

Supplementary information

Orchestrated experience-driven *Arc/Arg3.1* responses are disrupted in a mouse model of Alzheimer's disease

Nikita Rudinskiy¹; Jonathan M. Hawkes^{1,2}; Rebecca A. Betensky³; Megumi Eguchi⁴; Shun Yamaguchi⁴; Tara L. Spires-Jones¹; Bradley T. Hyman^{1*}

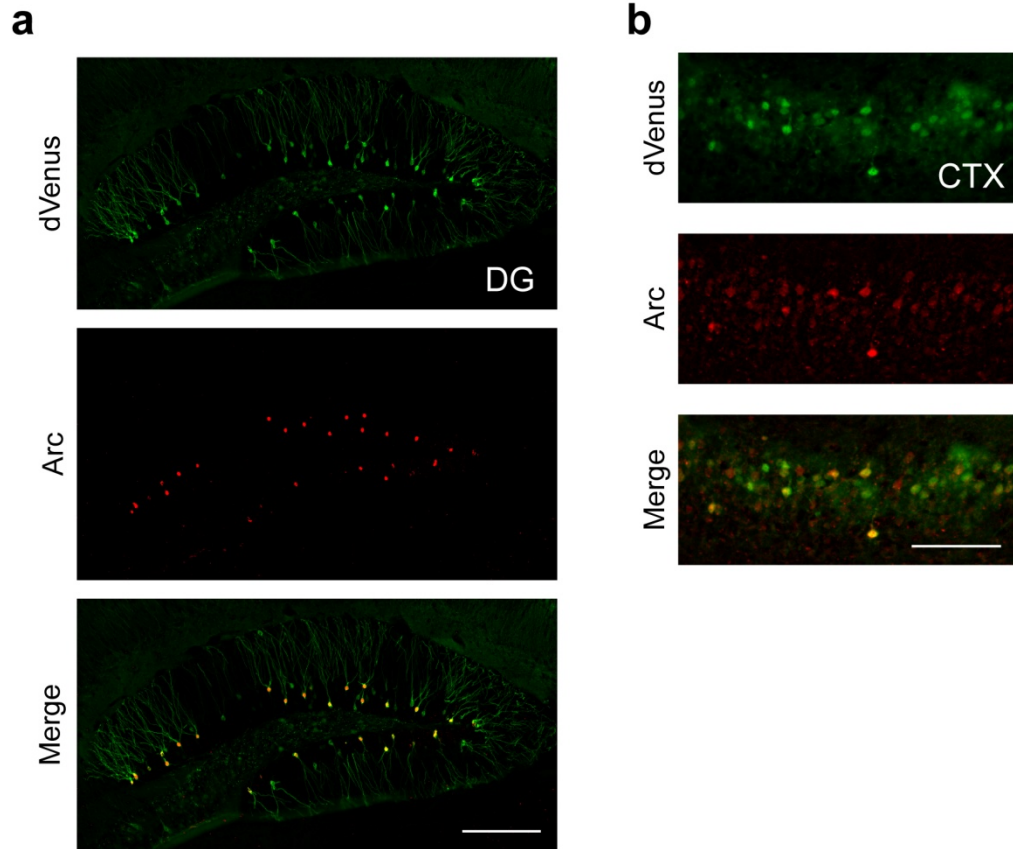
¹Alzheimer's Disease Research Laboratory, Department of Neurology, MassGeneral Institute for Neurodegenerative Disease, Massachusetts General Hospital / Harvard Medical School, Charlestown, Massachusetts 02129, USA. ²Behavioral Neuroscience Program, Northeastern University, Boston, Massachusetts 02115, USA. ³Department of Biostatistics, Harvard School of Public Health, Boston, Massachusetts 02115, USA. ⁴Division of Morphological Neuroscience, Gifu University Graduate School of Medicine, Gifu 501-1194, Japan.

