Supporting Information

Supporting Materials and Methods

Fixation of vital organs. Severe combined immunodeficient mice with MDA-MB-231 breast xenografts were humanely killed after 13 days of treatment with peptides or PBS using CO_2. Liver, lungs, and kidneys were excised and stored in zinc-based fixative (BD Biosciences) for 3 weeks, after which samples were placed in paraffin blocks. H&E staining was made to visualize tissue structure for pathology and normal tissue structure in comparison to the experimental control. Histologic samples were processed using Aperio Image Scope Software (Aperio Technologies) and compiled using Adobe Photoshop CS3 Software (Adobe Systems).

Flow cytometry analysis for receptor identification. Flow cytometry analysis was completed for receptor quantification on primary HUVECs and MDA-MB-231 breast cancer cells. HUVECs were acquired from a single donor were purchased from Cambrex. Cells were propagated in endothelial growth medium-2 (Lonza, Basel, Switzerland) using 2% FBS, growth factors (human basic fibroblast growth factor and VEGF), and antibiotics (gentamicin/amphotericin B). All cells used were from passage 3 to passage 6. MDA-MB-231 breast cancer cells were acquired from a single donor (ATCC) as described previously and propagated in RPMI-1640 medium (Cambrex) with 10% FBS and antibiotics (penicillin/streptomycin). Cells were gently dissociated from their flasks using PBS/EDTA (200 mg/ml), counted, and aliquoted at 5×10^5 cells/ml in medium. Heat-aggregated human IgG at 20 μg/ml was used to block nonspecific binding. Medium containing different monoclonal antibody solutions at 20 μl/1 × 10^6 cells were applied for β_1-integrins (FAB17781; R&D Systems, Minneapolis, MN), CXCR3 (FAB1685; R&D Systems), and CD36 (CB38/NL07; BD PharMingen). Cells were then incubated for 60 minutes at 4°C to allow binding of receptors to antibody, pelleted after centrifuging at 1000 RPM for 5 minutes at 4°C, and washed with a 200-μl medium of 1% FBS three times. Three hundred microliters of buffer (PBS, 1% BSA, 0.1% NaN_3; Sigma-Aldrich, Inc) was applied and transferred to FACS tubes. Flow cytometry analysis and protocols were followed using the FACSCalibur (BD Biosciences), and receptors were quantified using FlowJo Flow Cytometry Analysis Software (Tree Star, Inc, Ashland, OR).

