

Research Articles | Development/Plasticity/Repair

Microglial Presenilin-1 modulates neuronal function and memory-associated behaviors

<https://doi.org/10.1523/JNEUROSCI.0310-26.2026>

Received: 21 February 2026

Revised: 23 July 2026

Accepted: 1 October 2026

Copyright © 2026 the authors

This Early Release article has been peer reviewed and accepted, but has not been through the composition and copyediting processes. The final version may differ slightly in style or formatting and will contain links to any extended data.

Alerts: Sign up at www.jneurosci.org/alerts to receive customized email alerts when the fully formatted version of this article is published.

Microglial Presenilin-1 modulates neuronal function and memory-associated behaviors

Paula da Silva Frost ^{1,2,3}, Carley A. Martinez ^{1 *}, Alon Millet ^{4,5 *}, Joshua McWhirt ¹, Jia Cheng ⁶, Michael Bamkole ⁶, Yong Kim ^{7,8}, Hernandez M. Silva ⁹, Juan J. Lafaille ¹⁰, Estefania P. Azevedo ^{1,2,3}, Beth Stevens ^{11, 12, 13}, Jose Henrique Ledo ^{1, 2,3#}.

¹ Department of Neuroscience, Medical University of South Carolina, Charleston, South Carolina, USA.

² Institute for Behavioral Genetics, University of Colorado Boulder, Boulder, Colorado, USA.

³ Department of Integrative Physiology, University of Colorado Boulder, Boulder, Colorado, USA.

⁴ Tri-Institutional Program in Computational Biology and Medicine, The Rockefeller University, New York, New York, USA.

⁵ Laboratory of Systems Cancer Biology, The Rockefeller University, New York NY

⁶ Laboratory of Cellular and Molecular Neuroscience, The Rockefeller University, New York, New York, USA.

⁷ Department of Neurosurgery, Robert Wood Johnson Medical School, Rutgers University, Piscataway, NJ, USA.

⁸ Brain Health Institute, Rutgers University, Piscataway, NJ, USA.

⁹ Ragon Institute, Department of Biology, Massachusetts Institute of Technology, Cambridge, MA, USA.

¹⁰ Skirball Institute of Biomolecular Medicine, New York University, New York, New York, USA.

¹¹ Department of Neurology, F.M. Kirby Neurobiology Center, Boston Children's Hospital, Harvard Medical School, Boston, MA, USA.

¹² Stanley Center for Psychiatric Research, Broad Institute of MIT and Harvard, Cambridge, MA, United States.

¹³ Howard Hughes Medical Institute, Boston Children's Hospital, Boston, MA., USA.

* These authors contributed equally.

Correspondence to jose.ledo@colorado.edu (JHL)

Abstract

Microglial cells, the macrophages of the brain parenchyma, are increasingly recognized to play a key role in synaptic plasticity and function. Presenilin 1 (PS1) is an essential protein involved in learning and memory; the loss of neuronal Presenilin function causes cognitive deficits and impaired synapse plasticity in mice. Here we show memory enhancement in male mice lacking PS1 in microglia (PS1^{ΔMG}). Leveraging high-throughput RNA sequencing with single-cell deconvolution-analysis, we estimate that PS1^{ΔMG} increases Cd52-lo microglial population, a proliferative group enriched in AP-1 responsive genes; and increases chemokines *Ccl7* and *Ccl8*. In silico prediction of binding partners showed that microglial CCL7 may act via inhibition of neuronal CCR5, a receptor known to negatively regulate learning and memory. By profiling active neuronal populations via PhosphoTRAP the data showed a non-autonomous cell effect of microglial PS1 by modulating genes involved in neuronal excitability and synaptic plasticity. Ex vivo electrophysiology and in vivo calcium imaging in freely behaving mice confirm these effects with functional changes in CA1 hippocampus as seen by increased NMDA/AMPA ratios and increased amplitude of neuronal Ca²⁺ transients in freely behaving animals, respectively. Remarkably, PS1^{ΔMG} mice exhibited increased memory retention in contextual fear conditioning and novel object recognition. Together, these data suggest a novel mechanism where microglial PS1 negatively regulates neuronal activity through modulation of NMDAR and memory-associated behaviors.

Significance Statement

Microglia are increasingly recognized as active regulators of neuronal function, yet the molecular mechanisms that constrain their influence on synaptic activity remain incompletely defined. This study identifies microglial Presenilin-1 (PS1) as a key regulator of neuron-microglia signaling that shapes synaptic transmission and learning. We show that loss of microglial PS1 shifts microglia toward a transcriptional and signaling state that enhances NMDA receptor-dependent synaptic activity and improves memory, demonstrating that microglial molecular programs can directly modulate neuronal excitability. These findings establish microglial PS1 as a physiological brake on circuit plasticity and reveal that altering microglial signaling alone is sufficient to influence cognition. Targeting microglial regulatory pathways may therefore provide new strategies to enhance synaptic resilience and cognitive function in brain disorders.

Introduction

In recent years, many studies have reported that microglia, the predominant immune cells of the brain, modulate neuronal function through diverse roles, including the synaptic function and plasticity that could underlie learning and memory (Stevens et al., 2007; Schafer et al., 2012; Parkhurst et al., 2013; Zhan et al., 2014; Hong et al., 2016; Garré et al., 2017; Lehrman et al., 2018; Badimon et al., 2020; Favuzzi et al., 2021; Ayata et al., 2025). Presenilin 1 (*Psen1* gene – PS1 protein) is an essential protein for learning and memory (Yu et al., 2001; Marjaux et al., 2004;

Saura et al., 2004; Wines-Samuelson et al., 2010; Watanabe et al., 2012) and has been extensively studied in neurons in the context of cognition and Alzheimer's disease (AD). PS1 is the catalytic subunit of γ -secretase, an enzyme complex responsible for the formation of a plethora of products. To date 149 transmembrane proteins have been described as γ -secretase substrates (Haapasalo and Kovacs, 2011; Güner and Lichtenthaler, 2020). PS1 is a versatile protein and can regulate important biological processes (Bustos et al., 2017b, 2017a; Frost et al., 2017; Ledo et al., 2020b, 2020a). Interestingly, selective loss of Presenilin function in neurons impairs synaptic plasticity, resulting in cognitive deficits that are associated with reductions in both the number of synaptic NMDA receptors and in NMDA receptor-mediated responses (Saura et al., 2004). While the majority of research focuses on the role of PS1 in neurons, PS1 is also expressed in microglia from development through adulthood (Zhang et al., 2014). Given that PS1 serves as the catalytic core of γ -secretase and mediates the cleavage of myeloid substrates such as TREM2 and SIRP α (Londino et al., 2015; Glebov et al., 2016; Zhao et al., 2017; Steiner et al., 2020), its role in regulating microglial signaling and function warrants investigation. We previously reported that not only is γ -secretase active in microglia, but phosphorylation of microglial PS1 at serine 367 also plays an important role regulating autophagy-lysosomal pathways. Conversely, mice lacking S367 phosphorylation (PS1^{S367A}) showed synapse loss in the mouse hippocampus, due in part to defective spine elimination by microglia (Ledo et al., 2020a). Furthermore, we found that PS1^{S367A} alters microglial gene networks and the number of microglial progenitors during development (Ledo et al., 2020b), suggesting that loss of PS1 function in microglia can have profound consequences during development. Our previous work showed a novel function for PS1 by which it contributes to microglia development and function, suggesting that microglial PS1 might play a key role in brain development and neuronal function.

Here, we examined the role of microglial PS1 in neuronal plasticity and function in the adult mouse brain. Depletion of PS1 in microglia (PS1^{AMG}) altered microglial population structure

and was associated with changes in neuronal gene expression. PS1^{ΔMG} showed enrichment of sensome-, secretome-, and inflammatory-related pathways, consistent with altered microglia-neuron communication. Protein interaction network analysis predicted an interaction between CCL7 and neuronal CCR5. PhosphoTRAP profiling of active neurons revealed enrichment of metabolic pathways, including lipid and fatty acid catabolism, indicating transcriptional remodeling in active neuronal populations. Functionally, PS1^{ΔMG} mice exhibited enhanced NMDAR-mediated synaptic currents, and elevated excitatory neuronal activity measured by fiber photometry, accompanied by improved memory retention. Together, these findings identify microglial PS1 as a regulator of synaptic activity and learning-related neuronal function.

Materials and Methods

Mice

C57BL/6 (000664), Cx3Cr1^{CreERT2/+} (021160), Psen1^{loxP} (004825) mice were purchased from the Jackson Laboratories and maintained in our facilities. Psen1^{loxP/loxP} mice were first crossed to CX3CR1^{CreER/+}. Psen1 knockout (PS1^{ΔMG}) mice were generated by further crossing CX3CR1^{CreER/+}; Psen1^{loxP/+} to Psen1^{loxP/loxP}. CX3CR1^{CreER/+} mice littermates were used as controls. Both Control and PS1^{ΔMG} mice received 4-OHT. Mice were weaned, genotyped by Transnetyx, and kept on a 12h light/dark cycle with access to food and water ad libitum. Mice were maintained at The Rockefeller University and MUSC Animal facilities and used at 10-12 weeks of age for all experiments except when otherwise indicated. Littermates of the same sex were randomly assigned to experimental groups. Male mice were used for experiments. Animal care and experimentation were according to NIH guidelines and were approved by the Institutional Animal Care and Use Committee at The Rockefeller University (protocol #18035-H) and Medical University of South Carolina (protocol #2022-01513).

138

139 **4-Hydroxytamoxifen (4-OHT) treatment**

140 4-OHT (Sigma-Aldrich, H7904) was dissolved in ethanol at 100mg/mL, and then further diluted in
141 corn oil for intraperitoneal injection. Pregnant mouse received two doses of 0.4 g/g of 4-OHT at
142 embryonic day E9.5 and E11.5. Specificity of PS1 targeting after 4-OHT induced Cre
143 recombination was assessed in adult mice (see FigS1).