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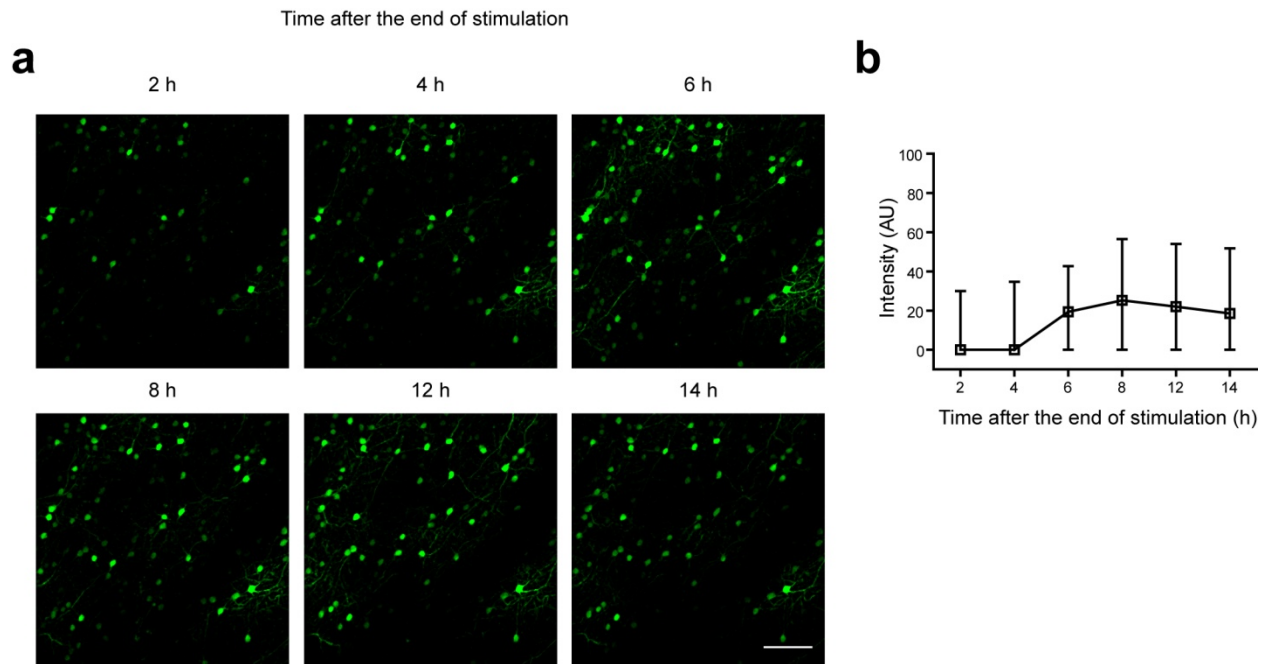
Supplementary Table 1

* To whom correspondence should be addressed. E-mail: bhyman@partners.org



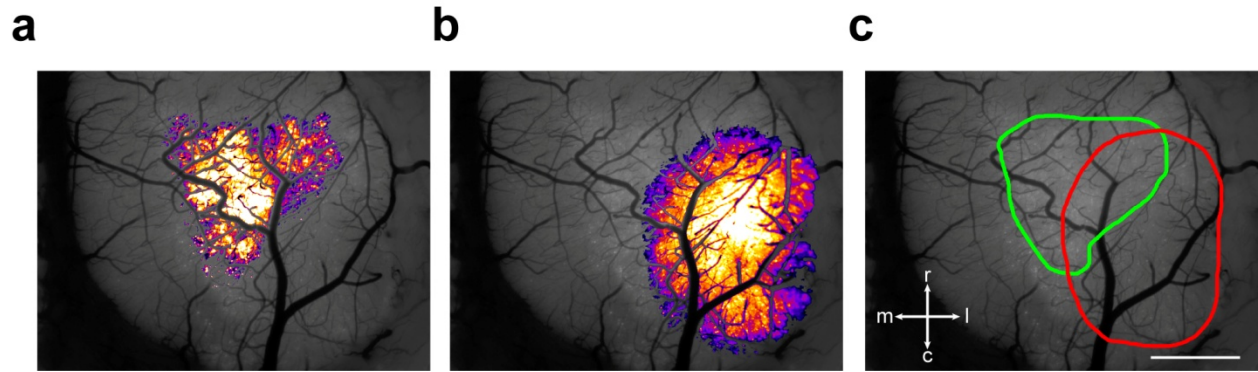
Supplementary Figure 1. Arc protein co-localizes with dVenus.

