

Supplementary information for

Pro-inflammatory polarization primes macrophages to transition into a distinct M2-like phenotype in response to IL-4

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Supplemental Table 1. RT-PCR primer sequences

Gene	Forward Sequence	Reverse Sequence
AXL	CCGTGGACCTACTCTGGCT	CCTTGGCGTTATGGGCTTC
CMKLR1	CAGTTACGGTGATGAATACCCTG	GACGATGCTGTAGACCACCAC
IL10RB	TACCACCTCCCGAAAATGTCA	CCCAGTCTGAATGCTCATCTG
MERTK	GTGCAGCGTTCAGACAATGG	TCGATGTAGATGGGATCAGACAC
NR3C1	TGCCGCTATCGAAAATGTCTT	GGGTAGGGGTGAGTTGTGGT
TLR2	TTATCCAGCACACGAATACACAG	AGGCATCTGGTAGAGTCATCAA
TNFR2	TTCATCCACGGATATTTGCAGG	GCTGGGGTAAGTGTACTGCC
GAPDH	TGCACCACCAACTGCTTAGC	GGCATGGACTGTGGTCATGAG

Supplemental Table 2. Custom Nanostring CodeSet – Dose response

Gene	Accession Number	Gene	Accession Number
ACAA2	NM_006111.2	GAPDH	NM_001256799.1
AKAP5	NM_004857.3	FAM198A	NM_001129908.2
ALOX15	NM_001140.3	HSPG2	NM_005529.5
ANGPTL4	NR_104213.1	IL10	NM_000572.2
AQP9	NM_020980.3	IL16	NM_004513.4
AXL	NM_021913.2	IL17RA	NM_014339.4

CACNB4	NM_000726.3	IL18	NM_001562.2
CASP4	NM_001225.3	IL1B	NM_000576.2
CASP8	NM_001080124.1	IL6	NM_000600.3
CBR1	NM_001757.2	IRF4	NM_002460.1
CCL17	NM_002987.2	ITGAV	NM_002210.2
CCL18	NM_002988.2	JAG1	NM_000214.2
CCL2	NM_002982.3	KLF4	NM_004235.4
CCL22	NM_002990.3	KRT1	NM_006121.2
CCL26	NM_006072.4	LY6E	NM_002346.2
CCL3	NM_002983.2	MARCO	NM_006770.3
CCL4	NM_002984.2	MMP10	NM_002425.1
CCL5	NM_002985.2	MMP27	NM_022122.2
CCL7	NM_006273.2	MMP8	NM_002424.2
CCL8	NM_005623.2	MMP9	NM_004994.2
CCR1	NM_001295.2	MRC1	NM_002438.2
CCR2	NM_001123041.2	MSR1	NM_002445.3
CCR3	NM_001837.2	NDUFA4	NM_002489.2
CCR7	NM_001838.2	NLRC3	NM_178844.2
CD163	NM_004244.4	NOS3	NM_000603.4
CD180	NM_005582.2	NOTCH4	NM_004557.3
CD209	NM_001144899.1	OTUB1	NM_017670.2
CD22	NM_001771.2	PARM1	NM_015393.3
CD274	NM_014143.3	PDCD1LG2	NM_025239.3
CD28	NM_001243078.1	PDGFB	NM_033016.2
CD33	NM_001177608.1	PDPN	NM_006474.4
CD37	NM_001774.2	CD31	NM_000442.3
CD38	NM_001775.2	PF4	NM_002619.2

