Supplementary Information

Human Placental-Derived Adherent Stromal Cells Co-Induced with TNF-α and IFN-γ Inhibit Triple-Negative Breast Cancer in Nude Mouse Xenograft Models

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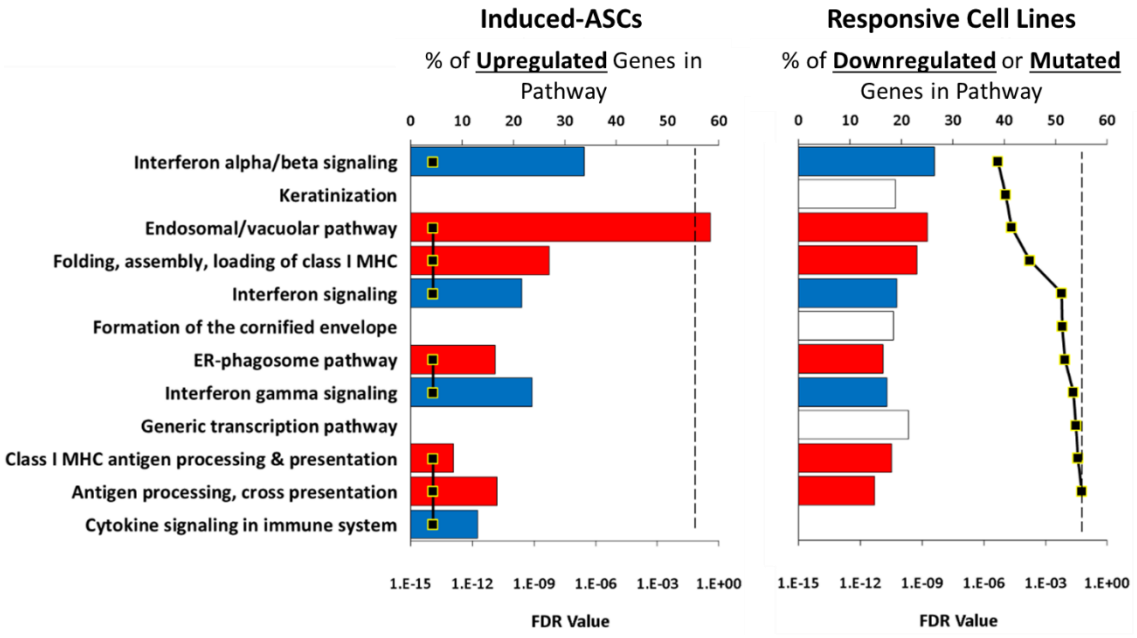
Supplementary Table S1: Time-Dependent Concentration of Cytokines in the Conditioned Medium of TNF-α/IFN-γ-Induced Human Placental-Derived ASCs

Cytokine	Days from Seeding						Non-Induced ASCs
	2	5	7	9	12	14	
CXCL8	16,887 ± 9,069	3,723 ± 2,644	4,546 ± 547	681 ± 71	2,268 ± 144	1,151 ± 239	313
CCL2	2,082 ± 448	1,109 ± 491	522 ± 241	281 ± 134	362 ± 168	1,330 ± 872	98
CCL5	16,549 ± 7,105	3,399 ± 755	627 ± 166	139 ± 46	27 ± 10	15 ± 02	0
GM-CSF	365 ± 107	28 ± 7	41 ± 10	08 ± 02	16 ± 03	19 ± 03	3
G-CSF	1,501 ± 358	40 ± 5	2 ± 2	2 ± 2	4 ± 4	5 ± 5	0
IL-6	5,604 ± 1,609	341 ± 94	203 ± 9	49 ± 6	79 ± 1	117 ± 12	8
CXCL10	4,941 ± 963	245 ± 63	2 ± 1	2 ± 0	2 ± 1	5 ± 2	0
IFN-γ	366 ± 44	91 ± 11	47 ± 10	9 ± 2	13 ± 2	10 ± 1	8
CXCL9	13,465 ± 2,632	4,163 ± 49	0	25 ± 7	0	0	0
HGF	133 ± 55	115 ± 45	16 ± 8	16 ± 9	22 ± 7	41 ± 22	158

