Supplementary material

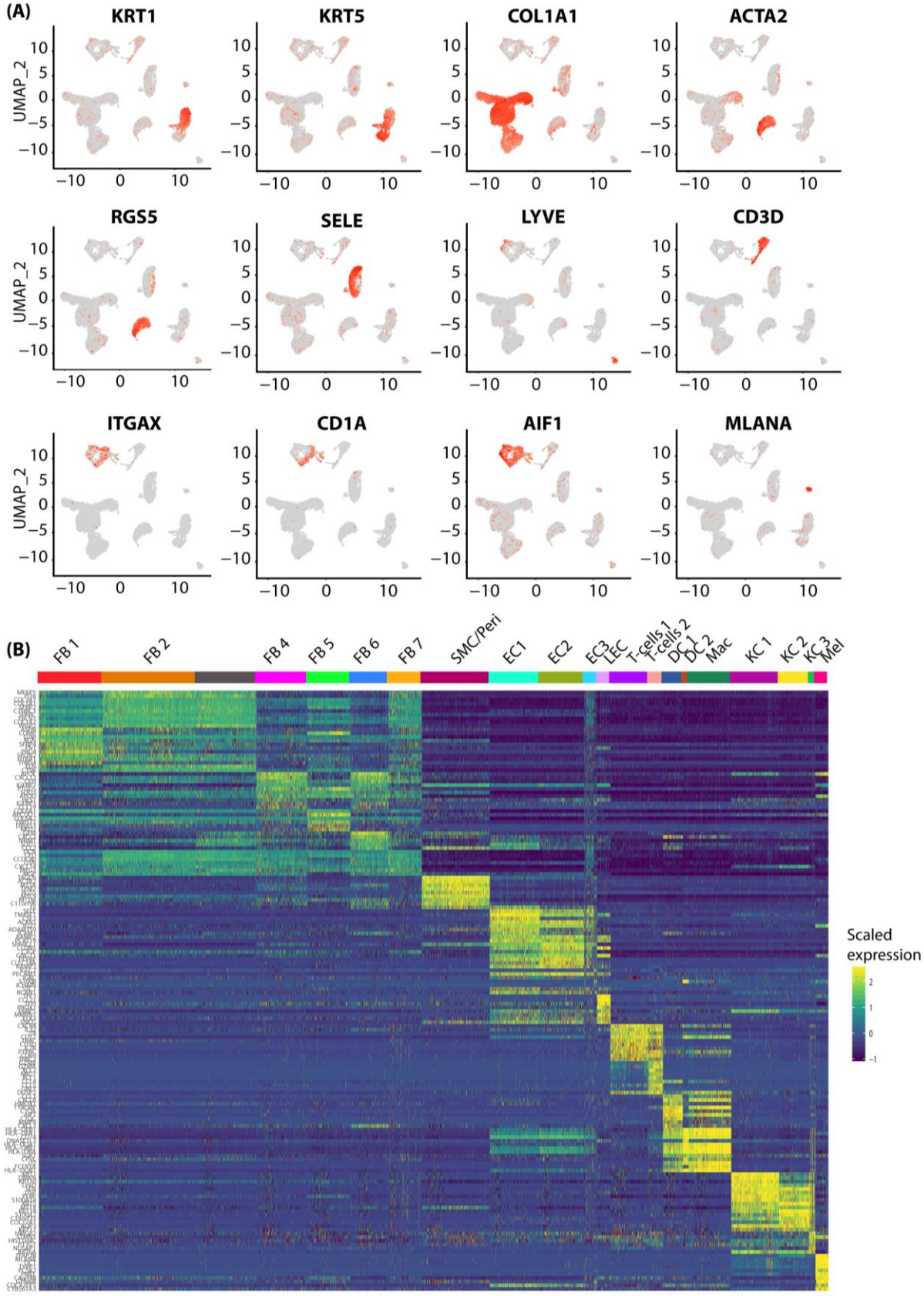


Figure S1: Identification of cell types by marker genes and marker gene expression patterns in human skin and scar

