

# Microglial responses to A $\beta$ rely on ATP release and on cell morphology changes requiring membrane lipid redistribution

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**TAKE HOME:** Microglial motility toward A $\beta$  requires cellular release of ATP from hemichannels and detection by P2RY12. In the microglial shape changes during activation, rearrangement of surface plasma membrane promotes lipid droplet formation through fatty acid activation and triacylglycerol synthesis.

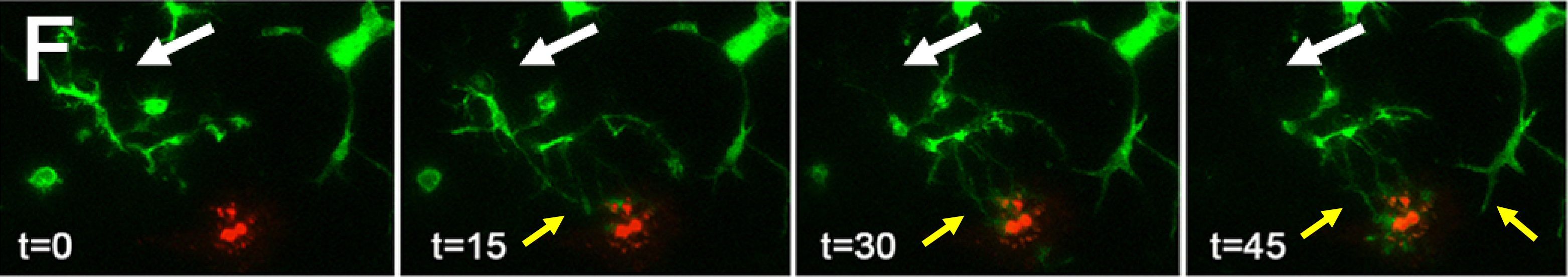


**Figure:** In ex vivo brain slices, newly injected fluorescently-labeled A $\beta$ 42 (in red) attracted microglia processes (in green due to CX3CR1-GFP expression) within 45 minutes. Yellow arrows indicate the extension of proximal processes toward A $\beta$  and white arrows indicate the concomitant retraction of distal processes. We hypothesize that the retraction of the distal processes is necessary due to the limited amount of surface plasma membrane available for the extension of other processes.

| INTRO/METHODS                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                          | RESULTS                                                                                                                                                                                                                                                                                                                                                                                                                                                             | MODEL                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                        | MORE RESULTS                                                                                                                                                                                                                                                                                                                                                                       | ANOTHER MODEL                                                                                                                                |
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| <p><b>Background:</b> Microglia respond acutely and chronically to brain damage signals, including extension/retraction of processes, dramatic changes to cell shape, and movement of the cell body toward the damage. Responses to A<math>\beta</math> deposits leads to plaque remodeling and clearance, as well as reseeding of new A<math>\beta</math> deposits.</p> <p><b>Methods:</b> Brains from CX3CR1<sup>GFP/+</sup> mice allowed analysis of microglial cell bodies and fine processes. We used confocal microscopy of mouse ex vivo brain slices to analyze the extension and retraction of microglial processes toward newly injected, fluorescently labeled A<math>\beta</math>42 over two hours. We also used fixed brain tissue from 5xFAD mice to measure microglial morphology (Branching Index) and total surface area based on skeletal reconstructions using Imaris software.</p> | <div><div><div><div><div>t = 0min</div></div><div><div>t = 60min</div></div></div><div><div>0 min</div></div><div><div>60 min</div></div></div></div> <p><b>Figure:</b> Movement of GFP-labeled microglial processes toward A<math>\beta</math>42 (in red) is completely blocked by 10<math>\mu</math>M PSB0739, an inhibitor of the ATP receptor P2RY12 (top panels) and by 300 mM carbenoxolone, an inhibitor of pannexin/connexon channels, (bottom panels).</p> | <div></div> <p><b>Model:</b> The detection of new A<math>\beta</math> deposits by microglia requires the release of ATP through hemichannels from cells directly in contact with the A<math>\beta</math> deposit. The ATP gradient established attracts microglia through its P2RY12.</p> <p><b>References:</b> Rebeck GW, Shautidze G, Sepulveda J, Healey GS, Narayan P, "Plasma membrane remodeling during microglial activation: a hypothesis linking microglial shape and lipid droplet formation," (Glia, in press). Sepulveda J, Kim JY, Binder J, Vicini S, Rebeck GW, 2024, "APOE4 genotype and aging impair injury-induced microglial behavior in brain slices, including toward A<math>\beta</math>, through P2RY12," Mol Neurodegener 19:24.</p> | <div></div> <p><b>Figure:</b> Microglia complexity (branching) and surface area (SA) are reduced near to plaques in 5XFAD mice. Based on analyses of existing transcriptomics, we propose that lipids from the plasma membrane are metabolized to triacylglycerols and stored in lipid droplets after membrane remodeling associated with activation (<b>cartoon at right</b>)</p> | <div></div> <div><p><b>Bill Rebeck</b><br/>Georgetown Univ.<br/>gwr2@georgetown.edu</p><p><b>ALZHEIMER'S ASSOCIATION   AAIC 26</b></p></div> |