144

145 **Brain microglia isolation from adult mice**

146 Mice were perfused with DPBS (Ca²⁺/Mg²⁺-free), and brains were placed in FACS buffer (PBS,
147 5% FBS and 10mM HEPES). Brain were minced and incubated with 4000U/mL of collagenase D
148 (Roche, 11088858001) at 37°C for 30min and inactivated by 10mM EDTA for 5min. Digested
149 brains were passed through a 70-µm cell strainer, washed in FACS buffer and submitted to
150 centrifugation at 2,000 r.p.m. in 38% Percoll for 30 min. Cell pellets were resuspended in FACS
151 buffer and incubated with a FcR blocker-specific antibody (BD-Pharmingen 553141) for 15min.
152 Cells were then washed and stained with the markers described below to certify cell population
153 specificity. Microglial cells isolated from mice brain were sorted using BD FACSAria Cell Sorters
154 from BD Biosciences.

155

156 **Bulk-sequencing of isolated microglia**

157 Isolated microglial cells (Cd11b⁺ CD45^{int}, MHC II⁻ CD206⁻, Ly6C⁻ c-kit⁻, CX3CR1⁺) were sorted
158 directly into Buffer RLT (Qiagen). Each brain was sorted into a separate tube, and RNA was
159 isolated for each sample according to manufacturer's instructions for RNeasy Micro Kit (Qiagen)
160 and used as input for library preparation using the SMART-Seq v4 Low Input workflow (Takara

Bio). Samples were sequenced on an Illumina NovaSeq SP.

RNA sequencing analysis

Raw reads were quality-checked with fastp and aligned to mm10 with STAR before using FeatureCounts to produce a counts matrix. Counts were variance-stabilized and normalized by limma-voom. For volcano plots, logFC values and t-tests were run on normalized counts after filtering to genes listed in the sensome or secretome databases (Hickman et al., 2013; Diaz-Aparicio et al., 2020). Normalized expression of key genes was defined as the ratio of raw counts against the mean expression computed across both control and PS1^{ΔMG} samples. For heatmaps, genes involved in inflammatory, or cytokine pathways were identified by association with the GO terms GO:0050729 and GO:0005125, respectively, before being scaled and seriated.

Deconvolution of bulk RNA sequencing data

The processed Seurat structure from Hammond et al. (Hammond et al., 2018) was loaded into R and reclustered with default parameters (30 PCA dimensions, resolution 0.8). The resulting clusters were manually annotated, with clusters with high similarity to clusters identified by the original authors indicated with the associated parenthetical. This annotated Seurat structure was coerced to a single cell expression set and used as the reference for Bisque to produce estimated decomposed frequencies in the bulk RNAseq data. An enrichment score for each cluster was defined as $\mathcal{S} = \sqrt{N} \frac{\bar{x}_{PS1\Delta MG} - \bar{x}_{Control}}{\bar{x}}$, where N is the size of the cluster in the original single cell data, $\bar{x}$ is the average frequency of the cluster across all samples, and $\bar{x}_{PS1\Delta MG}$ and $\bar{x}_{Control}$ are the average frequency of the cluster across PS1^{ΔMG} and control samples, respectively.

183

184 **Flow cytometry analysis**

185 Fluorescent-dye-conjugated antibodies were purchased from Biolegend (anti-Cx3Cr1, 149016;
186 anti-CD117 (c-kit), 105824; anti-Ly6c, 128033; anti-MHCII, 107643; anti-CD206, 141720),
187 Invitrogen (Anti-CD11b, 47-0112-82; anti-CD45, 56-0451-82) or BD-Pharmingen (anti-
188 CD16/CD32 (FC blocking); 553141). Live cells were verified using DAPI (4',6-Diamidino-2-
189 Phenylindole, Dilactate), (D3571, Thermofisher). Flow cytometry data were acquired on an LSR-
190 II flow cytometer (Becton Dickinson) and analyzed using FlowJo software (Tree Star).

191

192 **Real-time relative quantification PCR**

193 Microglial cells isolated from mice were sorted directly into RNA lysis buffer (Qiagen, 79216)
194 supplemented with 2 M dithiothreitol (DTT). RNA was isolated as described above. qPCR was
195 performed using Taqman reagents. ActB was used to normalize samples. Predesigned probes
196 used were purchased from IDT. mRNA levels are expressed using the $2^{-\Delta\Delta Ct}$ method (Schmittgen
197 and Livak, 2008).

198

199 **PhosphoTRAP**

200 pTRAP profiling was performed according to Knight et al 2012 (Knight et al., 2012). Briefly,
201 PS1^{ΔMG} and control mice had their hippocampi removed and pooled together. After
202 homogenization, ribosomal immunoprecipitation was obtained using pS6 antibody (Invitrogen,
203 #44-923G) previously conjugated to Protein-A coated magnetic beads (Thermofisher), following
204 manufacturer's protocol. Tissue RNA from before the immunoprecipitation was saved as input
205 for quality control. Both input and immunoprecipitated RNA were purified in RNeasy Mini kit

(QIAGEN). RNA quality was measured on a bioanalyzer using RNA PicoChip. Only RIN>7 was used in subsequent experiments. The samples had their cDNA amplified in SMARTer Ultralow Input RNA for Illumina Sequencing Kit and sequenced on an Illumina HiSeq2500 platform.

PhosphoTRAP data analysis

Raw reads were quality-checked with fastp and aligned to mm10 with STAR before being fed into FeatureCounts to produce a counts matrix. logFC values over background for each condition were computed and used as the normalized counts for downstream analysis. Data was filtered only to genes where the q-value for at least one condition was less than 0.01 and scaled prior to visualization. Pathway analysis was performed by computing the log-fold change of expression between PS1^{ΔMG} and control mice and ranked for the genelist in fGSEA against the GO Biological Process pathways.

Ligand-receptor interaction modeling

To define ligand-receptor interactions between microglia and neurons, putative ligands and receptors on neurons that might interact with microglia were identified by querying the CellPhoneDB v5 database for known interaction partners of proteins in the microglial sensome or secretome lists described above. Ligand expression in microglia was computed by taking the average expression of the corresponding transcript in bulk RNAseq of microglia from both PS1^{ΔMG} and control mice. To estimate receptor expression, we extracted raw counts data from single cells in cortical and hippocampal glutamatergic and GABAergic neuron clusters from the Allen Brain Atlas (Yao et al., 2023). We calculated the fraction of cells in which each transcript was detected and filtered putative interactions to only those in which the receptor was present in at least 0.1% of cells, thereby generating a list of biologically informed and feasible ligand-

receptor interactions between microglia and neurons. For visualization in Sankey-style plots, arrows were generated by computing an interaction score that defines the relative importance of the interaction in PS1^{ΔMG} mice versus control mice as the log-fold change between PS1^{ΔMG} and control in expression of the ligand from bulk RNAseq data. Interaction score values were then shifted to center around zero for interpretation purposes. Points representing ligands or receptors were colored by the average expression of the ligand in microglia of both PS1^{ΔMG} and control mice or by the fraction of neurons from the Allen Brain Atlas dataset in which transcripts of the given receptor were detected.

Immunoblotting and antibodies

Mouse CA1 hippocampus were lysed with a Syn-PER Synaptic Protein Extraction Reagent (Syn-PER™, 87793, Thermo Fisher Scientific) supplemented with a protease and phosphatase inhibitor cocktail (78442, Thermo Fisher Scientific). Synaptosomes-enriched extracts were prepared per manufacturer's instructions. Protein levels were measured by BCA. Samples were mixed with sample buffer and subjected to SDS-PAGE with 4–20% Novex Tris-Glycine gels (Thermo Fisher Scientific), followed by protein transfer onto a PVDF membrane. Immunoblotting was performed with a standard protocol using the following: anti-GluN2B (75-097, NeuroMab, 1:1,000), anti-GluN1 (75-272, NeuroMab, 1:1,000), anti-GluA1 (75-327, NeuroMab, 1:1000), anti-GluA2 (ab133477, Abcam, 1:1000), anti-Synaptophysin 1 (101004, Synaptic Systems, 1:1,000), anti-Vglut1 (135011, Synaptic Systems, 1:1000), anti-PSD95 (ab18258, Abcam, 1:1,000), anti-beta-Tubulin (2146, Cell Signaling, 1:1,000), anti-GAPDH (GTX89740, GeneTex, 1:1000), Presenilin 1 (MAB5232, Millipore Sigma, 1:500). Secondary antibodies were from Li-COR: IRDye® 800CW anti-Mouse (926-32212, 1:1000) IRDye® 800CW anti-Rabbit (926-32213, 1:1000), IRDye® 680RD anti-Rabbit (926-68073, 1:1000).

Electrophysiology

Transversal slices (400µm) were cut on Vibratome (Leica Microsystems, USA) in cutting solution (mM): 87 NaCl, 25 NaHCO₃, 2.5 KCl, 0.5 CaCl₂, 7 MgCl₂, 25 glucose, and 75 sucrose with 95%O₂ and 5%CO₂. Slices recovered for 45minutes at 35°C and 1h at RT in the recording solution. The extracellular recording solution contained (mM): 125 NaCl, 25 NaHCO₃, 2.5 KCl, 1.25 NaH₂PO₄, 2 CaCl₂, 1 MgCl₂ and 25 glucose (95%O₂ and 5%CO₂). The slice was placed in a recording chamber (RC-27L, Warner Instruments, USA) and constantly perfused with oxygenated aCSF at 24°C (TC-324B, Warner Instruments, USA) at a rate of 1.5–2.0ml/min. Dorsal CA1 pyramidal neurons were selected using a 40X water-immersion lens (Olympus BX51WI microscope, Japan) and recorded with the Multiclamp 700A amplifier (Molecular Devices). Whole-cell voltage-clamp recording technique was performed to record evoked EPSCs as described previously (Cheng et al., 2014). Glass pipettes (King Precision Glass, Inc, Glass type 8250) pulled in a pipette puller (Narishige) to a resistance of 3–4MΩ. Bicuculline (20µM) was added to aCSF. Patch electrodes contained internal solution (mM): 130 Cs-methanesulfonate, 10 CsCl, 4 NaCl, 10 HEPES, 1 MgCl₂, 5 EGTA, 2 QX -314, 12 phosphocreatine, 5 MgATP, 0.2 Na₃GTP, pH 7.2-7.3, 265-270mOsm. EPSC was evoked by stimulation of Schaffer-collaterals by a bipolar stimulating electrode (FHC). Stimulation pulse was generated by a S48 pulse generator (Grass Technologies). Evoked AMPAR-EPSC was elicited by a stimulation pulse (0.5ms, 70µA) when cells were clamped at -70mV. For evoked NMDAR-EPSC, cells were clamped at -70mV and then depolarized to +60mV for 3s to relieve the Mg²⁺ block. Same stimulation pulse was applied to elicit EPSC. eAMPA-EPSC was calculated as the peak amplitude. eNMDA-EPSC was calculated at 100ms after stimulation. Data analyses were performed with Clampfit. Statistical analysis was performed using the two-tailed unpaired Student's *t*-test (GraphPad Prism 6).