A) Feature Plots of cluster markers *KRT1* (Keratin1) for spinous and granular keratinocytes (KCs), *KRT5* (Keratin 5) for basal KCs, *COL1A1* (collagen I alpha 1) for fibroblasts (FBs),

ACTA2 (smooth muscle actin) for smooth muscle cells and myofibroblasts, *RGS5* (Regulator Of G Protein Signaling 5) for pericytes, *SELE* (E-selectin) for endothelial cells, *LYVE1* (Lymphatic Vessel Endothelial Hyaluronan Receptor 1) for lymphatic endothelial cells, *CD3D* (cluster of differentiation 3D) for T-cells, *ITGAX* (Integrin Subunit Alpha X, CD11C) and *CD1A* for dendritic cells, *AIF1* (Allograft Inflammatory Factor 1) for macrophages, and *MLANA (*Melan-A) for melanocytes. In feature plots, normalized log expression of the respective gene is mapped onto the UMAP-plot. B) Heatmap of top 10 clustermarker (differentially upregulated genes of each cluster compared to the rest of the dataset). Heatmap shows scaled expression values for genes, rows represent genes, columns represent individual cells. DEGs were calculated per cluster comparing scar versus skin using Wilcoxon rank sum test, including genes with average logarithmic fold change (avglogFC) of > 0.1 or < -0.1 and Bonferroni-adjusted p-value <0.05. Feature plot shows projection of nDEG onto the UMAP-plot, color intensity represents nDEG. UMAP, uniform manifold approximation and projection.

