

## SUPPORTING INFORMATION

**Figure S1. Experimental design and quality control.** (A) Schematic representation of the experimental design showing time points for generating the datasets and assays to confirm quiescence and senescence. Purple indicates quiescent cells treated with vehicle (PBS), whereas red indicates senescent cells treated with bleomycin. (B-C) Quality control assays confirming the induction of senescence. (B) Senescence-associated beta galactosidase (SA-B-gal) and EdU incorporation analysis of quiescent (PBS-treated) and senescent (bleomycin-treated) samples. (C) IL-6 ELISA confirming establishment of a SASP in senescent cells.

**Figure S2. Detection of gene transcripts varies by method and cell type.** (A) Rank ordered success rates by gene for all samples in our data sets. (B) Success rates by gene for C1 vs MI methods. (C) Success rates by gene for either senescent (SEN) or quiescent (QUI) cell populations.

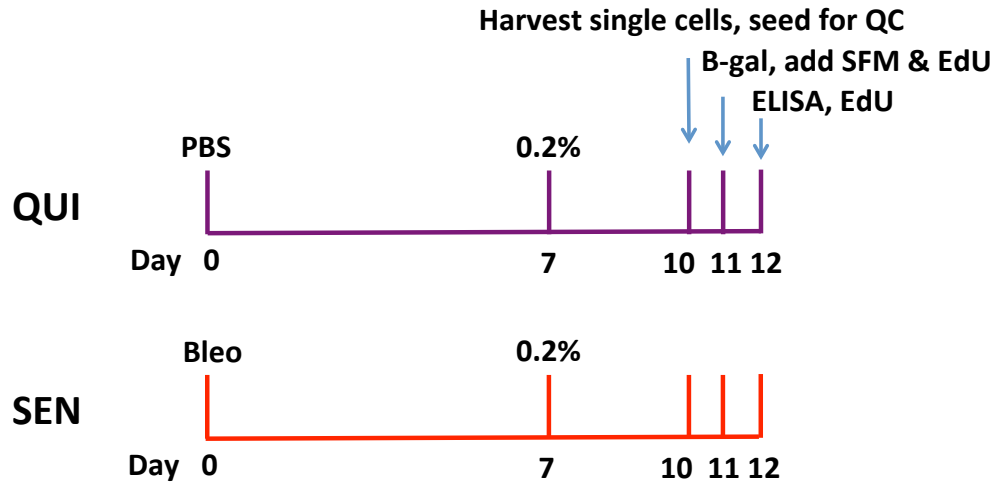
**Figure S3. Normalization of datasets.** Density maps for gene expression analyzed by C1 or MI before (A) and after (B) normalization.

**Figure S4. Examples of genes with strong positive or negative correlations.** Relative gene expression values for each single cell were plotted against each other. Each axis indicates gene expression values for each single cell; red dots indicate senescent cells, blue dots indicate quiescent cells. (A) Example of a strong positive correlation: GAPDH plotted against vimentin. (B) Example of a strong negative correlation: AGER plotted against GAPDH.

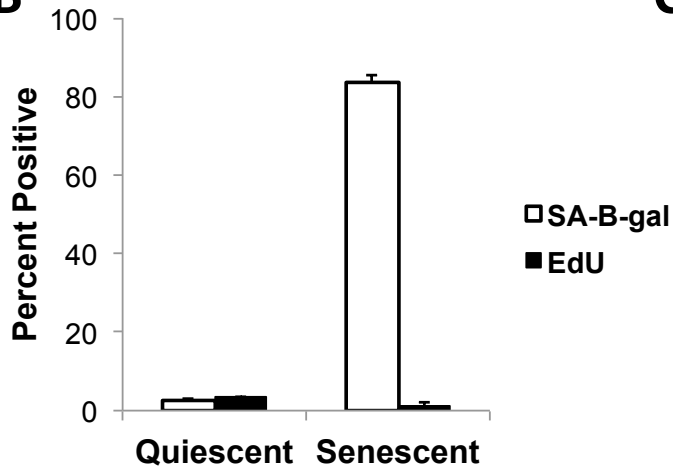
**Figure S5. Pathway analysis of Class 1 and Class 2 genes.** Pathway enrichment analysis of Class 1 (above) and Class 2 (below) genes, sorted by p-value.

