

Supplemental information

Topoisomerase I inhibition yields

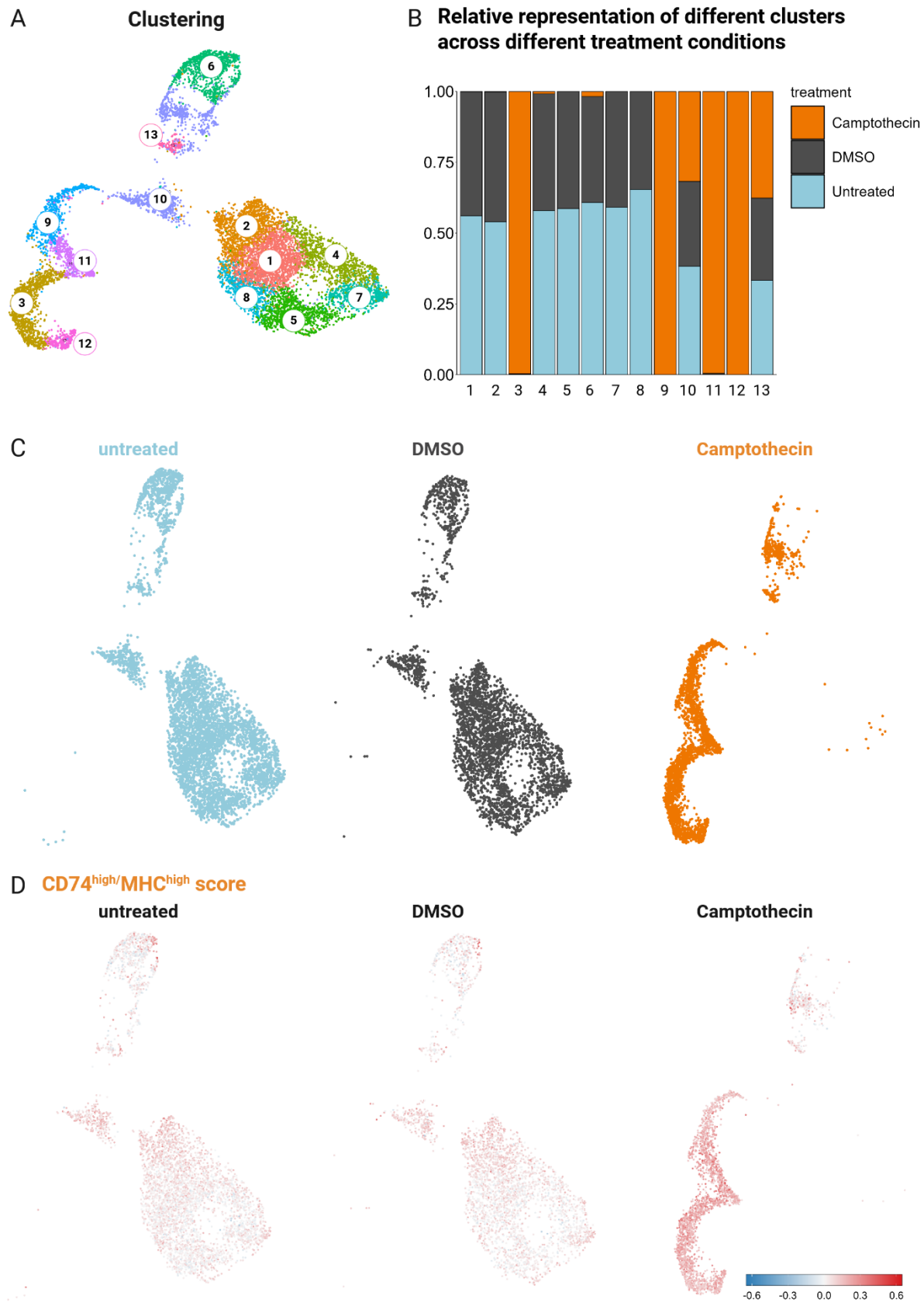
a CD74^{high}/MHC^{high} human microglial

subtype with enhanced A β uptake

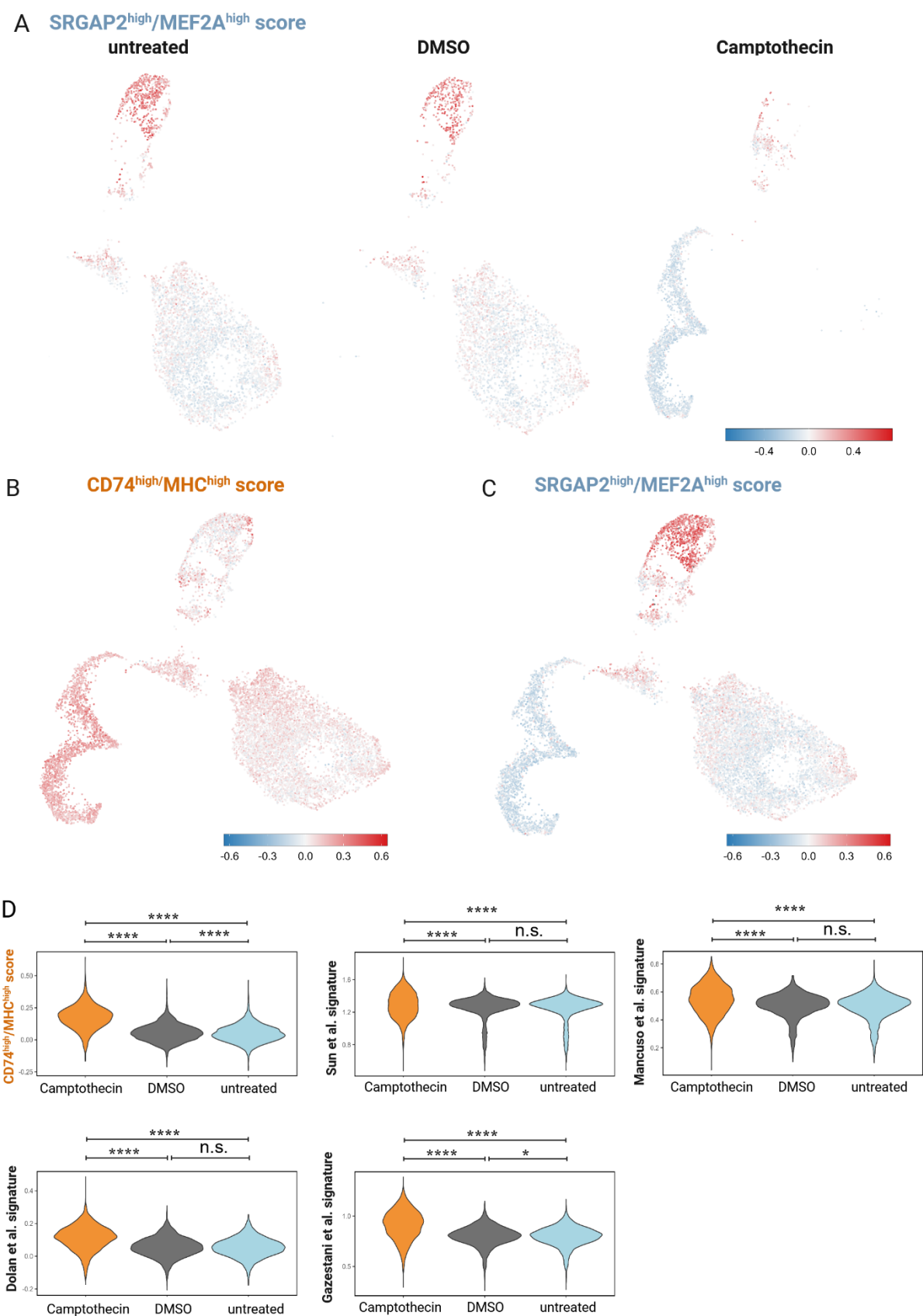
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1 **Supplementary Figures**

2 **Supplementary Figure S1**



Supplementary Figure S1. Overview of clustering, distribution and CD74^{high}/MHC^{high} signature of Camptothecin-treated HMC3 microglia-like cells. **A. UMAP of compound-treated HMC3 cells showing cluster identity (related to Figure 1).** Each dot is a single cell, colored by cluster identity ranging from clusters 1-13. n=1 biological replicate per condition. **B.** Bar plots showing the relative representation of different clusters across different treatment conditions, with each bar summing to 100% (untreated, DMSO, Camptothecin). **C.** Each UMAP depicts all HMC3 microglia-like cells belonging to a specific treatment group, including untreated (light blue, n=1, 4729 cells), DMSO-treated (dark grey, n=1, 3434 cells), or Camptothecin-treated (orange, n=1, 2790 cells). **D. CD74^{high}/MHC^{high} signature in Camptothecin-treated HMC3 cells.** Enrichment of the top 50 cluster genes was calculated on a per-cell basis compared to background genes with similar expression levels. Cells are colored by log-fold change of the CD74^{high}/MHC^{high} gene set. Each sample is represented separately based off the UMAP depicted in **C.**, including untreated, DMSO-treated control and Camptothecin-treated HMC3 cells.



