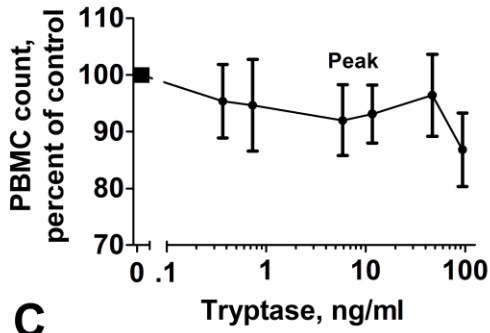
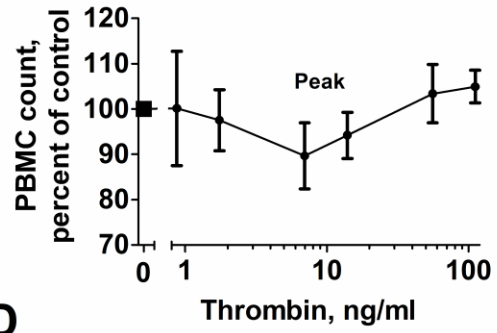
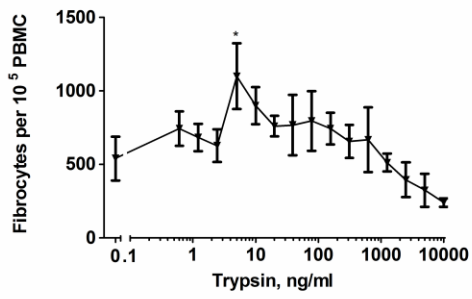
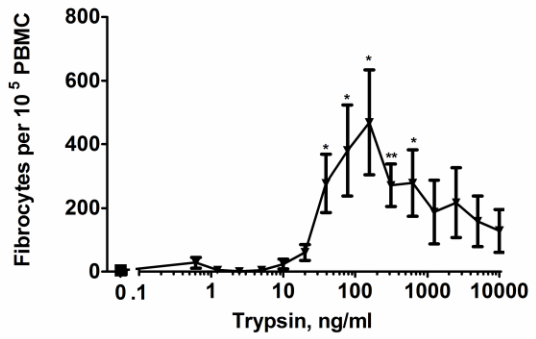
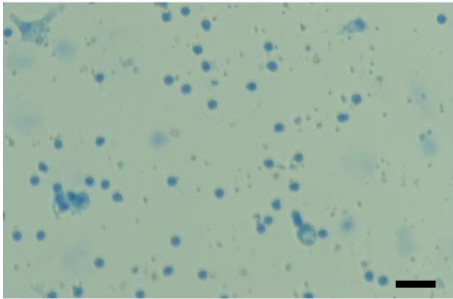
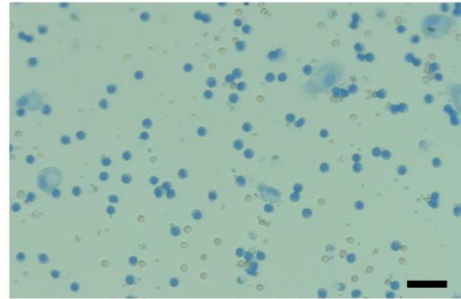
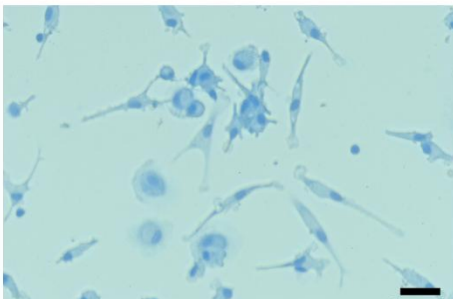