# Figure S1

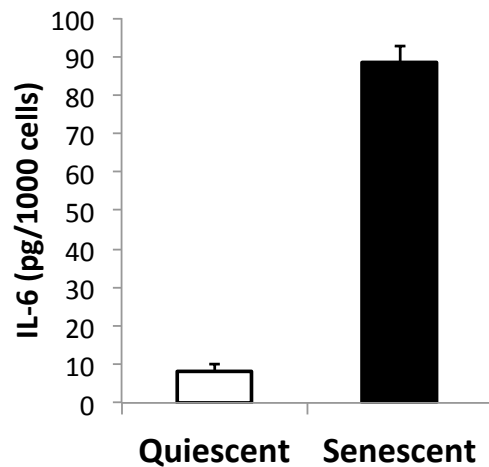
## A



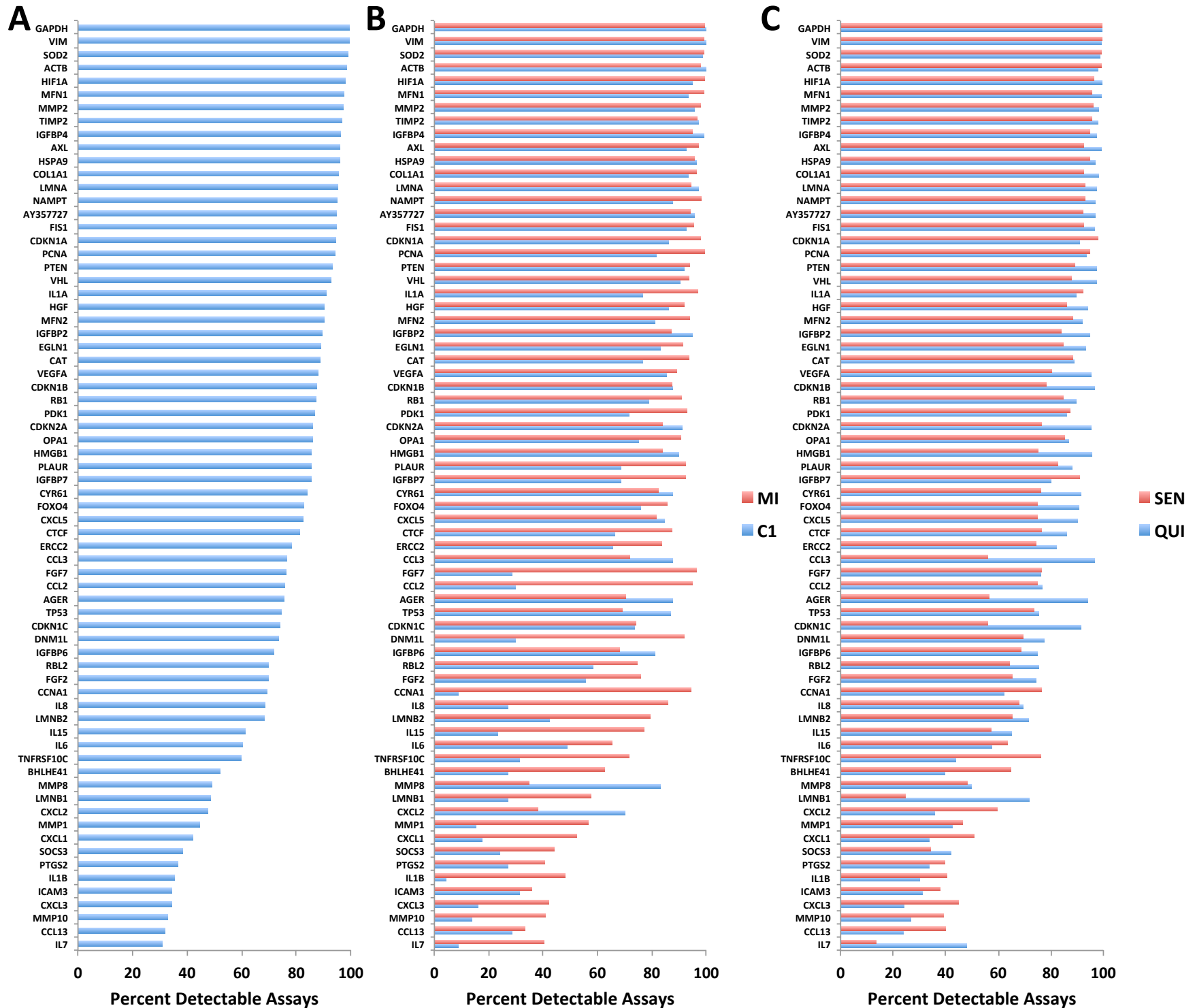
## B



