Supplementary figure legends:

Supplementary Figure1 (Related to Figure 1):

a-g. Immunofluorescence analysis of glial cell markers in the cortex following PBS, LPS_High, and LPS_Low_Repeated treatments. (a) Representative images showing IBA1 (red) staining and DAPI (blue) for nuclei. (b) Quantification of IBA1 expression levels. (c) Representative images showing F4/80 (green) staining and DAPI for nuclei. (d) Quantification of F4/80 expression levels. (e) Representative images showing GFAP (green) staining and DAPI for nuclei. (f) Quantification of GFAP expression levels. All data are presented as mean ± SEM, One-way ANOVA, Dunnett's post hoc test, *$p < 0.05$, **$p < 0.01$, and ***$p < 0.001$, (n = 6-8 mice (both male and female) in each group).

(g). Representative images showing co-expression of IBA1 (red) staining, F4/80 (green) and DAPI (blue) for nuclei.

Supplementary Figure 2 (Related to Figure 2):

a. Scatterplot matrices showing the FPKM distributions (histograms) and correlation (Corr) values of the RNA-seq data across the treatment groups PBS, LPS_High, and LPS_Low_Repeated. Each panel within shows pairwise comparisons between treatments, with the diagonal panels displaying density distributions of the log10(FPKM) values for each treatment group.

b. Venn diagram showing the overlap of DEGs between the LPS_High and LPS_Low_Repeated treatment groups compared to the PBS. The red circle represents the 4545 unique genes expressed in the LPS_High group. The green circle represents the 102 unique genes expressed in the LPS_Low_Repeated group. The overlapping section contains 241 genes that are commonly expressed between both treatment groups.

c. Four-way scatter plot comparing gene expression changes in LPS_High and LPS_Low_Repeated treatments relative to PBS. The x-axis represents the log fold change (LFC) of genes in the LPS_High treatment relative to PBS. The y-axis represents the LFC of genes in the LPS_Low_Repeated treatment relative to PBS. Each point represents a gene, color-coded based on statistical significance and fold change criteria. Green dots (1509) are genes with adjusted p-value < 0.05 and absolute $\log_2$ fold change > 1.5 only for LPS_High (significant in only one condition). Blue dots (101) are genes with adjusted p-value < 0.05 and absolute log fold change (LFC) > 1.5 for both LPS_High and LPS_Low_Repeated (significant in both conditions). Gray dots are genes with adjusted p-value > 0.05 (non-significant). Red dots (32) are genes with adjusted p-value < 0.05 and absolute LFC < 1.5 for LPS_High but significant for LPS_Low_Repeated (significant in only one condition). The triangle shapes represent genes with extreme fold changes. Right/Top triangles are genes with $\log_2$ fold change > 5. Left/Bottom triangles are genes with LFC < -5. Dashed lines mark LFC thresholds of ±1.5, and red dashed lines indicate LFC thresholds used to define significant gene expression changes in the analysis.

d-e. Perturbation plots for the Fc gamma R-mediated phagocytosis pathway (KEGG ID mmu:04666) (d) in LPS_High compared to PBS (e) in LPS_Low_Repeated compared to PBS. The perturbation of all genes in the pathway are depicted as a function of the log2 fold changes (left panel). Non differentially expressed genes are assigned 0 log2 fold-change. The null distribution of the net accumulated perturbation is also shown as a grey vertical line (right panel). The observed total accumulation (tA) with the actual data is shown as a red vertical line (right panel).

f-j. Heatmap showing the expression of genes (p.adj < 0.05 and LFC > 1.5) related to (f) A1 Astrocyte in LPS_High (red) and LPS_Low_Repeated (green) regimens compared to PBS (blue).

Supplementary Figure 3 (Related to Figure 3):

a. Representative flow cytometry plots showing the gating strategy used to identify the expression of TMEM119, F4/80, CD206 and TREM2 in CD45 low-mid CD11b+ and CD45 high CD11b+ cells.

b-c. Frequency of (b) CD45 low-mid CD11b+ TMEM119+ F4/80+ cells (c) CD45 high CD11b+ TMEM119+ F4/80+ cells as a percentage of CD45+ cells. Data are shown as mean ± SEM. one-way ANOVA, *p<0.05, **p<0.01, ***p<0.001.

