Selective Vulnerability of BEX1+ Excitatory Neurons Across Neocortical and Hippocampal Regions in Typical and Atypical Alzheimer's Disease

Liara Rizzi[1], Felipe L. Pereira[1,2], Caroline Lew[3], Song H. Li[3], Jonathan Chen[1,2], Kira Dewi[1], Alexander V. Soloviev[3], Vitor Paes[4], Sara D. Brooks[1], Salvatore Spina[3], Jessica E. Rexach[5,6], Kathy L. Newell[7], Renata E. P. Leite[4,8], William W. Seeley[3], Claudia K. Suemoto[9], Bernardino Ghetti[7], Melissa E. Murray[1], Lea T. Grinberg[1,4,8]

1 Department of Neuroscience, Mayo Clinic, Jacksonville, Florida, USA. 2 Department of Quantitative Health Sciences, Mayo Clinic, Jacksonville, Florida, USA. 3 Fein Memory and Aging Center, Weill Institute for Neurosciences, University of California San Francisco, San Francisco, USA. 4 Physiopathology in Aging Laboratory (LIM-22), Department of Internal Medicine, University of Sao Paulo Medical School, Sao Paulo, Brazil. 5 Program in Neurogenetics, Department of Neurology, David Geffen School of Medicine, University of California Los Angeles, Los Angeles, USA. 6 Department of Human Genetics, David Geffen School of Medicine, University of California Los Angeles, Los Angeles, USA. 7 Department of Pathology and Laboratory Medicine, Indiana University School of Medicine, Indianapolis, USA. 8 Department of Pathology, University of Sao Paulo Medical School, Sao Paulo, Brazil. 9 Division of Geriatrics, Department of Internal Medicine, University of Sao Paulo Medical School, Sao Paulo, Brazil.

BACKGROUND

- Alzheimer's disease (AD) is characterized by selective vulnerability of specific neurons.
- Prior snRNA-seq (>1.61M nuclei) revealed shared transcriptional changes in excitatory neurons.
- BEX1-positive excitatory neuron subpopulation (NRGN+BEX1) as consistently vulnerable in both neocortical (superior temporal gyrus, STG) and hippocampal (cornu ammonis sector 1, CA1) regions.

OBJECTIVE

To validate the vulnerability of the BEX1-positive excitatory subpopulation across CA1 and STG in amnestic AD, logopenic variant primary progressive aphasia (lvPPA), and posterior cortical atrophy (PCA) through quantitative neuropathological analysis.

METHODS

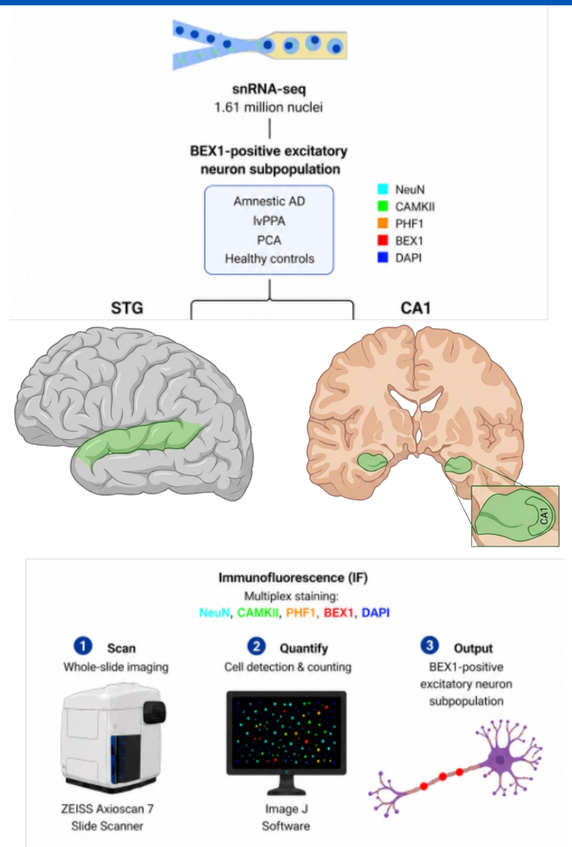
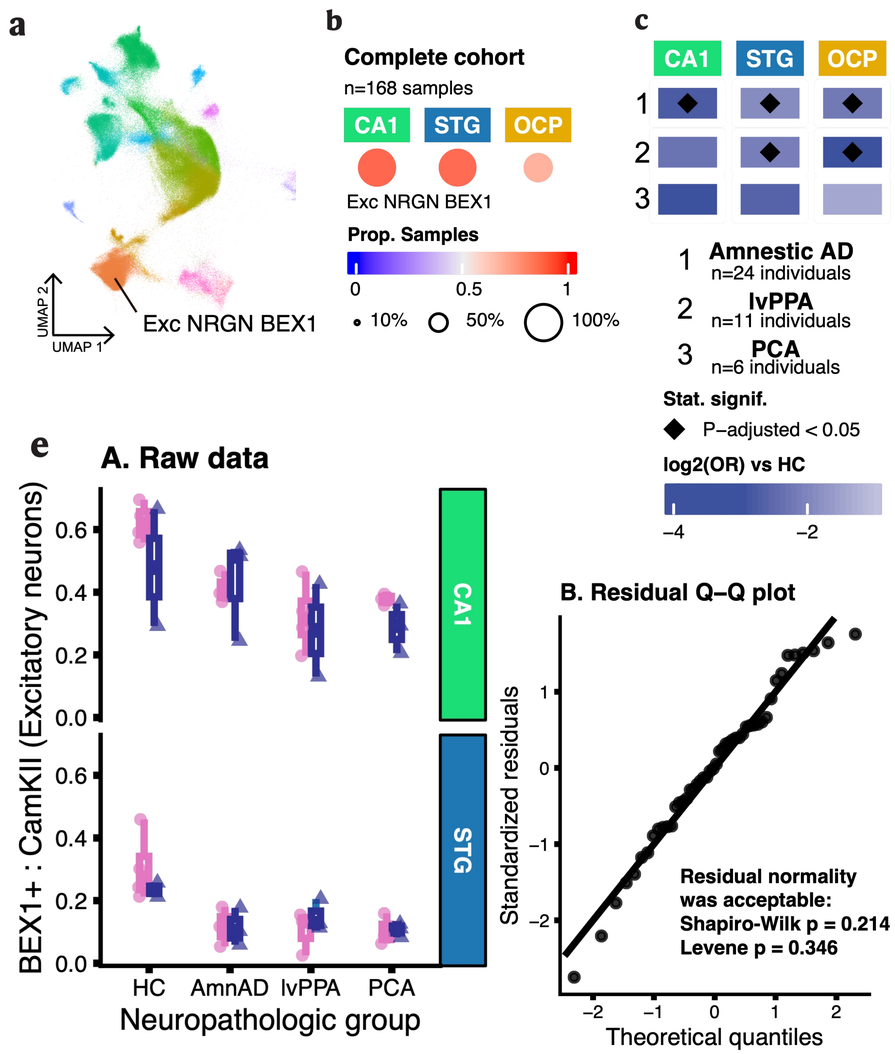


