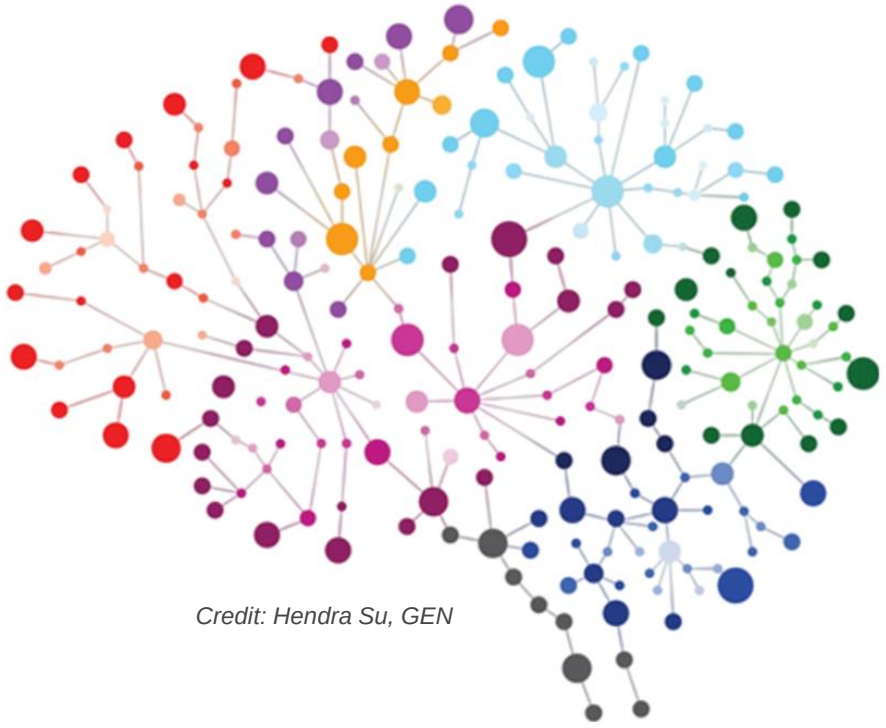
Microenvironment mapping of the PrP^{Sc} Interactome



Credit: Hendra Su, GEN

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CJD Foundation Family Conference
Washington, DC
July 19th-21st, 2024

Experimental Aims

1. To apply a novel technology, μMap, to discover proteins that interact with PrP^{Sc}
2. Interrogate “hits” using CRISPR mediated knockout and/or overexpression

Rationale: knowledge of PrP^C interactions = biological insights

Prion protein attenuates excitotoxicity by inhibiting NMDA receptors

Houman Khosravani,[1] Yunfeng Zhang,[1] Shigeki Tsutsui,[2] Shahid Hameed,[1] Christophe Altier,[1] Jawed Hamid,[1] Lina Chen,[1] Michelle Villemaire,[2] Zenobia Ali,[2] Frank R. Jirik,[2] and Gerald W. Zamponi[1]

The prion protein is an agonistic ligand of the G protein–coupled receptor Adgrg6

Alexander Küffer[1*], Asvin K. K. Lakkaraju[1*], Amit Mogha[2], Sarah C. Petersen[2], Kristina Airich[1], Cédric Doucerain[1], Rajlakshmi Marpakwar[1], Pamela Bakirci[1], Assunta Senatore[1], Arnaud Monnard[1], Carmen Schiavi[1], Mario Nuvolone[1], Bianka Grosshans[3], Simone Hornemann[1], Frederic Bassilana[3], Kelly R. Monk[2] & Adriano Aguzzi[1]

The 37-kDa/67-kDa Laminin Receptor Acts as a Receptor for Infectious Prions and Is Inhibited by Polysulfated Glycanes

Sabine Gauczynski,[1] Daphne Nikles,[1] Susanne El-Gogo,[1] Dulce Papy-Garcia,[3] Clémence Rey,[1] Susanne Alban,[2] Denis Barritault,[3,4] Corinne Ida Lasmézas,[4] and Stefan Weiss[1]

The Prion Protein Controls Polysialylation of Neural Cell Adhesion Molecule 1 during Cellular Morphogenesis

Mohadeseh Mehrabian[1,2], Dylan Brethour[1,2], Hansen Wang[1], Zhengrui Xi[1], Ekaterina Rogaeva[1,3], Gerold Schmitt-Ulms[1,2*]

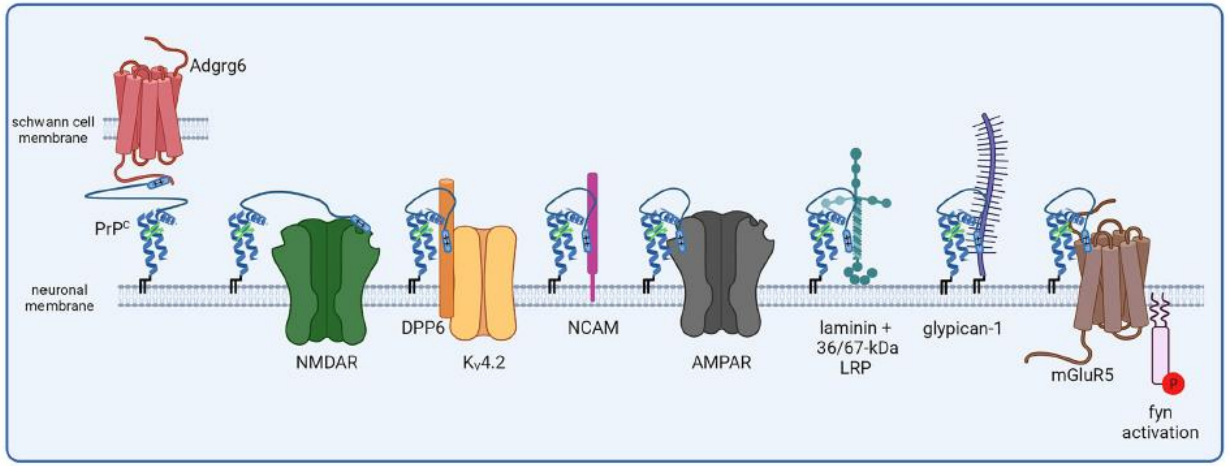
The Prion Protein Modulates A-type K^+ Currents Mediated by Kv4.2 Complexes through Dipeptidyl Aminopeptidase-like Protein 6*