Immunohistochemical staining for Arc and dVenus in different brain regions of *Arc::dVenus* mice sacrificed two hours after the end of visual stimulation. Time point of two hours was chosen as having sufficient levels of both Arc and dVenus proteins (Supplementary Fig. 4, ¹⁰). Green – dVenus, red – Arc. **(a)** Dentate gyrus region of hippocampal formation. Scale bar = 500 μm . **(b)** Neuronal layer II/III of medial extrastriate cortex. Scale bar = 200 μm . Occasional lack of colocalization of Arc::dVenus with endogenous Arc protein is expected due to different dynamics of *Arc::dVenus* and *Arc* mRNA expression in response to a stimulus as reported in ¹⁰.



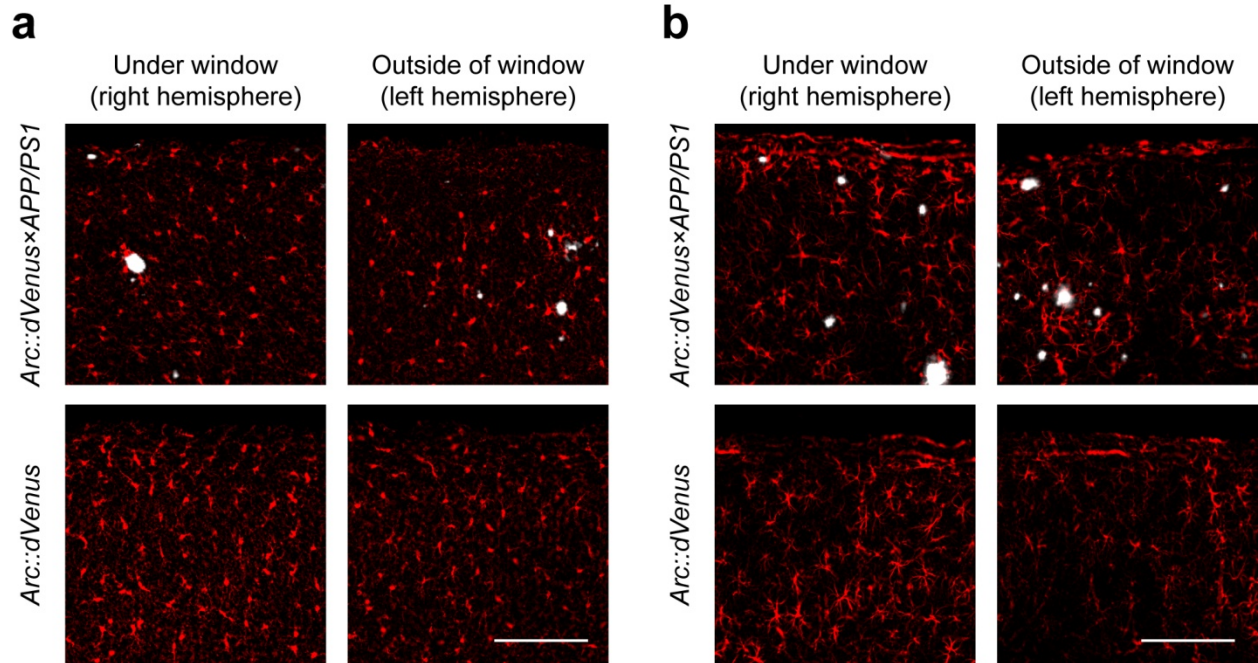
Supplementary Figure 2. Time course of Arc::dVenus expression in the visual cortex following 1-hour structured visual stimulation in the cylinder with vertical stripes.

(a) Maximum intensity projections of *in vivo* image stacks of the same region of medial extrastriate visual cortex imaged at several time points after the end of stimulation. Scale bar = 100 μ m. (b) Dynamics of Arc::dVenus fluorescence intensity in individual neurons over time. Data presented as medians with interquartile ranges. N = 4 mice, 2060 neurons.



Supplementary Figure 3. Low-resolution *in vivo* overview image of Arc::dVenus fluorescence in mouse right visual cortex.