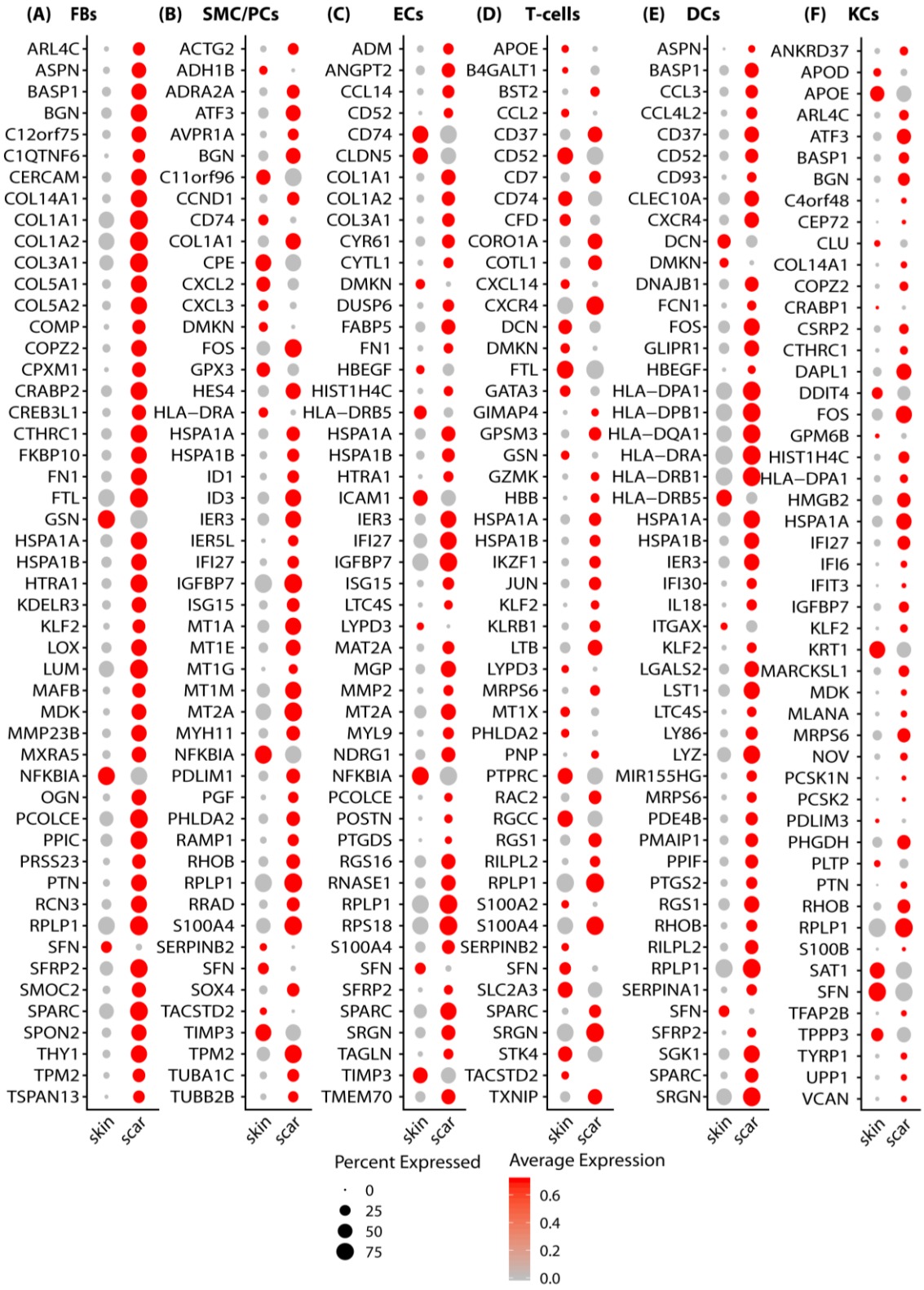


Figure S2: Top 50 regulated genes per cell group in scar compared to skin.
In cellgroups, i.e. A) fibroblasts (FBs), B) smooth muscle cells and pericytes (SMC/PCs), C) endothelial cells (ECs), D) T-cells, E) dendritic cells (DCs), and keratinocytes (KCs), differentially expressed genes (DEGs) were calculated comparing scar versus skin using Wilcoxon rank sum test, including genes with average logarithmic fold change (avglogFC) of > 0.1 or < -0.1 and Bonferroni-adjusted p-value <0.05. For each cell group, top 50 DEGs according to lowest adjusted p-value are displayed, split by skin and scar. Dot size represents percent of cells expressing the respective gene, color correlates with average expression.