Received for publication, May 24, 2013, and in revised form, November 8, 2013 Published, JBC Papers in Press, November 13, 2013, DOI 10.1074/jbc.M113.488650

Robert C. C. Mercer[‡§1,2], Li Ma[§¶1], Joel C. Watts[‖], Robert Strome[‖], Serene Wohlgemuth[‡§], Jing Yang[‡], Neil R. Cashman[**], Michael B. Coulthart[‡‡], Gerold Schmitt-Ulms[‖], Jack H. Jhamandas[§¶3], and David Westaway[‡§¶§§4]

Cellular prion protein mediates impairment of synaptic plasticity by amyloid-β oligomers

Juha Laurén[1], David A. Gimbel[1], Haakon B. Nygaard[1], John W. Gilbert[1] & Stephen M. Strittmatter[1]

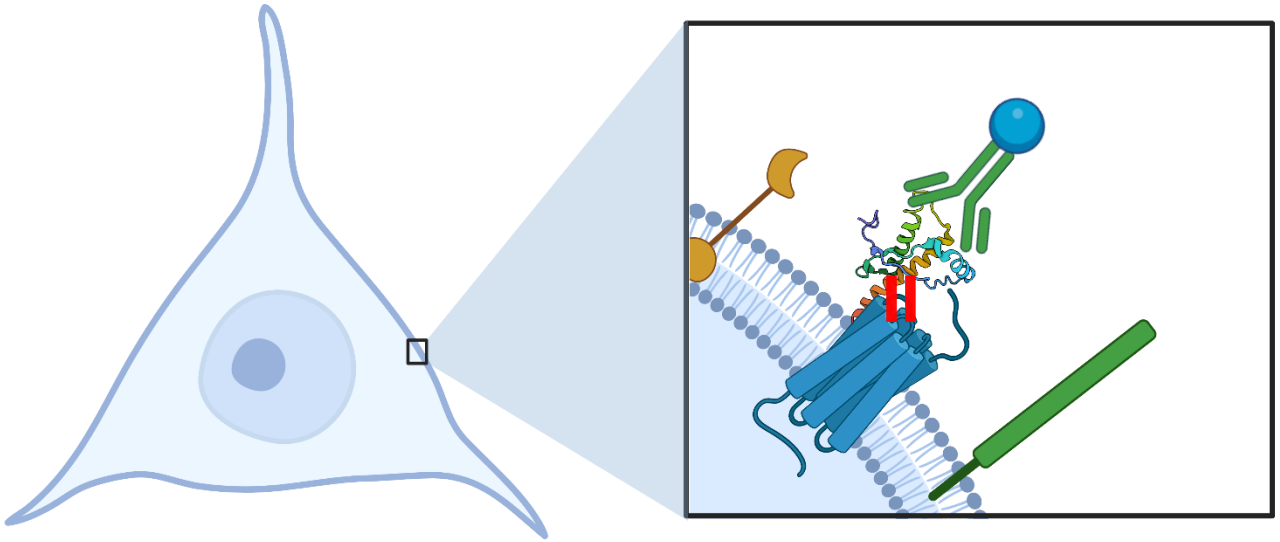


Mercer & Harris, 2023

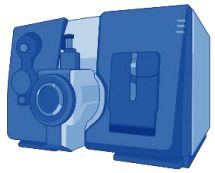
Interactomic studies of PrP^C

- formaldehyde (RED) sticks proteins together

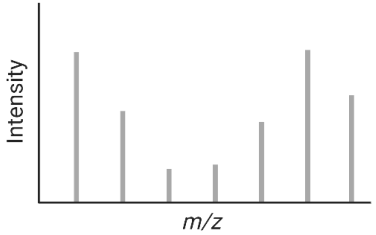
- Antibodies ( ) are used to isolate protein complexes for analysis



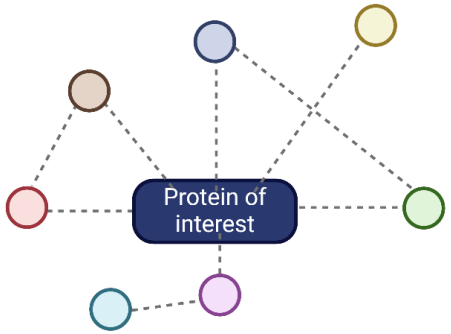
Analysis:



Mass spectrometry



Protein “fingerprint”



Interaction network

ARTICLES

nature biotechnology

Time-controlled transcardiac perfusion cross-linking for the study of protein interactions in complex tissues