Supplementary Figure 4 (Related to Figure 4):

a-c. Scatterplot matrices comparing gene expression (FPKM values) across different treatments PBS, LPS_High, LPS_Low_Repeated, and LPS_Low in the cell populations (a) CD45_low-mid_CD11b_pos (b) CD45_high_CD11b_pos and (c) CD45_neg_ACSA2_pos. Each panel within the matrices shows pairwise comparisons between treatments, with the diagonal panels displaying density distributions of the log10(FPKM) values for each treatment group.

d. PCA plot representing the comparison of cell type-specific markers between isolated populations CD45_low-mid_CD11b_pos, CD45_high_CD11b_pos, CD45_neg_ACSA2_pos and whole brain samples. The distinct clustering indicates clear separation between whole-brain (Prefrontal cortex with hippocampus) and isolated cell populations.

e. PCA plot showing cell type-specific clustering among the isolated populations CD45_low-mid_CD11b_pos, CD45_high_CD11b_pos, and CD45_neg_ACSA2_pos.

f-h. MA plots of the pairwise comparisons for LPS_Low treatment in the three cell types as compared to PBS. Log fold changes (LFCs) are plotted against the mean of normalized counts to determine the variance between two treatments in terms of gene expression. Red nodes on the graph represent statistically significant data points, i.e., p.adj < 0.05 and LFC > 1.5. Gray nodes are data points that are not statistically significant. Numerical values in parentheses for the significant legend indicate the number of genes that meet the prior condition. Dashed lines indicate the cutoff LFC values. (f) 577 genes are significantly differentially expressed in

CD45_low-mid_CD11b_positive_cells in the LPS_Low versus PBS comparison. (g) 2688 genes are significantly differentially expressed in CD45_high_CD11b_positive_cells in the LPS_Low versus PBS comparison. (h) 2539 genes are significantly differentially expressed in CD45_neg_ACSA2_positive_cells in the LPS_Low versus PBS comparison.

i-k. Venn diagrams illustrating the overlap of significant DEGs (p.adj < 0.05 and LFC > 1.5) across the three LPS treatment conditions LPS_High, LPS_Low_Repeated, and LPS_Low compared to PBS in the cell populations (i) CD45_low-mid_CD11b_pos, (j) CD45_high_CD11b_pos, and (k) CD45_neg_ACSA2_pos.

l. Upset plot illustrating the intersection of significant DEGs (p.adj < 0.05 and LFC > 1.5) across three treatments LPS_High, LPS_Low_Repeated, and LPS_Low as compared to PBS in CD45_low-mid_CD11b_pos cells. The horizontal bars on the left represent the total number of significant DEGs unique to each LPS treatment, with 6174 genes for LPS_High, 601 genes for LPS_Low_Repeated, and 95 genes for LPS_Low. The vertical bars at the top show the number of overlapping genes among different combinations of treatments. Notably, 217 genes are shared across all three treatments, while 949 genes are common between LPS_High and LPS_Low_Repeated, 200 genes between LPS_High and LPS_Low, and 65 genes between LPS_Low_Repeated and LPS_Low. The plot highlights both the shared and unique DEGs to LPS treatments in CD45_low-mid_CD11b_pos cells.

m. Upset plot illustrating the intersection of significant DEGs (p.adj < 0.05 and LFC > 1.5) across three treatments LPS_High, LPS_Low_Repeated, and LPS_Low as compared to PBS in CD45_high_CD11b_pos cells. The horizontal bars on the left represent the total number of significant DEGs unique to each LPS treatment, with 4091 genes for LPS_High, 1049 genes for LPS_Low_Repeated, and 208 genes for LPS_Low. The vertical bars at the top show the number of overlapping genes among different combinations of treatments. Notably, 1241 genes are shared across all three treatments, while 1067 genes are common between LPS_High and LPS_Low_Repeated, 1384 genes between LPS_Low_Repeated and LPS_Low, and 223 genes between LPS_High and LPS_Low. The plot highlights both the shared and unique DEGs to LPS treatments in CD45_high_CD11b_pos cells.