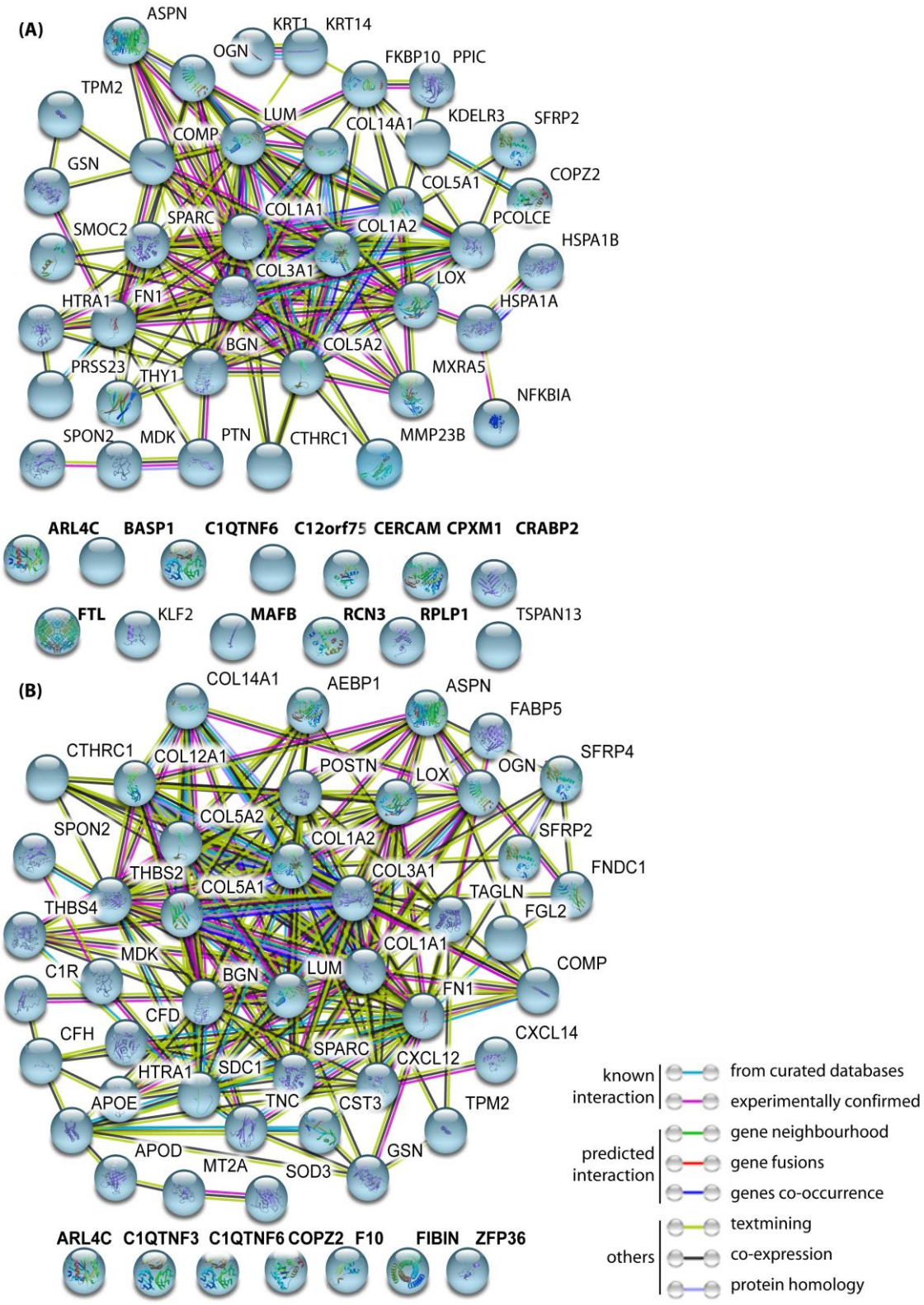


Figure S3: Gene interaction networks of upregulated genes in FBs
STRING networks of gene/protein interactions from A) top 50 regulated genes (according to lowest adjusted p-value) comparing skin FBs versus scar FBs and B) top 50 regulated genes (according to lowest adjusted p-value) cluster subFB1 to all other scar FBs. Lines indicate protein interaction with evidence level according to legend. Bold gene names indicate protein with no hitherto described relation to skin scarring.