Gerold Schmitt-Ulms[1,2,8], Kirk Hansen[3], Jialing Liu[4], Cynthia Cowdrey[5], Jian Yang[5], Stephen J DeArmond[1,5], Fred E Cohen[1,6,7], Stanley B Prusiner[1,2,7] & Michael A Baldwin[1,2,3]

OPEN ACCESS Freely available online

PLoS PATHOGENS

Interactome Analyses Identify Ties of PrP^C and Its Mammalian Paralogs to Oligomannosidic N-Glycans and Endoplasmic Reticulum-Derived Chaperones

Joel C. Watts[1,2☯a], Hairu Huo[1☯], Yu Bai[1☯b], Sepehr Ehsani[1,2☯], Amy Hye Won[1,2], Tujin Shi[1], Nathalie Daude[3], Agnes Lau[3], Rebecca Young[4], Lei Xu[4], George A. Carlson[4], David Williams[5], David Westaway[3], Gerold Schmitt-Ulms[1,2*]

SCIENTIFIC REPORTS

OPEN

The prion protein is embedded in a molecular environment that modulates transforming growth factor β and integrin signaling

Farinaz Ghodrati[1,2], Mohadeseh Mehrabian[1,2], Declan Williams[1], Ondrej Halgas[3], Matthew E. C. Bourkas[1,3], Joel C. Watts[1,3], Emil F. Pai[3,4] & Gerold Schmitt-Ulms[1,2]

RESEARCH ARTICLE

The cellular prion protein interacts with and promotes the activity of Na,K-ATPases

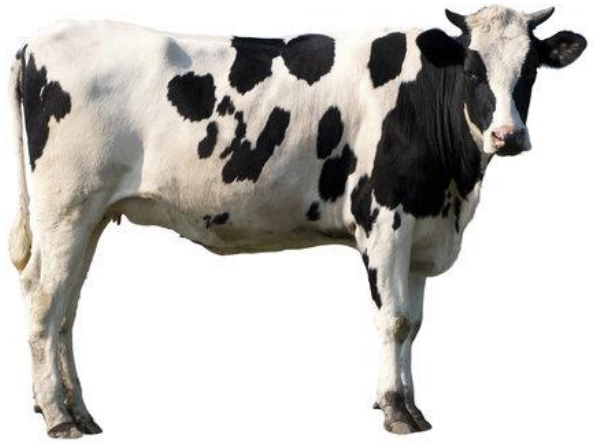
Declan Williams[1☯], Mohadeseh Mehrabian[1,2☯], Hamza Arshad[1,3☯], Shehab Eid[1,2], Christopher Sackmann[1], Wenda Zhao[1,2], Xinzhu Wang[1,2], Farinaz Ghodrati[1,2], Claire E. Verkuyl[1], Joel C. Watts[1,3], Gerold Schmitt-Ulms[1,2*]

Expression of PrP is required for the manifestation of prion disease…

Cell, Vol. 73, 1339–1347, July 2, 1993, Copyright © 1993 by Cell Press

Mice Devoid of PrP Are Resistant to Scrapie

H. Büeler,* A. Aguzzi,† A. Sailer,* R.-A. Greiner,‡
P. Autenried,‡ M. Aguet,* and C. Weissmann*



LETTERS

nature biotechnology

Production of cattle lacking prion protein

Jürgen A Richt[1,6], Poothappillai Kasinathan[2], Amir N Hamir[1], Joaquin Castilla[3], Thillai Sathiyaseelan[2], Francisco Vargas[1], Janaki Sathiyaseelan[2], Hua Wu[2], Hiroaki Matsushita[2], Julie Koster[2], Shinichiro Kato[4,5], Isao Ishida[4], Claudio Soto[3], James M Robl[2] & Yoshimi Kuroiwa[4–6]

