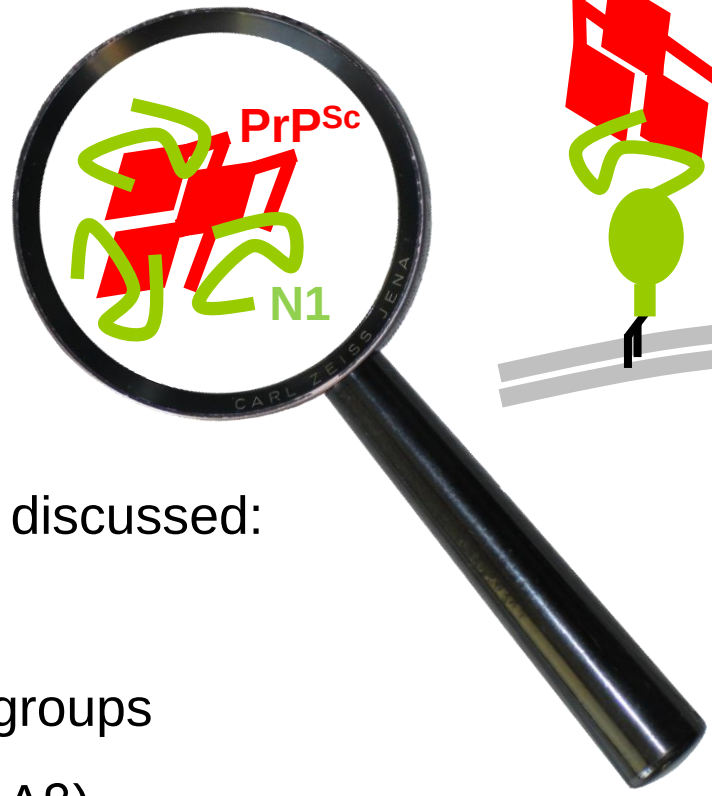
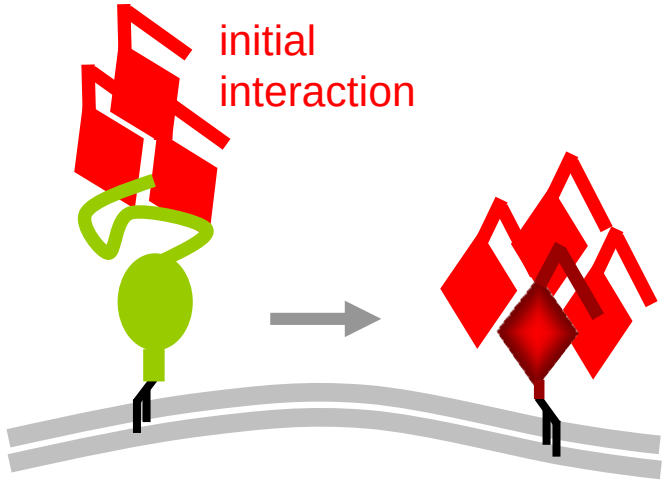
„Fighting prions with the prion protein“

The α-cleavage of PrP^C and the N1 fragment







- Responsible protease(s) unknown / controversially discussed:

  - no pharmacological target at the moment

- Protective effects of α-cleavage shown by several groups
- Blocking/neutralizing effects of N1 (mainly towards Aβ) convincingly demonstrated by several groups

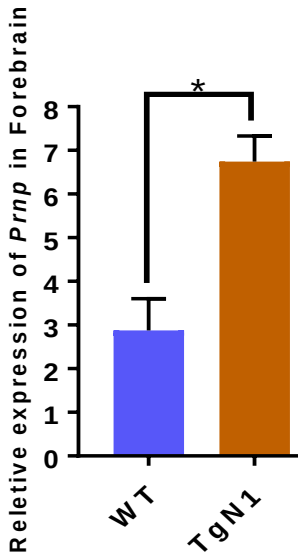
What about N1-associated protection in prion diseases?