Supplementary Methods for Table S1

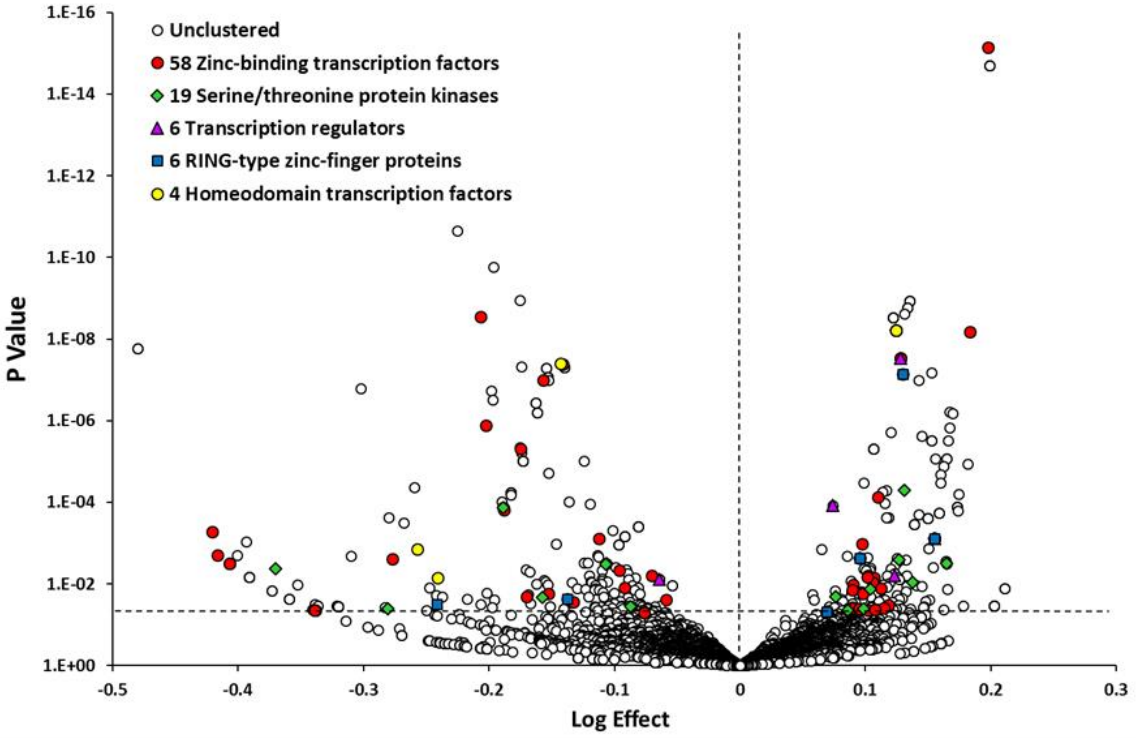
Induced-ASCs (3 different cell batches) were seeded $2x10^5$ cells/well in 6 well plates in DMEM low glucose supplemented with 2 mM L-glutamine, 10% FBS and 50 μg/ml gentamycin. Cells were passaged 5 and 9 days after seeding (when reaching 80-90% confluence). Two, five, seven, nine, 12 and 14 days from the day of seeding CM was collected from two wells as described, and cells were counted. Concentration of 10 highly expressed cytokines (CXCL8, CCL2, CCL5, GM-CSF, G-CSF, IL-6, CXCL10, IFN- γ, CXCL9 and HGF) were measured using custom made Luminex panels. Absolute concentrations as measured by Luminex in each well were divided by the number of cells and days since last medium renewal to calculate pg/10^6 cells/day. Concentrations are expressed as mean ± SEM (n=3).

Supplementary Figure S1: Interferon Signaling and MHC Class I Antigen Processing and Presentation Pathways Enriched in Key Gene Sets



The most statistically significant pathways enriched in genes upregulated in TNF-α/IFN-γ-induced placental-derived ASCs are shown on the left. Pathways enriched in genes downregulated and/or exclusively mutated in the responsive cell lines are shown on the right. Blue represents cytokine signaling in the immune system, and red represents MHC class I mediated antigen processing and presentation. The false discovery rate (FDR) value, the corrected enrichment probability for each pathway, is shown by the thick black line. Dashed vertical line represents FDR=0.05.

Supplementary Figure S2: Mutations in Transcription Factors and Protein Kinases Correlate with the Anti-Proliferative Response to CM from TNF-α/IFN-γ-Induced Placental-Derived ASCs



Mutation association plot showing somatic mutations in cancer cell lines that positively or negatively correlate with the anti-proliferative response to CM from induced placental-derived ASC. Each of the 6666 data points (whether colored or not) represent an individual mutated gene. Those with a positive log effect correlate with a pro-proliferative effect; those with a negative log effect correlate with an anti-proliferative effect. 611 genes appear above the horizontal dashed line, which represents p=0.05. The statistical significance of the five clusters found in these 611 genes are as follows: zinc-binding transcription factors (p=0.00013), serine/threonine protein kinases (p=0.0011), transcription regulators (p=0.0066), RING-type zinc-finger proteins (p=0.011), and homeodomain transcription factors (p=0.034).