Salvesen et al. Vet Res (2020) 51:1
https://doi.org/10.1186/s13567-019-0731-2

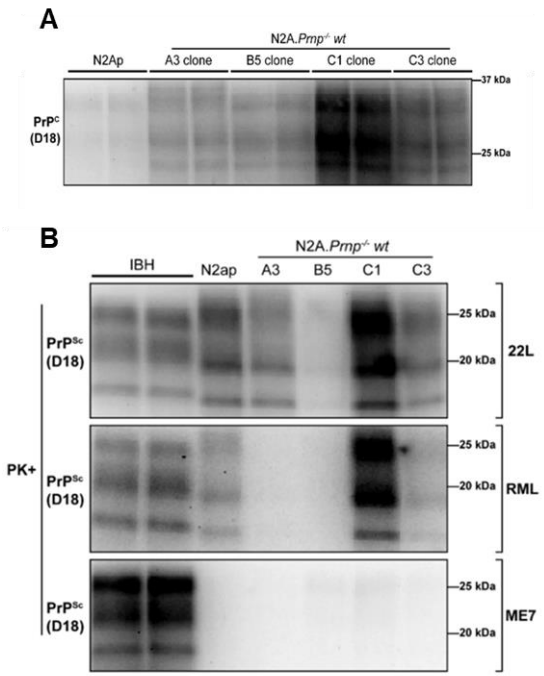
VETERINARY RESEARCH

RESEARCH ARTICLE Open Access

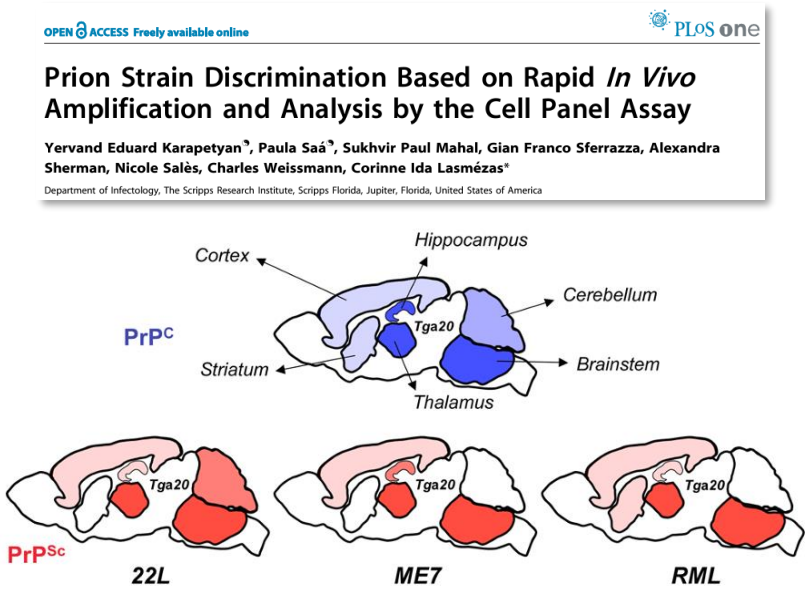
Goats naturally devoid of PrP^C are resistant to scrapie

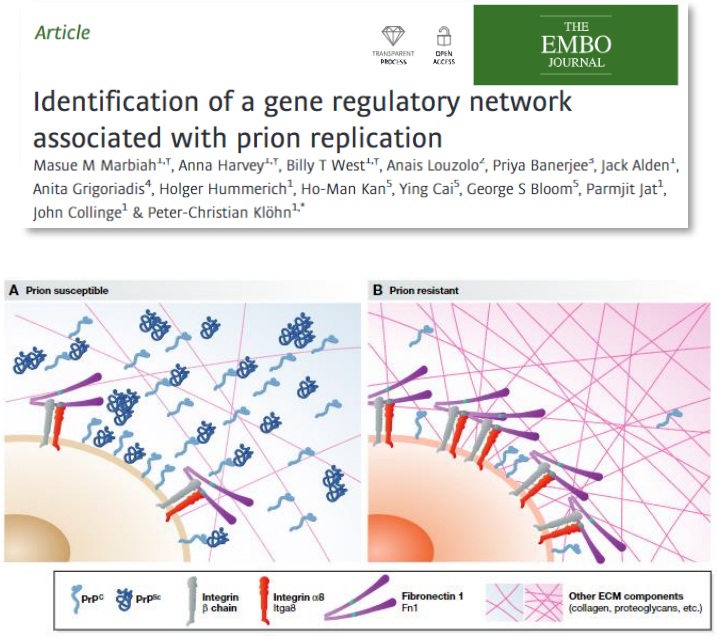
Øyvind Salvesen[1], Arild Espenes[2], Malin R. Reiten[2,3], Tram T. Vuong[3], Giulia Malachin[2], Linh Tran[3], Olivier Andréoletti[4], Ingrid Olsaker[2], Sylvie L. Benestad[3], Michael A. Tranulis[2] and Cecilie Ersdal[1*]

Expression of PrP is required for the manifestation of prion disease… but it is not sufficient.