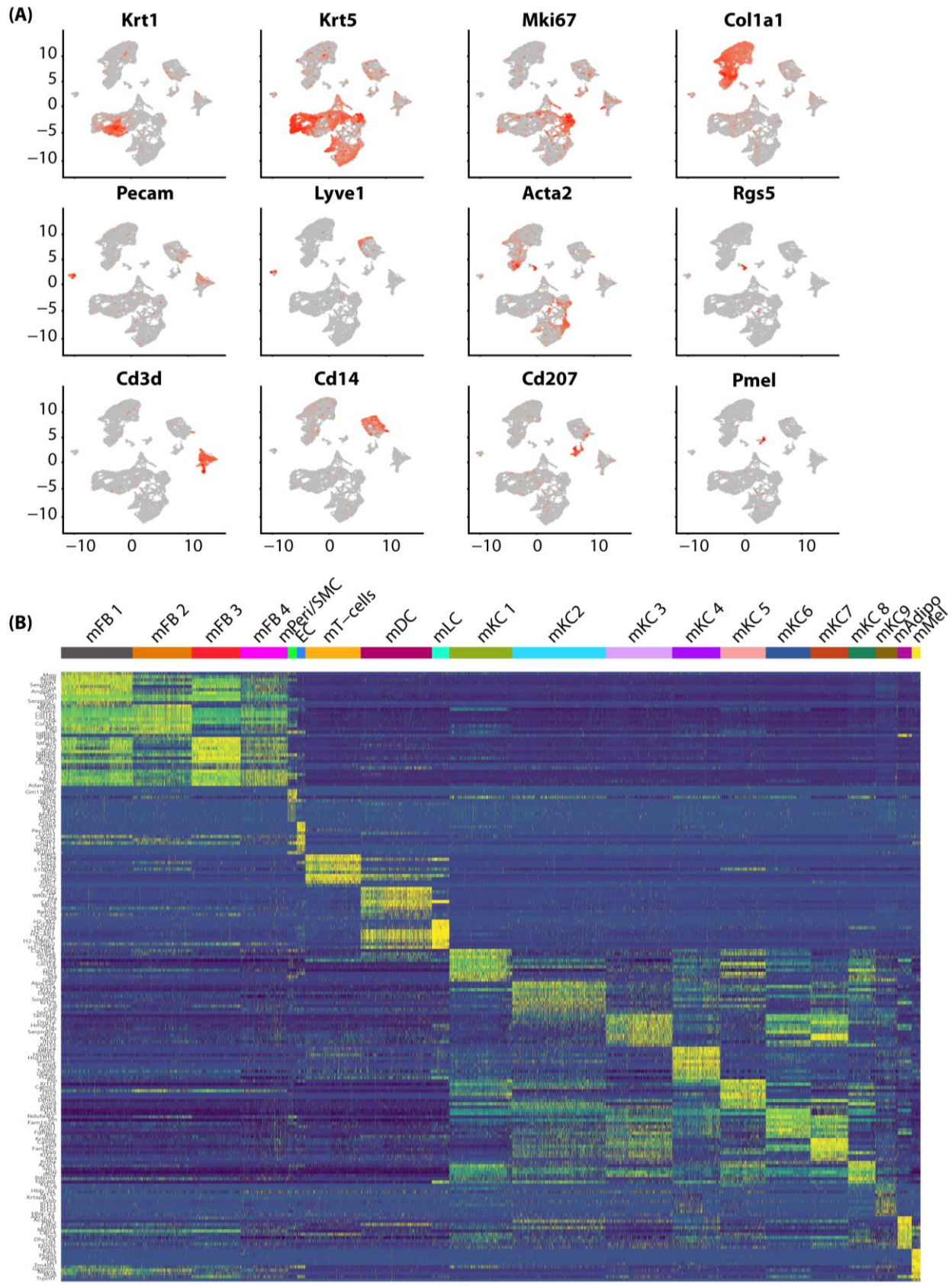


Figure S4: Identification of cell types by marker genes and gene expression patterns in mouse scars

A) Feature Plots of cluster markers *Krt1* (Keratin1) for spinous and granular keratinocytes (KCs), *Krt5* (Keratin 5) for basal KCs, *Mki67* (Marker Of Proliferation Ki-67) for proliferating cells, *Col1a1* (collagen I alpha 1) for fibroblasts; *Pecam* (Platelet And Endothelial Cell

Adhesion Molecule 1) for endothelial cells, *Lyve1* (Lymphatic Vessel Endothelial Hyaluronan Receptor 1) for lymphatic endothelial cells, *Acta2* (smooth muscle actin) for smooth muscle cells and myofibroblasts, *Rgs5* (Regulator Of G Protein Signaling 5) for pericytes, *Cd3d* (cluster of differentiation 3D) for T-cells, *Cd14* for dendritic cells, *Cd207* (Langerin) for Langerhans cells, and *Pmel* (Premelanosome Protein) for melanocytes. In feature plots, normalized log expression of the respective gene is mapped onto the UMAP-plot.
B) heatmap of top 10 upregulated clustermarker (differentially expressed genes of each cluster compared to the rest of the dataset). Heatmap showing scaled expression values for genes, rows represent genes, columns represent individual cells.

