An intact, molecularly active postmortem whole human brain model of Alzheimer's disease

Paul D. Wes, Congwei Wang, Scott Pope, Chunmin Ge, Nghia Nguyen, Machele Riccio, Josip Butkovic, Ivan Mumlek, Leah Rosales, Lindsay Slupczewski, Raghav Kansal, Mansi Vyas, Veronica Jove, Lucija Barisic, Bradley Parry, Sean Murphy, Zvonimir Vrselja

Bexorg, New Haven, CT, USA

INTRODUCTION

A major barrier to developing effective therapies for Alzheimer's disease is the limited ability of preclinical models to predict clinical efficacy. Animal models often fail to recapitulate key aspects of human disease biology and etiology, and human-derived in vitro systems, while valuable, typically lack the multicellular architecture and mature phenotypes of the adult aged brain. To address this gap, we established BrainEx, an ex vivo whole brain perfusion platform that supports physiological maintenance of molecular and cellular function in postmortem human brains, including tissue from donors with Alzheimer's disease. BrainEx enables preclinical drug discovery and translational validation directly in the human disease brain, supporting target validation, pharmacokinetic assessment and brain penetration, pharmacodynamic and functional readouts, biomarker discovery, and novel target identification.

ETHICAL CONSIDERATIONS

Acquisition of postmortem human brains adheres to the highest possible ethical standards, overseen by an independent board of world-renowned bioethicists. Brains are procured through Organ Procurement Organizations (OPOs) with enhanced levels of consent from patients and families that specifically cover the BrainEx platform. Measures are taken on the BrainEx device to ensure that there is no possibility of coordinated network activity associated with consciousness.

[1] THE BRAINEX PLATFORM

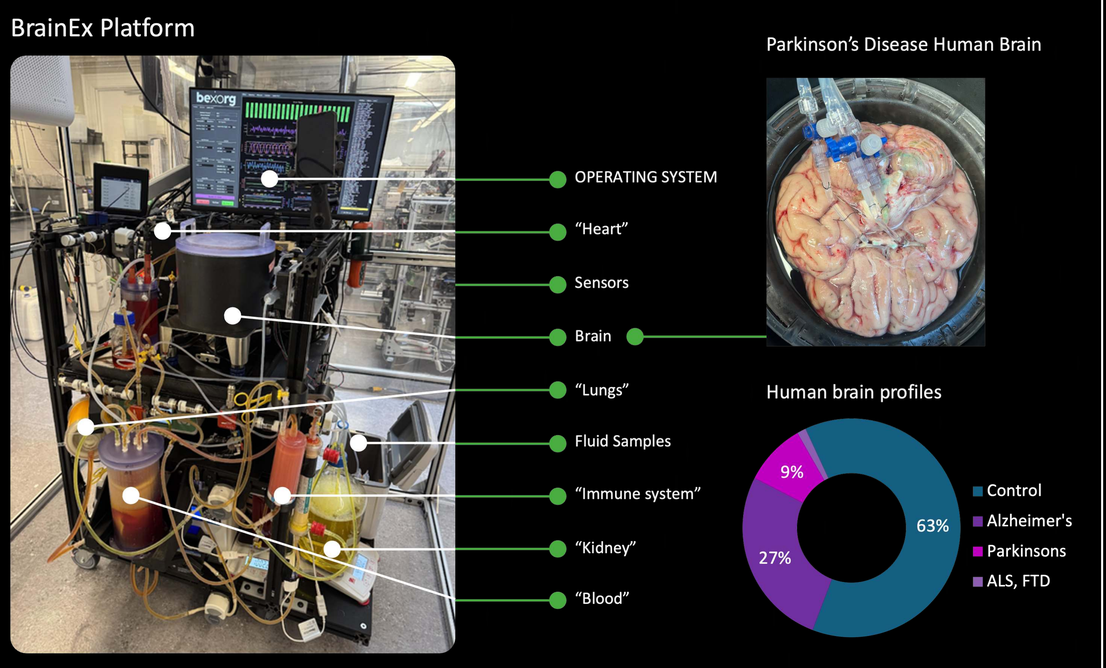


Figure 1. The BrainEx platform enables drug discovery in whole human disease brains. Bexorg has established a platform that maintains intact, molecularly and cellularly active postmortem human brains. The brain is connected to the BrainEx device via its endogenous vascular system. An acellular artificial perfusate supplies the brain with oxygen and nutrients, while a real-time operating system regulates physiological homeostasis. Drugs can be administered to the brain systemically, and pharmacokinetics, pharmacodynamics, and functional pharmacology can be assessed longitudinally in brain tissue and translational biofluids.