Arc::dVenus fluorescence was imaged 6 hours after standard 1-hour visual stimulation in an illuminated cylinder with vertical stripes (**a**) and after 7 hours of continuous visual stimulation with white light (**b**). In (**a**) Arc::dVenus signal is localized to the medial aspect of visual cortex, while in (**b**) it is concentrated more laterally. (**c**) Superimposed outlines of bright areas from (**b**) and (**c**). Scale bar = 1000 μm .



Supplementary Figure 4. Cranial window implantation causes no overt inflammatory response in the cortex of *Arc::dVenus* and *Arc::dVenus* \times *APP/PS1* mice.

(**a–b**) Immunohistochemical staining for microglia marker Iba1 (**a**) and activated astrocytes marker GFAP (**b**) of medial extrastriate visual cortex (layer II/III) of *Arc::dVenus* \times *APP/PS1* mouse and *Arc::dVenus* control littermate sacrificed 3 weeks after implantation of a cranial window. Left panels – right medial extrastriate visual area (covered by cranial window); right panels – left medial extrastriate visual area (contralateral to cranial window). Red – Iba1 (**a**) / GFAP (**b**) ; white – amyloid plaques stained with methoxy-X04. Scale bar = 300 μ m.

| Reference | Species / model | Brain region / cell type | AD-related pathology | Arc induction | Arc ↑ or ↓ in AD-related pathology | Protein/RNA |
|-------------------|--------------------------------|--------------------------------|---|--------------------------------|------------------------------------|-----------------------------------|
| ⁴⁵ | Ms brain | Hippocampus | Aged Tg2576 | Fear conditioning and baseline | n.c. | protein |
| ³³ | Hu brain ; ms neuronal culture | MFC; cortical primary cultures | Advanced AD; synthetic A β dimers | No | ↑ | protein |
| ⁴⁶ | Ms brain | Whole brain | Aged CRND8 | Basal and running wheel | n.c. | protein |
| ³⁹ | Ms brain | Hippocampus | Aged rTg4510 (tangles) | Env. enrichment | ↓ | RNA |
| ³¹ | Ms brain | Hippocampus | Young (preplaque) Tg2576 and APP/Lo | Basal, fear cond., Morris WM | ↑ | protein |
| ⁴³ | Ms brain | Hippocampus and neocortex | Young and aged APP/PS1, APPDutch, and APP23 | Env. enrichment | ↓ | protein |
| ²⁹ | Ms brain | Hippocampus and cortex | Young (preplaque) APP/PS1 | No | ↑ | Reporter under Arc promoter (RNA) |
| ^{35, 42} | Rat neuronal culture | Cortical primary cultures | Synthetic A β | BDNF | ↓ | protein |
| ³⁴ | Ms brain | Hippocampus | 4-7 mo hAPP-J20 | no | both ↑ and ↓ | protein |
| ³⁸ | Ms neuronal culture | Cortical primary cultures | Synthetic A β | BDNF | ↓ | protein |
| ⁴⁴ | Ms brain | Hippocampus and cortex | Young (preplaque) hAPP(low) | Env. enrichment | n.c. | protein |
| ⁴¹ | Ms brain | Hippocampus and cortex | Young (preplaque) hAPP(FAD) | Basal and env. enrichment | ↓ in DG, n.c. elsewhere | protein, RNA |
| ³² | Rat brain | Hippocampus | LPS infusion | Basal and env. enrichment | ↑ | protein, RNA |
| ³⁰ | Rat neuronal culture | Hippocampal primary cultures | Synthetic A β | No | ↑ | protein |
| ^{36, 37} | Ms brain | Hippocampus | Aged APP/PS1 | no | ↓ | RNA |
| ⁴⁰ | Hu brain | Hippocampus | Neurons with NFT | No | ↓ | RNA |

Supplementary Table 1. Summary of effects of AD-related pathologies on activity-induced or basal *Arc* expression reported in the literature.

Green entry – reported overall increase of *Arc* protein or RNA linked to AD; red – decrease; blue – no change or both directions. Ms, mouse; hu, human; DG, dentate gyrus of hippocampus; MFC, medial frontal cortex; NFT, neurofibrillary tangles; LPS, lipopolysaccharides.