CD44	NM_001001392.1	PPT1	NM_000310.3
CD47	NM_001777.3	PTGS1	NM_000962.2
CD80	NM_005191.3	RAMP1	NM_005855.2
CD93	NM_012072.3	RBM11	XM_017028384.2
CDH1	NM_004360.2	RETNLB	NM_032579.2
CD144	NM_001795.3	RPL4	NM_000968.2
CH25H	NM_003956.3	RPS23	NM_001025.4
CISH	NM_145071.2	SERPINB6	NM_004568.4
CLEC10A	NM_182906.2	SIGLEC12	NM_033329.1
CRB2	NM_173689.6	SIGLEC6	NM_001177548.1
CX3CR1	NM_001337.3	SIGLEC8	NM_014442.2
CXCL2	NM_002089.3	SOCS1	NM_003745.1
IL8	NM_000584.2	ST8SIA6	NM_001004470.1
CXCL9	NM_002416.1	TAL1	NM_003189.2
CXCR3	NM_001504.1	TBP	NM_001172085.1
CXCR4	NM_003467.2	TEK	NM_000459.3
CYFIP1	NM_014608.2	TFRC	NM_003234.1
DACT1	NM_001079520.1	TGFA	NM_003236.2
DUOX1	NM_175940.1	TGM2	NM_004613.2
DUOXA1	NM_001276265.1	TLR3	NM_003265.2
EGFL7	NM_016215.3	TLR6	NM_006068.2
EHF	NM_012153.3	TLR7	NM_016562.3
ESPNL	NM_194312.2	TLR8	NM_016610.2
FAM110B	NM_147189.2	TNF	NM_000594.2
FCER2	NM_002002.4	VEGFA	NM_001025366.1
GALNT18	NM_198516.1	VEGFB	NM_003377.3

Supplemental Table 3. Custom Nanostring CodeSet – M1→M2 phenotyping

Gene	Accession Number	Gene	Accession Number
ACTA2	NM_001613.1	FOXO4	NM_001170931.1
AGGF1	NM_018046.3	FST	NM_006350.2
ANG	NM_001145.4	FYN	NM_002037.3
BGN	NM_001711.3	GAPDH	NM_001256799.1
BTG1	NM_001731.2	HSPG2	NM_005529.5
CABLES1	NM_001100619.2	IDO1	NM_002164.5
CCL15	NM_032965.4	IGF1	NM_000618.3
CCL17	NM_002987.2	IL1B	NM_000576.2
CCL18	NM_002988.2	IL6	NM_000600.3
CCL2	NM_002982.3	JAG1	NM_000214.2
CCL22	NM_002990.3	LUM	NM_002345.3
CCL24	NM_002991.2	MARCO	NM_006770.3
CCL26	NM_006072.4	MMP2	NM_004530.2
CCL5	NM_002985.2	MMP9	NM_004994.2
CCL8	NM_005623.2	MRC1	NM_002438.2
CTGF	NM_001901.2	PDGFA	NM_002607.5
CCR7	NM_001838.2	PDGFB	NM_033016.2
CD163	NM_004244.4	PDGFC	NM_016205.2
CD200R1	NM_138806.3	PDGFRA	NM_006206.3
CD80	NM_005191.3	PDGFRB	NM_002609.3
CLEC10A	NM_182906.2	RAMP1	NM_005855.2
COL1A1	NM_000088.3	SPP1	NM_000582.2
COL3A1	NM_000090.3	STAT3	NM_003150.3
COL5A1	NM_000093.3	STAT6	NM_003153.3

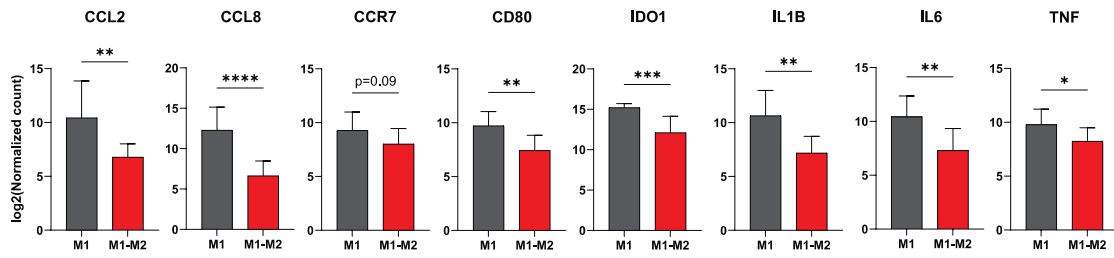
CTNNB1	NM_001098210.1	TBP	NM_001172085.1
CXCL12	NM_199168.3	TGFB1	NM_000660.3
CXCR4	NM_003467.2	TIE1	NM_005424.2
DACT1	NM_001079520.1	TIMP3	NM_000362.4
DCN	NM_001920.3	TNF	NM_000594.2
EGFL7	NM_016215.3	TNFRSF11A	NM_003839.3
ETS1	NM_005238.3	VCAN	NM_004385.3
FGF2	NM_002006.4	VEGFA	NM_001025366.1
FLT1	NM_002019.4	VEGFB	NM_003377.3
FN1	NM_212482.1	VEGFC	NM_005429.2
FOXO1	NM_002015.3	VIM	NM_003380.2
FOXO3	NM_001455.2	WNT5A	NM_003392.3