[2] PHARMACOKINETICS (PK)

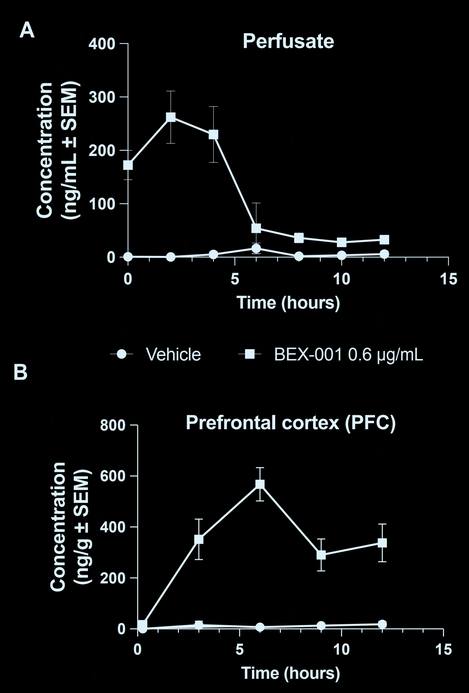


Figure 2. Pharmacokinetics of small molecule (BEX-001) on the BrainEx platform. BEX-001 (0.6 µg/mL) or vehicle control was administered systemically via the perfusate circuit and quantified longitudinally over 12 h by LC-MS in (A) circulating perfusate (ng/mL) and (B) brain parenchyma (ng/g).

[3] FUNCTIONAL PHARMACOLOGY

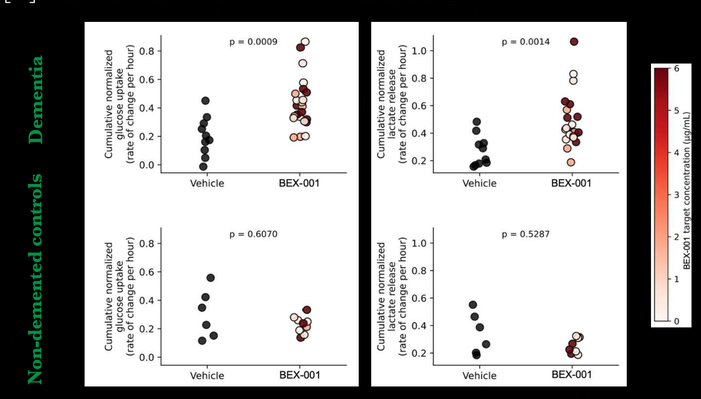


Figure 3. BrainEx enables functional pharmacology assessment of energy metabolism in intact postmortem human brains. Cumulative normalized glucose uptake (left) and lactate release (right) were measured during BrainEx perfusion after systemic dosing with BEX-001 or vehicle. Data are stratified by donor diagnosis (Dementia, top; Non-demented controls, bottom). Each point is one perfusion run; BEX-001 points are colored by achieved target concentration (0-6 µg/mL). In dementia brains, BEX-001 significantly increased glucose uptake ($p = 0.0009$) and lactate release ($p = 0.0014$). No significant effects were observed in non-demented control brains (glucose uptake $p = 0.6070$; lactate release $p = 0.5287$).