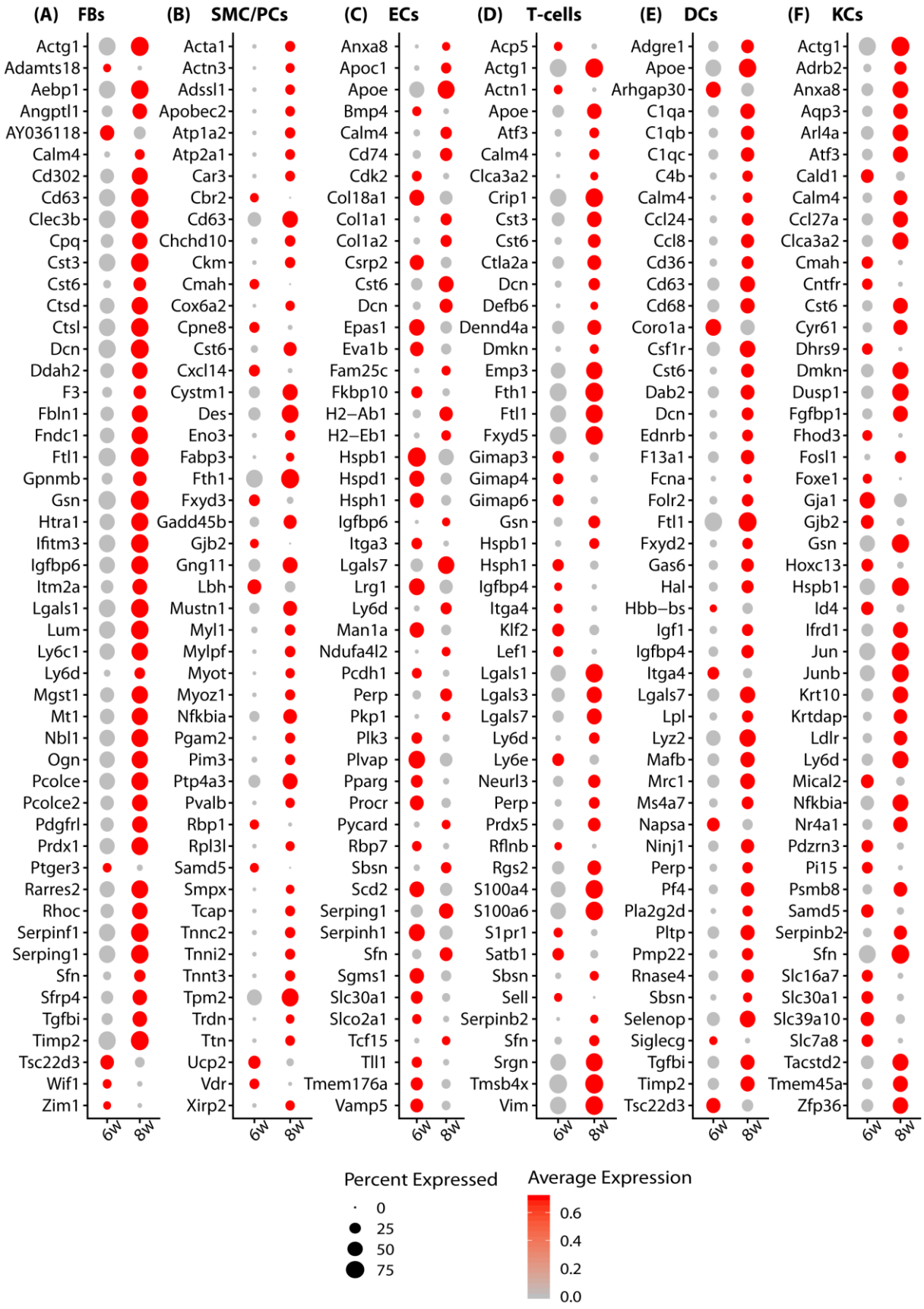


Figure S5: Top 50 regulated genes per cell group in 6 weeks and 8 weeks old murine scars. In A) fibroblasts (FBs), B) smooth muscle cells and pericytes (SMC/PCs), C) endothelial cells (ECs), D) T-cells, E) dendritic cells (DCs), and keratinocytes (KCs), differentially expressed genes (DEGs) were calculated comparing 8 weeks versus 6 weeks old mouse scars using