Need for a reliable mouse model

„Fighting prions with the prion protein“

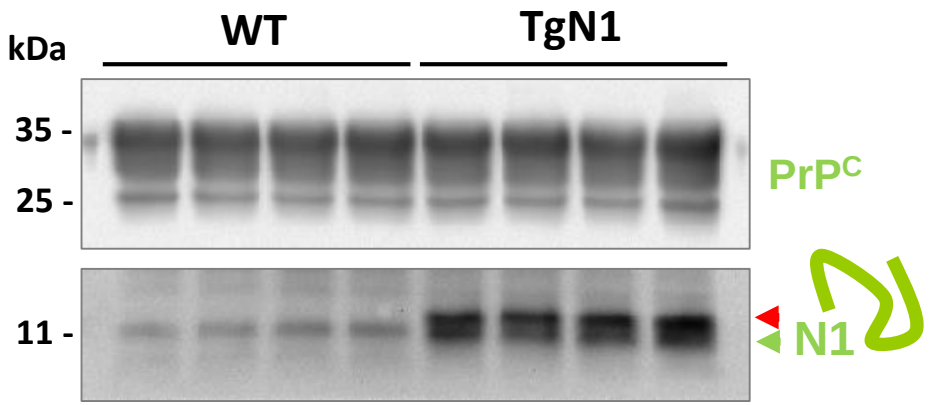
Generation of N1-overexpressing mice (TgN1)

mRNA level in forebrain:



TgN1 mice show higher mRNA transcript levels

Biochemical assessment (western blot) of mouse brain homogenates:



TgN1 mice show increased N1 amounts at the protein level

Everything so far looked promising, but…

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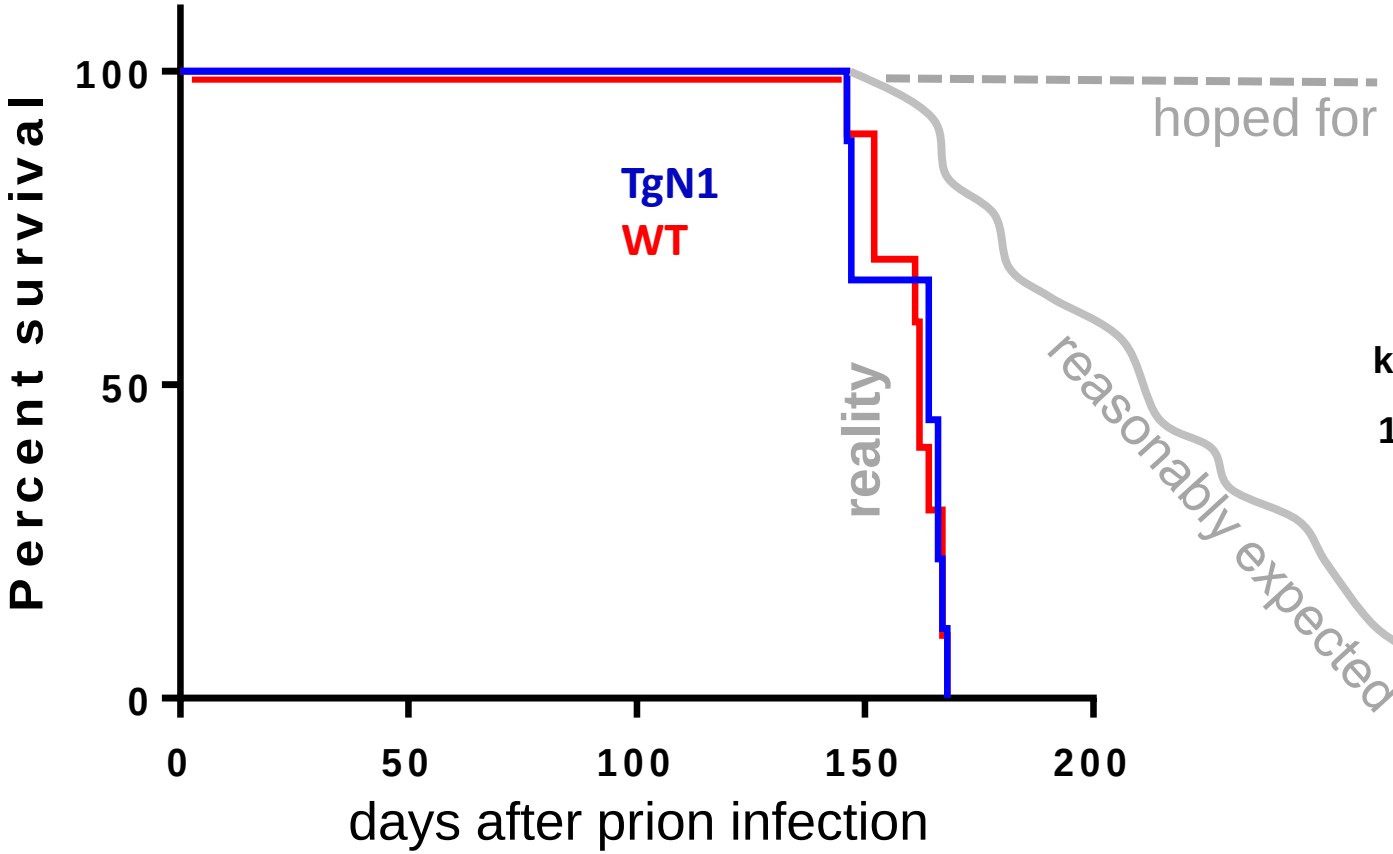
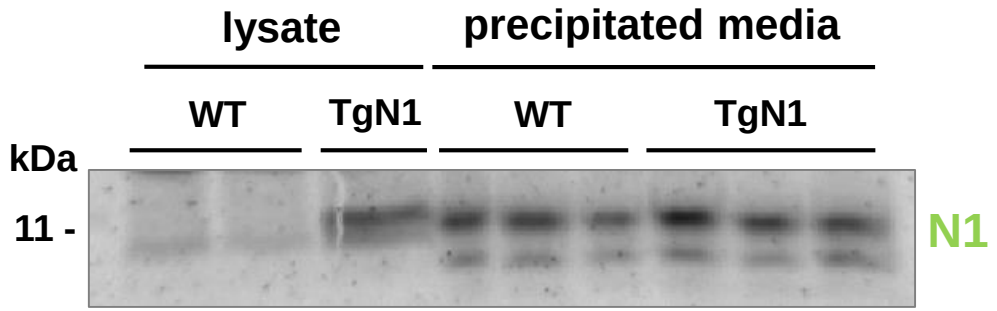
Intracerebral infection of TgN1 mice with prions