Figure 1. Quantitative neuropathological analysis of STG and CA1 excitatory neurons: FFPE postmortem CA1 and STG tissues from amnestic AD, lvPPA, PCA, and healthy controls (n = 6/group) obtained from the UCSF Neurodegenerative Disease Brain Bank and the Brazilian Biobank for Aging Studies at the University of São Paulo were analyzed. Six-micrometer sections underwent multiplex immunofluorescence for NeuN, CAMKII, PHF1, and BEX1. Whole-slide images were acquired using a ZEISS Axioscan 7 and quantified with ImageJ, with 700–1,000 neurons counted per case and region. (Figure created with BioRender.)

©2026 Mayo Foundation for Medical Education and Research

RESULTS



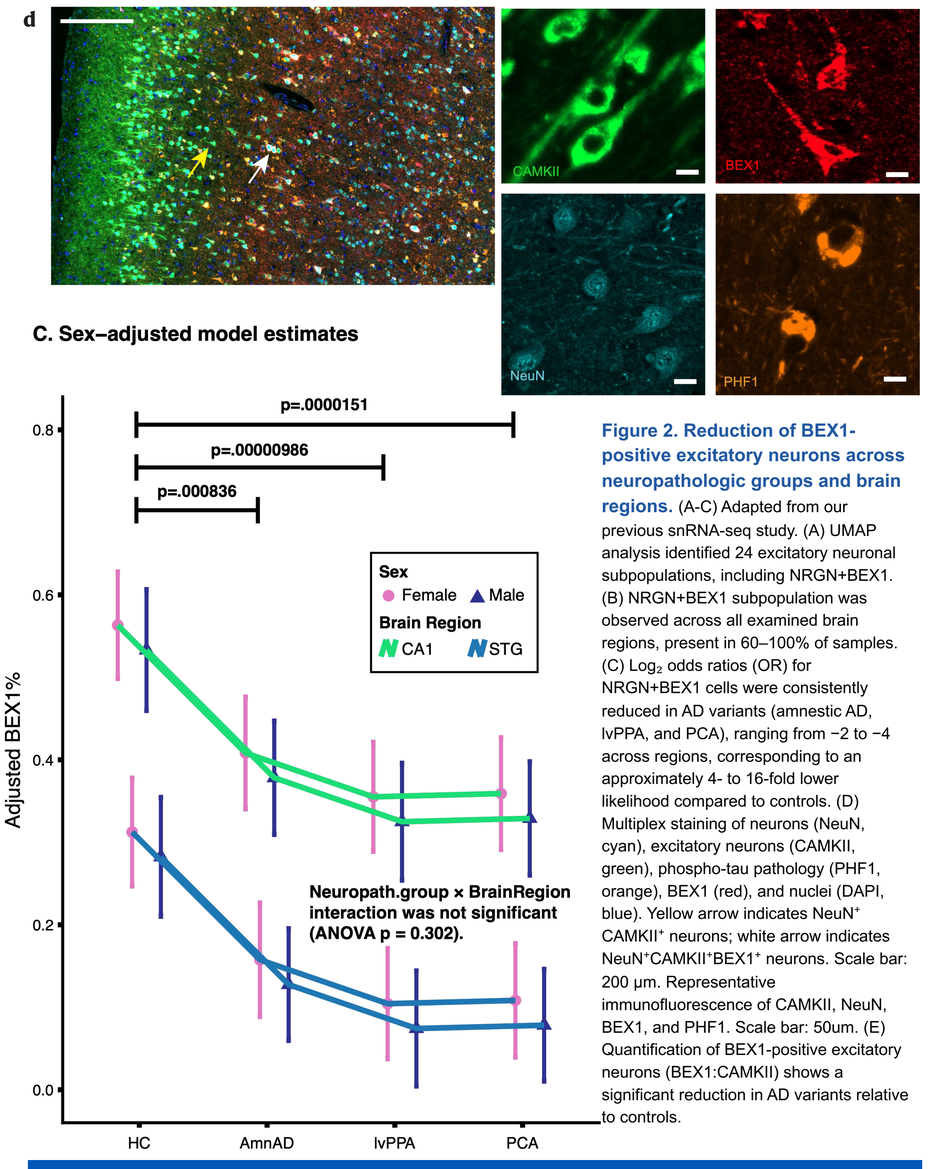


Figure 2. Reduction of BEX1-positive excitatory neurons across neuropathologic groups and brain regions. (A-C) Adapted from our previous snRNA-seq study. (A) UMAP analysis identified 24 excitatory neuronal subpopulations, including NRGN+BEX1. (B) NRGN+BEX1 subpopulation was observed across all examined brain regions, present in 60–100% of samples. (C) Log_2 odds ratios (OR) for NRGN+BEX1 cells were consistently reduced in AD variants (amnestic AD, lvPPA, and PCA), ranging from −2 to −4 across regions, corresponding to an approximately 4- to 16-fold lower likelihood compared to controls. (D) Multiplex staining of neurons (NeuN, cyan), excitatory neurons (CAMKII, green), phospho-tau pathology (PHF1, orange), BEX1 (red), and nuclei (DAPI, blue). Yellow arrow indicates $NeuN^+$ $CAMKII^+$ neurons; white arrow indicates $NeuN^+CAMKII^+BEX1^+$ neurons. Scale bar: 200 µm. Representative immunofluorescence of CAMKII, NeuN, BEX1, and PHF1. Scale bar: 50um. (E) Quantification of BEX1-positive excitatory neurons (BEX1:CAMKII) shows a significant reduction in AD variants relative to controls.