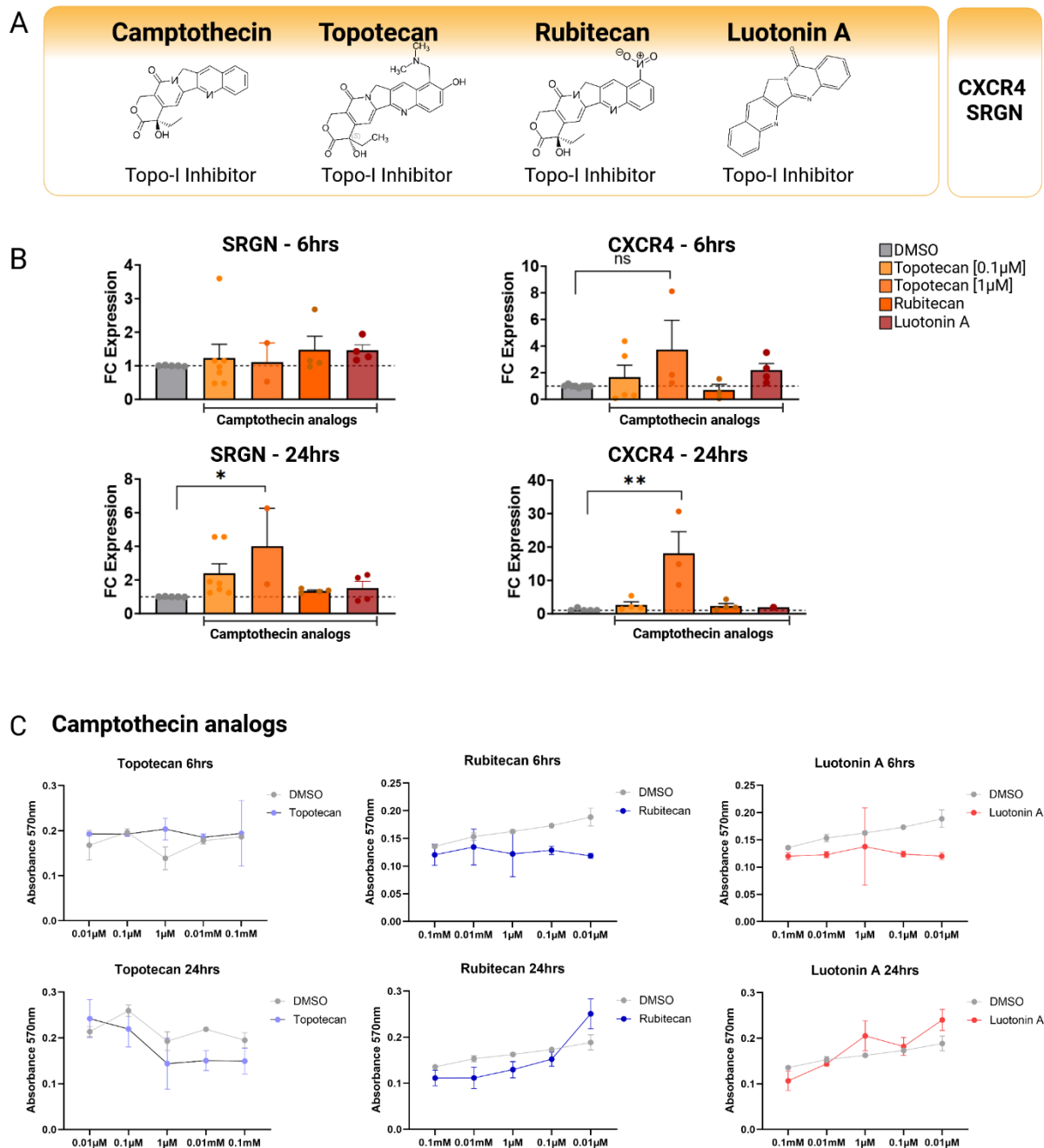
Supplementary Figure S2. Distribution and statistical analysis of CD74^{high}/MHC^{high}, SRGAP2^{high}/MEF2A^{high} signatures and additional MHC^{high} microglia signatures in Camptothecin-treated HMC3 microglia-like cells.

A. SRGAP2^{high}/MEF2A^{high} signature in compound-treated HMC3 microglia-like cells (related to Figure 1). Enrichment of the top 50 cluster genes was calculated on a per-cell basis compared to background genes with similar expression levels. Cells are colored by log-fold change of the SRGAP2^{high}/MEF2A^{high} gene set. Each sample is represented separately based off the UMAP depicted in **Supplementary Figure 1C.**, including untreated, DMSO-treated control and Camptothecin-treated HMC3 cells.

B. CD74^{high}/MHC^{high} signature in Camptothecin-treated HMC3 microglia-like cells. Enrichment of the top 50 cluster genes was calculated on a per-cell basis compared to background genes with similar expression levels. Cells are colored by log-fold change of the CD74^{high}/MHC^{high} gene set.

C. SRGAP2^{high}/MEF2A^{high} signature in compound-treated HMC3 microglia-like cells. Enrichment of the top 50 cluster genes was calculated on a per-cell basis compared to background genes with similar expression levels. Cells are colored by log-fold change of the SRGAP2^{high}/MEF2A^{high} gene set.

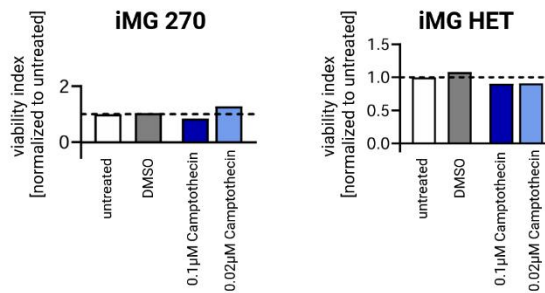
D. Violin plots depict the CD74^{high}/MHC^{high} module score or respective HLA^{high}- or MHC^{high}- scores from the following studies: Dolan et al.¹, Sun et al.², Gazestani et al.³, Mancuso et al.⁴, assessed in HMC3-treated cells, depicted for each treatment group (untreated - light blue ; DMSO-treated - dark grey; Camptothecin-treated - orange). For statistical analysis pairwise Wilcoxon rank-sum tests were performed; ns > 0.05, *p.adj ≤ 0.05; **p.adj ≤ 0.01; ***p.adj ≤ 0.001; ****p.adj ≤ 0.0001. CD74^{high}/MHC^{high} module: untreated vs. Camptothecin - p ≤ 0.0001; DMSO vs. Camptothecin – p ≤ 0.0001; untreated vs. DMSO – p = 0,0000277. Dolan et al. module: untreated vs. Camptothecin - p = 8,04E-269; DMSO vs. Camptothecin – p = 1,08E-220; untreated vs. DMSO – p = 0,136. Sun et al. module: untreated vs. Camptothecin - p = 1,4E-12; DMSO vs. Camptothecin – p = 3,36E-07; untreated vs. DMSO – p = 0,122. Gazestani et al. module: untreated vs. Camptothecin - p = 5,82E-292; DMSO vs. Camptothecin – p = 1,13E-236; untreated vs. DMSO – p = 0,016. Mancuso et al. module: untreated vs. Camptothecin - p = 1,21E-77; DMSO vs. Camptothecin – p = 6,56E-60; untreated vs. DMSO – p = 0,073.

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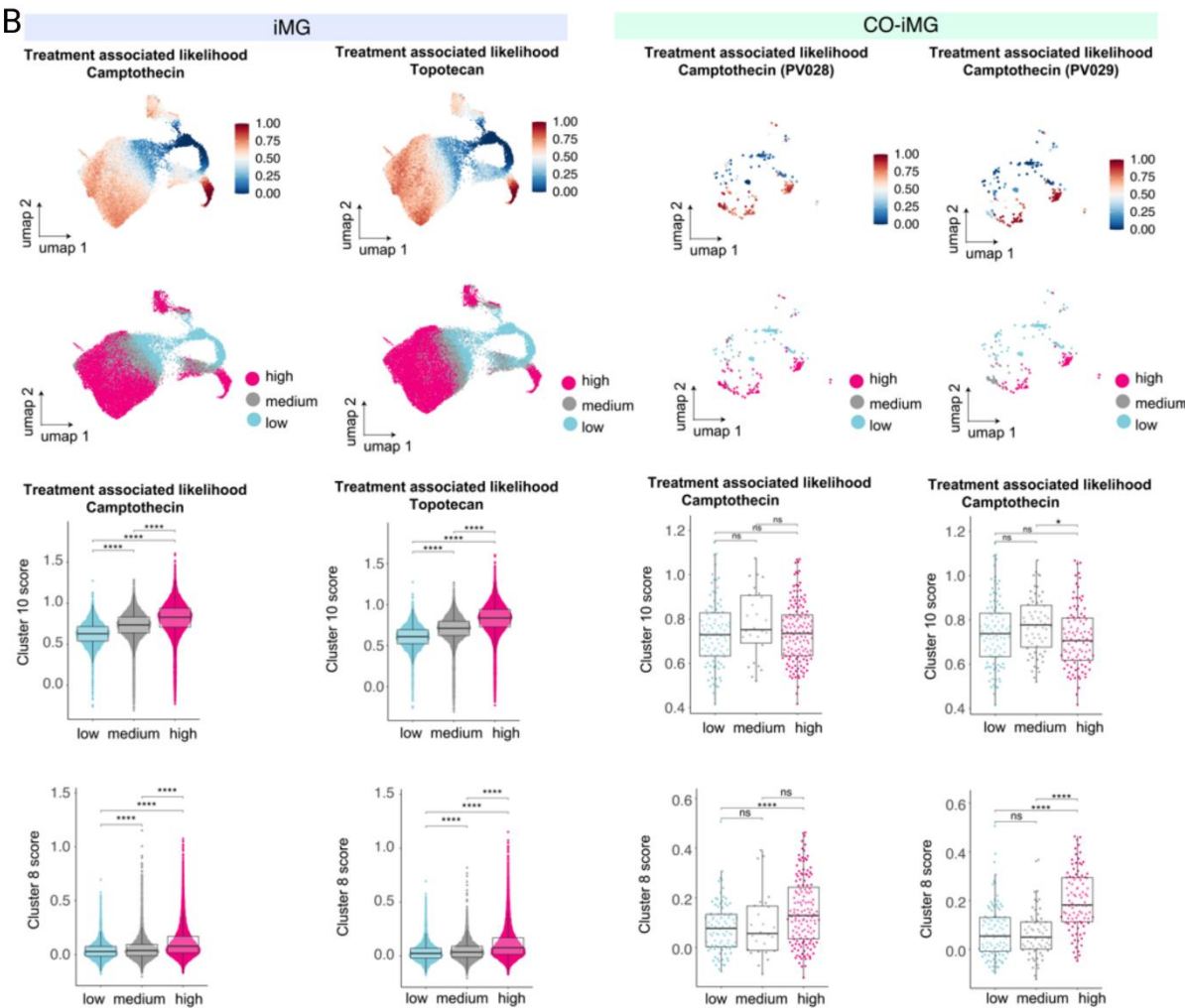
83 **Supplementary Figure S3. Structure activity relationship (SAR) analysis of HMC3**
 84 **microglia-like cells treated with Camptothecin analogs. A. Overview of chemical**
 85 **structures and functional targets** of selected compounds for structure activity relationship
 86 analysis (related to Figure 2). **B. Marker gene expression (SRGN, CXCR4) in HMC3**
 87 **microglia-like cells treated with Camptothecin analogs (6hrs and 24hrs)** assessed via RT-
 88 qPCR. CT values were normalized to *HPRT1*. Bars represent fold change expression (mean
 89 $\pm$ SEM (Standard Error of the Mean)) in relation to DMSO control. For statistical analysis, one-
 90 way ANOVA followed by Dunnett's multiple comparisons test was performed. ns > 0.05, *p.adj

≤ 0.05; **p.adj ≤ 0.01; ***p.adj ≤ 0.001; ****p.adj ≤ 0.0001. SRGN 6hrs (DMSO – n= 5; Topotecan 0.1μM - n= 7 ; Topotecan 1μM - n= 2 ; Rubitecan - n= 4; Luotonin A - n= 4), SRGN 24hrs (DMSO – n= 5; Topotecan 0.1μM - n= 7; Topotecan 1μM - n= 2; Rubitecan - n= 4; Luotonin A - n= 4), CXCR4 6hrs (DMSO – n= 8; Topotecan 0.1μM - n= 5; Topotecan 1μM - n= 3; Rubitecan - n= 3; Luotonin A - n= 4), CXCR4 24hrs (DMSO – n= 5; Topotecan 0.1μM - n= 4; Topotecan 1μM - n= 3; Rubitecan - n= 4; Luotonin A - n= 1). **C. Titration experiments of SAR (structure activity relationship analysis).** Each compound was titrated on HMC3 cells covering a range from 0.1mM to 0.01μM (0.1mM, 0.01mM, 1μM, 0.1μM, 0.01μM). Following 6hrs and 24hrs treatment, cell viability was assessed using MTT viability assay. MTT viability assay for Camptothecin analogs (Topotecan, Rubitecan, Luotonin A) 6hrs and 24hrs following treatment, in comparison to DMSO control. Data points/concentration are derived from n=1 biological replicates of treatment in technical triplicates as mean with 95% CI. Compound-treatment is depicted in color, DMSO controls are visualized in grey.