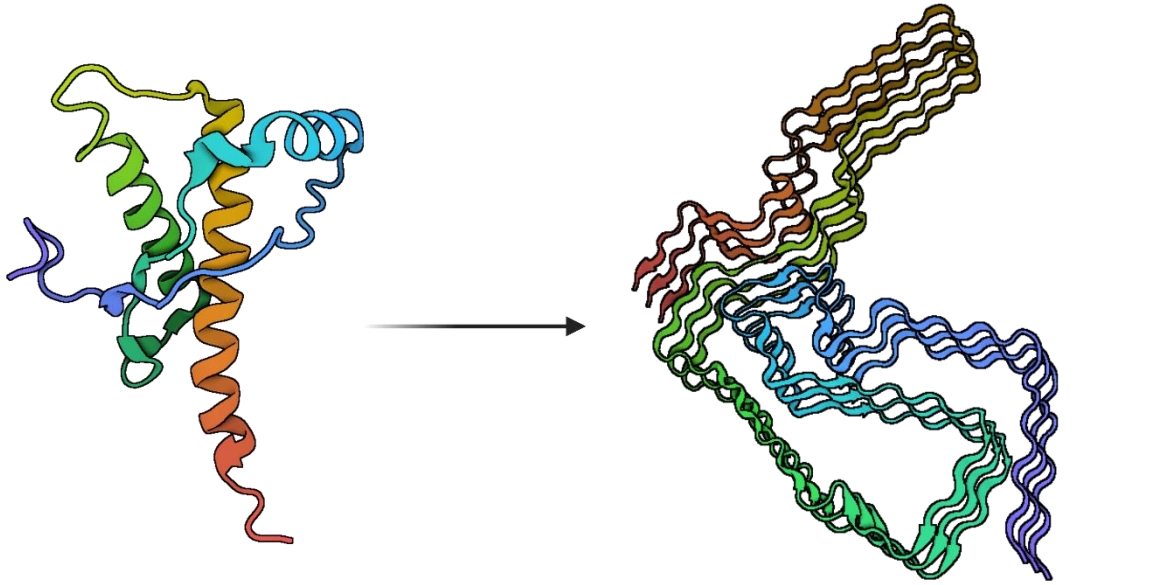
Jean R.P. Gatdula





Imberdis & Harris, 2014

The problem with PrP^{Sc}



PrP^{C}

Riek *et al.*, 1996

PrP^{Sc}

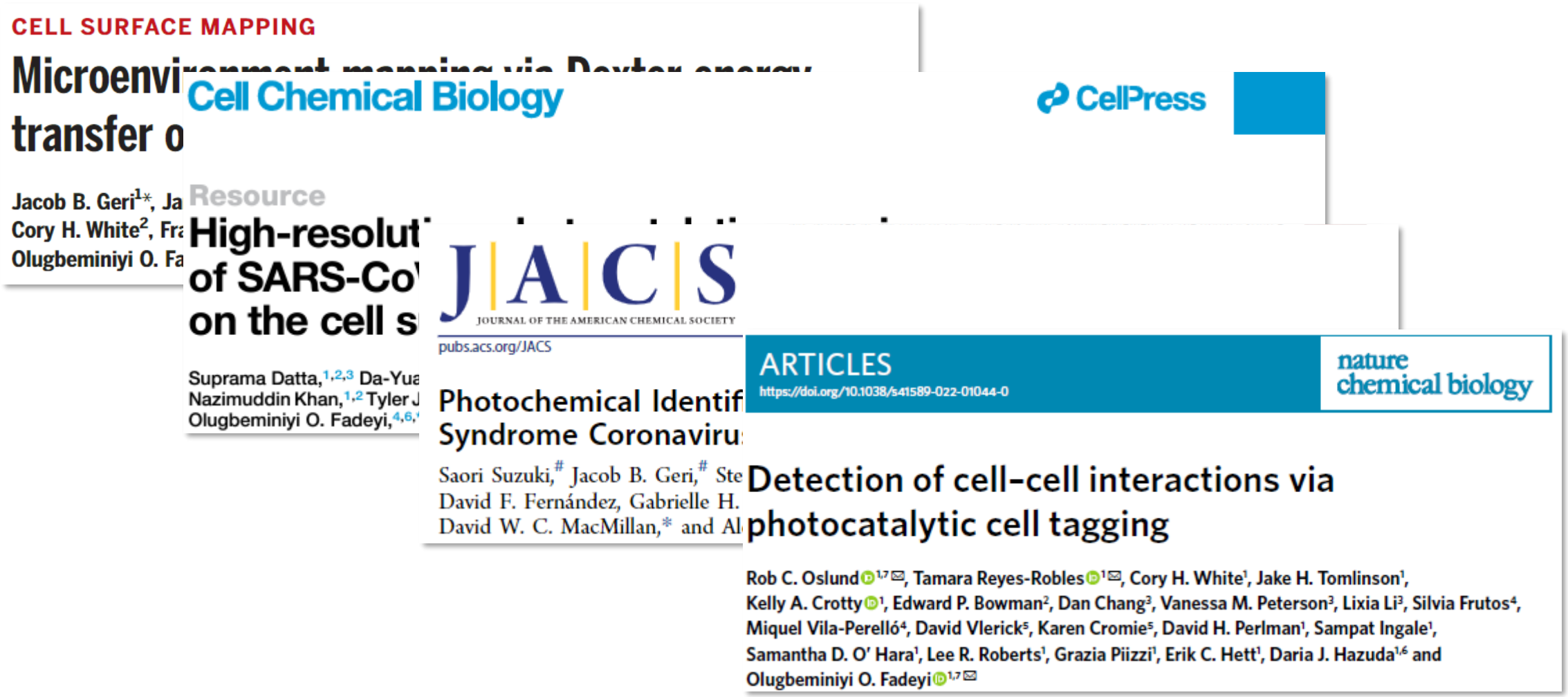
Kraus *et al.*, 2021

PrP^{C} and PrP^{Sc} are *very* different structurally….

But they are the same protein.

This makes traditional techniques unsuitable for looking at the PrP^{Sc} interactome