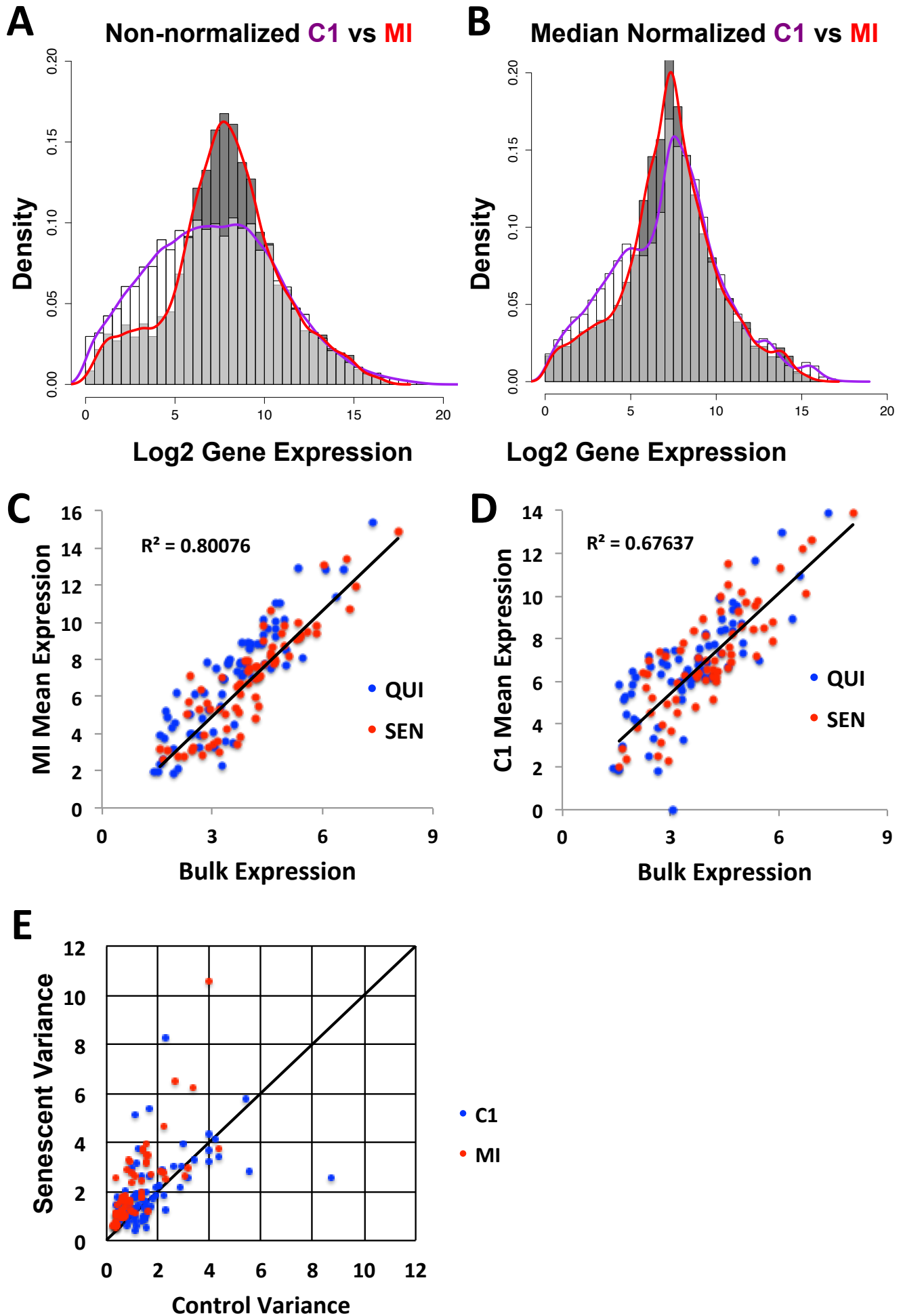
## C



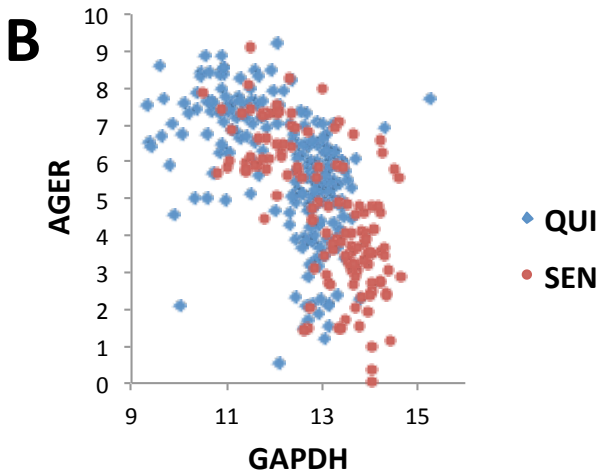
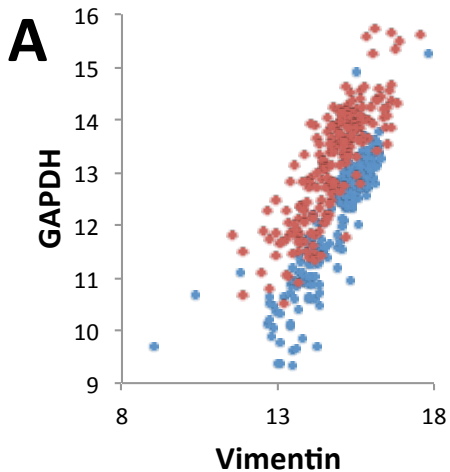
# Figure S2