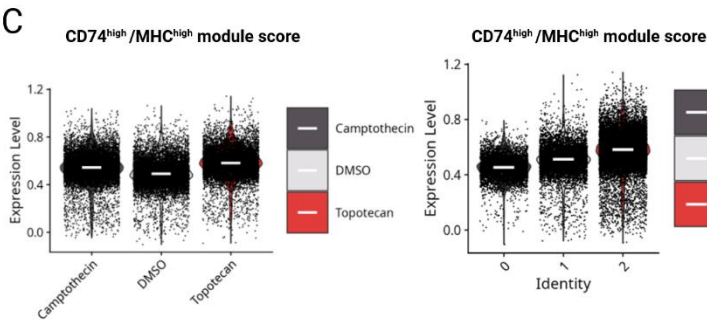
A Alamar Blue Assay iMG



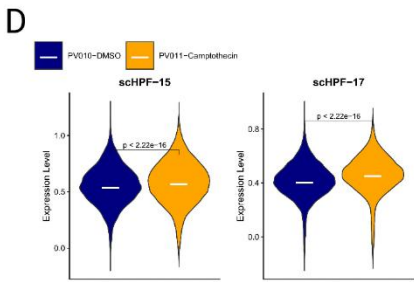
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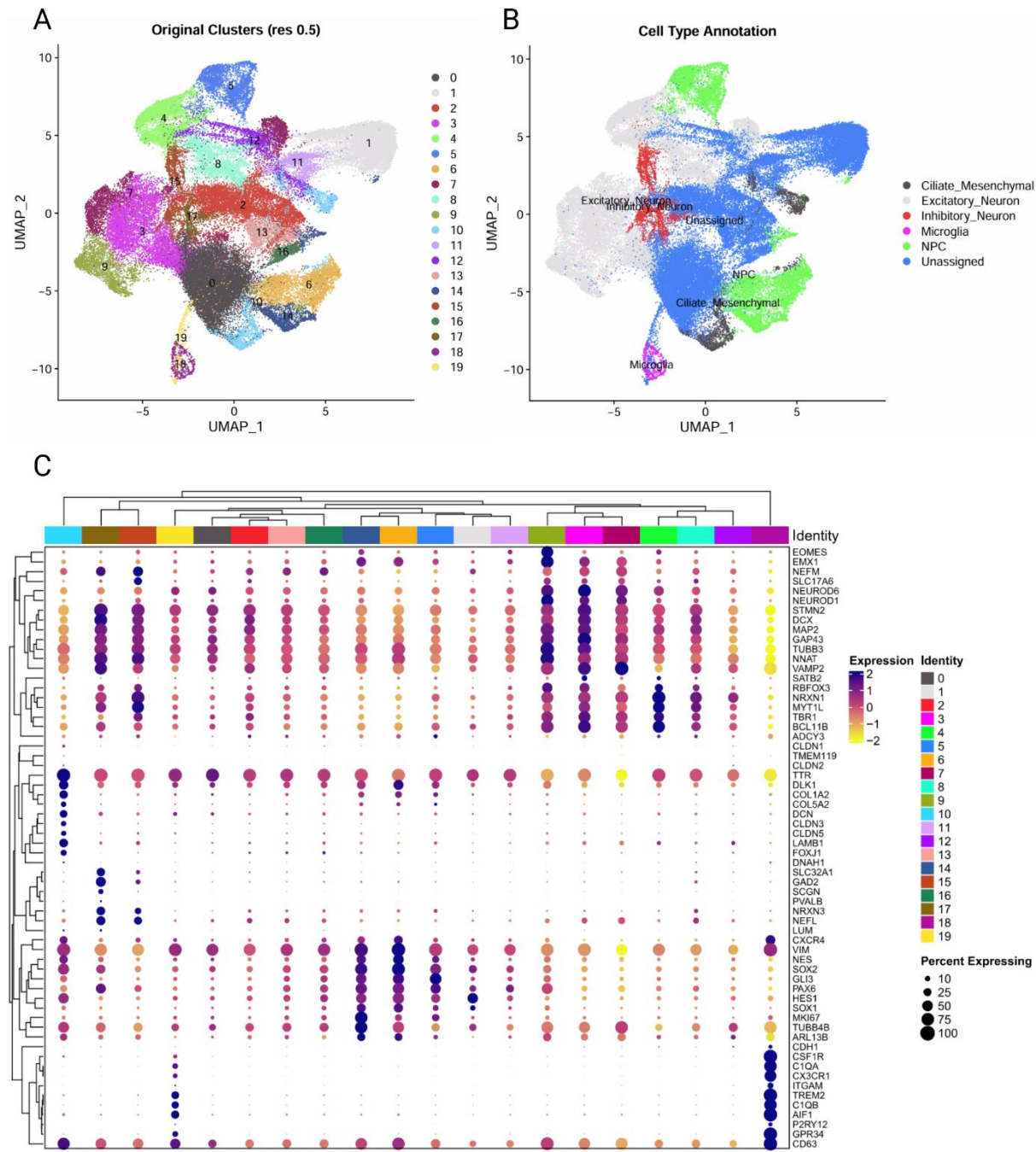
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Supplementary Figure S4. sc-RNA sequencing of compound-treated iMG and CO-iMG indicates upregulation of the CD74^{high}/MHC^{high} signature following treatment with Camptothecin and Topotecan. A. Alamar blue assay assessing viability of human iPSC-derived microglia (iMG) after 24hrs of treatment with Camptothecin or DMSO-treated and untreated cells as control (Camptothecin (0.1 μM; 0.02 μM; related to Figure 2). Graphs depict assessed viability index for n=1 of two distinct iPSC-derived microglia (iMG) lines (270 and HET). B. Individual iMGs (left) and iMG co-cultured with cerebral organoids (CO-iMG) (right) colored by treatment-associated likelihood in the joint UMAP. Upper row shows coloring based on MELD score, lower row shows coloring based on relative treatment-associated likelihood scores (red:high, blue:low) calculated by MELD for treatment conditions: Camptothecin- and Topotecan treatment in iMG, and Camptothecin treatment at 0.1 μM and 0.5 μM in CO-iMG (iMG – n=1 differentiation per condition; CO-iMG - n= 1 differentiation from two independent iPSC-lines per condition). Graphs depict the CD74^{high}/MHC^{high} score in iMG and CO-iMG across different levels of treatment-associated likelihood. Cells are colored by three different levels (low, medium, high) of treatment-associated likelihood for each drug-treatment condition. Upper row – depicts cluster 10 gene module score⁵ across the three levels of treatment-associated likelihood inferred from the indicated treatment conditions. Each boxplot highlights the median, lower and upper quartiles. Whiskers in boxplots indicate 1.5 times interquartile ranges. Lower row - single-cell distributions of the Cluster 8 gene module score⁵ across the three levels of treatment-associated likelihood inferred from the indicated treatment conditions. Each boxplot highlights the median, lower and upper quartiles. Whiskers in boxplots indicate 1.5 times interquartile ranges. Statistical analysis was performed using unpaired t-test; ns > 0.05, *p.adj ≤ 0.05; **p.adj ≤ 0.01; *p.adj ≤ 0.001; ****p.adj ≤ 0.0001. (iMG – n=1 differentiation per condition; CO-iMG - n= 1 differentiation from two independent iPSC-lines per condition). C. Graphs depict expression level of CD74^{high}/MHC^{high} signature across different treatment groups as defined by the experiment (DMSO (light grey), Camptothecin (dark grey), Topotecan (red); left). Each dot represents a single cell; each boxplot highlights the median, lower, and upper quartiles. Right graph depicts single-cell distribution of the CD74^{high}/MHC^{high} module scores across the three levels of Camptothecin treatment-associated likelihood (low - 0, medium -1, high- 2) for Camptothecin. Each dot represents a single cell. Each boxplot highlights the median, lower, and upper quartiles. Whiskers indicate 1.5 times interquartile ranges. D. Projection of DMSO- and Camptothecin-treated iPSC-derived human microglia into the 23-factor model published by Marshe et al.⁶. Depicted are projections into factors schPF-15 (HLA^{high}/APC) and sc-HPF17 (C1Q^{high}). For projections, the top50 genes from each of the factors (schPF-15, schPF-17) were taken and projected into the DMSO- (blue) or Camptothecin-(yellow) treated iPSC-derived microglia. X axis depicts samples, y-axis depicts samples, y-axis depicts expression levels of the**