Novel proximity labeling technique: µMapping



µMapping: PrP^C

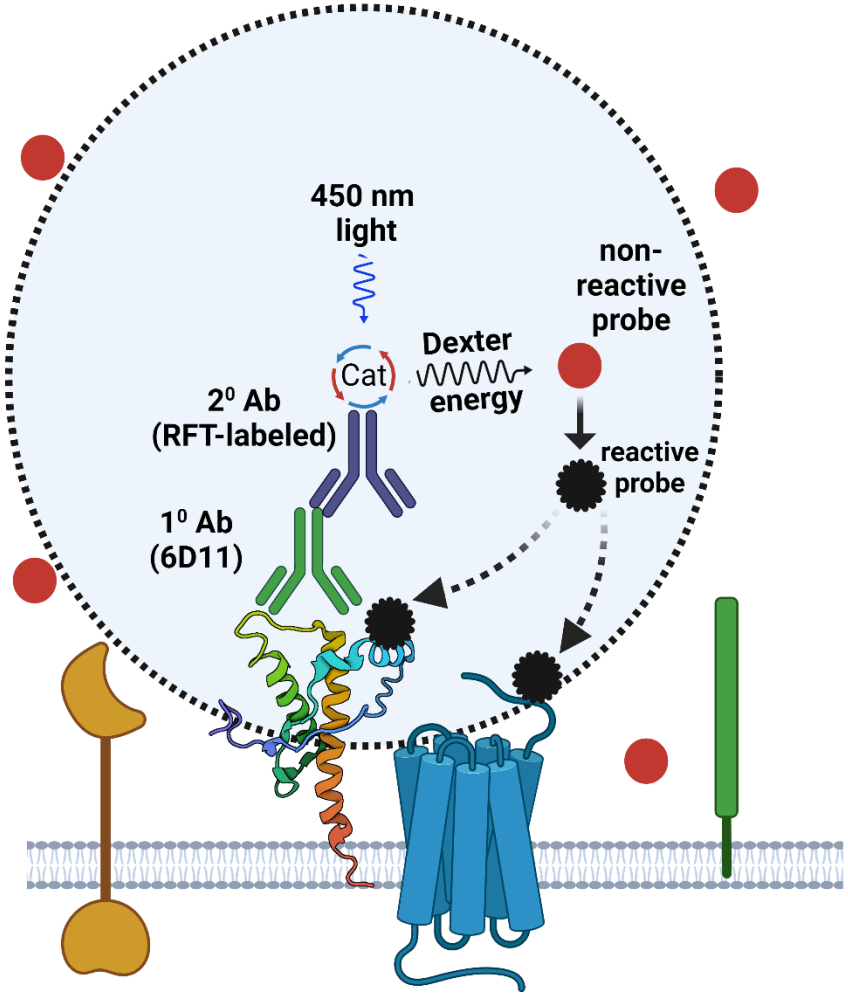
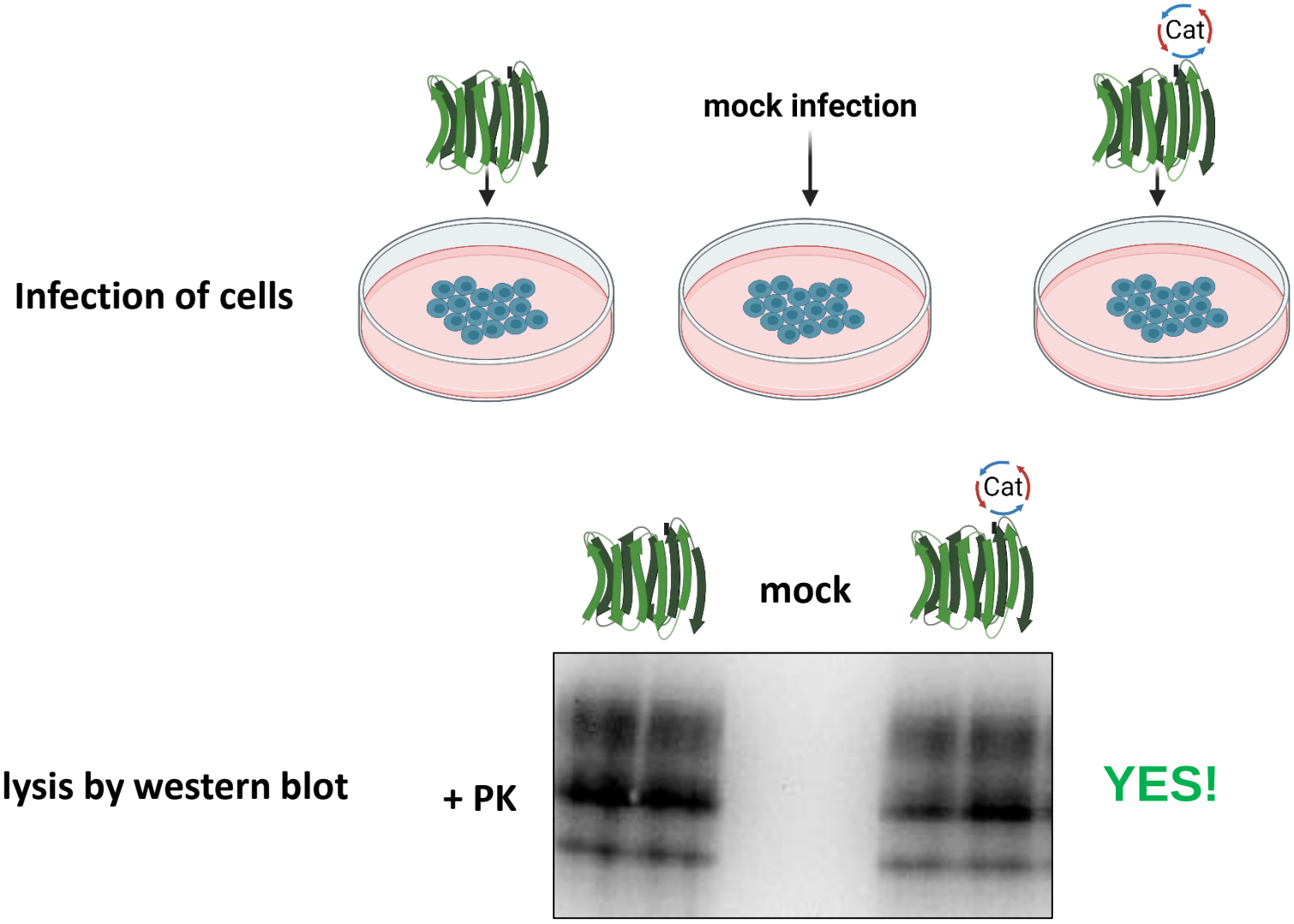