Stereotaxic injections and Fiber Implantation

Mice were anesthetized with 2% isoflurane, placed in a stereotaxic frame (Kopft Instruments). Subcutaneous injection of meloxicam (2mg/kg) was given to each mouse before and after surgery for up to 2 days. The craniotomy was performed using a dental drill (Dremel). AAV was infused using a Hamilton syringe, 10^{11} titer viral preparations of AAV5-hSyn-GCaMP6s were purchased (Addgene) and injected unilaterally: AP-1.82mm, ML+0.5mm, DV-1.7mm. Mice were monitored for 72h to ensure full recovery and used in experiments 2-4 weeks later. Optic fiber implants (400mm, Doric) for photometry experiments were implanted to the same coordinates as the viral injections with a -0.2mm difference. Implants were inserted slowly and secured to the mouse skull with Metabond (Parkell Inc).

Fiber photometry

Analysis of the signal was done using fiber photometry system and processor (RZ5P) from TDT (Tucker-Davis Technologies), and the Synapse software (Tucker-Davis Technologies). Change in fluorescence (ΔF) was calculated as (473nm signal – fitted 405nm signal), adjusted so that a $\Delta F/F$ was calculated by dividing each point in ΔF by the 405nm curve at that time. Z-score and plot traces were calculated using MATLAB. Peak analysis to determine frequency and amplitude of the signals was done by determining the median average deviation (MAD) of the corrected/normalized data set and peaks were identified as events that exceeded the MAD by 2.91 deviations (Calipari et al., 2016). Behavioral variables (nose-to-object) were timestamped in the signaling traces via the real-time processor as TTL signals from Noldus (Ethovision) software. This allowed for precise determinations of the temporal profile of signals in relation to specific behaviors. All behavior was manually scored by a blind, second investigator.

Novel-object recognition (NOR) memory

The NOR task was conducted as described (Tang et al., 1999; Stefanko et al., 2009). After acclimation, mice were habituated for 5min in a 28x28cm open field arena. During the training phase, a 3min training session was used as this does not typically lead to changes in short-term memory or long-term memory in wildtype mice (Stefanko et al., 2009; McQuown et al., 2011). Briefly, two identical objects are placed in opposite sides of the arena and mice are free to explore the arena and objects for 3min. No preference for a specific object was observed during the training session. 24h afterwards mice performed the testing session; in which one of the objects (familiar object) is replaced by a novel object and the mice were free to explore the arena and objects for 5min. For training and test condition a discrimination index was calculated as the ratio between the time spent exploring the novel object and the sum of time spent exploring the familiar object and novel object.

Contextual Fear Conditioning

We used a fear conditioning shock chamber (MedAssociates) coupled with shock-grid floors (Coulbourn Instruments). During training (5min), mice were placed in the chamber, and a foot shock was delivered (0.7mA for 2s). After, mice were allowed to stay in the chamber for 1min and then removed to their home cage. During the retention test, each mouse was placed back into the shock chamber, and the freezing response was recorded for 5min (contextual conditioning). Freezing was measured manually by a blind second experimenter.

Locomotion Analysis

Mice were placed in an open field arena for the duration of ten minutes and video recordings were

taken. Videos were analyzed using Any-Maze software, in which the dimensions of the arena are recorded, and the animal is tracked throughout the duration of the task. Overall total distance travelled by each mouse is then given in meters (m).

Quantification and statistical analysis

Statistical analysis was performed in GraphPad Prism, MATLAB, or pClamp10 software. Fiber photometry raw data was recorded using TDT Synapse software (Tucker-Davis Technologies) and analyzed using custom written codes in MATLAB software. Animal behavior was time-locked with Ca²⁺ recording using digital timestamps Noldus Ethovision 9.0 software. Normality tests were performed before choosing the statistical test.

Data availability

Data supporting the findings of this study are available from the corresponding author upon reasonable request.

Results

Microglial PS1 loss promotes cell-autonomous microglial remodeling.

Global deletion of PS1 is lethal in mice, and neuron-specific deletion impairs learning and memory (Saura et al., 2004). However, the role of PS1 in microglial cells is poorly understood. To examine the role of Presenilin specifically in microglia, we generated a conditional microglial PS1 knockout (PS1^{ΔMG}) (Figure S1). First, we performed bulk RNA sequencing (Figure 1A) following microglial isolation from adult PS1^{ΔMG} and control animals. The knockout did not impact microglial

number or PS1 expression in neurons (Figure S1 F-I). We then mapped our bulk sequencing data onto single cell data generated by Hammond, T. R. et al. (Hammond et al., 2018) to investigate changes in microglial cell state. We re-analyzed the clustering from Hammond et al., expanding the populations originally defined (Figure 1B and Figure S2). Based on the results of single-cell deconvolution of our bulk counts, we computed the enrichment score for each microglial subpopulation, reflecting both the relative frequency of each cluster between control and PS1^{ΔMG} animals as well as the overall frequency of the cluster. Out of 18 clusters represented in Hammond et al., 13 were detected in our deconvolution analysis (Figure 1C). Nnat-hi and Nnat-lo Embryonic, Fabb5-hi (3), Ccl4-hi (8) and Monocyte/Macrophage cluster are not represented in our dataset. Bulk deconvolution estimates of cluster abundance against this reference showed that PS1^{ΔMG} induced a shift in population frequency, increasing Cd52-hi and Cd52-lo populations and decreasing the Rrm2-hi population compared to control (Figure 1D, Figure S3).

Cd52-hi, Cd52-lo and Rrm2-hi populations are characterized as proliferating microglia, based on gene enrichment profile (Table S1). The Rrm2-hi proliferative population decreased in PS1^{ΔMG} is not enriched in immune genes, while CD52-lo and CD52-hi have an increased AP-1 activation or phagocytosis related genes respectively. This suggests proliferative microglia enrichment is now functionally primed in PS1^{ΔMG}, although more experiments are necessary to confirm this. Projecting our enriched populations into Hammond et al. dataset does not show sex as a variable that affects population clusters in control mice (Figure S3).

Next, we identified differentially expressed genes between control and PS1^{ΔMG} (Figure 1E) and specifically examined genes annotated as being members of the microglial sensome (Hickman et al., 2013), microglial secretome (Diaz-Aparicio et al., 2020), and the general inflammatory cascade (defined by gene ontology) to evaluate how PS1^{ΔMG} influence microglial interaction with their local stroma. We found significant alterations in the microglial sensome, including decreases in two major chemokine receptors, *Ccr5* and *Cd74* (Figure 1F). This was

mirrored by accompanying changes in the microglial secretome and inflammatory cascade, consistent with the predicted enrichment of the CD52-lo population. Notably, *Ccl8* and *Ccl7*, two chemokines that are known to signal through CCR5, were among the most upregulated in PS1^{ΔMG} microglia (Figure 1G and H). Collectively, this data suggests that loss of microglial PS1 is sufficient to drive changes in microglial cell state and chemokine secretome that could affect microglial-neuronal interactions in the mouse brain. We also leveraged computational methods to estimate ligand-receptor interactions between microglia and neurons using the most upregulated genes found in PS1^{ΔMG} (Figure 2A). We identified genes from the microglial secretome with the biggest differential expression between PS1^{ΔMG} and control animals and queried the CellPhoneDB ligand-receptor interaction database to identify putative receptors for these ligands. We surveyed hippocampal single-cell data from the Allen Brain Atlas (Yao et al., 2023) to compile a targeted list of receptors with measurable expression in neurons, yielding a final set of ligand-receptor pairs that are likely relevant in this biological system. Finally, we computed relative interaction strength of each ligand-receptor pair on the basis of the ratio of expression of the ligand between PS1^{ΔMG} and control animals. Among the cytokines shown in analysis, both CCL8 and CCL7 are predicted to have increased biologically relevant interactions with CCR5 in PS1^{ΔMG} compared to control (Figure 2A). CCL7 is an inhibitor of neuronal CCR5, and inhibition of CCR5 signaling has been shown to increase neuronal excitability, improving performance in cognitive tasks (Zhou et al., 2016). This binding-partner predictive data suggests transcriptional alterations found in PS1^{ΔMG} have binding partners in neurons and are thus in direct capacity to affect neuronal activity and culminate in behavioral alterations.

Microglial PS1 loss is associated with non-cell-autonomous transcriptional remodeling of active neuronal ensembles.

To determine whether PS1^{ΔMG}-associated transcriptional changes were accompanied by

alterations in neuronal gene expression, we profiled active neuronal ensembles in control and PS1^{ΔMG} mice using PhosphoTRAP pulldown followed by mRNA sequencing. This approach enables activity-dependent molecular profiling of neurons in vivo by isolating transcripts from recently activated cells. Comparative analysis revealed marked differences in the transcriptional profiles of active neurons between control and PS1^{ΔMG} animals, particularly in genes related to neuronal activity (Figure 2B). Among the most upregulated neuronal genes from PS1^{ΔMG} mice are those associated with synaptic plasticity. Notably, we observed upregulation of adhesion molecules (*MarvelD*) (Liu et al., 2018), genes that increase synapse formation (*Rpph1*, *Ephb4* and *Notch4*) (Wang et al., 2004; Cai et al., 2017; Rasool and Jahani-Asl, 2024), and a translation initiation factor (*Eif4EBP3*) (Moon et al., 2009). Pathway enrichment analysis revealed distinct functional profiles between groups (Figure 2C). Active neurons from control animals showed greater enrichment of synaptic signaling pathways, including glutamate receptor signaling, whereas active neurons from PS1^{ΔMG} mice were preferentially enriched for metabolic processes. Because PhosphoTRAP quantifies transcripts enriched in active neurons relative to the total tissue transcriptome (IP / Input), pathway enrichment reflects the transcriptional composition of activated neuronal populations rather than absolute differences in signaling between control and PS1^{ΔMG} groups. In predominantly glutamatergic hippocampal circuits, enrichment of glutamatergic signaling pathways is therefore expected under baseline conditions. At the gene level, differential expression analysis identified selective changes in glutamatergic signaling components, including reduced transcription of *Grin2a* and *Grin2d* and increased expression of *Grin2c*, *Grina*, *Camk2g*, and *Grid1* in PS1^{ΔMG} relative to control. Together, these findings indicate transcriptional remodeling of functional pathways and glutamatergic signaling components in active neuronal populations following microglial PS1 deletion.