n. Upset plot illustrating the intersection of significant DEGs (p.adj < 0.05 and LFC > 1.5) across three treatments LPS_High, LPS_Low_Repeated, and LPS_Low as compared to PBS in CD45_neg_ACSA2_pos cells. The horizontal bars on the left represent the total number of significant DEGs unique to each LPS treatment, with 3814 genes for LPS_High, 687 genes for LPS_Low_Repeated, and 383 genes for LPS_Low. The vertical bars at the top show the number of overlapping genes among different combinations of treatments. Notably, 907 genes are shared across all three treatments, while 703 genes are common between LPS_High and LPS_Low_Repeated, 1009 genes between LPS_High and LPS_Low, and 240 genes between LPS_Low_Repeated and LPS_Low. The plot highlights both the shared and unique DEGs to LPS treatments in CD45_neg_ACSA2_pos cells.

o. Scatter plot depicting gene expression changes in CD45_low-mid_CD11b_pos cells comparing LPS_Low and LPS_High treatments relative to PBS. The x-axis shows the LFC for LPS_High versus PBS, while the y-axis shows the LFC for LPS_Low versus PBS. Colored dots

represent genes categorized based on their significance (adjusted p-value < 0.05) and fold change thresholds (LFC > 1.5). Blue dots (179 genes) are significant in both comparisons, with LFC > 1.5 in either direction. Green dots (2562 genes) are significant in both comparisons but with varying LFC between comparisons. Red dots (63 genes) are significant in both comparisons, but with varying LFC between comparisons. Gray dots are non-significant genes (adjusted p-value > 0.05). Shapes represent genes with extreme LFC values. Triangles represent genes with LFC beyond ±12 for the x-axis and ±4.9 for the y-axis, marking extreme upregulation or downregulation. Circles represent genes with more moderate LFC values. Dashed lines indicate the thresholds for fold change (LFC > 1.5).

p. Scatter plot depicting gene expression changes in CD45_high_CD11b_pos cells comparing LPS_Low and LPS_High treatments relative to PBS. The x-axis shows the LFC for LPS_High versus PBS, while the y-axis shows the LFC for LPS_Low versus PBS. Blue dots (624 genes) are significant in both comparisons, with LFC > 1.5 in either direction. Green dots (2271 genes) are significant in both comparisons but with varying LFC between comparisons. Red dots (1023 genes) are significant in both comparisons but with varying LFC between comparisons. Gray dots are non-significant genes (adjusted p-value > 0.05). Triangles represent genes with extreme LFC values beyond ±11 for the x-axis and ±8.5 for the y-axis. Dashed lines indicate fold change thresholds (LFC > 1.5).

q. Scatter plot depicting gene expression changes in CD45_neg_ACSA2_pos cells comparing LPS_Low and LPS_High treatments relative to PBS. The x-axis shows the LFC for LPS_High versus PBS, while the y-axis shows the LFC for LPS_Low versus PBS. Blue dots (832 genes) are significant in both comparisons, with LFC > 1.5 in either direction. Green dots (1602 genes) are significant in both comparisons but with varying LFC between comparisons. Red dots (219 genes) are significant in both comparisons, both comparisons but with varying LFC between comparisons. Gray dots are non-significant genes (adjusted p-value > 0.05). Triangles represent genes with extreme LFC values beyond ±9.7 for the x-axis and ±7.1 for the y-axis. Dashed lines indicate fold change thresholds (LFC > 1.5).

r. Scatter plot depicting gene expression changes in CD45_low-mid_CD11b_pos cells comparing LPS_Low and LPS_Low_Repeated treatments relative to PBS. The x-axis shows the LFC for LPS_Low_Repeated versus PBS, while the y-axis shows the LFC for LPS_Low versus PBS. Colored dots represent genes categorized based on their significance (adjusted p-value < 0.05) and fold change thresholds (LFC > 1.5). Blue dots (133 genes) are significant in both comparisons, with LFC > 1.5 in either direction. Green dots (453 genes) are significant in both comparisons but with varying LFC between comparisons. Red dots (96 genes) are significant in both comparisons but with varying LFC between comparisons. Gray dots are non-significant genes (adjusted p-value > 0.05). Shapes represent genes with extreme LFC values, with triangles marking extreme upregulation or downregulation (LFC beyond ±6.3 for the x-axis and ±4.9 for the y-axis). Dashed lines indicate fold change thresholds (LFC > 1.5), with red dashed lines indicating limits on the y-axis (LPS_Low).