**A****B****C****D****E** Control

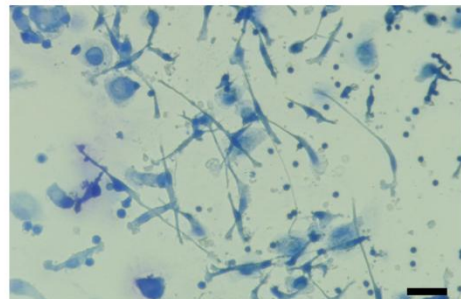
Trypsin



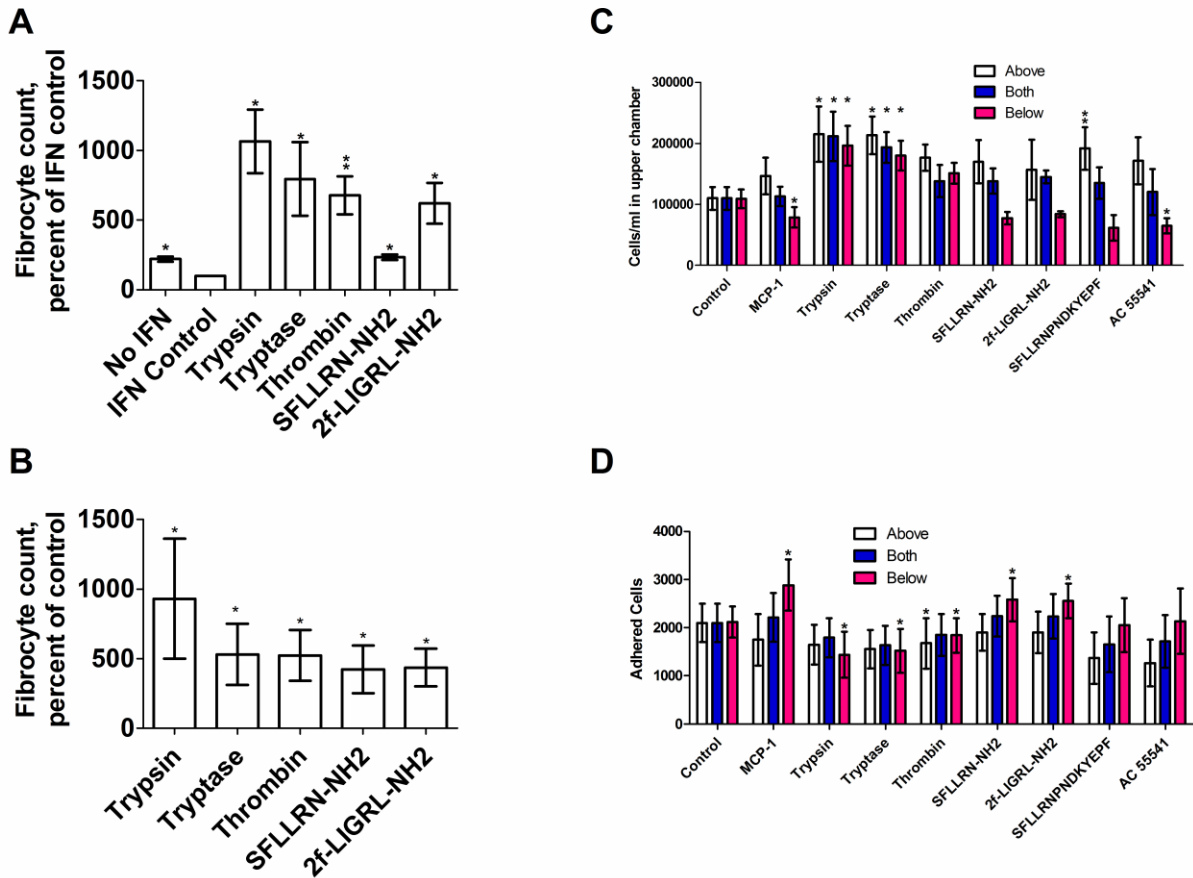
Thrombin



Tryptase

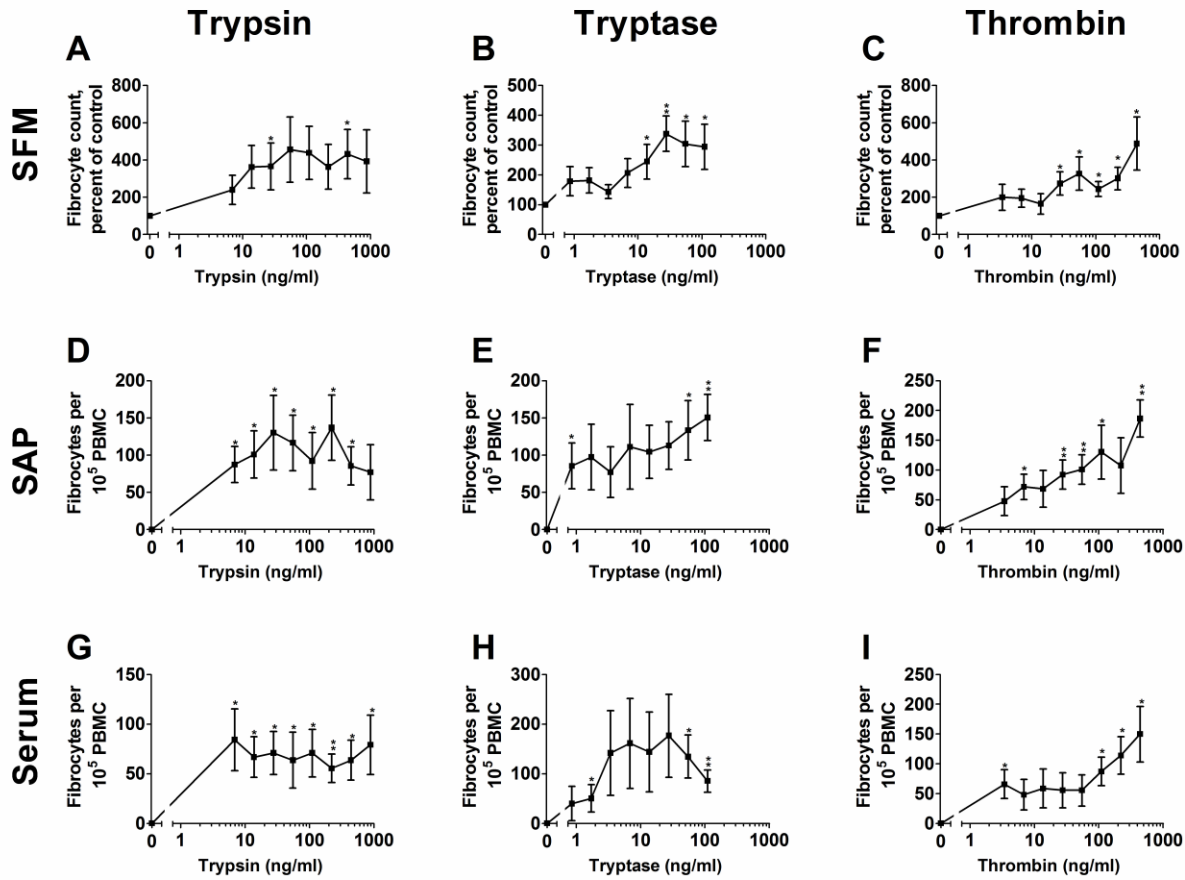


**Supplementary Figure 1. Trypsin and thrombin do not significantly affect the total number of adhered PBMC, and trypsin potentiates fibrocyte differentiation in the presence of fish gelatin or skim milk.** (A) The PBMC populations in Figure 1 were counted for the total number of PBMC adhered to the plate following fixing and staining for trypsin and (B) thrombin. Counts were normalized for each donor to the no-protease control. There were no significant differences in the numbers of adhered PBMC following fixing and staining. The protease concentrations causing peak fibrocyte counts are indicated. (C) PBMC were cultured in the presence of 500  $\mu\text{g/ml}$  fish gelatin or (D) powdered skim milk in the presence of the indicated concentrations of trypsin. After 5 days, fibrocytes were counted as in Figure 1. Values are mean  $\pm$  SEM, n=6. \* indicates  $p < .05$ , \*\*  $p < .01$  compared to the no-protease control (t-test). (E) Images of PBMC after incubating for 5 days with 10  $\mu\text{g/ml}$  SAP and either no protease (control), 12.5 ng/ml trypsin, 12.5 ng/ml thrombin, or 55 ng/ml trypsin. Bar is 40  $\mu\text{m}$ .