Microglial PS1 depletion remodels NMDA receptor composition, elevates NMDA synaptic

currents, and enhances hippocampal activity in vivo.

While PhosphoTRAP profiling provides insight into activity-dependent transcriptional programs, transcriptomic changes do not necessarily predict synaptic receptor composition or synaptic function. Therefore, we examined the levels of synaptic proteins in synaptosome-enriched hippocampal brain extracts. After generating the synaptosome-enriched fractions from PS1^{ΔMG} and control mice, we investigated the levels of the various synaptic proteins via western blot.

We report no differences in pre-synaptic vesicular glutamate transporter (VGlut), synaptic markers synaptophysin 1 and PSD95 protein levels in PS1^{ΔMG} compared to control (Figure 3A and B). Likewise, we did not see differences in the levels of AMPA subunits GluA1 and GluA2 (Figure 3C and D). We did, however, observe significant increases in GluN2B and GluN1 NMDA subunit levels in PS1^{ΔMG} mice (Figure 3E). Collectively, these data show the absence of microglial PS1 is sufficient to induce neuronal increase of NMDA glutamate receptors in synaptic structures.

To assess the functional consequences of the PS1^{ΔMG}-mediated changes in synaptic NMDA protein level, we examined the ratio of NMDA-to-AMPA excitatory postsynaptic currents (EPSC), which provides a measure of NMDAR activity relative to AMPAR, regardless of the number of presynaptic afferents recruited and the position of stimulating electrode. As shown in Figure 3F the NMDA/AMPA EPSC ratio was significantly increased in PS1^{ΔMG} CA1 pyramidal neurons compared to age-matched controls (control: 0.27 ± 0.06 , $n = 11$, PS1^{ΔMG}: 0.82 ± 0.19 , $n = 10$; $p < 0.05$, t -test), showing that NMDAR activity is enhanced in CA1 neurons of PS1^{ΔMG} mice. These data indicate that PS1^{ΔMG} mice experience a selective increase in CA1 NMDAR activity when measured by ex vivo electrophysiology.

To further investigate the effects of PS1^{ΔMG} on hippocampal activity, we sought to examine neuronal activity in freely behaving mice using fiber photometry (Gunaydin et al., 2014) (Figure

447 4). We injected an AAV-hSyn-GCaMP6s construct into the hippocampus to label CA1 pyramidal
448 neurons and validated the placement of the optic fiber and viral expression (Figure 4A). We then
449 performed peak analysis to determine the number of events per minute (frequency of Ca²⁺
450 transients) (Calipari et al., 2016) and area under the curve was plotted for each event (amplitude
451 of Ca²⁺ transients). Neuronal activity was assessed as the total frequency of events measured
452 from Ca²⁺ transients and compared to control littermates. This effect was observed both in home-
453 cage recordings (Figure 4B and C) and during an open field task (Figure 4D and E). Next, we
454 asked whether microglial PS1^{ΔMG} altered neuronal activity when mice were engaged in object
455 exploration. To analyze the dynamics of CA1 neurons during memory encoding, we recorded CA1
456 neuronal activity in freely moving mice during object exploration. When neuronal activity
457 recordings were time-locked with behavior (nose-to-object exploration), activity was increased, as
458 indicated by greater amplitude of Ca²⁺ transient signals in PS1^{ΔMG} compared to controls (Figure
459 4F and G). Together, our data showed that CA1 neuronal activity increases during object
460 exploration in PS1^{ΔMG} mice, indicating that modulation of microglial PS1 can alter the function of
461 these neurons and increase engagement during an object exploration task.

463 **Depletion of Microglial PS1 improves memory in mice.**

464 Finally, we assessed associative emotional memory through contextual fear conditioning
465 in PS1^{ΔMG} mice (Figure 5). Surprisingly, results revealed increased freezing in PS1^{ΔMG} mice
466 compared to control mice at the 24h retention test (Figure 5B).

467 To further validate our results, we tested PS1^{ΔMG} mice in another memory task. We used
468 a modified version of novel object recognition test, which is used to evaluate memory
469 enhancement. In this task, mice were introduced to 2 objects and allowed to explore for 3 minutes.
470 Typically, this approach does not result in changes in short- or long-term memory in WT mice, as

the 3 minutes training period is insufficient for memory consolidation (Haettig et al., 2011, 2013; McQuown et al., 2011). Memory retention was evaluated 24 hours later by replacing one of the familiar objects with a novel object and allowing 5 minutes for exploration (Figure 5C-E). As expected, neither group showed preference for a specific object during the 3-minute training period (Figure 5D). However, in the 24h retention test, the PS1^{ΔMG} mice showed an increased discrimination index for the novel object (Figure 5E). Distance travelled within arena shows no difference between groups, suggesting the results are not a consequence of hyperactivity or locomotion deficits (Figure 5F). Overall, our data showed that PS1^{ΔMG} mice demonstrate memory enhancement, in opposition to what has been reported for mice lacking neuronal PS1.

Discussion

Collectively, our findings identify microglial PS1 as a regulator of microglial transcriptional state, synaptic function, and learning-related behavior. PS1 deletion induces persistent shifts in microglial populations, including enrichment of Cd52-hi and Cd52-lo clusters characterized by proliferative gene expression. Although this population remains incompletely defined, its predicted association with developmental periods of heightened neuronal plasticity suggests a potential link between microglial state and synaptic maturation. NMDA receptor composition is tightly regulated during development, with GluN2B-dominant receptors in early stages transitioning toward GluN2A-containing receptors as synapses mature (Sheng et al., 1994; Roberts and Ramoa, 1999; Zhang et al., 2012; Paoletti et al., 2013; Jacobs et al., 2015). Whether PS1-dependent microglial states influence this developmental trajectory, and whether Cd52-hi microglia contribute to this process, remains an important question for future investigation.

Transcriptional profiling of active neuronal populations revealed broad remodeling of pathways related to synaptic signaling and metabolism. Although pathway-level analyses capture

global transcriptional shifts rather than specific functional directionality, differential expression patterns suggest reorganization of glutamatergic signaling components in PS1^{ΔMG} mice. These transcriptional changes are accompanied by altered NMDA receptor composition and increased NMDA-mediated synaptic currents in hippocampal CA1 neurons, consistent with enhanced synaptic responsiveness. Because microglia and neurons communicate through bidirectional signaling, we cannot exclude reciprocal neuronal feedback contributing to microglial transcriptional remodeling.

One limitation of this study is the sole use of male mice. Our analyses did not identify sex-specific enrichment of PS1^{ΔMG}-associated clusters. Nonetheless, sex-dependent effects on the mechanisms described here remain possible and warrant further investigation. Another limitation is the implications to the Alzheimer's disease field. Age is the strongest risk factor for the development of Alzheimer's disease, and disease progression is accompanied by a significant change in the brain environment. The microglial transcriptional states we describe in PS1^{ΔMG} mice exist in a healthy young brain environment, which is fundamentally different than the chronic amyloid/Tau and inflammatory burden present in the aged AD brain. Therefore, either the use of aged mice or Alzheimer's disease models is necessary to assess the role of PS1 deletion in microglia in AD context, and this presents another limitation of our study. PS1 deletion in microglia alters expression of genes within the microglial sensome and secretome, supporting a model in which microglial transcriptional changes influence neuronal activity through altered intercellular signaling. Among the most upregulated factors are chemokines *Ccl7* and *Ccl8*, predicted to interact with neuronal CCR5, a receptor known to negatively regulate learning and memory (Zhou et al., 2016; Shen et al., 2022). Because reduced CCR5 signaling enhances neuronal excitability and modulates NMDA receptor function (Musante et al., 2008; Zhou et al., 2016; Shen et al., 2022), altered CCL7-CCR5 signaling provides a plausible pathway linking microglial PS1 deletion

to synaptic and behavioral changes. However, this interaction remains a computational prediction and should be considered a testable hypothesis rather than a demonstrated mechanism.

PS1^{ΔMG} groups are predicted to shift microglial populations, enriching CD52-hi and CD52-lo microglial clusters, while reducing Rrm2-hi proliferative microglia. The Cd52-hi proliferative cluster retains expression of canonical brain-resident microglial homeostatic markers including *P2ry12*, *Hexb*, *Fcrls*, *Cx3cr1*, and *Trem2* (Figure S3A), alongside complement components *C1qb* and *C1qc*, which mediate complement-dependent synaptic pruning (Stevens et al., 2007; Schafer et al., 2012). CD52 expression in myeloid cells has been studied in the context of peripheral macrophage biology, CD52-high monocytes exhibit enhanced phagocytic capacity and cytokine responsiveness (Geng et al., 2025), in T-cells CD52 glycoprotein is often cleaved and when soluble acts as an anti-inflammatory signal (Rashidi et al., 2018). Because transcript abundance does not distinguish membrane-bound to shed CD52, these observations are not mutually exclusive. Based on peripheral myeloid biology, the enrichment of Cd52-hi microglia in PS1^{ΔMG} and its genetic signature predicts an amplification of phagocytic capacity while retaining homeostatic identity.

The Cd52-lo proliferative cluster presents a functionally distinct profile. The cluster is defined by the immediate early response transcription factors *Jun* and *Junb*, NF-κB pathway activation (*Nfkbia*), antigen presentation machinery (*H2-DMa*, *H2-D1*, *B2m*), and myeloid activation genes (*Cd34*, *Lat2*, *Cd14*, *Csf2ra* and *Mafb*). This transcriptional state is consistent with an activated myeloid population, sustaining inflammatory gene expression (Bahrami and Drabløs, 2016; Keren-Shaul et al., 2017; Hammond et al., 2019). The enrichment of AP-1 transcripts in the absence of an overt immunological challenge suggests a non-resolving activation state. Immediate-early gene transcription is a defining feature of Terminally Inflammatory Microglia (TIMs), a terminal population we previously described, which co-express AP-1 and cellular stress markers (Millet et al., 2024). This suggests PS1^{ΔMG} could favor accumulation of TIM-like

immunological exhausted microglia over time, although further work is required to test this relationship.

