SUPPLEMENTAL MATERIALS

Figure S1. Morphological assessment of Iba1-positive BV2 microglia after exposure to TEMNAP [100nM]

(A) (a-c) Confocal microscopic images displaying three representative morphological categories of $lba1^{(+)}$ -positive cells: resting (a), primed (b) and hyperactivated (c). Nuclei were stained by the nuclear dye Hoechst-33258. Scale bars: 5µm.

(B) Quantitative morphological analysis of Iba1⁽⁺⁾-positive microglia in control condition or exposed to LPS in presence respectively of: (i) self-assembling nanoparticle (SANP); (ii) nanoencapsulated-TEMNAP [100nM] (SANP-TEMNAP); (iii) TEMNAP [100nM] non-nanoencapsulated. Iba1⁽⁺⁾-positive cells were scored in three categories accordingly to their morphological complexity: (i) resting; (ii) primed; and (iii) hyperactivated. In each category data were normalized on the total number of Iba1⁽⁺⁾-positive cells. The values represent the mean±SEM; (n=3 indipendent experimental replicates); *p<0.05 versus respective LPS-SANP condition; #p<0.05 versus respective LPS-TEMNAP [100nM] condition.



Figure S2. Kinetics of two selective siRNA targeting TSPO in BV2 microglial cells

(A-B) Images showing two representative blotting of TSPO protein expression levels in BV2 microglial cells exposed respectively to two different RNAi molecules targeting TSPO gene: (A) The Ambion[™] Silencer[™] Pre-Designed TSPO siRNA (#1) and (B) the mm.Ri.Tspo.13.1 siRNA (#2) at the concentration of [100nM] or to siControl [100nM]. BV2 microglial cells were harvested at the indicated time points (48h and 72h). The vinculin was used as housekeeping protein.

Α а b 124 124 Vinculin Vinculin KDa KDa 18 18 TSPO TSPO KDa KDa 48h 72h 48h 72h siCtl siCtl Control Control siTSPO (#1) siTSPO (#2) [100nM] [100nM] [100nM] [100nM]

Supp. Figure 2

Figure S3. Example of Chromatograms obtained from brain cortex homogenates deriving form mice intraperitoneally infused with: (A) Negative Control; (B) Only Nanoparticles; (C) SANP-TF-TEMNAP.