[4] TARGET ENGAGEMENT

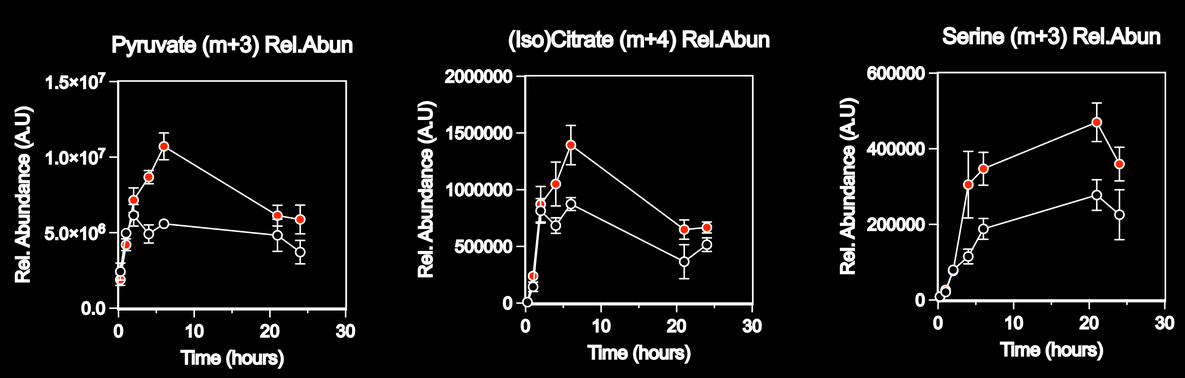


Figure 4. Pharmacodynamic (PD) biomarkers measured longitudinally on BrainEx using stable-isotope tracing. Brains were co-dosed with ^{13}C-glucose and either vehicle (black) or BEX-001 (red), and serial prefrontal cortex (PFC) biopsies were collected at the indicated timepoints during perfusion. Targeted mass spectrometry quantified ^{13}C incorporation into pathway metabolites-pyruvate (m+3), (iso)citrate (m+4), and serine (m+3) (relative abundance, A.U.; m+x denotes the number of ^{13}C atoms incorporated).

[5] BRAIN PENETRATION

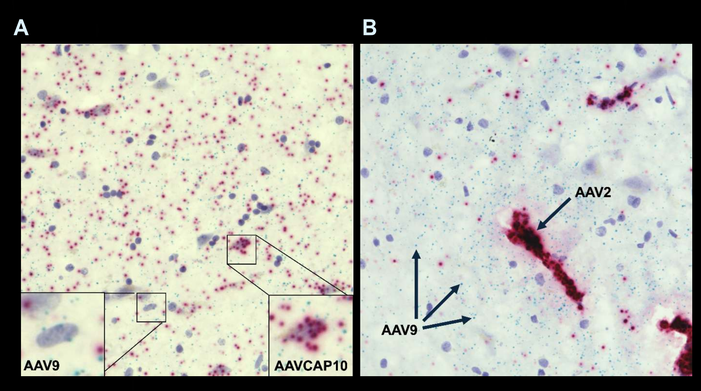


Figure 5. BrainEx preserves blood-brain barrier selectivity and predicts AAV vector brain biodistribution. Adeno-associated viral (AAV) vectors were delivered systemically to human brains maintained ex vivo using the BrainEx perfusion platform. Transgene expression was assessed 24 h post-delivery by RNAscope in situ hybridization. (A) AAV9 (blue) and AAV-CAP10 (red), serotypes with demonstrated ability to efficiently cross the blood-brain barrier (BBB). (B) AAV2 (red) and AAV9 (blue); AAV2 is a serotype with limited capacity to traverse the BBB, whereas AAV9 is BBB-permeable.

[6] BIOMARKER IDENTIFICATION

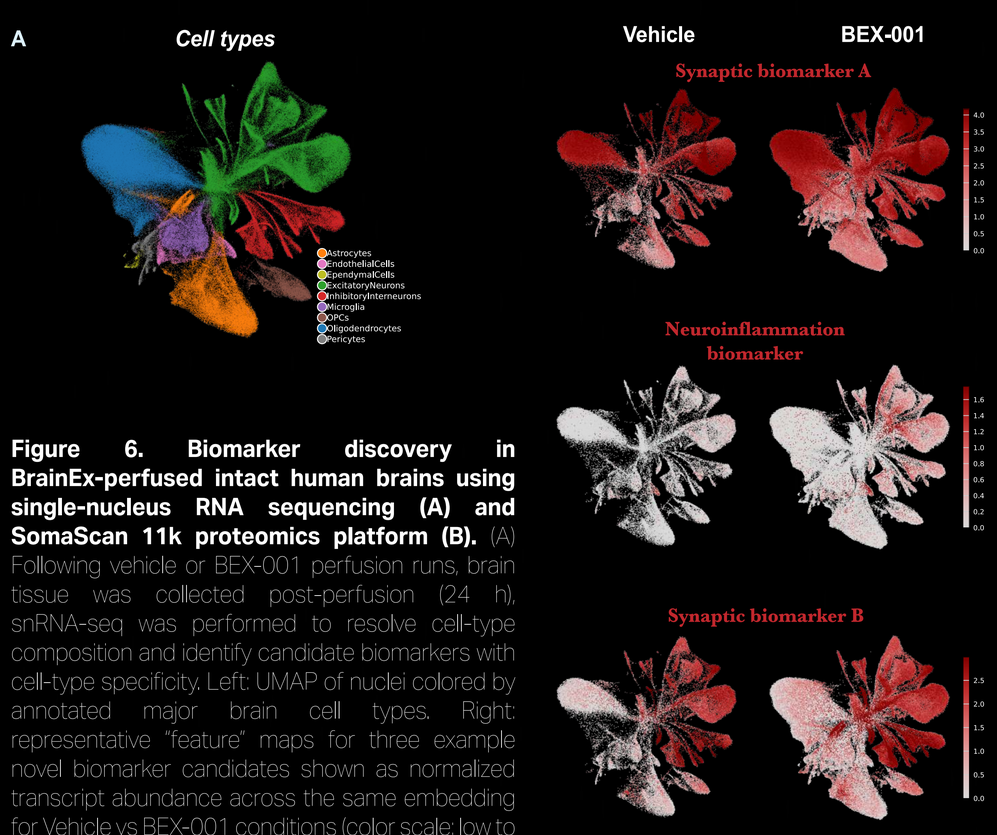
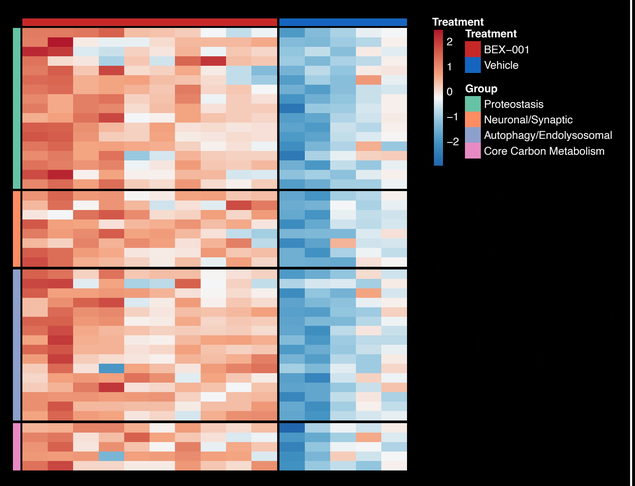