The Rrm2-hi cluster, which is decreased in PS1^{ΔMG}, is defined exclusively by core DNA replication machinery (*Pcna*, *Mcm2-7*, *Fen1*, *Rrm2*; Figure S3A) and has no immune effector function (Hammond et al., 2019). Its reduction in PS1^{ΔMG} therefore represents a shift away from a transcriptionally quiescent dividing state toward functionally active microglial states with direct capacity to influence their environment.

Absence of PS1 in microglial enhances learning and memory, in contrast to the cognitive impairments observed following global or neuronal PS1 loss. These findings highlight cell type specific functions of PS1 in the regulation of synaptic plasticity and cognition. While neuronal PS1 supports synaptic integrity, our results suggest that microglial PS1 constrains synaptic responsiveness under homeostatic conditions. This distinction underscores the importance of cellular context when interpreting PS1 function. Selective modulation of microglial PS1 signaling may represent a more targeted strategy to influence synaptic function.

More broadly, these findings support a model in which microglial transcriptional state regulates neuronal excitability through soluble signaling mechanisms. Under physiological conditions, microglial PS1 appears to limit this influence, maintaining synaptic stability. Its loss shifts microglia toward a signaling state that enhances NMDA-dependent synaptic activity and improves memory performance. Notably, this heightened synaptic responsiveness is accompanied by transcriptional enrichment of metabolic pathways in active neurons, suggesting increased energetic demand. Given the substantial metabolic cost associated with sustained excitatory synaptic transmission, these findings raise the possibility that microglial PS1 functions as a regulatory constraint that helps balance synaptic activity with metabolic capacity. Together, our results reinforce the emerging view that microglia are active regulators of neuronal circuit function, and that modulation of microglial transcriptional programs alone is sufficient to influence

cognition. Targeting microglial regulatory pathways may therefore offer new approaches to enhance synaptic resilience.

Author Contribution

J.H.L., B.S. and Paul Greengard conceived; J.H.L. and B.S., supervised this study; J.H.L. and B.S. designed experiments; P.S.F., C.A.M., J.M., J.C., Y.K., A.M., J.H.L., E.P.A and H.M.S. performed and analyzed experiments; P.S.F., C.A.M., J.M. and J.H.L. prepared figures. P.S.F., C.A.M., J.M., J.H.L. and B.S. wrote the paper.

Acknowledgments

This work was supported by funds received from Fisher Center for Alzheimer's Research Foundation and Cure Alzheimer's Fund. J.H.L. is supported by the Alzheimer's Association. We thank Rada Norinsky and the team of the Transgenic and Reproductive Services Center at the Rockefeller University for their valuable assistance with IVF experiments.

Conflict of interest

E.P.A is a co-founder at Kaidos, Inc. J.H.L is a co-founder at AzoraX, Inc. The other authors declare no conflict of interest.

Figure Legends

Figure 1. Deconvolution Analysis of bulk RNA sequencing into single-cell RNA seq of

PS1^{ΔMG} in microglia. (A) Schematic diagram of microglial isolations for bulk sequencing and deconvolution analysis into single-cell RNAseq. Dataset used in deconvolution was a reanalysis of Hammond, T.R. et al 2019. (B) UMAP of microglial populations in single-cell RNAseq, reanalyzed from Hammond, T.R. et al 2019. (C) UMAP showing cluster representation detected versus non-detected in bulk RNAseq superimposed to Hammond, T. R. et al 2019 single-cell RNAseq. (D) Volcano plot of differentially expressed genes between microglia of PS1^{ΔMG} and control mice. Log-2-fold change in expression is plotted on the x-axis while the log-10 adjusted p-value is plotted on the y-axis. (E) Estimated enrichment score of microglial populations in bulk RNA sequencing. Score is a composite function that reflects both the size (number of cells) and the relative frequency of the cluster between PS1^{ΔMG} and control mice. (F-H) Lollipop plots of significantly differentially expressed genes annotated in microglial sensome, (F) microglial secretome (G) and cytokine and inflammatory pathways (H). Normalized expression is computed as the mean counts of the gene in PS1^{ΔMG} samples divided by the mean counts of the gene in control samples.

Figure 2. PS1^{ΔMG} affects active neuronal ensembles gene expression. (A) Analysis of predicted microglial-neuronal ligand-receptor interactions. Ligands were selected as the most differentially expressed genes between PS1^{ΔMG} and control animals identified in the microglial secretome (Diaz-Aparicio et al., 2020). Receptors were selected first by identifying putative receptors for microglial ligands and then by filtering to genes expressed in hippocampal scRNA-seq from the Allen Brain Atlas. Ligands are colored by the log-2 value of the average expression of the gene across both control and PS1^{ΔMG}, while receptors are colored by the percentage of

cells in which at least one read of the gene was detected. The width of arrows indicating pairwise interactions is defined as the log-2-fold change of the gene between PS1^{ΔMG} and control. (B) Heatmap of Z-scores of genes expressed by neurons pulled from active ensembles in both PS1^{ΔMG} and control. Highly differential genes are annotated at right, with genes associated with immunity and inflammation highlighted in orange text. (C) Lollipop plot of pathway analysis of pTRAP data from control and PS1^{ΔMG} neurons. The top ten pathways enriched in each group as defined by normalized enrichment score as computed by fGSEA are shown. Because pTRAP quantifies transcripts enriched in active neurons relative to the total tissue transcriptome (IP / Input), pathway enrichment reflects the transcriptional composition of activated neuronal populations rather than absolute differences in signaling between control and PS1^{ΔMG} groups.

Figure 3: Loss of microglial PS1 significantly alters NMDAR protein levels and activity.

Synaptosome-enriched fractions from control and PS1^{ΔMG} CA1 brains probed with indicated antibodies by Western blot. Protein levels of (A) presynaptic proteins VGlut1 and Synaptophysin, (B) postsynaptic protein PSD-95, (C) AMPAR subunits GluA1, (D) GluA2, and (E) NMDAR subunits GluN2B and GluN1 from synaptosome-enriched extractions from PS1^{ΔMG} and control. Densitometric quantification of the western blots **p < 0.01 using unpaired two-tail student's t-test. Each graph bar represents ± SEM. (F) Representative traces and histogram showing mean of the NMDAR- to AMPAR-EPSC ratio of control versus PS1^{ΔMG} mouse. Scale bar, 50 pA, 100 ms. *p < 0.05 using unpaired student's t-test. N=10-11 neurons per group, 3 mice per group. Each graph bar represents ± SEM.

Figure 4. PS1^{ΔMG} Drives Hippocampal Hyperactivity. (A) Enhanced neuronal activity in freely behaving mice lacking microglial PS1. Left panel, experimental scheme representing fiber

placement in the mouse brain. Mice were attached to a fiber photometry setup (Doric) through patch cords and GCaMP6s was excited with a 473 nm LED (green) and a 405 nm LED (blue). Middle and right panels, experimental scheme and representative images showing AAV5-hSyn-GCaMP6s expression in mouse CA1 hippocampus. (B, C) PS1^{ΔMG} mice show enhanced neuronal activity during baseline and (D, E) open field exploration. Neuronal activity recording at baseline or during open field test were performed for 5 min in control and PS1^{ΔMG} mice. Representative Ca²⁺ traces (represented as the percent change in fluorescence over the mean fluorescence [ΔF/F]) for each group is shown in the middle panels. Peak analysis of Ca²⁺ traces. PS1^{ΔMG} in increased events / min (left panel). Histogram showing area under the curve (AUC) measurement for each animal trial (right panel). (F) Neuronal activity was recorded during novel object exploration. (G) Left panel average GCaMP6s z-score (black line, control; gray line, PS1^{ΔMG}) across all recording sites time-locked to nose-to-object touch, dotted lines represent standard error (SE). Right panel graph showing area under the curve (AUC) measurement for each nose-to-object time-locked Ca²⁺ transient for each animal trial. Data are represented as mean ± SEM. N=4-6 mice per group *p<0.05 using unpaired student's *t*-test.

Figure 5. Enhanced memory in mice lacking microglial PS1. (A) Schematic of contextual fear conditioning (B) Mice were trained in contextual fear conditioning and tested 24h after training. Compared with control mice, PS1^{ΔMG} mice showed increased freezing. Data are represented as mean ± SEM, n=12 mice per group. ***p < 0.001, unpaired student's *t*-test. (C) Schematic of object exploration task used for both PS1^{ΔMG} and control. (D) Exploratory preference in the training session. A discrimination index of 0.5 (dashed lines) represents performance at chance (50%). The amount of time spent exploring both objects was the same for all mice. We used a 3-min training protocol (see Methods) as this does not typically lead to changes in short-term memory or long-term memory in wild-type mice. (E) Exploratory preference in control and PS1^{ΔMG} mice at

24-hour retention test. Data are represented as mean $\pm$ SEM, $n \geq 6$ -10 mice per group. **** $p < 0.0001$ using unpaired Student's t -test. (F) Distance travelled in open field control vs PS1 ^{Δ MG} mice.