s. Scatter plot depicting gene expression changes in CD45_high_CD11b_pos cells comparing LPS_Low and LPS_Low_Repeated treatments relative to PBS. The x-axis shows the LFC for LPS_Low_Repeated versus PBS, while the y-axis shows the LFC for LPS_Low versus PBS.

Blue dots (1814 genes) are significant in both comparisons, with LFC > 1.5 in either direction. Green dots (386 genes) are significant in both comparisons but with varying LFC between comparisons. Red dots (118 genes) are significant in both comparisons but with varying LFC between comparisons. Gray dots are non-significant genes (adjusted p-value > 0.05). Triangles represent genes with extreme LFC values beyond ±9.2 for the x-axis and ±8.1 for the y-axis. Dashed lines indicate fold change thresholds (LFC > 1.5).

t. Scatter plot depicting gene expression changes in CD45_neg_ACSA2_pos cells comparing LPS_Low and LPS_Low_Repeated treatments relative to PBS. The x-axis shows the LFC for LPS_Low_Repeated versus PBS, while the y-axis shows the LFC for LPS_Low versus PBS. Blue dots (654 genes) are significant in both comparisons, with LFC > 1.5 in either direction. Green dots (528 genes) are significant in both comparisons but with varying LFC between comparisons. Red dots (397 genes) are significant in both comparisons but with varying LFC between comparisons. Gray dots are non-significant genes (adjusted p-value > 0.05). Triangles represent genes with extreme LFC values beyond ±7.7 for the x-axis and ±7.1 for the y-axis. Dashed lines indicate fold change thresholds (LFC > 1.5).

u. Venn diagram illustrating the intersection of significant DEGs (p.adj < 0.05 and LFC > 1.5) across CD45_low-mid_CD11b_pos, CD45_high_CD11b_pos, and CD45_neg_ACSA2_pos cells in LPS_High treatment compared to PBS. 1885 genes are uniquely differentially expressed in CD45_low-mid_CD11b_pos cells, 1306 genes are uniquely differentially expressed in CD45_high_CD11b_pos cells, and 3007 genes are uniquely differentially expressed in CD45_neg_ACSA2_pos cells. A total of 1946 genes are commonly differentially expressed across all three cell types. Additionally, 862 genes are shared between CD45_low-mid_CD11b_pos and CD45_neg_ACSA2_pos cells, 618 genes are shared between CD45_high_CD11b_pos and CD45_neg_ACSA2_pos cells, and 2847 genes are shared between CD45_low-mid_CD11b_pos and CD45_high_CD11b_pos cells. This diagram highlights both the unique and overlapping DEGs in the three cell types for LPS_High treatment relative to PBS.

v. Upset plot illustrating the intersection of significant DEGs (p.adj < 0.05 and LFC > 1.5) in the three cell types CD45_low-mid_CD11b_pos, CD45_high_CD11b_pos, and CD45_neg_ACSA2_pos for the LPS_High treatment compared to PBS. The horizontal bars on the left represent the total number of significant DEGs unique to each cell type, with 3007 DEGs in CD45_neg_ACSA2_pos, 1885 DEGs in CD45_low-mid_CD11b_pos, and 1306 DEGs in CD45_high_CD11b_pos. The vertical bars at the top show the number of overlapping DEGs across different cell types. Notably, 1946 genes are shared across all three cell types, while 862 genes are shared between CD45_low-mid_CD11b_pos and CD45_neg_ACSA2_pos, 2847 genes are shared between CD45_low-mid_CD11b_pos and CD45_high_CD11b_pos, and 618 genes are shared between CD45_high_CD11b_pos and CD45_neg_ACSA2_pos. This plot highlights both the shared and unique DEGs in the three cell types for the LPS_High treatment compared to PBS.