Figure 6. Biomarker discovery in BrainEx-perfused intact human brains using single-nucleus RNA sequencing (A) and SomaScan 11k proteomics platform (B). (A) Following vehicle or BEX-001 perfusion runs, brain tissue was collected post-perfusion (24 h), snRNA-seq was performed to resolve cell-type composition and identify candidate biomarkers with cell-type specificity. Left: UMAP of nuclei colored by annotated major brain cell types. Right: representative "feature" maps for three example novel biomarker candidates shown as normalized transcript abundance across the same embedding for Vehicle vs BEX-001 conditions (color scale: low to high expression).



(B) Following vehicle or BEX-001, brain tissue was collected and profiled using the SomaScan 11k. Heatmaps display row-scaled log2 protein expression (z-score, capped at +/-3) for curated functional protein groups: Proteostasis, Autophagy/Endolysosomal, Neuronal/Synaptic, and Core Carbon Metabolism. Columns represent individual samples separated by treatment group (BEX-001 vs. Vehicle).

[7] TARGET DISCOVERY

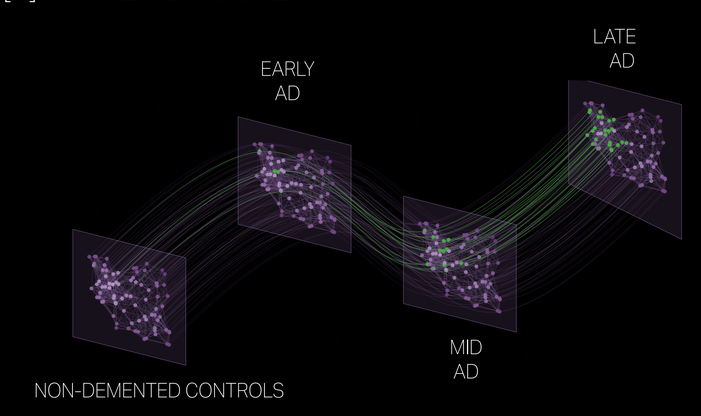


Figure 7. Neurolens learns patient-stratified disease trajectories using BrainEx derived ground truth multiomic data. Multiomic profiles generated from physiologically maintained postmortem human brains on the BrainEx perfusion platform are embedded into a low dimensional disease manifold spanning Healthy, Early, Mid, and Late Alzheimer's disease states. Each point represents an individual human brain omic sample (or aggregated donor profile), and connecting streamlines depict inferred transitions along the disease continuum, enabling data driven ordering of molecular states over progression. This trajectory framework supports discovery of stage specific pathway shifts, nomination of mechanistically distinct therapeutic target classes, and identification of biomarkers that bridge brain tissue biology to biofluid readouts such as CSF and plasma.

References
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