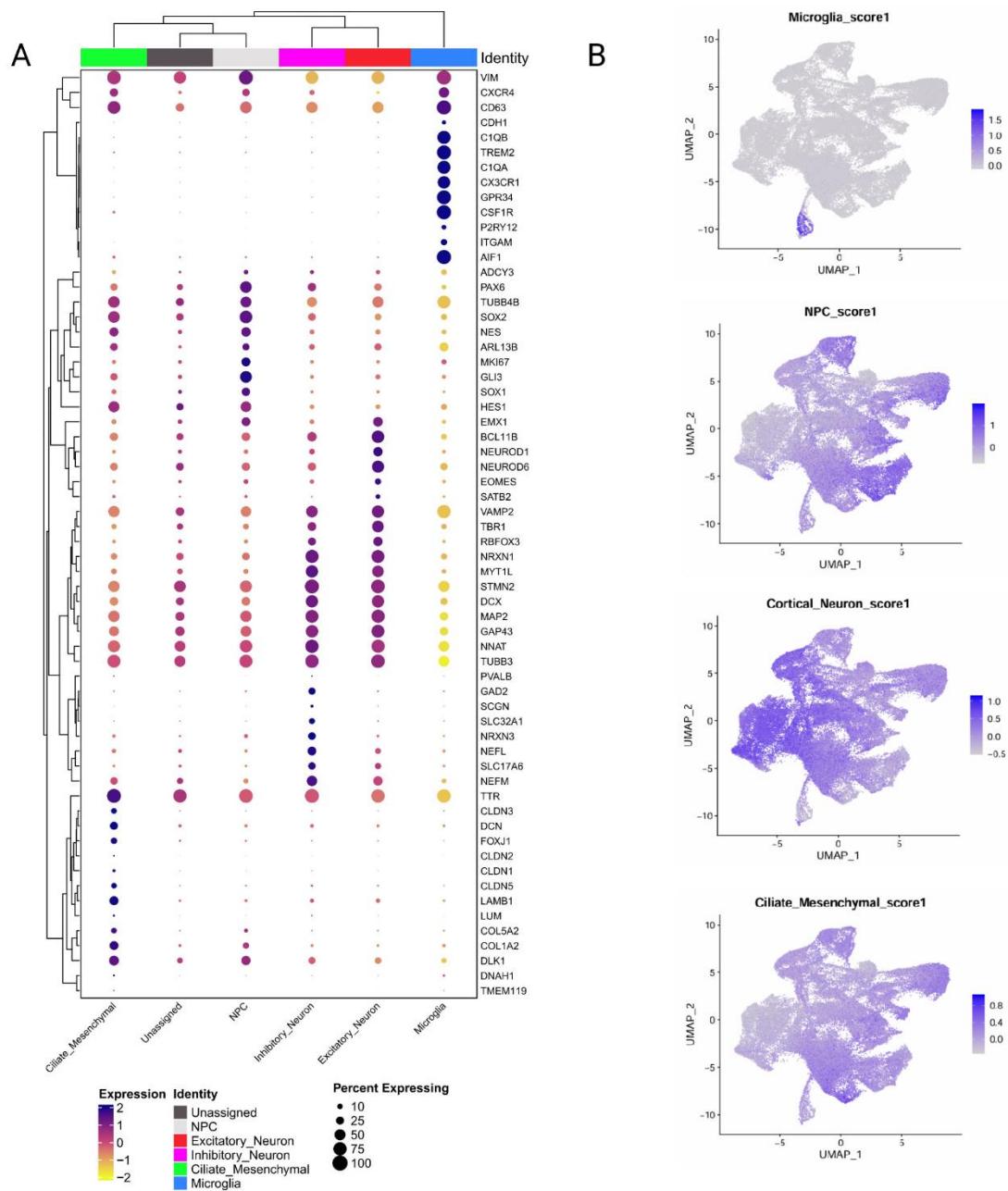
respective factors. Center line of each distribution indicates median. For statistical analysis, unpaired Wilcoxon test was performed. ns > 0.05, *p.adj ≤ 0.05; **p.adj ≤ 0.01; ***p.adj ≤ 0.001; ****p.adj ≤ 0.0001.

Supplementary Figure S5



Supplementary Figure S5. Clustering, cell type annotation and marker expression of cerebral organoid samples co-cultured with iPSC-derived human microglia. A. UMAP of

all cells present in the cerebral organoid samples co-cultured with iPSC-derived human microglia (iMG; related to Figure 2). Organoids derived from n = 2 donors (one male, one female donor) were pooled (n= 33915 cells total). Hierarchical clustering analysis resulted in 19 clusters, each depicted in a different color. Each dot is a single cell. **B.** UMAP from A. depicting the distribution of the different cell types annotated as per marker gene expression. Dark grey = ciliate mesenchymal cells; light grey = excitatory neurons; red = inhibitory neurons; pink = microglia; green = neural progenitor cells (NPCs); blue = unassigned cells. **C.** Expression of a set of marker genes across the 19 identified clusters. X axis represents the 19 different clusters (top: colors, bottom: cluster number), Y axis represents the different genes assessed. Dots size represents percentage of cells expressing each gene and Z-scored expression as depicted by color (yellow – low; purple - high). Each dot is a single cell.

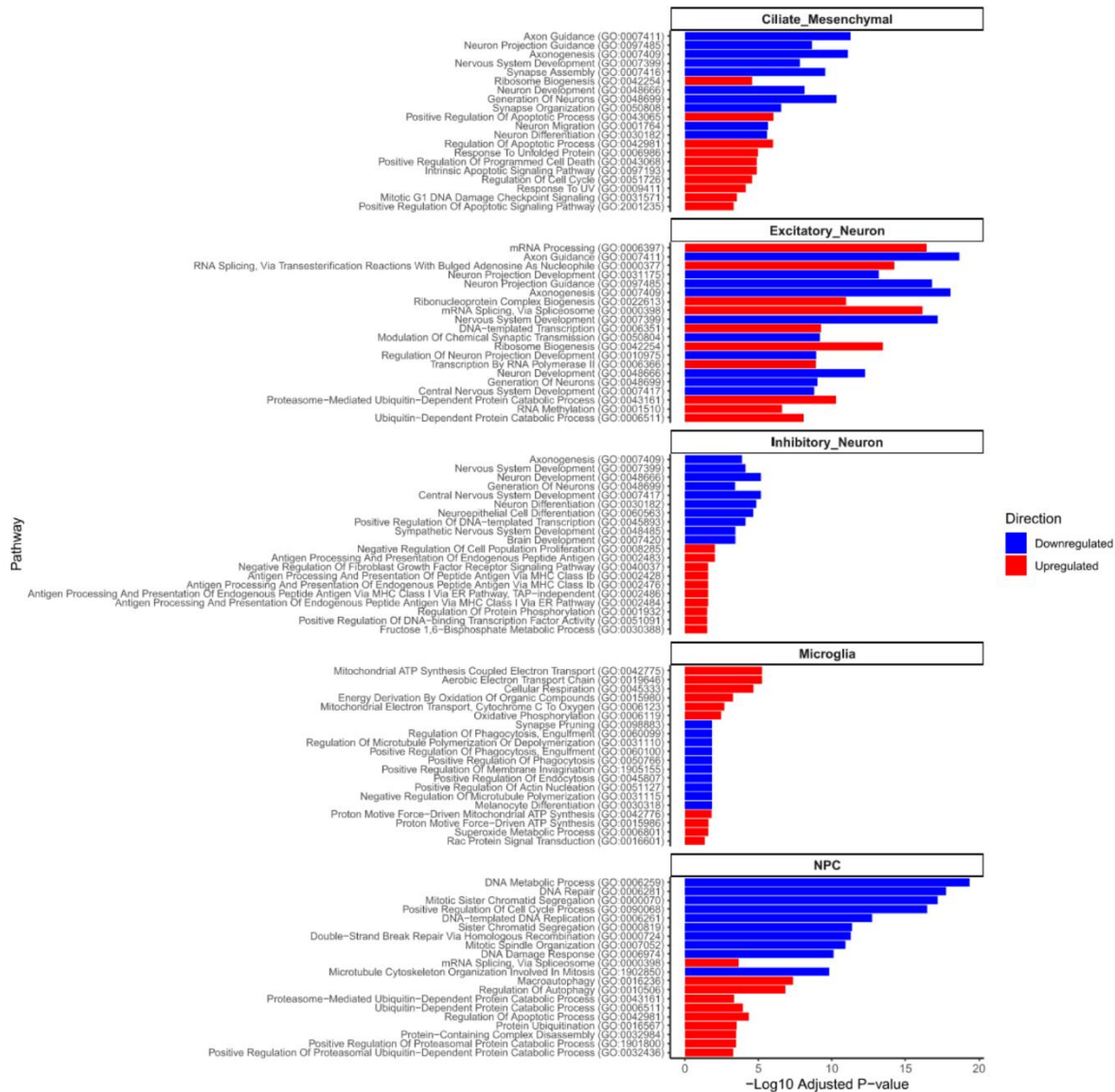


Supplementary Figure S6. Marker expression and cell type distribution across scRNA Seq data derived from cerebral organoid samples co-cultured with iPSC-derived human microglia. A. Expression of a set of marker genes across the assigned cell types (related to Figure 2). X axis represents the different cell types (top: colors (green – ciliate mesenchymal cells; dark grey- unassigned; light grey – unassigned; pink – inhibitory neurons; red – excitatory neurons; blue - microglia), bottom: cell type name), Y axis represents the different genes assessed. Dots represent percentage of cells expressing each gene and colors indicate Z-scored expression as depicted by color (yellow – low; purple - high). B. UMAP from Supplementary Figure S5A. UMAPs represent cells colored by their expression level of

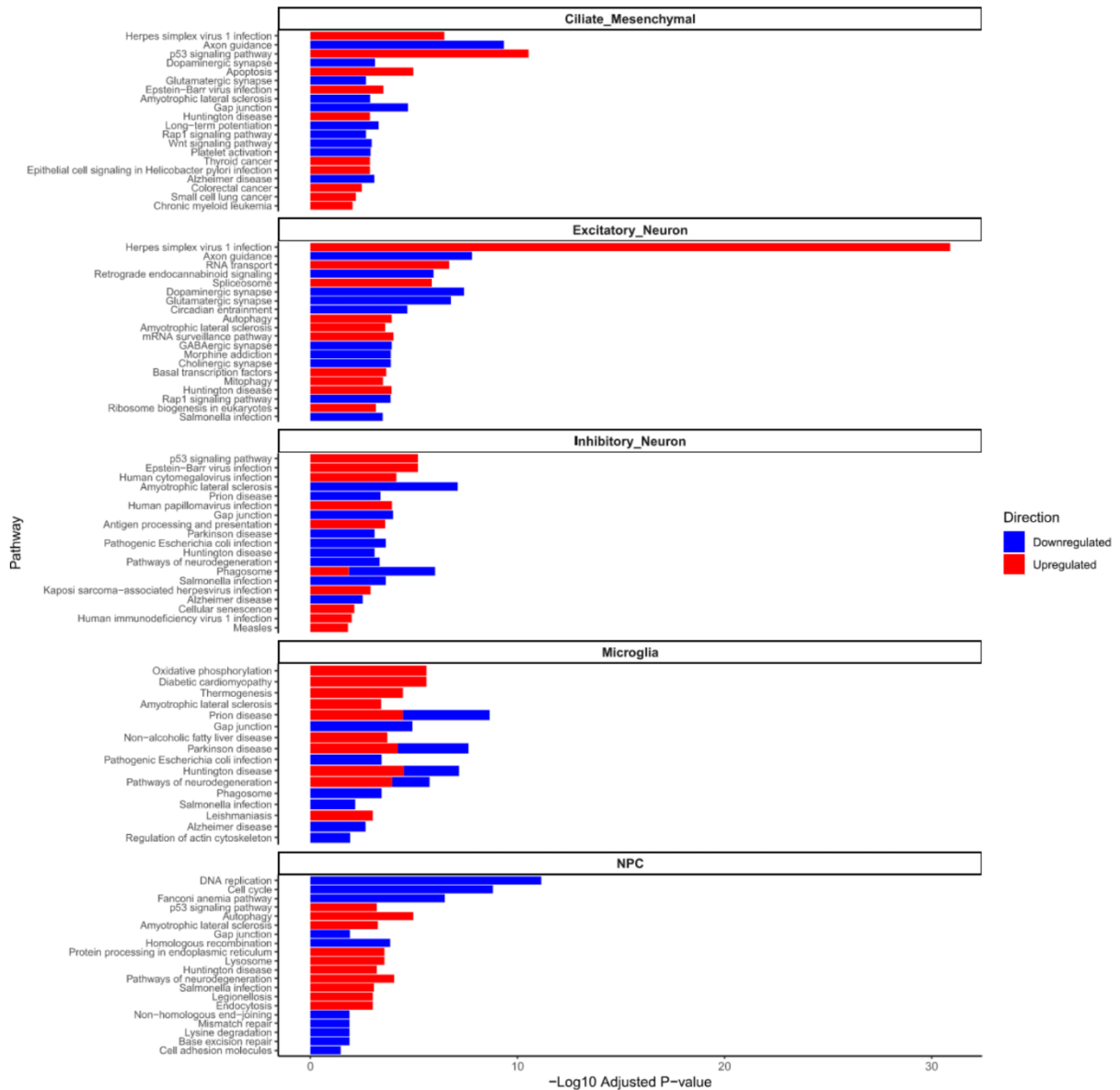
different cell-type defining signatures (microglia, neural precursor cells (NPC), cortical neurons, ciliate/mesenchymal cells). Each dot is a single cell.