No protection observed in prion diseases

Have we been wrong with our strategy?

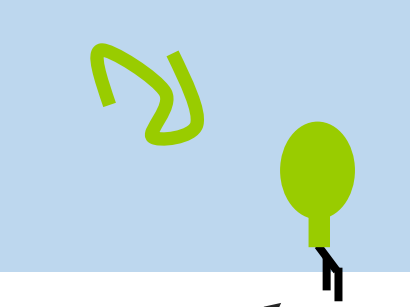
Primary neurons provide further insight:



Similar N1 secretion in TgN1 and WT mice

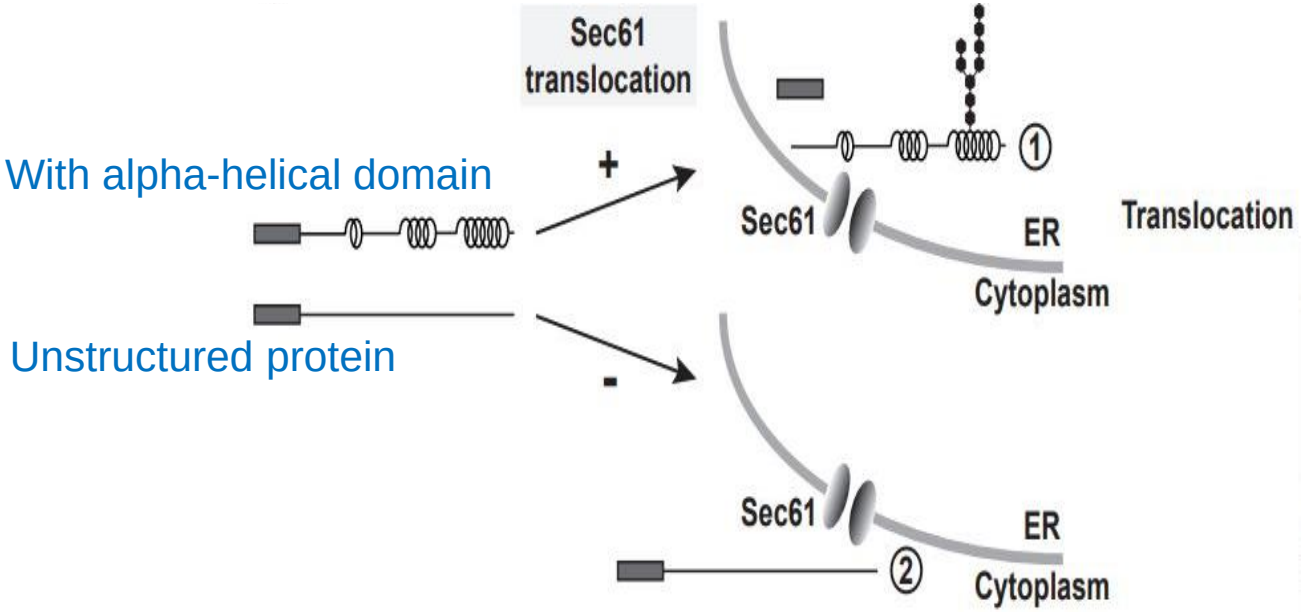
How can this be explained?

A new finding with general relevance for protein biosynthesis published in parallel to our project

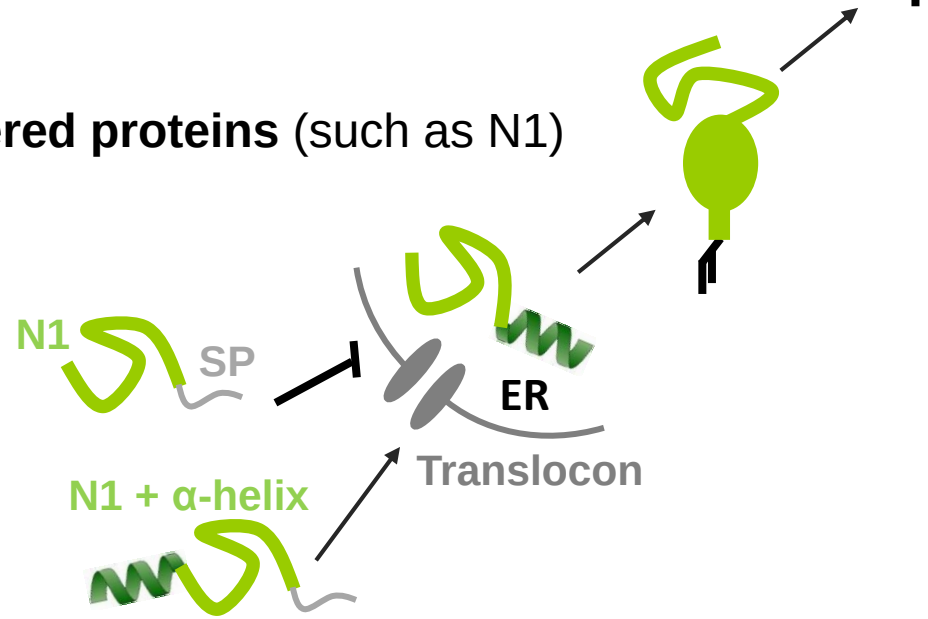


Gonsberg *et al*. (J Biol Chem, Dec. 2017)

Inefficient ER-import and secretion of **intrinsically disordered proteins** (such as N1)



modified from Gonsberg *et al*. (JBC, 2017)