References

- Ayata P et al. (2025) Lymphoid gene expression supports neuroprotective microglia function. *Nature* 648:157–165.
- Badimon A et al. (2020) Negative feedback control of neuronal activity by microglia. *Nature*:1–7.
- Bahrami S, Drabløs F (2016) Gene regulation in the immediate-early response process. *Adv Biol Regul* 62:37–49.
- Bustos V, Pulina MV, Bispo A, Lam A, Flajolet M, Gorelick FS, Greengard P (2017a) Phosphorylated Presenilin 1 decreases β -amyloid by facilitating autophagosome-lysosome fusion. *Proc National Acad Sci* 114:201705240.
- Bustos V, Pulina MV, Kelahmetoglu Y, Sinha SC, Gorelick FS, Flajolet M, Greengard P (2017b) Bidirectional regulation of A β levels by Presenilin 1. *P Natl Acad Sci Usa* 114:201705235.
- Cai Y, Sun Z, Jia H, Luo H, Ye X, Wu Q, Xiong Y, Zhang W, Wan J (2017) Rpph1 Upregulates CDC42 Expression and Promotes Hippocampal Neuron Dendritic Spine Formation by Competing with miR-330-5p. *Front Mol Neurosci* 10:27.
- Calipari ES, Bagot RC, Purushothaman I, Davidson TJ, Yorgason JT, Peña CJ, Walker DM, Pirpinias ST, Guise KG, Ramakrishnan C, Deisseroth K, Nestler EJ (2016) In vivo imaging identifies temporal signature of D1 and D2 medium spiny neurons in cocaine reward. *P Natl Acad Sci Usa* 113:2726–2731.
- Cheng J, Xiong Z, Duffney LJ, Wei J, Liu A, Liu S, Chen G-J, Yan Z (2014) Methylphenidate exerts dose-dependent effects on glutamate receptors and behaviors. *Biol Psychiat* 76:953–962.
- Diaz-Aparicio I et al. (2020) Microglia Actively Remodel Adult Hippocampal Neurogenesis through the Phagocytosis Secretome. *J Neurosci* 40:1453–1482.
- Favuzzi E, Huang S, Saldi GA, Binan L, Ibrahim LA, Fernández-Otero M, Cao Y, Zeine A, Sefah A, Zheng K, Xu Q, Khlestova E, Farhi SL, Bonneau R, Datta SR, Stevens B, Fishell G (2021) GABA-receptive microglia selectively sculpt developing inhibitory circuits. *Cell* 184:5686.
- Frost GR, Wong E, Li Y-M (2017) Versatility of presenilin 1. *P Natl Acad Sci Usa* 114:6885–6887.
- Garré JM, Silva HM, Lafaille JJ, Yang G (2017) CX3CR1⁺ monocytes modulate learning and learning-dependent dendritic spine remodeling via TNF- α . *Nat Med* 23:714–722.
- Geng A, Robert GB, Julien R, Mechthild L, Hung-Wei C, Emilio E. F, Paul OGL, et al. 2025. “Circulating Monocytes Upregulate CD52 and Sustain Innate Immune Function in Cirrhosis Unless Acute Decompensation Emerges.” *Journal of Hepatology* 83 (1): 146–60.

719 Glebov K, Wunderlich P, Karaca I, Walter J (2016) Functional involvement of γ -secretase in
720 signaling of the triggering receptor expressed on myeloid cells-2 (TREM2). *J Neuroinflamm*
721 13:17.

722 Gunaydin LA, Grosenick L, Finkelstein JC, Kauvar IV, Fenno LE, Adhikari A, Lammel S,
723 Mirzabekov JJ, Airan RD, Zalocusky KA, Tye KM, Anikeeva P, Malenka RC, Deisseroth K
724 (2014) Natural neural projection dynamics underlying social behavior. *Cell* 157:1535–1551.

725 Güner G, Lichtenthaler SF (2020) The substrate repertoire of γ -secretase/presenilin. *Semin Cell*
726 *Dev Biol*.

727 Haapasalo A, Kovacs DM (2011) The many substrates of presenilin/ γ -secretase. *Journal of*
728 *Alzheimer's disease : JAD* 25:3–28.

729 Haettig J, Stefanko DP, Multani ML, Figueroa DX, McQuown SC, Wood MA (2011) HDAC
730 inhibition modulates hippocampus-dependent long-term memory for object location in a
731 CBP-dependent manner. *Learn Mem Cold Spring Harb N Y* 18:71–79.

732 Haettig J, Sun Y, Wood MA, Xu X (2013) Cell-type specific inactivation of hippocampal CA1
733 disrupts location-dependent object recognition in the mouse. *Learn Mem Cold Spring Harb N*
734 *Y* 20:139–146.

735 Hammond TR, Dufort C, Dissing-Olesen L, Giera S, Young A, Wysoker A, Walker AJ, Gergits F,
736 Segel M, Nemesh J, Marsh SE, Saunders A, Macosko E, Ginhoux F, Chen J, Franklin RJM,
737 Piao X, McCarroll SA, Stevens B (2018) Single-Cell RNA Sequencing of Microglia
738 throughout the Mouse Lifespan and in the Injured Brain Reveals Complex Cell-State
739 Changes. *Immunity* 50:253-271.e6.

740 Hickman SE, Kingery ND, Ohsumi TK, Borowsky ML, Wang L, Means TK, Khoury JE (2013)
741 The microglial sensome revealed by direct RNA sequencing. *Nat Neurosci* 16:1896–1905.

742 Hong S, Beja-Glasser VF, Nfonoyim BM, Frouin A, Li S, Ramakrishnan S, Merry KM, Shi Q,
743 Rosenthal A, Barres BA, Lemere CA, Selkoe DJ, Stevens B (2016) Complement and
744 microglia mediate early synapse loss in Alzheimer mouse models. *Science* 352:712–716.

745 Jacobs S, Wei W, Wang D, Tsien JZ (2015) Importance of the GluN2B carboxy-terminal domain
746 for enhancement of social memories. *Learn Mem Cold Spring Harb N Y* 22:401–410.

747 Keren-Shaul H, Spinrad A, Weiner A, Matcovitch-Natan O, Dvir-Szternfeld R, Ulland TK, David
748 E, Baruch K, Lara-Astaiso D, Toth B, Itzkovitz S, Colonna M, Schwartz M, Amit I (2017)
749 A Unique Microglia Type Associated with Restricting Development of Alzheimer's
750 Disease. *Cell* 169:1276-1290.e17.

751 Knight ZA, Tan K, Birsoy K, Schmidt S, Garrison JL, Wysocki RW, Emiliano A, Ekstrand MI,
752 Friedman JM (2012) Molecular profiling of activated neurons by phosphorylated ribosome
753 capture. *Cell* 151:1126–1137.

754 Ledo JH, Liebmann T, Zhang R, Chang JC, Azevedo EP, Wong E, Silva HM, Troyanskaya OG,
755 Bustos V, Greengard P (2020a) Presenilin 1 phosphorylation regulates amyloid- β
756 degradation by microglia. *Mol Psychiatr*:1–16.

757 Ledo JH, Zhang R, Mesin L, Mourão-Sá D, Azevedo EP, Troyanskaya OG, Bustos V,
758 Greengard P (2020b) Lack of a site-specific phosphorylation of Presenilin 1 disrupts
759 microglial gene networks and progenitors during development. *Plos One* 15:e0237773.

760 Lehrman EK, Wilton DK, Litvina EY, Welsh CA, Chang ST, Frouin A, Walker AJ, Heller MD,
761 Umemori H, Chen C, Stevens B (2018) CD47 Protects Synapses from Excess Microglia-
762 Mediated Pruning during Development. *Neuron* 100:120-134.e6.

763 Liu W, Han F, Qu S, Yao Y, Zhao J, Akhtar ML, Ci Y, Zhang H, Li H, Zhao Y, Yue L, Zhang Y,
764 Wang C, Li Y (2018) MARVELD1 depletion leads to dysfunction of motor and cognition via
765 regulating glia-dependent neuronal migration during brain development. *Cell Death Dis*
766 9:999.

767 Londino JD, Gulick D, Isenberg JS, Mallampalli RK (2015) Cleavage of Signal Regulatory
768 Protein α (SIRP α) Enhances Inflammatory Signaling. *J Biological Chem* 290:31113–31125.

769 Marjaux E, Hartmann D, Strooper BD (2004) Presenilins in Memory, Alzheimer's Disease, and
770 Therapy. *Neuron* 42:189–192.

771 Millet A, Ledo JH, Tavazoie SF (2024) An exhausted-like microglial population accumulates in
772 aged and APOE4 genotype Alzheimer's brains. *Immunity* 57:153-170.e6.

773 McQuown SC, Barrett RM, Matheos DP, Post RJ, Rogge GA, Alenghat T, Mullican SE, Jones
774 S, Rusche JR, Lazar MA, Wood MA (2011) HDAC3 is a critical negative regulator of long-
775 term memory formation. *J Neurosci Official J Soc Neurosci* 31:764–774.

776 Moon IS, Cho S-J, Seog D-H, Walikonis R (2009) Neuronal activation increases the density of
777 eukaryotic translation initiation factor 4E mRNA clusters in dendrites of cultured hippocampal
778 neurons. *Exp Mol Med* 41:601–610.

779 Musante V, Longordo F, Neri E, Pedrazzi M, Kalfas F, Severi P, Raiteri M, Pittaluga A (2008)
780 RANTES Modulates the Release of Glutamate in Human Neocortex. *J Neurosci* 28:12231–
781 12240.

782 Paoletti P, Bellone C, Zhou Q (2013) NMDA receptor subunit diversity: impact on receptor
783 properties, synaptic plasticity and disease. *Nat Rev Neurosci* 14:383–400.

784 Parkhurst CN, Yang G, Ninan I, Savas JN, Yates JR, Lafaille JJ, Hempstead BL, Littman DR,
785 Gan W-B (2013) Microglia promote learning-dependent synapse formation through brain-
786 derived neurotrophic factor. *Cell* 155:1596–1609.

787 Rashidi M, Bandala-Sanchez E, Lawlor KE, Zhang Y, Neale AM, Vijayaraj SL, O'Donoghue R,
788 Wentworth JM, Adams TE, Vince JE, Harrison LC (2018) CD52 inhibits Toll-like receptor

789 activation of NF- κ B and triggers apoptosis to suppress inflammation. *Cell Death Differ*
790 25:392–405.

791 Rasool D, Jahani-Asl A (2024) Master regulators of neurogenesis: the dynamic roles of Ephrin
792 receptors across diverse cellular niches. *Transl Psychiatry* 14:462.

793 Roberts EB, Ramoa AS (1999) Enhanced NR2A Subunit Expression and Decreased NMDA
794 Receptor Decay Time at the Onset of Ocular Dominance Plasticity in the Ferret. *J*
795 *Neurophysiol* 81:2587–2591.

796 Saura CA, Choi S-Y, Beglopoulos V, Malkani S, Zhang D, Rao BSS, Chattarji S, Kelleher RJ,
797 Kandel ER, Duff K, Kirkwood A, Shen J (2004) Loss of Presenilin Function Causes
798 Impairments of Memory and Synaptic Plasticity Followed by Age-Dependent
799 Neurodegeneration. *Neuron* 42:23–36.