Supplementary Figure S7

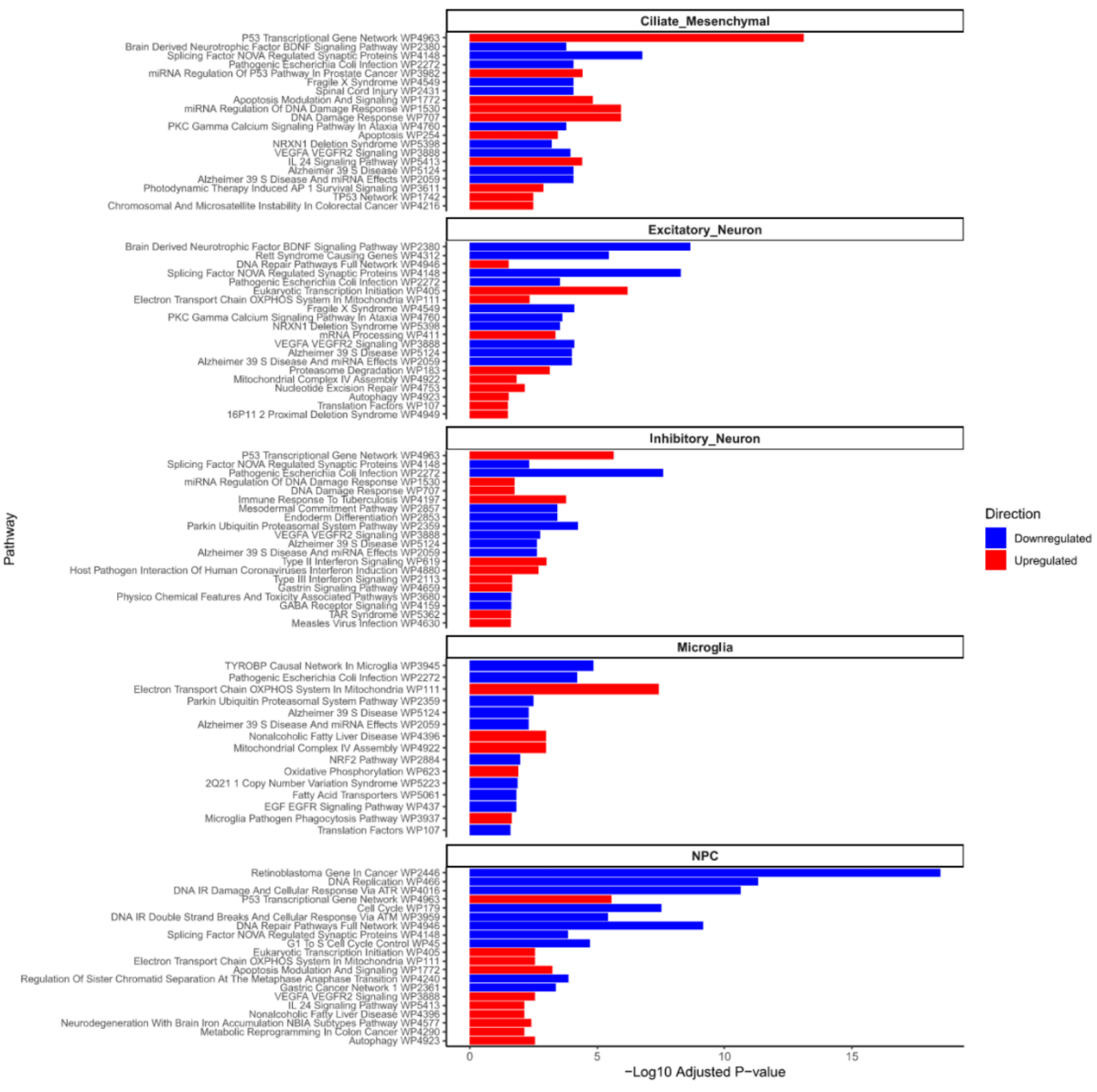
A Top Enriched Pathways - GO - Biological Process



B Top Enriched Pathways - KEGG



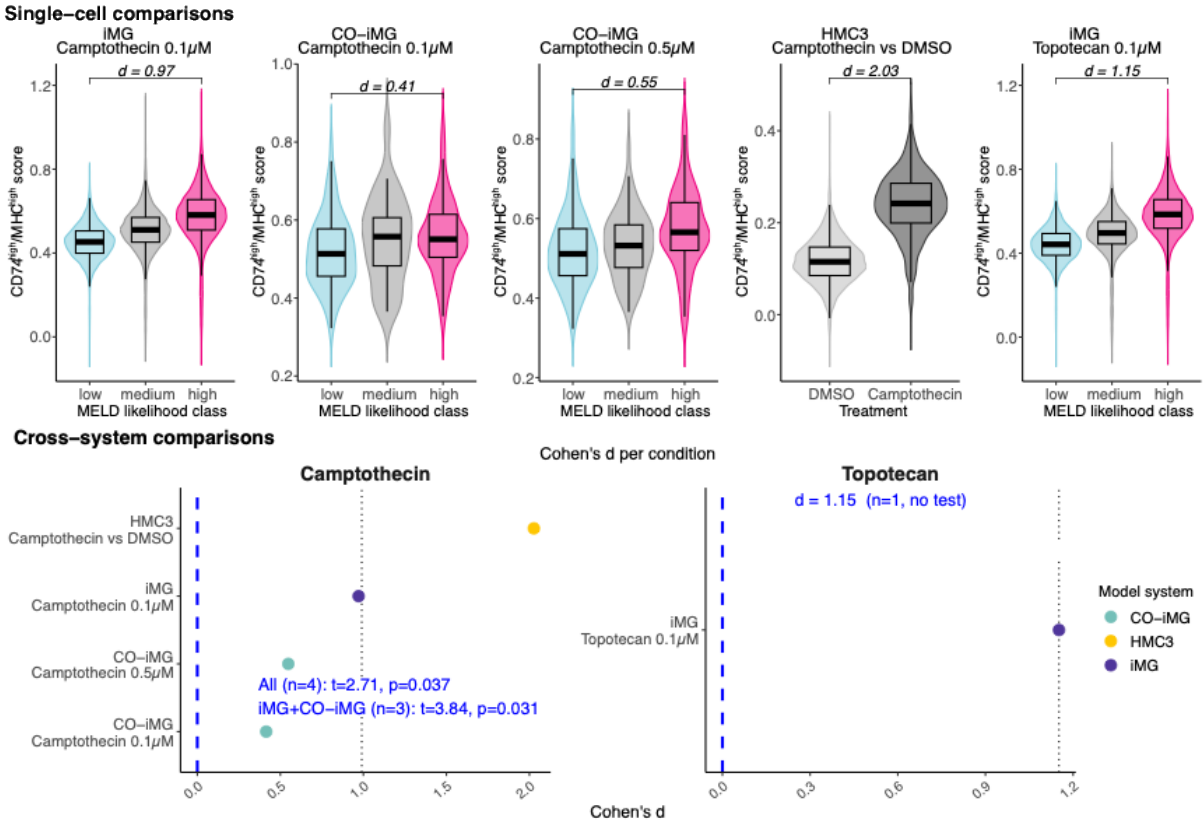
C Top Enriched Pathways - WikiPathway



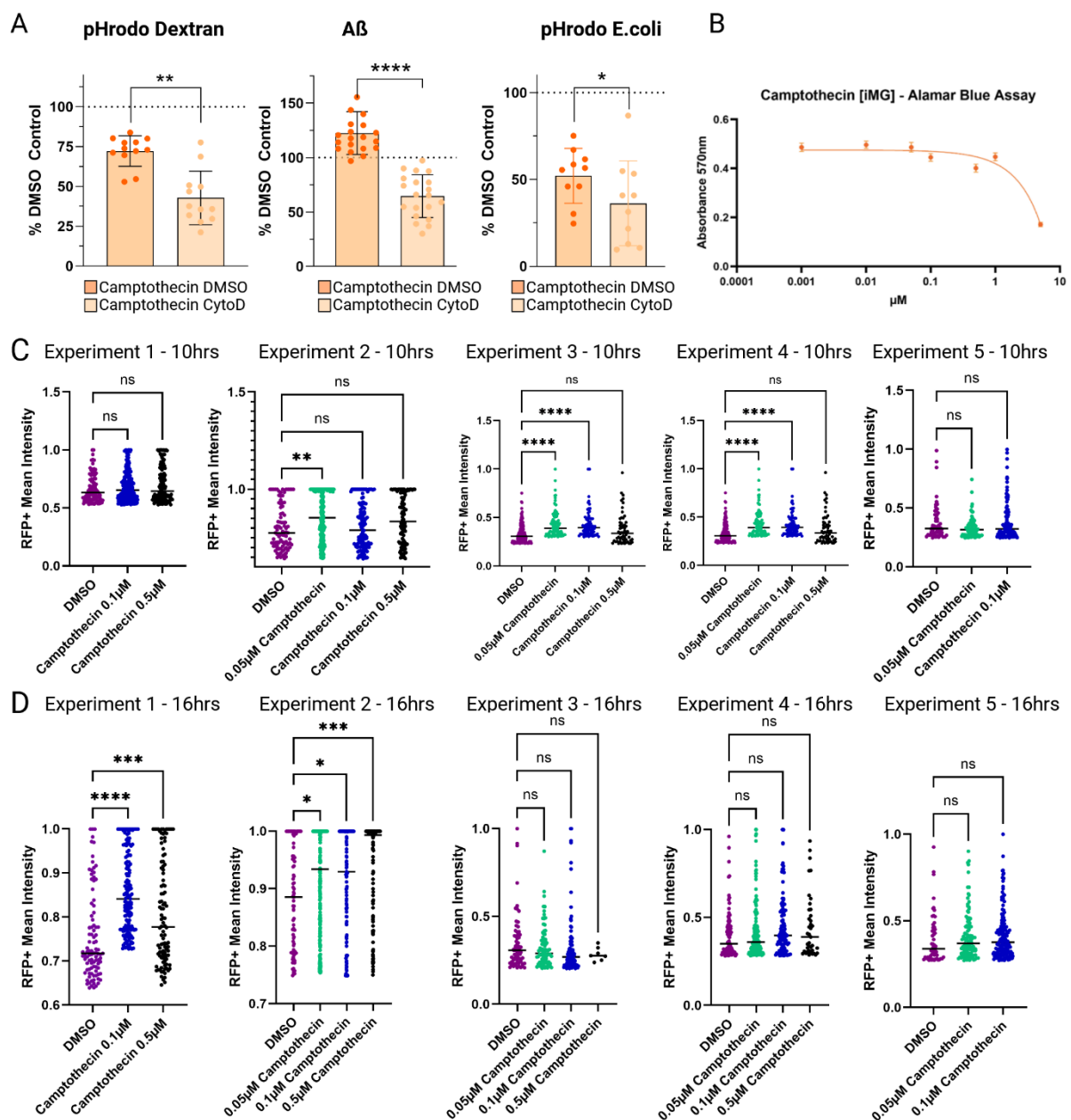
Supplementary Figure S7. Results of GO Pathway analysis, KEGG pathway analysis and Wikipathway analysis for each of the assigned cell types in CO-iMG (microglia, ciliate mesenchymal cells, inhibitory neurons, excitatory neurons, neural precursor cells (NPCs). A. GO Pathway analysis (Biological Process) for all cell types present in the treated cerebral organoids. Pathway analysis of control-treated vs. Camptothecin-treated cells for each of the assigned cell types (microglia, ciliate mesenchymal cells, inhibitory neurons, excitatory neurons, neural precursor cells (NPCs)) using GO pathway analysis and Biological processes. X-axis shows -log10 adjusted p-value, y-axis shows identified pathways. Red bars depict significantly upregulated pathways for each of the cell types; blue bars depict significantly downregulated pathways for each of the cell types. B. KEGG Pathway analysis