w. Venn diagram illustrating the intersection of significant DEGs (p.adj < 0.05 and LFC > 1.5) across CD45_low-mid_CD11b_pos, CD45_high_CD11b_pos, and CD45_neg_ACSA2_pos cells in LPS_Low_Repeated treatment compared to PBS. 936 genes are uniquely differentially

expressed in CD45_low-mid_CD11b_pos cells, 3206 genes are uniquely differentially expressed in CD45_high_CD11b_pos cells, and 1813 genes are uniquely differentially expressed in CD45_neg_ACSA2_pos cells. A total of 147 genes are commonly differentially expressed across all three cell types. Additionally, 414 genes are shared between CD45_high_CD11b_pos and CD45_neg_ACSA2_pos cells, 163 genes are shared between CD45_low-mid_CD11b_pos and CD45_neg_ACSA2_pos cells, and 586 genes are shared between CD45_low-mid_CD11b_pos and CD45_high_CD11b_pos cells. This diagram highlights both the unique and overlapping DEGs in the three cell types for LPS_Low_Repeated treatment compared to PBS.

x. Upset plot illustrating the intersection of significant DEGs (p.adj < 0.05 and LFC > 1.5) in the three cell types CD45_low-mid_CD11b_pos, CD45_high_CD11b_pos, and CD45_neg_ACSA2_pos for the LPS_Low_Repeated treatment as compared to PBS. The horizontal bars on the left represent the total number of significant DEGs unique to each cell type, with 3206 DEGs in CD45_high_CD11b_pos, 1813 DEGs in CD45_neg_ACSA2_pos, and 936 DEGs in CD45_low-mid_CD11b_pos. The vertical bars at the top show the number of overlapping DEGs across different cell types. Notably, 147 genes are shared across all three cell types, while 414 genes are shared between CD45_high_CD11b_pos and CD45_neg_ACSA2_pos, 163 genes between CD45_low-mid_CD11b_pos and CD45_neg_ACSA2_pos, and 586 genes are shared between CD45_low-mid_CD11b_pos and CD45_high_CD11b_pos cells. This plot highlights both the shared and unique DEGs in the three cell types for the LPS_Low_Repeated treatment compared to PBS.

y. Venn diagram illustrating the intersection of significant DEGs (p.adj < 0.05 and LFC > 1.5) across CD45_low-mid_CD11b_pos, CD45_high_CD11b_pos, and CD45_neg_ACSA2_pos cells in LPS_Low treatment compared to PBS. 322 genes are uniquely differentially expressed in CD45_low-mid_CD11b_pos cells, 1838 genes are uniquely differentially expressed in CD45_high_CD11b_pos cells, and 1706 genes are uniquely differentially expressed in CD45_neg_ACSA2_pos cells. A total of 108 genes are commonly differentially expressed across all three cell types. Additionally, 660 genes are shared between CD45_high_CD11b_pos and CD45_neg_ACSA2_pos cells, and 65 genes are shared between CD45_low-mid_CD11b_pos and CD45_neg_ACSA2_pos cells and 82 genes are shared between CD45_low-mid_CD11b_pos and CD45_high_CD11b_pos cells. This diagram highlights both the unique and overlapping DEGs in the three cell types for LPS_Low treatment compared to PBS.

(z) Upset plot illustrating the intersection of significant DEGs (p.adj < 0.05 and LFC > 1.5) in the three cell types CD45_low-mid_CD11b_pos, CD45_high_CD11b_pos, and CD45_neg_ACSA2_pos for the LPS_Low treatment compared to PBS. The horizontal bars on the left represent the total number of significant DEGs unique to each cell type, with 1838 DEGs in CD45_high_CD11b_pos, 1706 DEGs in CD45_neg_ACSA2_pos, and 322 DEGs in CD45_low-mid_CD11b_pos. The vertical bars at the top show the number of overlapping DEGs across different cell types. Notably, 108 genes are shared across all three cell types, while 660 genes are shared between CD45_high_CD11b_pos and CD45_neg_ACSA2_pos, 65 genes are shared between CD45_low-mid_CD11b_pos and CD45_neg_ACSA2_pos, and 82 genes are shared between CD45_low-mid_CD11b_pos and CD45_high_CD11b_pos cells. This plot

highlights both the shared and unique DEGS in the three cell types for the LPS_Low treatment compared to PBS.