Wilcoxon rank sum test, including genes with average logarithmic fold change (avglogFC) of > 0.1 or < -0.1 and Bonferroni-adjusted p-value <0.05. For each cellgroup, top 50 DEGs according to lowest adjusted p-value are displayed, split by timepoint. Dot size represents percent of cells expressing the respective gene, color correlates with average expression.

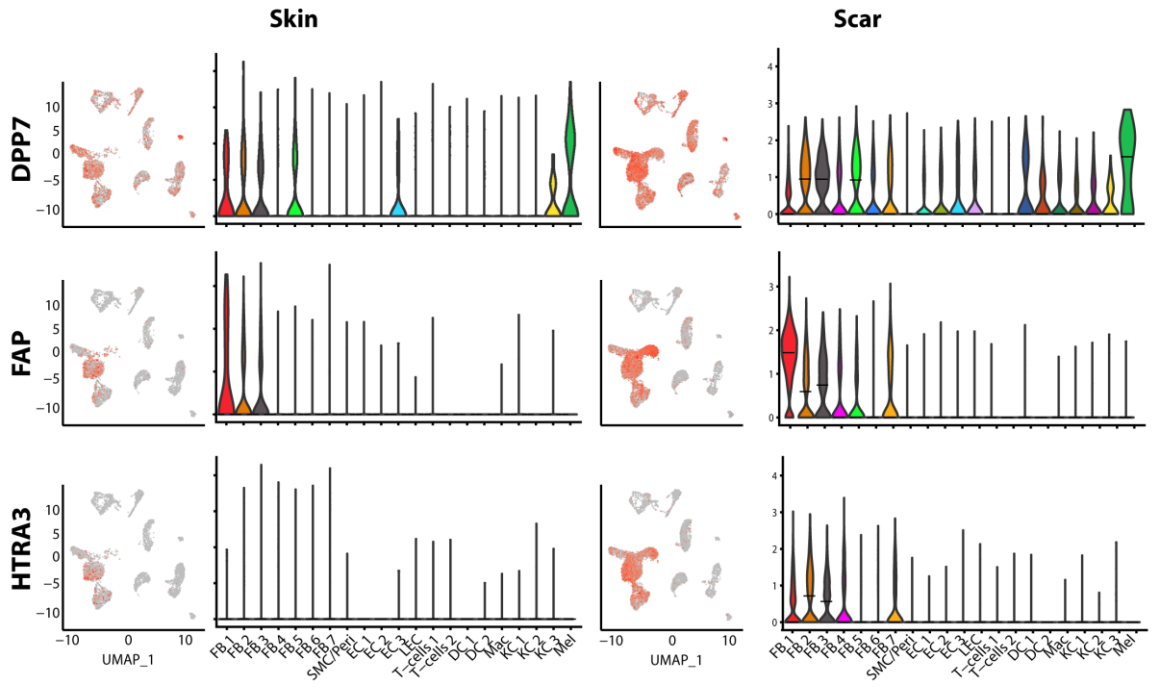
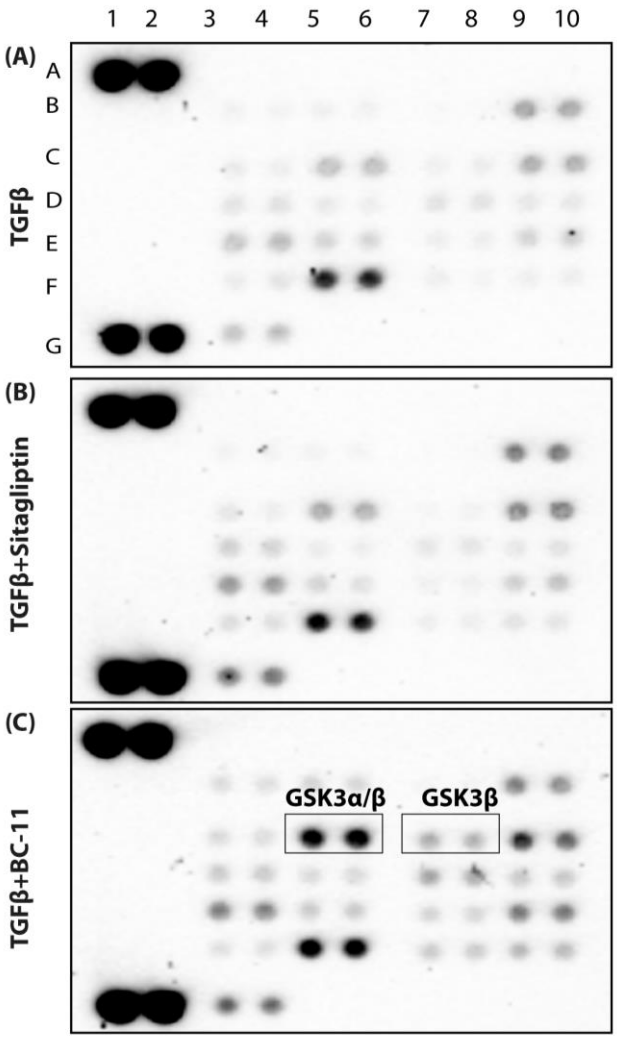