800 Schafer DP, Lehrman EK, Kautzman AG, Koyama R, Mardinly AR, Yamasaki R, Ransohoff RM,
801 Greenberg ME, Barres BA, Stevens B (2012) Microglia sculpt postnatal neural circuits in an
802 activity and complement-dependent manner. *Neuron* 74:691–705.

803 Schmittgen TD, Livak KJ (2008) Analyzing real-time PCR data by the comparative C(T) method.
804 *Nature protocols* 3:1101–1108.

805 Shen Y et al. (2022) CCR5 closes the temporal window for memory linking. *Nature* 606:146–
806 152.

807 Sheng M, Cummings J, Roldan LA, Jan YN, Jan LY (1994) Changing subunit composition of
808 heteromeric NMDA receptors during development of rat cortex. *Nature* 368:144–147.

809 Stefanko DP, Barrett RM, Ly AR, Reolon GK, Wood MA (2009) Modulation of long-term memory
810 for object recognition via HDAC inhibition. *P Natl Acad Sci Usa* 106:9447–9452.

811 Steiner A, Schlepckow K, Brunner B, Steiner H, Haass C, Hagn F (2020) γ -Secretase cleavage
812 of the Alzheimer risk factor TREM2 is determined by its intrinsic structural dynamics. *Embo J*
813 39:e104247.

814 Stevens B, Allen NJ, Vazquez LE, Howell GR, Christopherson KS, Nouri N, Micheva KD,
815 Mehalow AK, Huberman AD, Stafford B, Sher A, Litke AM, Lambris JD, Smith SJ, John
816 SWM, Barres BA (2007) The Classical Complement Cascade Mediates CNS Synapse
817 Elimination. *Cell* 131:1164–1178.

818 Tang YP, Shimizu E, Dube GR, Rampon C, Kerchner GA, Zhuo M, Liu G, Tsien JZ (1999)
819 Genetic enhancement of learning and memory in mice. *Nature* 401:63–69.

820 Wang Y, Chan SL, Miele L, Yao PJ, Mackes J, Ingram DK, Mattson MP, Furukawa K (2004)
821 Involvement of Notch signaling in hippocampal synaptic plasticity. *Proc Natl Acad Sci*
822 101:9458–9462.

823 Watanabe H, Xia D, Kanekiyo T, Kelleher RJ, Shen J (2012) Familial frontotemporal dementia-
824 associated presenilin-1 c.548G>T mutation causes decreased mRNA expression and
825 reduced presenilin function in knock-in mice. *J Neurosci Official J Soc Neurosci* 32:5085–
826 5096.

827 Wines-Samuelson M, Schulte EC, Smith MJ, Aoki C, Liu X, Kelleher RJ, Shen J (2010)
828 Characterization of age-dependent and progressive cortical neuronal degeneration in
829 presenilin conditional mutant mice. *Plos One* 5:e10195.

830 Yao Z et al. (2023) A high-resolution transcriptomic and spatial atlas of cell types in the whole
831 mouse brain. *Nature* 624:317–332.

832 Yu H, Saura CA, Choi SY, Sun LD, Yang X, Handler M, Kawarabayashi T, Younkin L, Fedeles
833 B, Wilson MA, Younkin S, Kandel ER, Kirkwood A, Shen J (2001) APP processing and
834 synaptic plasticity in presenilin-1 conditional knockout mice. *Neuron* 31:713–726.

835 Zhan Y, Paolicelli RC, Sforazzini F, Weinhard L, Bolasco G, Pagani F, Vyssotski AL, Bifone A,
836 Gozzi A, Ragozzino D, Gross CT (2014) Deficient neuron-microglia signaling results in
837 impaired functional brain connectivity and social behavior. *Nat Neurosci* 17:400–406.

838 Zhang Y, Chen K, Sloan SA, Bennett ML, Scholze AR, O’Keeffe S, Phatnani HP, Guarnieri P,
839 Caneda C, Ruderisch N, Deng S, Liddelw SA, Zhang C, Daneman R, Maniatis T, Barres
840 BA, Wu JQ (2014) An RNA-sequencing transcriptome and splicing database of glia, neurons,
841 and vascular cells of the cerebral cortex. *J Neurosci Official J Soc Neurosci* 34:11929–
842 11947.

843 Zhang Z, Peterson M, Liu H (2012) Essential role of postsynaptic NMDA receptors in
844 developmental refinement of excitatory synapses. *P Natl Acad Sci Usa* 110:1095–1100.

845 Zhao Y, Li X, Huang T, Jiang L-L, Tan Z, Zhang M, Cheng IH-J, Wang X, Bu G, Zhang Y-W,
846 Wang Q, Xu H (2017) Intracellular trafficking of TREM2 is regulated by presenilin 1. *Exp Mol*
847 *Medicine* 49:e405–e405.

848 Zhou M, Greenhill S, Huang S, Silva TK, Sano Y, Wu S, Cai Y, Nagaoka Y, Sehgal M, Cai DJ,
849 Lee Y-S, Fox K, Silva AJ (2016) CCR5 is a suppressor for cortical plasticity and hippocampal
850 learning and memory. *eLife* 5:e20985.