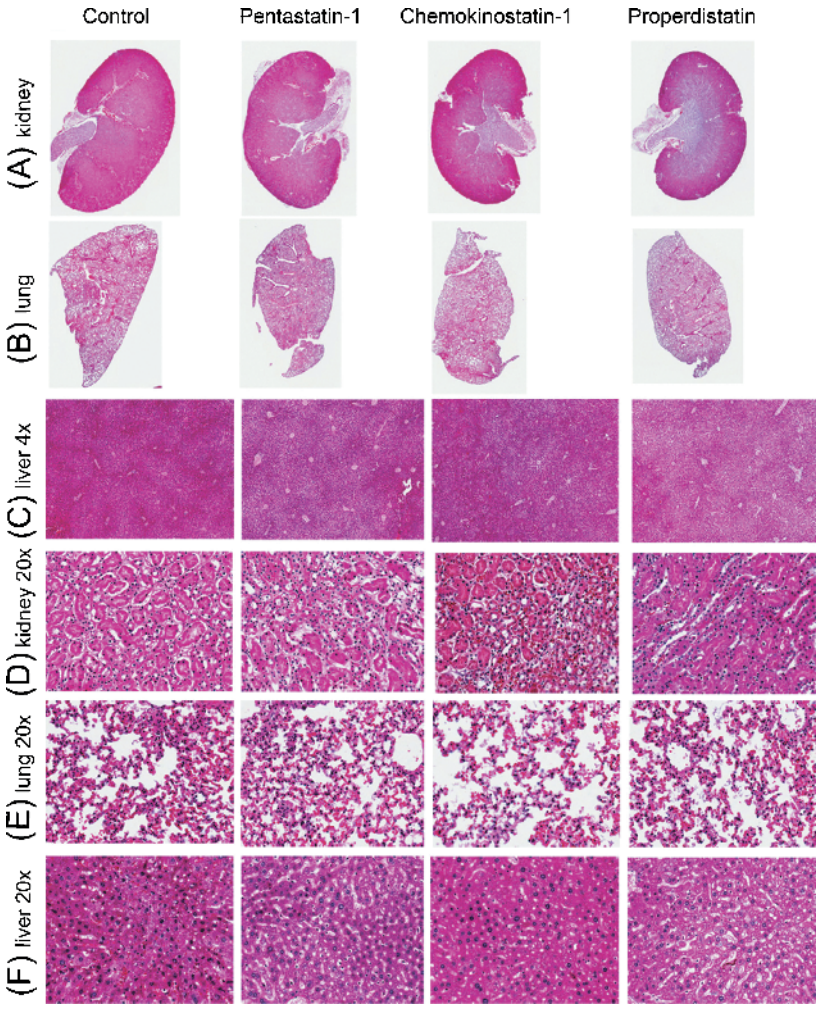
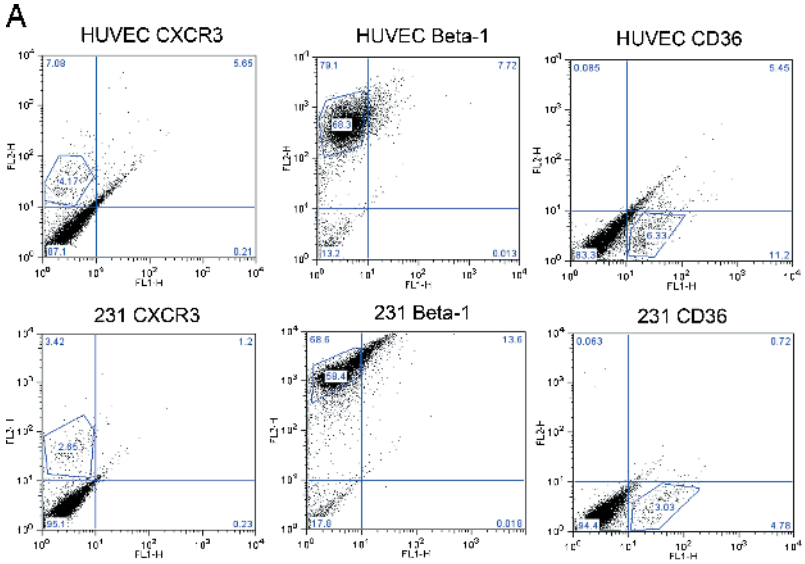


Figure W1. Peptide-treated tumors and their respective vital organs using H&E staining on day 13. Gross tissue morphology is shown in (A) for kidney at 1×, and (B) normal lung structure at 1×. (C) Liver sections were taken and magnified at 4× to show consistent morphology in control and peptide-treated tumors. (D) Kidney, (E) lung, and (F) liver were magnified at 20× to show normal cell structure and the absence of pathologies due to peptide delivery.

A



B

Receptor	Cell Type	Quadrant	Event	% Gated	Ratio (HUVEC/231)
Beta-1	HUVEC	UL	4165	84.5	1.1
	231	UL	5052	78.8	
CXCR3	HUVEC	UL	272	5.3	1.9
	231	UL	86	2.7	
CD36	HUVEC	LR	316	4.4	1.5
	231	LR	92	2.9	

Figure W2. Flow cytometry analysis for receptor identification on HUVECs and MDA-MB-231 cells. (A) Phycoerythrin conjugation for CXCR3 identification on HUVECs and MDA-MB-23s, showing 5.3% cells contain the receptor in HUVECs and 2.9% in MDA-MB-23s. Similar analysis shows HUVECs contain the β_1-integrin receptor in 84.5% of cells, and 78.8% of MDA-MB-231 cells. Fluorescein isothiocyanate conjugation for CD36 revealed 4.4% of HUVECs and 2.9% of MDA-MB-231 cells contain the receptor. (B) The corresponding table summarizes the ratios of these receptors in HUVEC/MDA-MB-231 cells as 1.9 for CXCR3, 1.1 for β_1-integrins, and 1.5 for CD36.