for all cell types present in the treated cerebral organoids. Pathway analysis of control-treated vs. Camptothecin-treated cells for each of the assigned cell types (microglia, ciliate mesenchymal cells, inhibitory neurons, excitatory neurons, neural precursor cells (NPCs)) using KEGG pathway analysis. X-axis shows $-\log_{10}$ adjusted p-value, y-axis shows identified pathways. Red bars depict significantly upregulated pathways for each of the cell types; blue bars depict significantly downregulated pathways for each of the cell types. **C. WikiPathways analysis** for all cell types present in the treated cerebral organoids Pathway analysis of control-treated vs. Camptothecin-treated cells for each of the assigned cell types (microglia, ciliate mesenchymal cells, inhibitory neurons, excitatory neurons, neural precursor cells (NPCs)) using WikiPathways pathway analysis. X-axis shows $-\log_{10}$ adjusted p-value, y-axis shows identified pathways. Red bars depict significantly upregulated pathways for each of the cell types; blue bars depict significantly downregulated pathways for each of the cell types.

Supplementary Figure S8



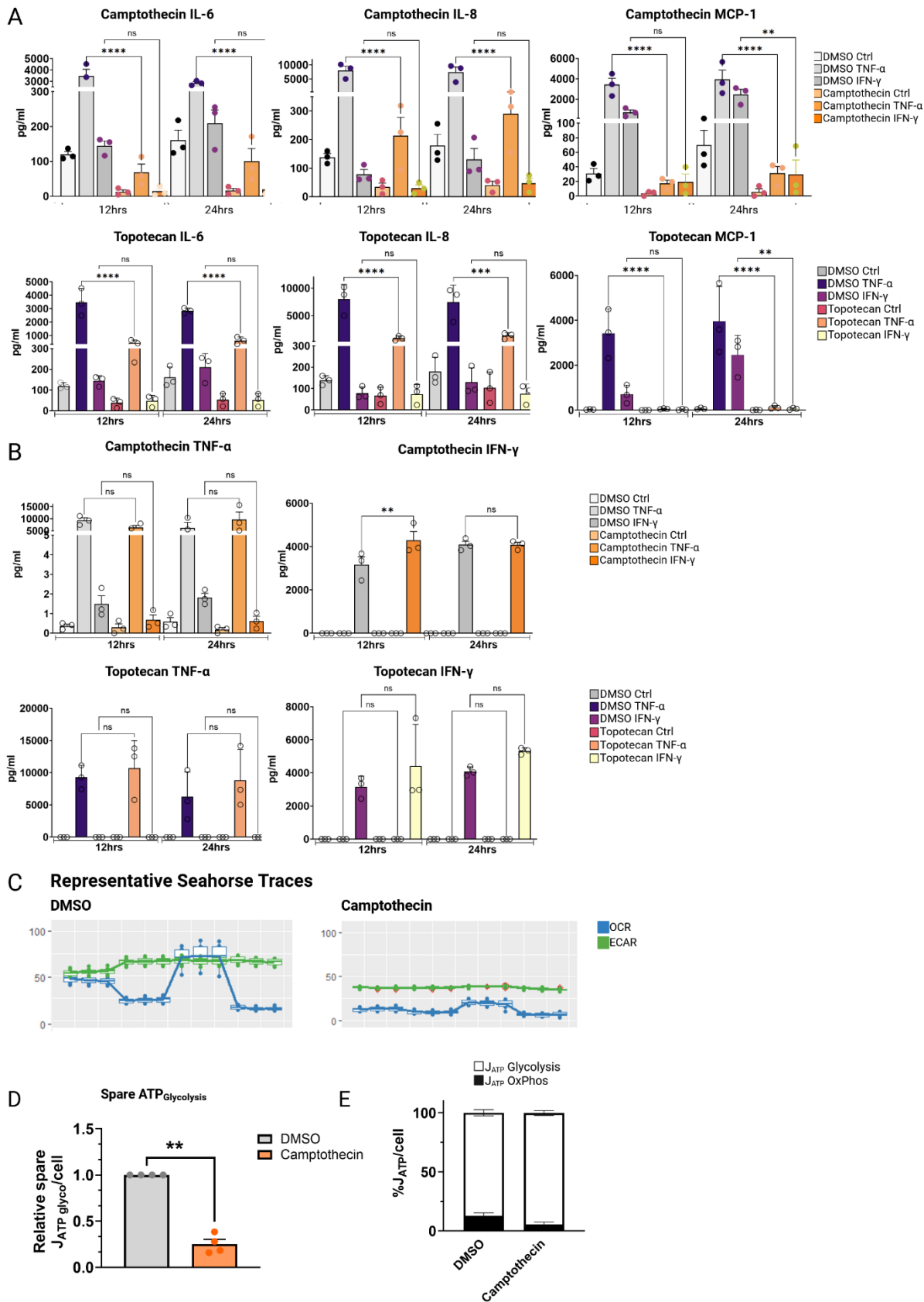
Supplementary Figure S8. Addressing the concerns for pseudoreplication from the small number of single cell data (related to Figure 2). (Top panel) We present violin plots for each of the four scRNAseq datasets from the iMG, CO-iMG, and HMC3 profiles following Camptothecin stimulation. There is also one set of data from Topotecan-treated iMG to the right of the panel. The iPSC-derived data are presented after partitioning using the likelihood that a given cell belongs to a low, medium, or high likelihood group of cells that respond to Camptothecin or Topotecan. Cells are colored by three different levels (low, medium, high) of treatment-associated likelihood for each drug-treatment condition. Each boxplot highlights the median, lower and upper quartiles. Whiskers in boxplots indicate 1.5 times interquartile ranges. (Lower panel) The Cohen's d measure for each of the four Camptothecin-treated samples are shown to the left, and all four are consistent with greater expression of the CD74^{high}/MHC^{high} signature. There is only one Topotecan profile (right side), but its results are consistent with the Camptothecin effects.



Supplementary Figure S9. Compound-treated HMC3 cells and iPSC-derived human microglia exhibit substrate-specific endocytic and phagocytic phenotypes. A. Complete figures of endocytosis and phagocytosis assays following compound treatment (with Camptothecin or DMSO as control) using different substrates (related to Figure 3). For all three assays, HMC3 microglia-like cells were pretreated with Camptothecin (1 μ M) or DMSO as control for 24hrs, subsequently exposed to pHrodo-labeled Dextran (left), Alexa 647-labelled A β 1-42 (middle) or pHrodo-labeled E.coli (right), containing either 5 μ M Cytochalasin D or DMSO as control for Cytochalasin D treatment. After 1hr the uptake of the respective substrate was assessed using flow cytometry. Individual experiments are depicted as individual dots in the bar graphs depicting mean $\pm$ SEM. Phagocytosis was normalized to %