Table 1: PrP^C interacting proteins in N2a cells identified by µMapping	
Gene	**Protein**
Prnp*	**PrP**
Gpc1*	**Glypican 1**
Pcdh1	Protocadherin 1
Cntfr	Ciliary neurotrophic factor receptor
Cadm1	Cell adhesion molecule 1
Marcksl1	MARCKS-like protein 1
Pcdhgb1	Protocadherin Gamma Subfamily B, 1
Slc39a10*	**Solute Carrier Family 39 Member 10**
Igf1r	Insulinlike growth factor1 receptor
L1cam	L1 Cell Adhesion Molecule
Ncam1*	**Neural Cell Adhesion Molecule 1**
Nes	Nestin
Gprin1	G protein regulated inducer of neurite outgrowth 1
Dbn1	Drebrin 1
Epn1	Epsin 2
Ldlr	low-density lipoprotein receptor
Dgl3	Discs Large MAGUK Scaffold Protein 3
Lrch2	Leu Rich Repeats & Calponin Homo Dom Cont 2

* previously reported PrP interacting protein

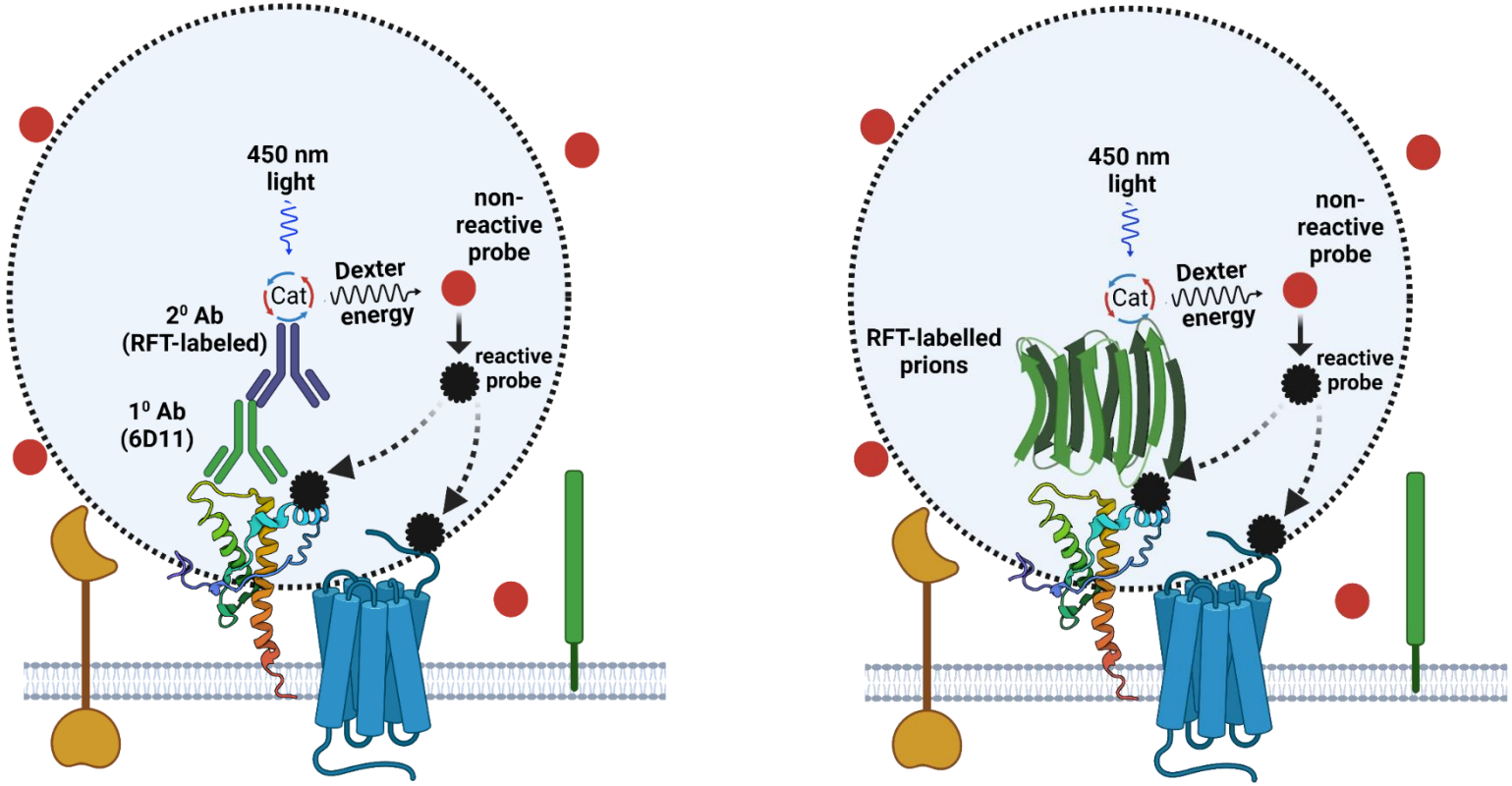
Ladan Amin

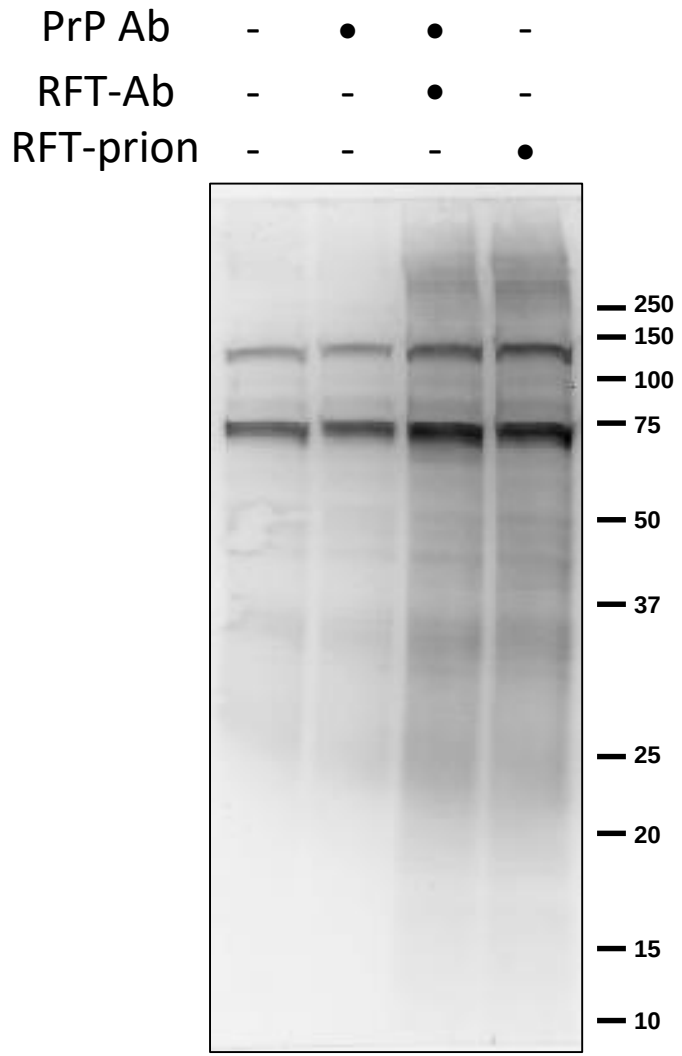
Are catalyst labeled prions infectious?



Comparing labeling efficiencies

Antibody based **vs** **prion based**

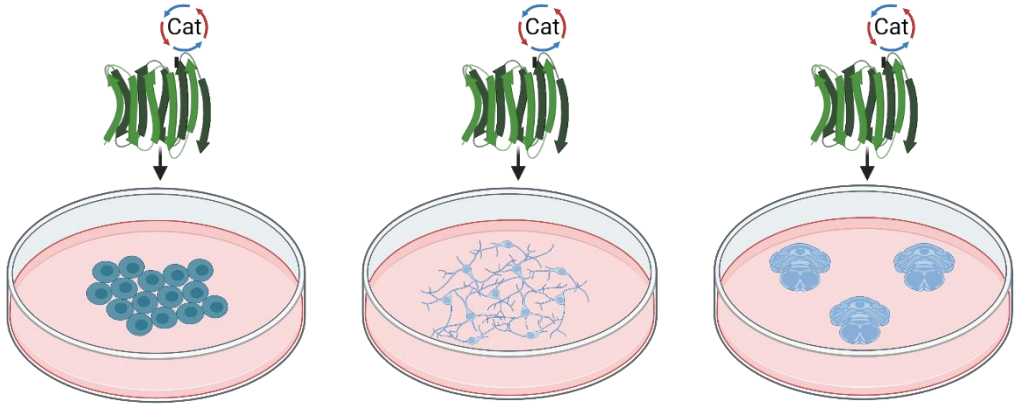




biotin immunoblot

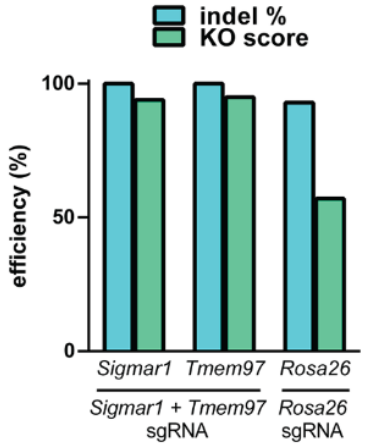
Current experimentation