Supplementary Figure 5 (Related to Figure 5):

a-b. Perturbation plots for the apoptosis pathway (KEGG ID mmu:04210) (a) in CD45_low-mid_CD11b_pos cells comparing LPS_Low vs PBS (b) in CD45_high_CD11b_pos cells comparing LPS_Low vs PBS.

c. Gene Set Enrichment Analysis (GSEA) plot for the neurodegeneration pathway (KEGG ID mmu:05022) in CD45_low-mid_CD11b_pos cells comparing LPS_High vs PBS.

d-e. Perturbation plots for the Fc gamma R-mediated phagocytosis pathway (KEGG ID mmu:04666) (d) in CD45_low-mid_CD11b_pos cells comparing LPS_High vs PBS (e) in CD45_high_CD11b_pos cells comparing LPS_High vs PBS

f-k. Perturbation plots for the NF-kappa B signaling pathway (KEGG ID mmu:04064) (f) in CD45_low-mid_CD11b_pos cells comparing LPS_High vs PBS (g) in CD45_high_CD11b_pos cells comparing LPS_High vs PBS (h) in CD45_low-mid_CD11b_pos cells comparing LPS_Low_Repeated vs PBS (i) in CD45_high_CD11b_pos cells comparing LPS_Low_Repeated vs PBS) (j) in CD45_low-mid_CD11b_pos cells comparing LPS_Low vs PBS (k) in CD45_high_CD11b_pos cells comparing LPS_Low vs PBS. The perturbation of all genes in the pathway are depicted as a function of the log2 fold changes (left panel). Non differentially expressed genes are assigned 0 log2 fold-change. The null distribution of the net accumulated perturbation is also shown as a grey vertical line (right panel). The observed total accumulation (tA) with the actual data is shown as a red vertical line (right panel).

Supplementary Figure 6 (Related to Figure 6):

a. Perturbation plot for the apoptosis pathway (KEGG ID mmu:04210) in CD45_neg_ACSA2_pos cells comparing LPS_High vs PBS.

b. Perturbation plot for the NF-kappa B signaling pathway (KEGG ID mmu:04064) in CD45_neg_ACSA2_pos cells comparing LPS_High vs PBS.

c. Heatmaps showing the expression of genes associated with (c) A1 Astrocytes in CD45_neg_ACSA2_pos cells in PBS (blue), LPS_High (red), LPS_Low (magenta), and LPS_Low_Repeated (green) treatment conditions.

d. Line graph showing the weight (in grams) of WT mice treated with PBS (blue) and LPS_Low_Repeated (green) over time. The x-axis represents the days post-treatment (Day 0 to Day 90), and the y-axis represents the weight of the animals. Data are shown from Day 0 to Day 5, with a follow-up measurement on Day 90. All data are presented as mean ± SEM, t test, *$p < 0.05$, **$p < 0.01$, and ***$p < 0.001$, (n = 6-8 mice (both male and female) in each group).

e. Line graph showing the time to reach the hidden platform (in seconds) during the hidden platform training phase of the water maze test for different groups of PS19 mice: PS19 Non Tg PBS (blue squares), PS19 Tg PBS (red triangles), PS19 Tg LPS_Low_Repeated (pink diamonds), and PS19 Non Tg LPS_Low_Repeated (green triangles). The x-axis represents the number of days of training (Day 1 to Day 7), and the y-axis represents the time taken to reach the platform.

f-h. Bar plots showing the results from the probe trial of the water maze behavior test for different groups of PS19 mice: PS19 Non Tg PBS (blue), PS19 Tg PBS (red), PS19 Non Tg LPS_Low_Repeated (green), and PS19 Tg LPS_Low_Repeated (pink). f. Total time duration spent in the target quadrant. All data are presented as mean ± SEM, One-way ANOVA, Dunnett's post hoc test, *$p < 0.05$, **$p < 0.01$, and ***$p < 0.001$, ($n = 6$-8 mice (both male and female) in each group).