Figure 1

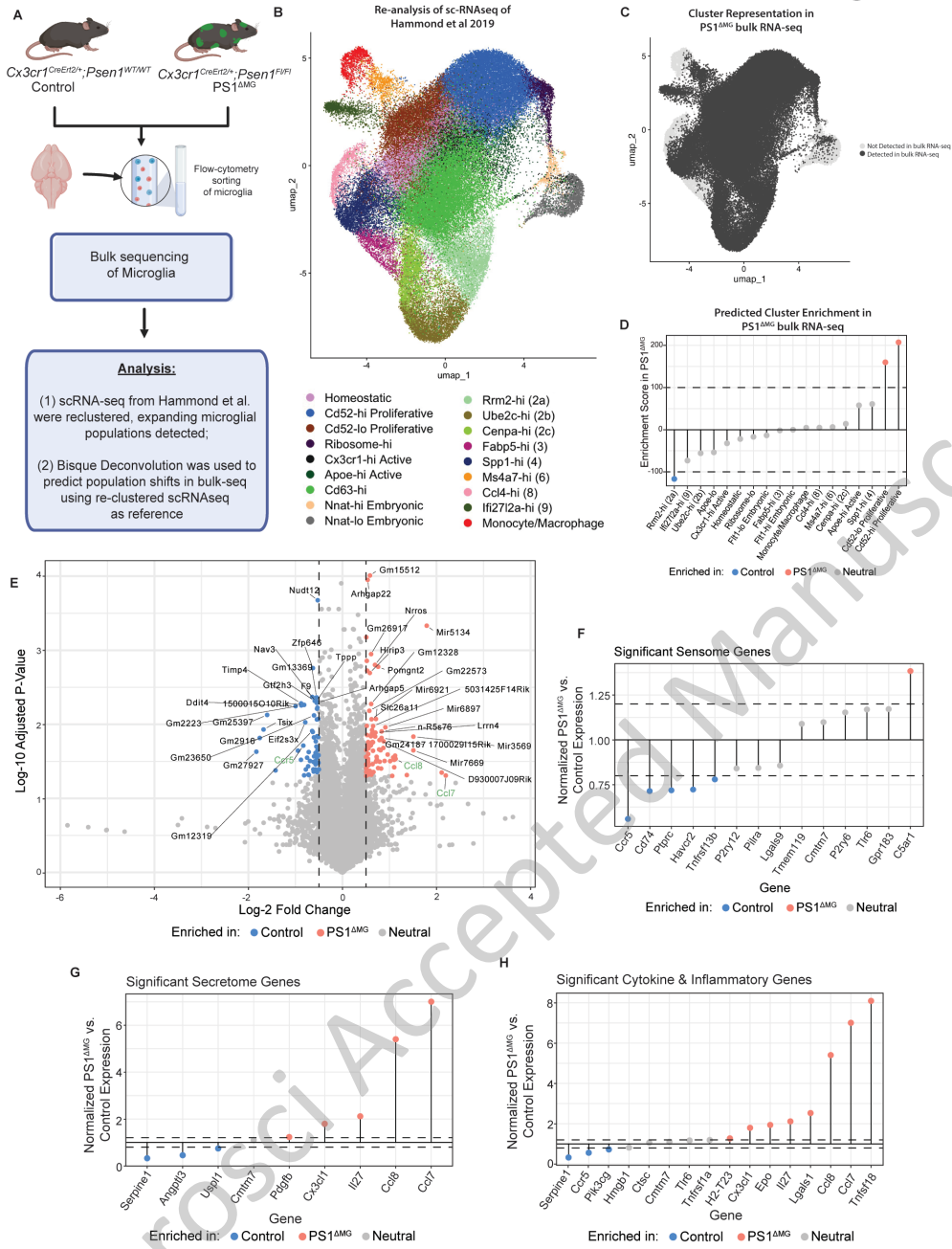


Figure 2

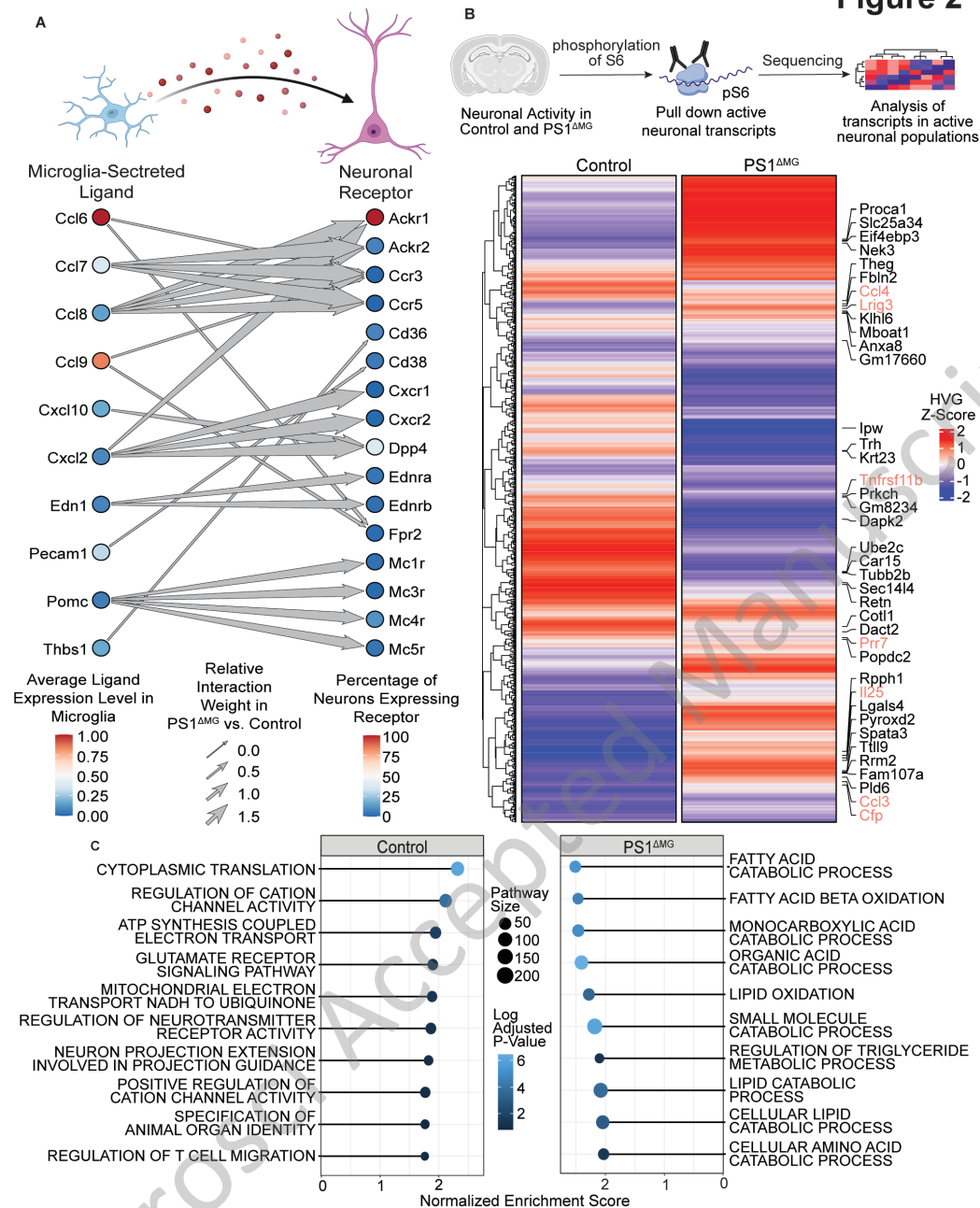


Figure 3

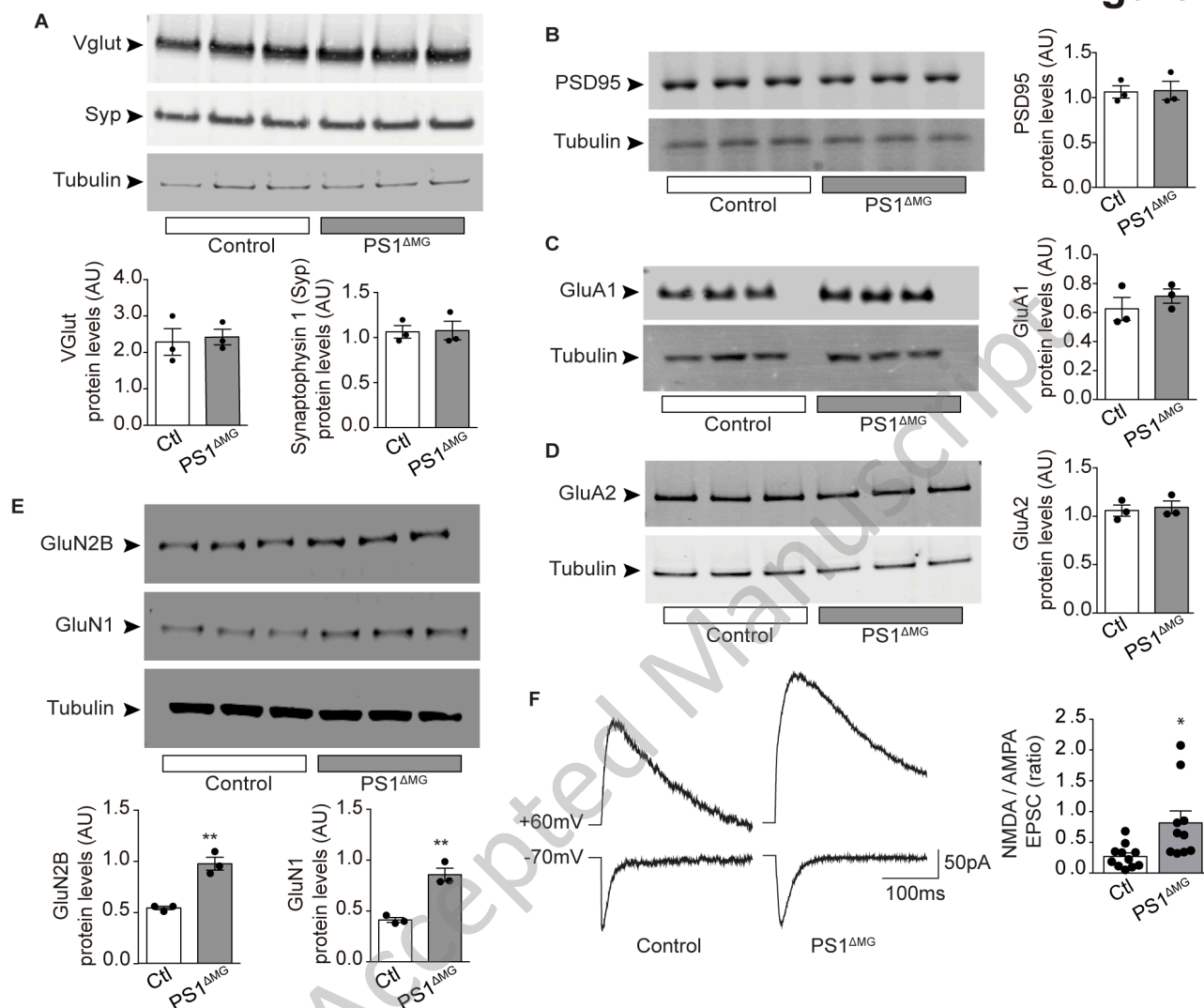


Figure 4

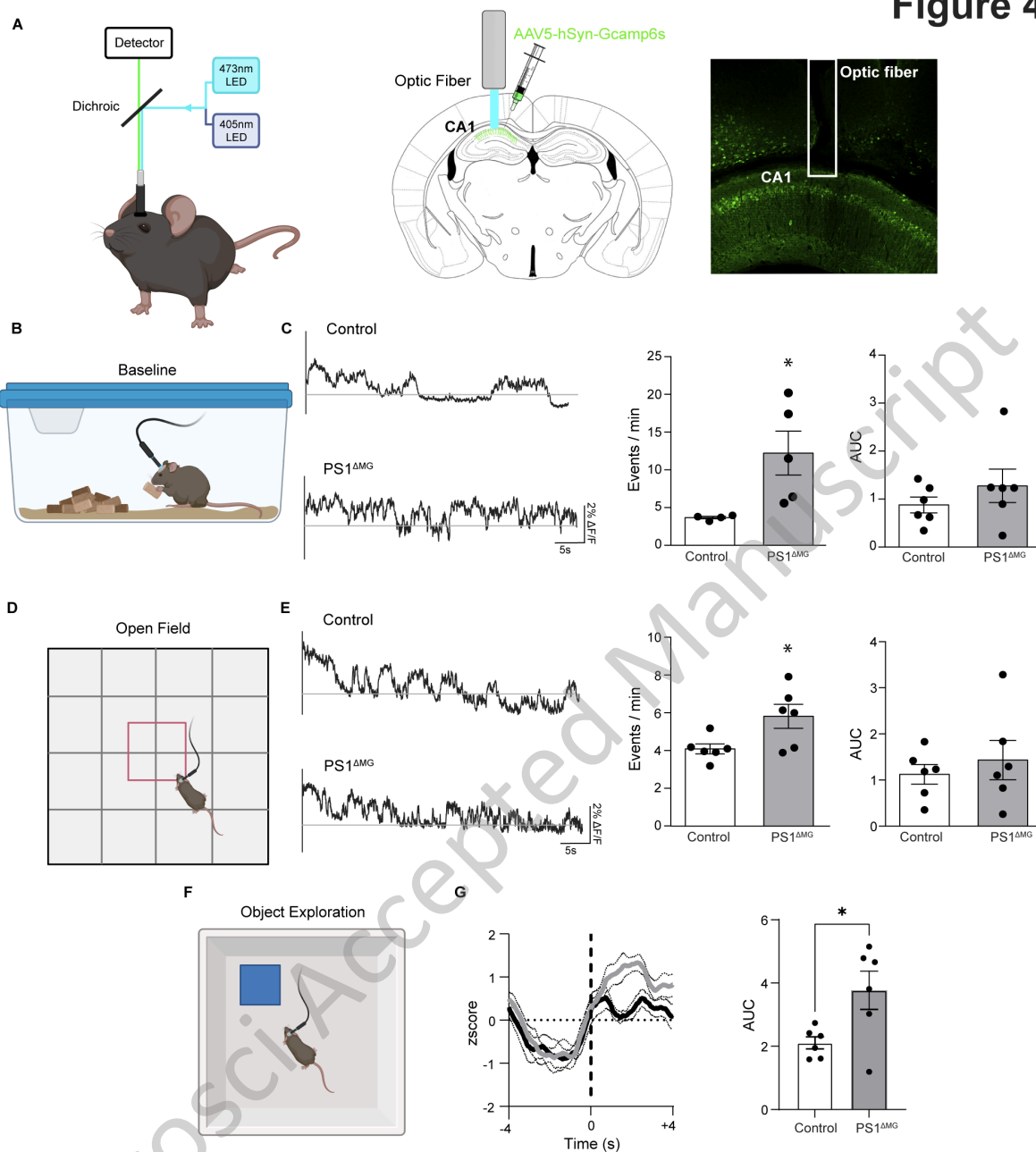


Figure 5