Figure S6: Feature Plots and Violin plots of serine proteases

A-C) Feature plots and violin plots of serine proteases in human skin and scar. *DPP7* (dipeptidyl-peptidase 7), *FAP* (Fibroblast Activation Protein Alpha), *HTRA3* (High-Temperature Requirement A Serine Peptidase 3). In violin plots, dots represent individual cells, *y*-axis represents log2 fold change of the normalized genes and log-transformed single-cell expression. Vertical lines in violin plots represent maximum expression, shape of each violin represents all results, and width of each violin represents frequency of respective expression level. In feature plots, normalized log expression of the respective gene is mapped onto the UMAP-plot. Color intensity indicates level of gene expressions. UMAP, uniform manifold approximation and projection.



(D)

A1,2	A3,4	A5,6	A7,8	A9,10
Reference spots				
B1,2	B3,4	B5,6	B7,8	B9,10
	CREB	EGFR	eNOS	ERK1/2
C1,2	C3,4	C5,6	C7,8	C9,10
	Fgr	GSK3α/β	GSK3β	HSP27
D1,2	D3,4	D5,6	D7,8	D9,10
	JNK1/2/3	Lck	Lyn	MSK1/2
E1,2	E3,4	E5,6	E7,8	E9,10
	p38a	PDGF Rβ	PLCγ1	Src
F1,2	F3,4	F5,6	F7,8	F9,10
	STAT2	STAT5a/b	WNK1	Yes
G1,2	G3,4	G5,6	G7,8	G9,10
Reference spots	β-Catenin			Negative control (PBS)

Figure S7: A proteome profiler-assisted identification of signaling pathways after stimulation with TGFβ and serine protease inhibitors.
Proteome profiler analysis of signaling pathways of primary human skin FBs stimulated with A) TGFβ1, B) TGFβ1 and DPP4-inhibitor Sitagliptin, and C) TGFβ1 and PLAU-inhibitor BC-11. D) Legend table for proteome profiler data points.