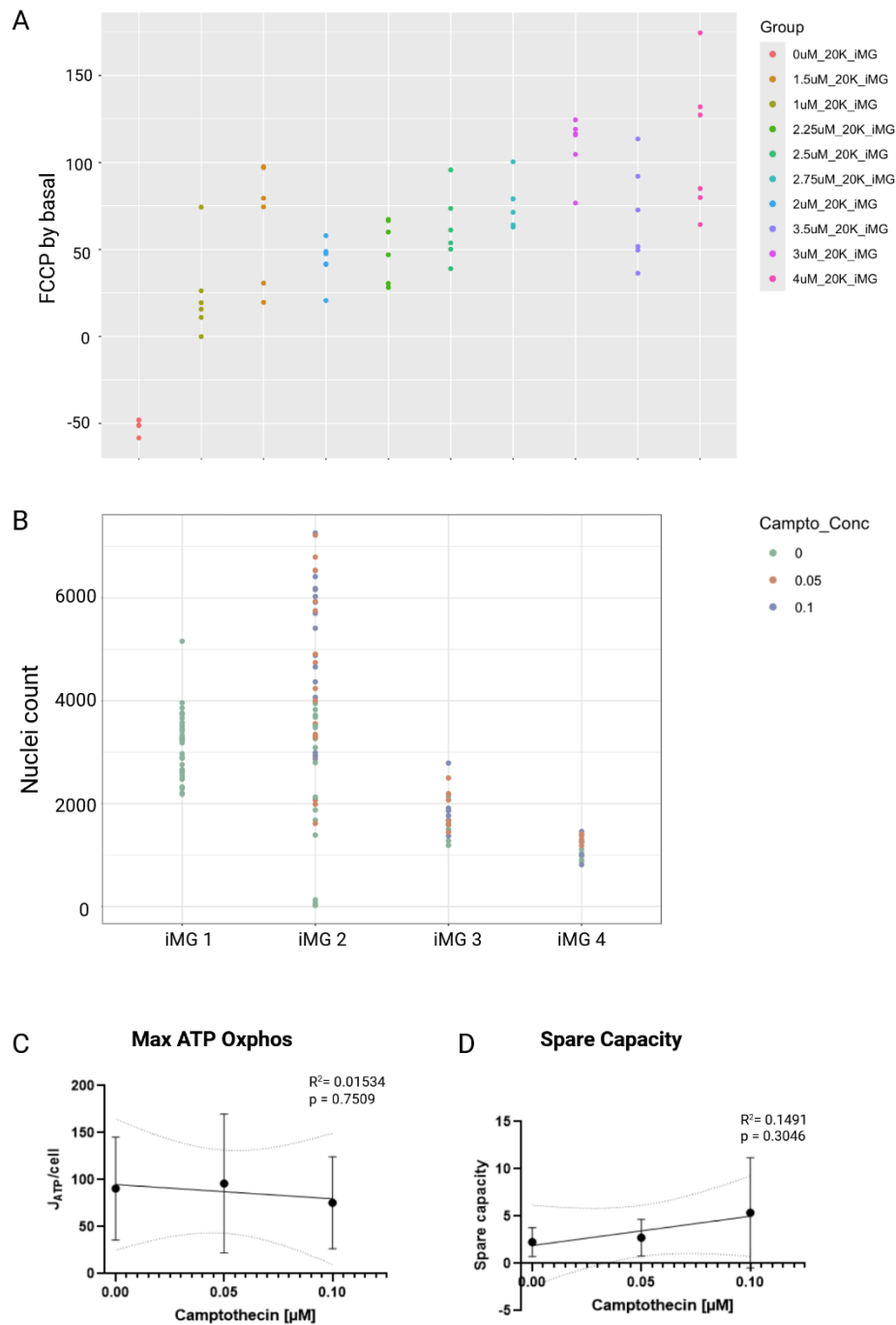
DMSO control and for statistical analysis, log-fold change values of compound- and Cytochalasin D - treated cells in comparison to compound-treated samples was analyzed using Mann-Whitney U test. ns > 0.05, *p.adj ≤ 0.05; **p.adj ≤ 0.01; ***p.adj ≤ 0.001; ****p.adj ≤ 0.0001. pHrodo Dextran – n=9; Aβ – n=20; pHrodo E. coli – n= 10. **B.** Titration experiments with Camptothecin (0μM, 0.001μM, 0.01μM, 0.05μM, 0.1μM, 0.5μM, 1μM, 5μM) were performed on one differentiation batch of iMG (human iPSC-derived microglia). Readout was performed via Alamar Blue assay, capturing absorbance at 570nm. Dots represent mean of 8 technical replicates per concentration ± SD. **C.-D. Graphs depicting results of 4-5 independent amyloid-beta phagocytosis assays using human iPSC-derived microglia** (iMG; 0.05μM Camptothecin– n=4; 0.1μM Camptothecin – n=5; 0.5μM Camptothecin – n=4). **Assays were analyzed after 10hrs (C) and were performed over a total time of 16hrs (D) with hourly readout of fluorescence signal.** Each graph represents the mean RFP intensity of all cells above the DMSO mean + 2 SD (standard deviations) across conditions (DMSO – purple; 0.05μM Camptothecin – green; 0.1μM Camptothecin – blue; 0.5μM Camptothecin - black) for each of the experiments analyzed. For statistical analysis, one-way ANOVA followed by Dunnett's multiple comparison test was performed. ns > 0.05, *p.adj ≤ 0.05; **p.adj ≤ 0.01; ***p.adj ≤ 0.001; ****p.adj ≤ 0.0001.

Supplementary Figure S10



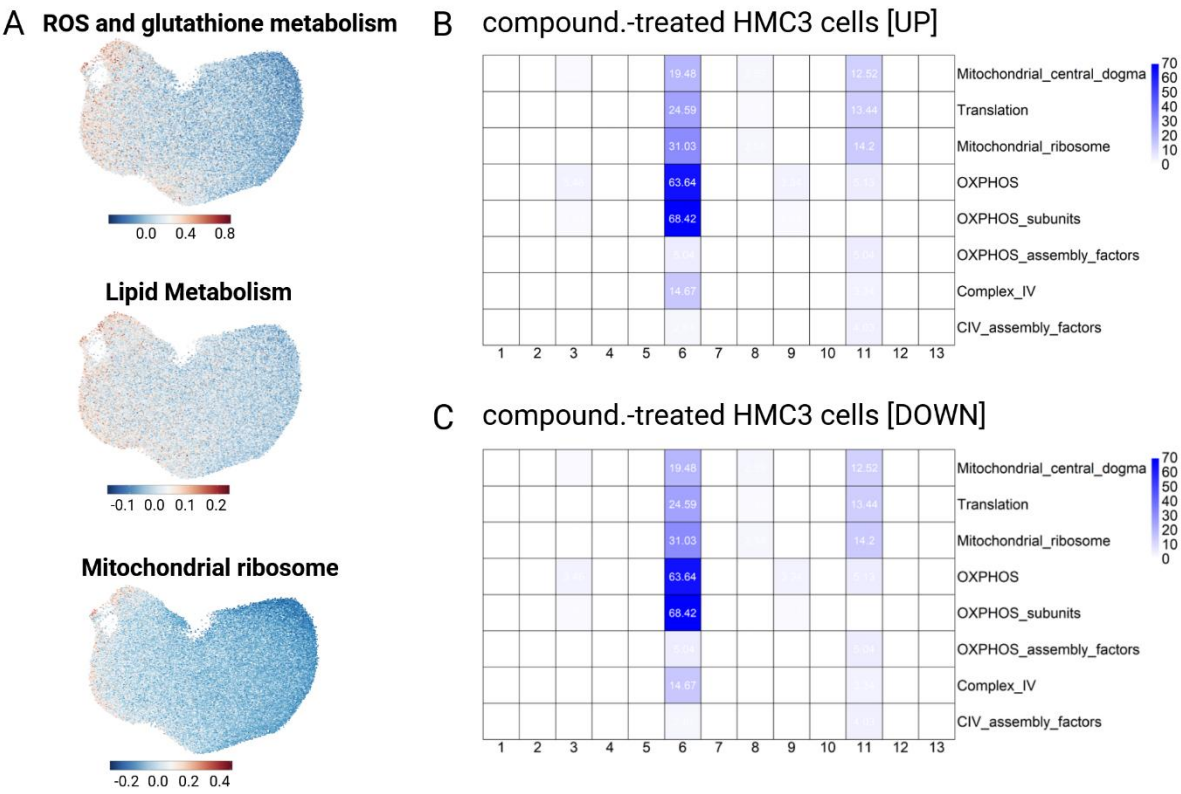
Supplementary Figure S10. Extended analysis of cytokine secretion and MitoStress test on HMC3 cells treated with Camptothecin. A-B. Cytokine secretion of Camptothecin-,

Topotecan- or DMSO-pretreated HMC3 cells (24hrs) followed by stimulation with either TNF-
a (0.3 µg/mL), IFN-γ (0.3 µg/mL) or H₂O as control for 12 or 24hrs (**related to Figure 3**). Pro-
inflammatory cytokine secretion was assessed using a human pro-inflammatory cytokine
discovery assay. Bar graphs depict measured amount of cytokines IL-6, IL-8, MCP-1 (**A**) or
TNF-α and IFN-γ (**B**) (mean ± SEM) in pg/ml for DMSO control-treated samples (white, light
grey, grey) or Camptothecin-treated samples (1µM, orange). Each dot represents a biological
replicate (n= 3 per condition). For statistical analysis, one-way ANOVA followed by Tukey's
multiple comparisons test with a single pooled variance was performed. ns > 0.05, *p.adj ≤
0.05; **p.adj ≤ 0.01; ***p.adj ≤ 0.001; ****p.adj ≤ 0.0001. **C-E. MitoStress test on HMC3 cells**
treated with Camptothecin or DMSO (n= 4 biological replicates per condition), depicting
representative traces (**C**; OCR – Oxygen Consumption Rate; ECAR - Extracellular Acidification
Rate), ATP Glycolysis (**D**; p= 0.0047), ATP generated by Glycolysis or OxPhos relative to
DMSO (**E**). Data shows mean ± SEM. For statistical analysis, Welch's t-test was performed.
ns > 0.05, *p.adj ≤ 0.05; **p.adj ≤ 0.01; ***p.adj ≤ 0.001; ****p.adj ≤ 0.0001.

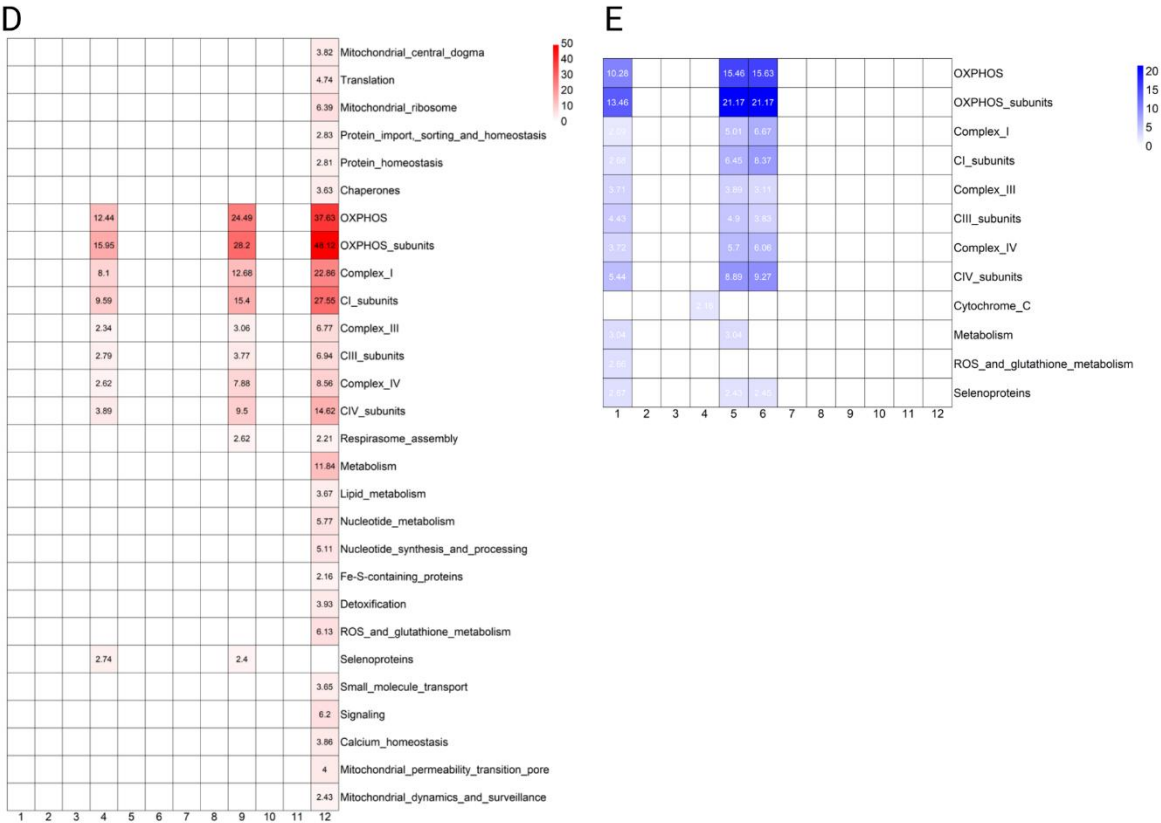


Supplementary Figure S11. Seahorse optimization and analysis in iPSC-derived human microglia (related to Figure 3). **A.** Titration curve for FCCP tested in 20K seeded iPSC-derived human microglia of iMG (human iPSC-derived microglia) differentiation batch 1 (iMG1). Concentrations tested: 1 μ M, 1.5 μ M, 2 μ M, 2.25 μ M, 2.5 μ M, 2.75 μ M, 3 μ M, 3.5 μ M, 4 μ M. **B.** Graph shows nuclei count across the different iMG batches used for seahorse assay (iMG1 – FCCP titration; iMG2-4 – seahorse assay). y-axis depicts nuclei counts in thousands; x-axis depicts the iMG batches. Colored dots depict counts based on their treatment identity (green

– DMSO control/ 0 μ M Camptothecin concentration = 0; red – 0.05 μ M Camptothecin; blue – 0.1 μ M Camptothecin). **C-D**. Linear regression analysis for seahorse parameters assessed in iMG (Max ATP Oxphos (**C**) and Spare Capacity (**D**)). Datapoints indicate mean $\pm$ SD for each treatment condition (DMSO, 0.05 μ M Camptothecin, 0.1 μ M Camptothecin). Simple linear regression was performed and dotted lines indicate 95% confidence bands of the best-fit line (continuous line), goodness of fit is indicated by R^2 value and p-value indicates whether slope is significantly non-zero. All statistical results are depicted in **Supplementary Table S5**.