Table 1. Demographic characteristics across neuropathologic groups (STG and CA1 cohorts).

Variable	HC N = 12	Amnestic N = 12	lvPPA N = 12	PCA N = 12	P-value[1]
Age of onset, median [IQR]		76.0 [56.0, 77.0]	56.0 [53.0, 65.0]	56.5 [51.0, 61.0]	0.013
Age at death, median [IQR]	66.0 [64.0, 77.5]	87.5 [68.0, 93.0]	65.5 [62.5, 74.5]	68.0 [61.0, 71.0]	0.009
Sex, n (%)					0.818
F	8 (66.7%)	6 (50.0%)	7 (58.3%)	6 (50.0%)	
M	4 (33.3%)	6 (50.0%)	5 (41.7%)	6 (50.0%)	

[1] P-values from Kruskal-Wallis tests for AOO and AAD, and chi-squared test for sex across neuropathologic groups.

Table 2. Proportion of BEX1-positive excitatory neurons across neuropathologic groups in STG and CA1.

Neuropathologic group	Mean Prop [95%CI]	Reduction [95%CI]	P-adjusted[1]
HC	0.423 [0.367, 0.479]	Reference	
AmnAD	0.268 [0.213, 0.323]	-0.155 [-0.252, -0.058]	8.36e-04
lvPPA	0.215 [0.159, 0.270]	-0.208 [-0.305, -0.112]	9.86e-06
PCA	0.219 [0.163, 0.274]	-0.204 [-0.301, -0.107]	1.51e-05

[1] P-adjusted value from the model 1 = Proportion BEX1:Exc ~ Neuropath.group + Brain Region + Sex. Residual normality was acceptable: Shapiro-Wilk p = 0.214 Levene p = 0.346; and Neuropath.group × BrainRegion interaction was not significant (ANOVA p = 0.302).

CONCLUSIONS

- Our findings provide quantitative evidence that BEX1-positive excitatory neurons are selectively vulnerable across both typical and atypical Alzheimer's disease, and across distinct brain regions.
- The convergence of this vulnerability across clinical phenotypes and anatomical sites highlights a shared, cell type-specific signature of AD-related neurodegeneration and reinforces the importance of determining neuronal vulnerability in AD.

ACKNOWLEDGMENTS / CONTACT

NIH 5R01AG075802-05

Rizzi.Liara@mayo.edu
Grinberg.Lea@mayo.edu