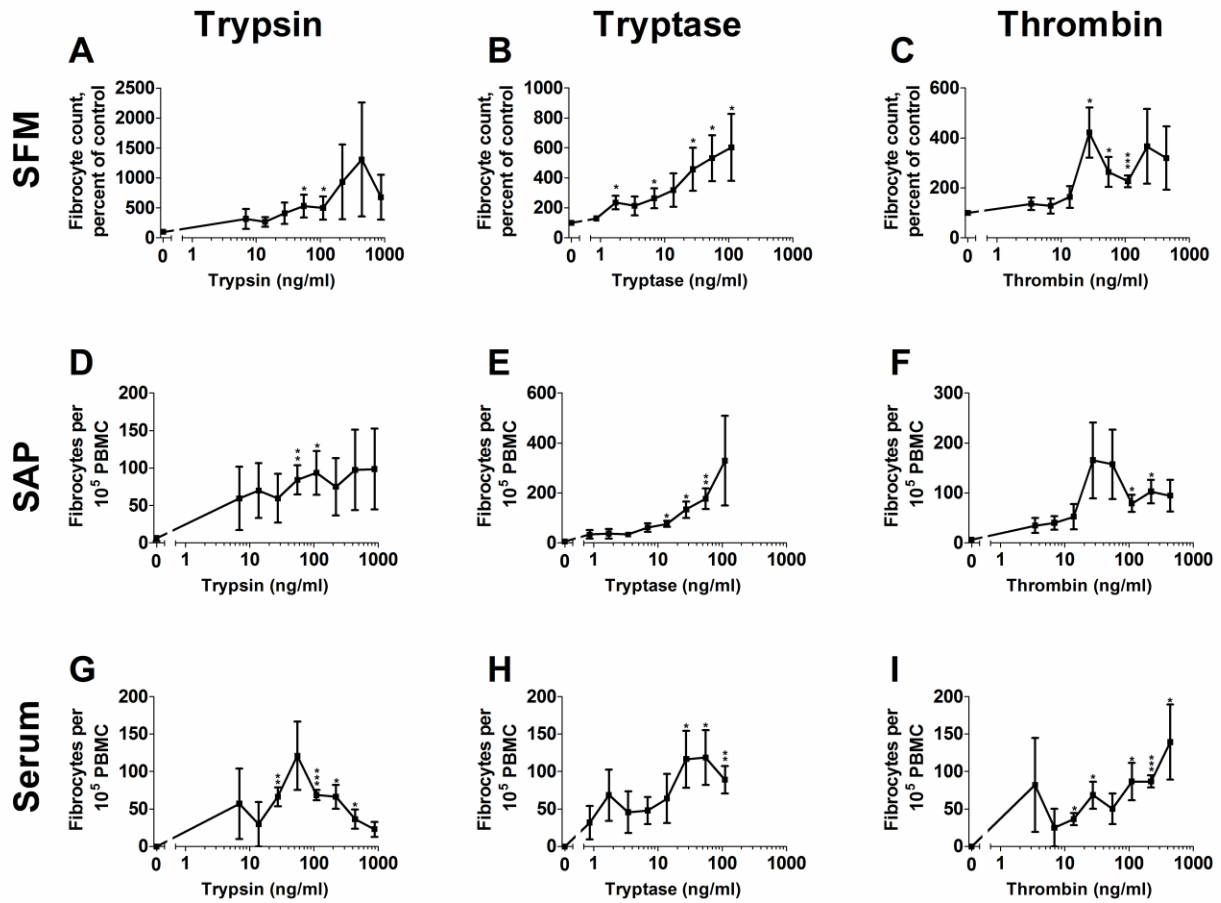


**Supplementary Figure 2. Trypsin, tryptase, thrombin, PAR-1 agonist, and PAR-2 agonist potentiate fibrocyte differentiation in the presence of interferon-gamma (IFN- $\gamma$ ), and act as chemoattractants or chemostatic agents for monocytes. (A)** PBMC were co-incubated with no IFN- $\gamma$  or 10 ng/ml IFN- $\gamma$  and either no protease or agonist (IFN- $\gamma$  control), or 12.5 ng/ml trypsin, tryptase, or thrombin, or 0.1  $\mu$ g/ml PAR-1 agonist (SFLLRN-NH2) or PAR-2 agonist (sf-LIGRL-NH2). After 5 days, fibrocytes were counted as in Figure 1, and counts were normalized to the IFN- $\gamma$  control. Values are mean  $\pm$  SEM, n=4. \* indicates  $p < .05$ , \*\*  $p < .01$ , and \*\*\*  $p < .001$  compared to the IFN- $\gamma$  control (t-test). **(B)** Monocytes were co-incubated with 20 ng/ml IFN- $\gamma$  and either no protease or agonist (IFN- $\gamma$  control), or 12.5 ng/ml protease or 0.1  $\mu$ g/ml PAR-1 or -2 agonist. Monocytes were incubated and counted as in Figure 3, and counts were

normalized to the IFN control. Values are mean  $\pm$  SEM, n=3. \* indicates  $p < .05$  compared to the IFN- $\gamma$  control (t-test). (C) PAR-1 (SFLLRN-NH2 or SFLLRNPNDKYEPF) and PAR-2 (2f-LIGRL-NH2 or AC 55541) agonists were added at 10  $\mu$ M, proteases were added at 12.5 ng/ml, and the monocyte chemoattractant MCP-1 (Peprotech) was added at 50 ng/ml, into SFM either above, below, or on both sides of an 8  $\mu$ m pore size insert in a 24-well plate well. PBMC were added to the insert chamber and after 12 hours cells in the insert were counted. (D) PBMC that adhered to the plate were stained, imaged, and counted. Values are mean  $\pm$  SEM, n=3. \* indicates  $p < .05$  and \*\*  $p < .01$  compared to the SFM control (paired t-test).



**Supplementary Figure 3. A 12 hour exposure to trypsin, trypsin, and thrombin potentiates fibrocyte differentiation.** Cells were incubated as in Figure 8, with the exception that media were changed at 12 hours instead of at 4 hours.



**Supplementary Figure 4. A 24 hour exposure to trypsin, trypsin, and thrombin potentiates fibrocyte differentiation.** Cells were incubated as in Figure 8, with the exception that media were changed at 24 hours instead of at 4 hours.