Supplemental Table 4. GO Enrichment Analysis of M1→M2 Phenotype

Biological Process	M1→M2 Fold Enrichment
Basophil chemotaxis	>100
Metanephric glomerular mesangial cell development	>100
Chemoattraction of serotonergic neuron axon	>100
Planar cell polarity pathway involved in midbrain dopaminergic neuron differentiation	>100
Negative regulation of cell proliferation in midbrain	>100
Positive regulation of vascular associated smooth muscle cell dedifferentiation	>100
Endocardial cushion cell development	>100
Planar cell polarity pathway involved in pericardium morphogenesis	>100
Planar cell polarity pathway involved in cardiac muscle tissue morphogenesis	>100
Planar cell polarity pathway involved in cardiac right atrium morphogenesis	>100
Planar cell polarity pathway involved in ventricular septum morphogenesis	>100
Planar cell polarity pathway involved in outflow tract morphogenesis	>100
Non-canonical Wnt signaling pathway involved in heart development	>100

Positive regulation of protein kinase D signaling	>100
Regulation of protein kinase D signaling	>100
Negative regulation of endothelial cell differentiation	>100
Positive regulation of protein kinase C signaling	>100
Positive regulation of T cell apoptotic process	>100
Positive regulation of T cell chemotaxis	>100
Regulation of T cell chemotaxis	>100
Dendritic cell chemotaxis	>100
Positive regulation of lymphocyte apoptotic process	>100
Regulation of protein kinase C signaling	>100
Positive regulation of lymphocyte chemotaxis	>100
Regulation of cell-cell adhesion mediated by cadherin	>100
Dendritic cell migration	>100
Cell surface receptor signaling pathway involved in heart development	>100
Positive chemotaxis	>100
Regulation of lymphocyte chemotaxis	>100
Monocyte chemotaxis	>100
Positive regulation of leukocyte apoptotic process	>100
Positive regulation of protein autophosphorylation	>100
Positive regulation of T cell migration	>100
Dopaminergic neuron differentiation	>100
Regulation of axon extension involved in axon guidance	>100
Activation of protein kinase B activity	>100
Epithelial cell differentiation involved in kidney development	>100
Platelet-derived growth factor receptor signaling pathway	>100
Regulation of endothelial cell differentiation	>100
Positive regulation of lymphocyte migration	>100
Regulation of T cell apoptotic process	>100
Positive regulation of smooth muscle cell migration	>100
Regulation of T cell migration	>100
Negative regulation of epithelial cell differentiation	>100
Outflow tract morphogenesis	>100
Cell differentiation involved in kidney development	>100
Negative regulation of fat cell differentiation	>100

Regulation of protein autophosphorylation	>100
Morphogenesis of an epithelial sheet	>100
Lymphocyte chemotaxis	>100
Positive regulation of fibroblast proliferation	>100
Positive regulation of cytokine-mediated signaling pathway	>100
Regulation of morphogenesis of a branching structure	>100



Supplemental Figure 1: Downregulation of M1 markers in M1-M2 macrophages. Data presented as mean \pm SD, n=10 donors. Two-tailed unpaired t-test, *p<0.05, **p<0.01, ***p<0.001, ****p<0.0001.