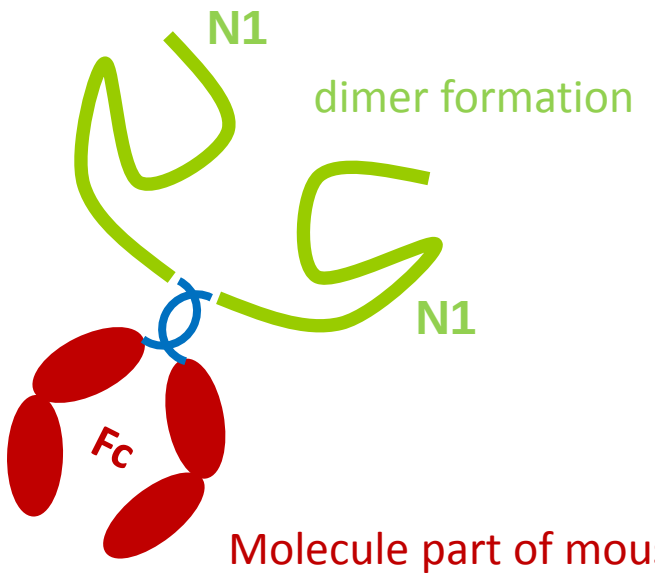
- signal peptide remains attached (double band!)
- accumulation in the cytosol / altered signaling
- first *in vivo* proof YET not what we aimed for

Can we overcome this problem?

„Keeping up the fight“

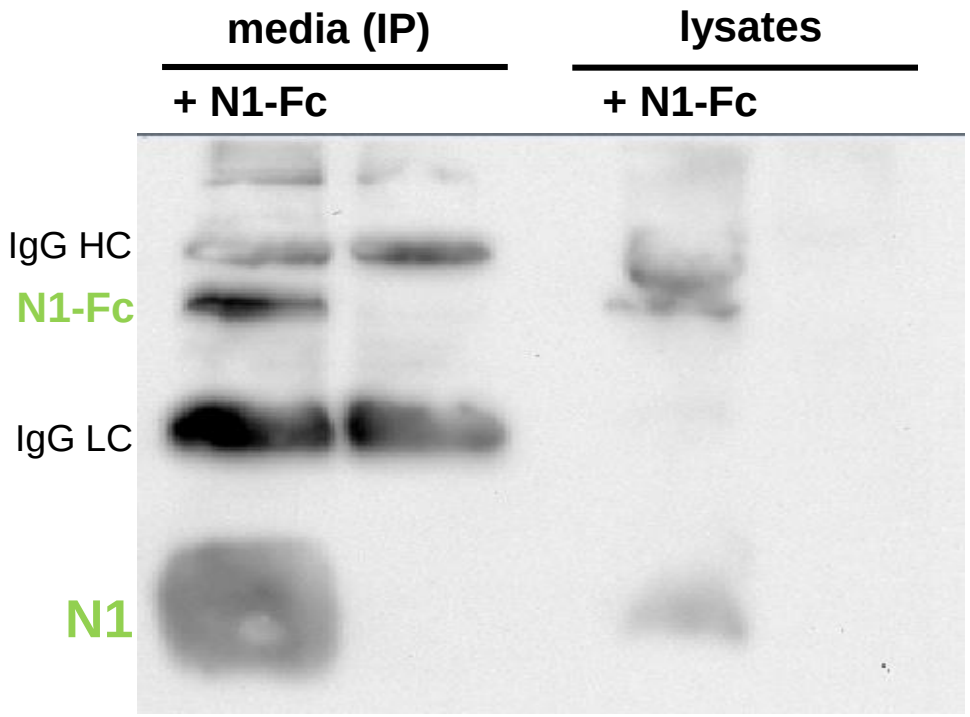
A new mouse model for increased N1 secretion

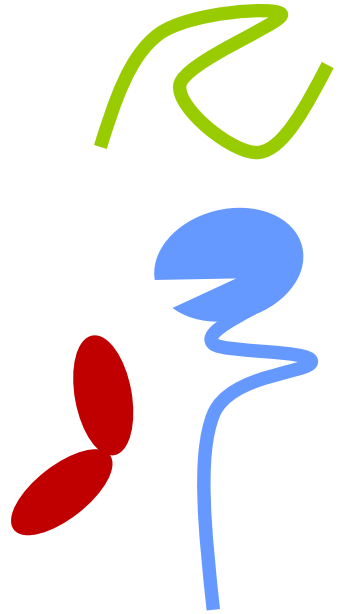
Work in progress: Generation of transgenic mice expressing the fusion protein N1-Fc



