**Supplemental Figures Legends.**

Supplemental Legends are shown in this section. In addition, supplemental movie files, along with a separate file of legends describing the movies, have been uploaded, as well as one supplemental document describing source codes used for image analysis.

**Figure S1: SILAC proteomics of Fibroblasts.**

a) Ingenuity pathway analysis shows the number of proteins significantly differentially regulated in the top 5 pathways altered during aging. b) Fibroblasts were tested for cell death and viability using Calcein AM (live viable cells) and Topro-3 (dead cells) under conditions used for collection of conditioned media for LC-MS/MS analysis. c) SDS-PAGE analysis of concentrated conditioned media collected for LC-MS/MS analysis. d) Very few significant changes (less than 10) were found in the secreted proteins when the same young cell line was SILAC labeled for heavy or light proteins. e) Significant changes were found in 90 secreted proteins in the SILAC experiment comparing heavy labeled aged vs unlabeled young fibroblasts.

**Figure S2: Collagen contractility and ECM thickness**

a) Collagen was embedded with young and aged fibroblasts and layered in 48 well plates and assessed for contractility over 3 days. b) Quantification of the collagen contractility wells shown in (a). c) Young and aged fibroblasts were allowed to deposit matrices for 5 days and the thickness was measured using SP5 LASII confocal microscope. d) Young and aged fibroblasts were allowed to deposit matrices and were lysed to test for levels for fibronectin by qPCR analysis. ANOVA was performed to determine statistical significance among different young and aged cell lines (p<0.0001).

**Figure S3: HAPLN1 levels in melanoma cells are affected by microenvironment**

a) Nine young and nine aged fibroblasts were used to collect conditioned media and tested for levels of HAPLN1 by ELISA. b) Young fibroblasts were treated with recombinant TGFβ1 and recombinant TGFβ2 and tested for HAPLN1 levels by western blotting. c) Aged fibroblasts were treated with recombinant TGFβ1 and recombinant TGFβ2 and tested for HAPLN1 levels. d) TCGA analysis of melanoma patients for HAPLN1 expression by increasing decade of age. e, f) Melanoma cells were treated with unconditioned, young and aged conditioned media for 48 hours and measured for HAPLN1 expression by western blotting and q-PCR analysis (ANOVA, p=0.0002).

**Figure S4: HAPLN1 effects on the ECM.**

a) Additional aged fibroblasts were allowed to form matrices and were treated with rHAPLN1 during matrix production. rHAPLN1 decreased matrix alignment. b) graph showing significant decrease in the percentage of fibers aligned within a 30° range after rHAPLN1 treatment (ANOVA p<0.0001). c) HAPLN1 knockdown in young 2003-071-032 fibroblasts as tested by western blotting and qPCR analysis (ANOVA p<0.0001). d Young fibroblasts with HAPLN1 knockdown (sh3400) were allowed to form matrices with or without rHAPLN1 (25ng/ml) and assessed for matrix alignment. rHAPLN1 decreased matrix alignment e) graph for young fibroblast matrices with HAPLN1 knockdown with or without rHAPLN1 reconstitution showing decrease in the percentage of fibers oriented within 30° range (ANOVA, p<0.0001). f) Matrices were prepared using aged fibroblasts treated with varying concentrations of rHAPLN1. Thickness of the matrix was measured and plotted (ANOVA p<0.0001). g) Matrices produced by young fibroblasts with HAPLN1 knockdown were assessed for matrix thickness and plotted (ANOVA p<0.0001). h) Matrices were prepared using young fibroblasts with HAPLN1 knockdown as well as aged fibroblasts with rHAPLN1 treatment and stained with α-SMA to determine fibroblast activation. i) Aged mice were treated with rHAPLN1 intradermally and skin was assessed for collagen fiber by second harmonic generation using two photon microscopy. Multiple areas were imaged and quantified as shown. j) Aged Yumm1.7 tumor bearing mice were treated with rHAPLN1 and tumor/skin interface was assessed for matrix orientation. Fibers oriented within the 30° range are graphed.

