Supplementary Material

Reduced TSPO expression after AMD3100 treatment in mouse stroke model:

a PET study with [¹⁸F]DPA-714

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Running title: PET of Stroke Treatment

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Supplemental methods

2, 3, 5-Triphenyl-2H-tetrazolium chloride (TTC) staining

Mouse brain was cut into 5 slices with a thickness of 2 mm using a mouse brain mold. All slices were stained with 2 % TTC solution and put in dark for 20 min at 37 °C.

[¹⁸F]FDG positron emission tomography

[¹⁸F]FDG PET was performed on day 1, 3, 10 and 16 after MCAO (n=3-6). Another four mice were used as sham control. A single dose of 0.37 MBq (100 μ Ci) [¹⁸F]FDG was injected into animals via tail vein under isoflurane anesthesia. One hour after tracer injection, a 15-min static PET scan was performed with a heating pad to keep animal body temperature at 37°C. All PET acquisition was performed with an Inveon small animal PET scanner (Siemens Preclinical Solutions, PA, USA). All the images were reconstructed using a two-dimensional ordered-subset expectation maximum algorithm (2D OSEM). Imaging analysis was performed with Inveon Research Workplace (Siemens Preclinical Solution). Three-dimensional ellipsoidal regions of interest (ROIs) were manually defined on the ischemic region in the ipsilateral hemisphere under the guidance of T₂-weighted MR images of each mouse obtained at the same time points of PET imaging. Another ROI with the same shape was drawn on the corresponding region in contralateral brain sphere as normal background signals.



Supplementary Figure 1. A, TTC staining result indicates the compromised area on brain slices. **B,** T₂-weighted MR confirm the success of MCAO model establishment. White dash circles show the quantitative ROIs on both two hemispheres. **C,** Representative PET image shows the ROI.



Supplementary Figure S2. A, Representative coronal PET images using [¹⁸F]FDG at different time points after MCAO surgery. White dash line portrays the brain area and the lesion area is indicated by white arrow. **B**, Quantification of [¹⁸F]FDG uptake in the brain tissue after surgery over time, expressed as lesion-to-normal ratio (*p < 0.05).



Supplementary Figure 3. Quantification of [¹⁸F]DPA-714 uptake over time of sham group.

There is no significant difference between all the time points.



Supplementary Figure 4. Co-localization of macrophage marker and TSPO expression in injured brain tissue on day 7 after MACO.