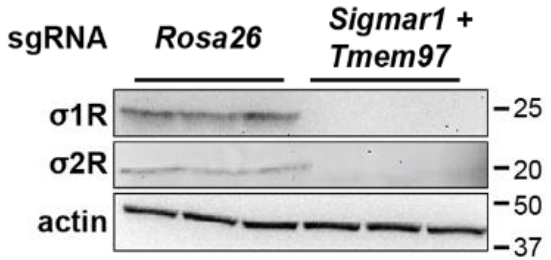
µMapping:



N2a & CAD5 cells **Cultured neurons** **Brain slices**

CRISPR knockout:





Mercer *et al.*, 2024

Acknowledgements

The Patrice Haggerty Memorial Grant
contributed by William Haggerty

The Laura McElmurray Memorial Grant
contributed by Mike McElmurray

The Lynda Morris Memorial Grant
contributed by David Morris and Family

The Nic Ziccardi Memorial Grant
contributed by Kandi Ziccardi

The Joanne (Jody) Atchison Memorial Research Grant
contributed by Dick Atchison and Family

The Robert Dodd Memorial Grant
contributed by Kathleen Dodd and Family

The Sherry Maxwell Fabian Memorial Grant
contributed by Tom Fabian

The Chuck Fear Memorial Research Grant
contributed by Pamela Fear and Family

The Strides for CJD Grant
contributed by the Families of the CJD Foundation

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contributed by the Families of the CJD Foundation

National Institute of Neurological Disorders and Stroke

U.S. DEPT OF DEFENSE

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