**Figure S5: HAPLN1 effects on melanoma cell invasion**

a) Young and aged fibroblasts were used to produce matrices, and then extracted and matrices were seeded with melanoma cells. Time lapse microscopy of melanoma cells indicated cell motility on various matrices (bar=25microns). b) Matrices were produced from aged fibroblasts treated with varying concentrations of rHAPLN1. Fibroblasts were extracted and matrices were plated with melanoma cells and imaged using timelapse microscopy to assess cellular motility. ANOVA analysis was used to generate p values shown. c) Matrices produced by young fibroblasts with HAPLN1 knockdown were extracted and seeded with melanoma cells to assess cellular motility ANOVA analysis was used to generate p values shown. d) Melanoma cells expressing mCherry were allowed to form spheroids and embedded in collagen along with aged fibroblasts and varying concentrations of rHAPLN1. Spheroids were imaged in mCherry channel and invasion was quantified ANOVA analysis was used to generate p values shown. e) HAPLN1 knockdown fibroblasts expressing GFP and melanoma cells expressing mCherry were mixed 1:1 and allowed to form spheroids. Spheroids were embedded in collagen and the invasive area was quantified, normalized to core size and plotted (ANOVA p<0.0001). f) WM3918 melanoma cells were used to form spheroids and embedded in collagen with varying concentrations of rHAPLN1 (0-200ng/mL). Invasive area was quantified and normalized to the core and plotted (ANOVA p<0.0001).

**Figure S6: HAPLN1 effects on cellular proliferation**

a) 1205lu melanoma cells were treated with aged media in presence of varying concentrations of rHAPLN1 for 48 hours. Apoptosis was measured by staining for AnnexinV and PI positivity. b) Melanoma cells were treated with aged media and varying concentrations of rHAPLN1 for 48 hours and tested for apoptosis by AnnexinV/PI staining. c) Aged fibroblasts were used to prepare skin reconstructs and treated with varying concentrations of rHAPLN1 during reconstruct generation. Reconstructs were stained for Ki67 positivity to determine cellular proliferation (ANOVA p=0.2061). d) Primary tumors from aged mice either treated with PBS or with rHAPLN1 were embedded in paraffin, sectioned and stained for Ki67 positivity to determine cellular proliferation (ANOVA p=0.7727). e) Examples of Ki67 staining in mouse tumor treated with either PBS or rHAPLN1 (bar=100microns).

**Figure S7: HAPLN1 affects immune cell motility in vitro**

a) Melanoma cells were allowed to form spheroids, mixed with T cells and embedded in collagen plugs. Collagen plugs were imaged for localization of T cells (CD4+ positivity) and collagen I by immunofluorescence (bar=25microns). b) Schematic of the reconstructs used to study motility of T cells and melanoma cells in collagen. c) Melanoma cells were layered in collagen followed by layering with T cells and aged fibroblasts treated with varying concentrations of HAPLN1 and allowed to set over 5 days. H&E staining was used to visualize cellular interactions and infiltration and activity of T cells into the melanoma layer. Black arrows show regions of T cell activity marked by absence of cells and collagen in the presence of rHAPLN1.

**Figure S8: HAPLN1 affects immune cell motility in vivo**

a) Aged mice were injected with Yumm1.7 allografts and treated with rHAPLN1. Tumors were harvested and analyzed for percentage of CD45 positivity by flow cytometry. b) Aged mice with Yumm1.7 allografts were treated with rHAPLN1 and assessed for percentage of CD8+ cells and T helper cells and compared across treatments. c) Yumm1.7 allografts in aged mice treated with rHAPLN1 were assessed for percentage of CD4 positive cells within the tumor. d) Yumm1.7 allografts were implanted in young (6 weeks) and aged (52 weeks) mice and assessed for infiltrating cells by flow cytometry. Graph shows relative proportions of PMN-MDSC, M-MDSC and macrophages with respect to each other.