# Figure S3



# Figure S4



# Figure S5

## Class 1 pathways

Pathway name	Entities pValue	Entities FDR	Submitted entities found
Activation of Matrix Metalloproteinases	1.11E-16	1.48E-14	MMP2;TIMP2
Degradation of the extracellular matrix	1.11E-16	1.48E-14	COL1A1;MMP2;TIMP2
Collagen degradation	1.11E-16	1.48E-14	COL1A1;MMP2
Extracellular matrix organization	1.11E-13	1.11E-11	COL1A1;MMP2;TIMP2
Interleukin-4 and 13 signaling	3.64E-11	2.91E-09	IL6;MMP2;VIM;HIF1A;VEGFA
Cellular responses to stress	7.07E-09	4.66E-07	EGLN1;HSPA9;RB1;IL6;CAT;IGFBP7;VHL;SOD2;HIF1A;VEGFA
Cellular responses to external stimuli	5.12E-08	2.92E-06	EGLN1;HSPA9;RB1;IL6;CAT;IGFBP7;VHL;SOD2;HIF1A;VEGFA
Signaling by Interleukins	3.20E-07	1.60E-05	FGF7;IL6;MMP2;VIM;VHL;HIF1A;VEGFA
Immune System	5.19E-06	2.18E-04	COL1A1;IL6;FGF7;MMP2;CAT;TIMP2;PTEN;VHL;VIM;HIF1A;PDK1;VEGFA
Cytokine Signaling in Immune system	5.46E-06	2.18E-04	FGF7;IL6;MMP2;VIM;VHL;HIF1A;VEGFA
Assembly of collagen fibrils and other multimeric structures	7.38E-06	2.66E-04	COL1A1;MMP2
Regulation of Hypoxia-inducible Factor (HIF) by oxygen	2.17E-05	6.52E-04	EGLN1;VHL;HIF1A;VEGFA
Cellular response to hypoxia	2.17E-05	6.52E-04	EGLN1;VHL;HIF1A;VEGFA
Regulation of gene expression by Hypoxia-inducible Factor	3.83E-05	1.07E-03	HIF1A;VEGFA
Collagen formation	5.96E-05	1.55E-03	COL1A1;MMP2
Cellular Senescence	1.36E-04	3.41E-03	RB1;IL6;IGFBP7
Regulation of Insulin-like Growth Factor (IGF) transport and uptake	1.51E-04	3.47E-03	IL6;MMP2;IGFBP2;IGFBP7
PIP3 activates AKT signaling	3.15E-04	4.82E-03	FGF7;PTEN;PDK1
PI3K/AKT activation	3.44E-04	4.82E-03	FGF7;PTEN;PDK1
GAB1 signalosome	3.44E-04	4.82E-03	FGF7;PTEN;PDK1
Senescence-Associated Secretory Phenotype (SASP)	4.41E-04	6.18E-03	IL6;IGFBP7
Downstream signaling events of B Cell Receptor (BCR)	1.97E-03	2.56E-02	FGF7;PTEN;PDK1
Oxygen-dependent proline hydroxylation of HIF1-Alpha	2.76E-03	3.41E-02	EGLN1;VHL;HIF1A
Role of LAT2/NTAL/LAB on calcium mobilization	2.84E-03	3.41E-02	FGF7;PTEN;PDK1
Pink/Parkin Mediated Mitophagy	3.52E-03	4.23E-02	MFN1;MFN2

## Class 2 pathways

Pathway name	Entities pValue	Entities FDR	Submitted entities found
Chemokine receptors bind chemokines	1.15E-08	1.38E-06	IL8;CXCL2;CXCL5
Interleukin-10 signaling	2.29E-07	1.37E-05	IL8;PTGS2;CXCL2
Peptide ligand-binding receptors	1.07E-05	4.29E-04	IL8;CXCL2;CXCL5
Interleukin-4 and 13 signaling	1.64E-05	4.92E-04	SOCS3;IL8;PTGS2
Signaling by Interleukins	5.81E-05	1.16E-03	SOCS3;IL8;PTGS2;CXCL2
Unfolded Protein Response (UPR)	1.04E-04	1.77E-03	IL8;LMNA
G alpha (i) signalling events	1.31E-04	1.97E-03	IL8;CXCL2;CXCL5
Class A/1 (Rhodopsin-like receptors)	3.28E-04	3.28E-03	IL8;CXCL2;CXCL5
Cytokine Signaling in Immune system	6.12E-04	4.90E-03	SOCS3;IL8;PTGS2;CXCL2
Regulation of Insulin-like Growth Factor (IGF) transport and uptake	1.00E-03	8.00E-03	IGFBP4;IGFBP6;CYR61