Molecule part of mouse antibodies (IgG)

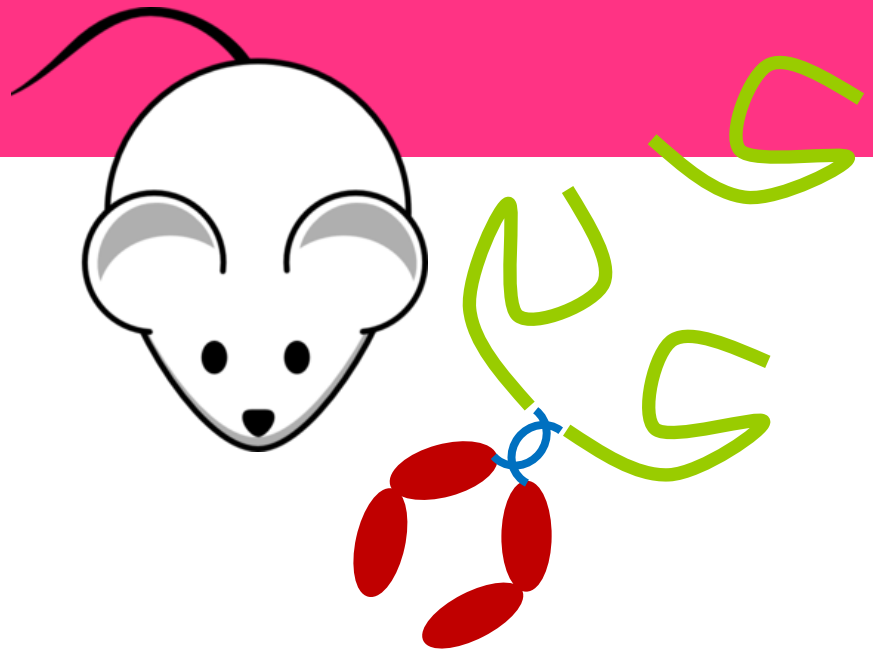
Expression in PrP^C knockout cells:





An α-cleavage-like event still occurs

Outlook



- detailed characterization of the N1-Fc mouse model
- prion inoculation study using N1-Fc mice and controls
- continue identification of the responsible protease(s)

- further assessment of the protective shedding by ADAM10 in neurodegenerative diseses
- substrate-specific stimulation of the shedding
- influence of physiological/pathological binding partners

Push forward translational aspects and novel therapeutic approaches

part of future combination therapies?

Summary

- **There is an urgent need for novel therapeutic strategies against prion diseases**
- **Under normal conditions, a fraction of PrP^C molecules is cleaved by proteases**
- **Increasing evidence suggests that at least two cleavage events are protective**
- **Mechanistic details of how the resulting prion protein fragments act protective are currently unknown and have to be investigated in detail in order to device novel treatment options (e.g. stimulation of the proteases; administration of PrP fragments)**
- **New transgenic mice overexpressing the N1 fragment of the prion protein have been/will be generated that will show whether N1 also acts protective in prion diseases**
- **Promising advances also have been made with the shedding of PrP^C** (grant 2016)

Acknowledgements

Thank you for your continued support and motivation!

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Prof. Dr. M. Glatzel

Behnam Mohammadi

Luise Linsenmeier

Dr. B. Puig

B. Szalay, Dr. G. Galliciotti

Dr. S. Krasemann

K. Hartmann (Mouse Pathology Core Unit)

all coworkers in the Institute of Neuropathology

all national and international collaboration partners

Universitätsklinikum Hamburg-Eppendorf

PROTEIN CLEAVAGE FOR REGULATION

KIEL BIOMEDICINE

WERNER OTTO STIFTUNG

STIFTUNG DES BÜRGERLICHEN RECHTS