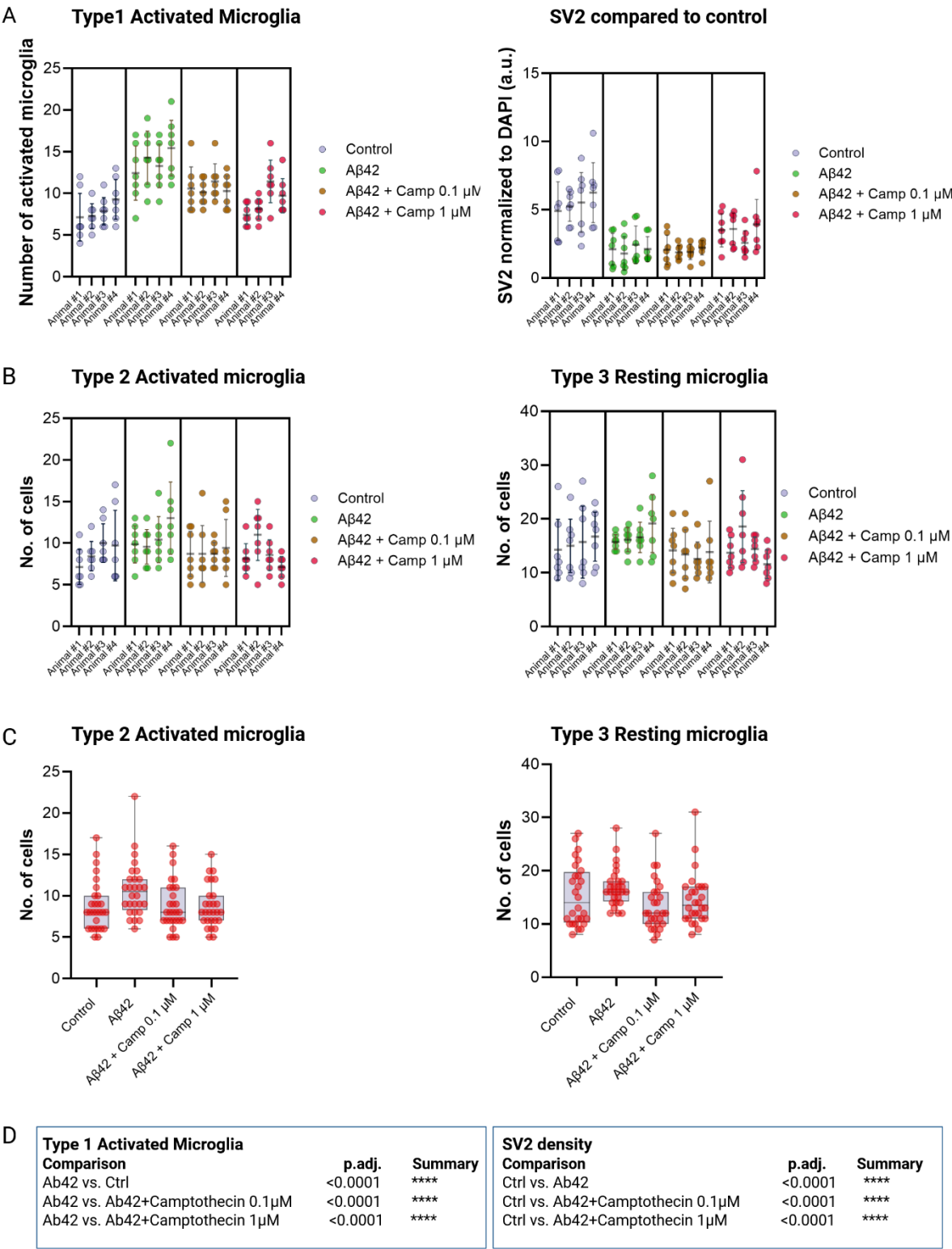


Freshly isolated human microglia [Tuddenham et al. 2024]



Supplementary Figure S12. Mitotype analysis in human microglia (related to Figure 4).

A. Selected results of GSEA for specific mitotypes in human microglial subtypes are shown. Legend depicts module scores, showing average expression for module genes vs. a background of similarly expressed control genes. **B. Mitochondrial phenotyping identifies clusters enriched in different aspects of respiratory metabolism (compound-treated HMC3 microglia-like cells).** Enrichment of modules of genes associated with different aspects of mitochondrial activity was performed in upregulated gene lists associated with each cluster (compound-treated HMC3 cells) using the hypergeometric test with Benjamini-Hochberg correction. Bars are colored if they have an FDR < 0.01. **C. Mitochondrial phenotyping identifies clusters downregulated in different aspects of respiratory metabolism (compound-treated HMC3 microglia-like cells).** Enrichment of modules of genes associated with different aspects of mitochondrial activity was performed in downregulated gene lists associated with each cluster (compound-treated HMC3 cells) using the hypergeometric test with Benjamini-Hochberg correction. Bars are colored if they have an FDR < 0.01. **D. Mitochondrial phenotyping identifies clusters enriched in different aspects of respiratory metabolism (freshly isolated human microglia; ⁵).** Enrichment of modules of genes associated with different aspects of mitochondrial activity was performed in upregulated gene lists associated with each cluster (freshly isolated human microglia;⁵) using the hypergeometric test with Benjamini-Hochberg correction. Bars are colored if they have an FDR < 0.01. **E. Mitochondrial phenotyping identifies clusters downregulated in different aspects of respiratory metabolism.** Enrichment of modules of genes associated with different aspects of mitochondrial activity was performed in downregulated gene lists associated with each cluster (freshly isolated human microglia; ⁵) using the hypergeometric test with Benjamini-Hochberg correction. Bars are colored if they have an FDR < 0.01.



474 **Supplementary Figure S13. Analysis of dose-dependent effects of Camptothecin on**
475 **microglial activation and synaptic density in a zebrafish amyloidosis model (related to**
476 **Figure 5). A.** Quantification of Type 1 microglia (left) and SV2 immunoreactivity (right) per
477 animal (n = 4 per group) presented as nested plot with individual data points from each animal.
478 n = 4 independent animals per condition with 7 images analyzed per animal. Each dot
479

represents an independent image analyzed. Conditions: controls (light blue), A β (green), A β + 0.1 μ M Camptothecin (brown), A β + 1 μ M Camptothecin (pink). All data represents mean $\pm$ SEM. **B.** Quantification of Type 2 microglia (left) and type 3 microglia (right) per animal (n = 4 per group) presented as nested plot with individual data points from each animal. n = 4 independent animals per condition with 7 images analyzed per animal. Each dot represents an independent image analyzed. Conditions: controls (light blue), A β (green), A β + 0.1 μ M Camptothecin (brown), A β + 1 μ M Camptothecin (pink). All data represents mean $\pm$ SEM. **C.** Quantification of Type 2 microglia (left) and type 3 microglia (right) per animal (n = 4 per group) presented as grouped boxplot. All data represents mean $\pm$ SEM. n = 4 independent animals per condition with 7 images analyzed per animal. Two-way ANOVA followed by Tukey's post hoc test was used for statistical comparisons. Significance is indicated as: ns > 0.05, *p.adj $\leq$ 0.05; **p.adj $\leq$ 0.01; ***p.adj $\leq$ 0.001; ****p.adj $\leq$ 0.0001. **D. Table summarizing statistical results of quantifications of Type 1 activated microglia and synaptic density.** For statistical analysis, two-way ANOVA followed by Tukey's post hoc test was performed. ns > 0.05, *p.adj $\leq$ 0.05; **p.adj $\leq$ 0.01; ***p.adj $\leq$ 0.001; ****p.adj $\leq$ 0.0001.

References

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Supplementary Tables

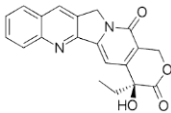
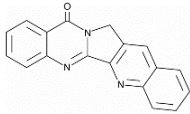
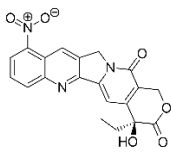
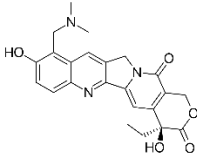
Supplementary Table S11. Overview of compounds and concentrations used for iMG treatment.

Compound	Vendor	Cat#	Cluster signature	Concentration used for <i>in vitro</i> iMG experiments
Camptothecin	EMD Millipore	390238	CD74 ^{high} /MHC ^{high}	0.1µM
Topotecan	APExBIO	B4982	CD74 ^{high} /MHC ^{high}	0.1µM

Supplementary Table S12. Overview of compounds used for zebrafish experiments.

Compound	Treatment concentration	Manufacturer	Cat #:
Camptothecin	1 µM and 0.1 µM	EMD Millipore	390238

537 **Supplementary Table S13. Overview of compounds assessed in structure activity**
538 **relationship experiments.**

Compound	Structure	Vendor	Cat#	Cluster signature	Final concentration used for <i>in vitro</i> experiments
Camptothecin		EMD Millipore	390238	CD74 ^{high} / MHC ^{high}	1μM
Luotonin A		Sigma-Aldrich	L4045- 10MG	CD74 ^{high} / MHC ^{high}	1μM
Rubitecan		Sigma-Aldrich	R3655- 10MG	CD74 ^{high} / MHC ^{high}	0.01μM
Topotecan		APExBIO	B4982	CD74 ^{high} / MHC ^{high}	0.1μM