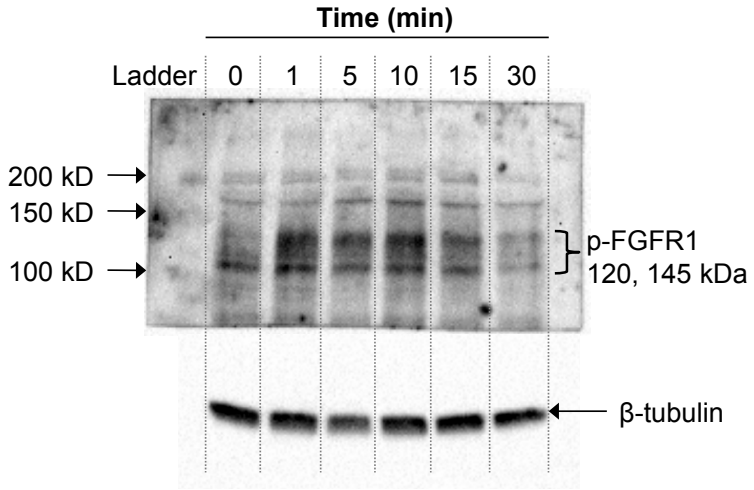
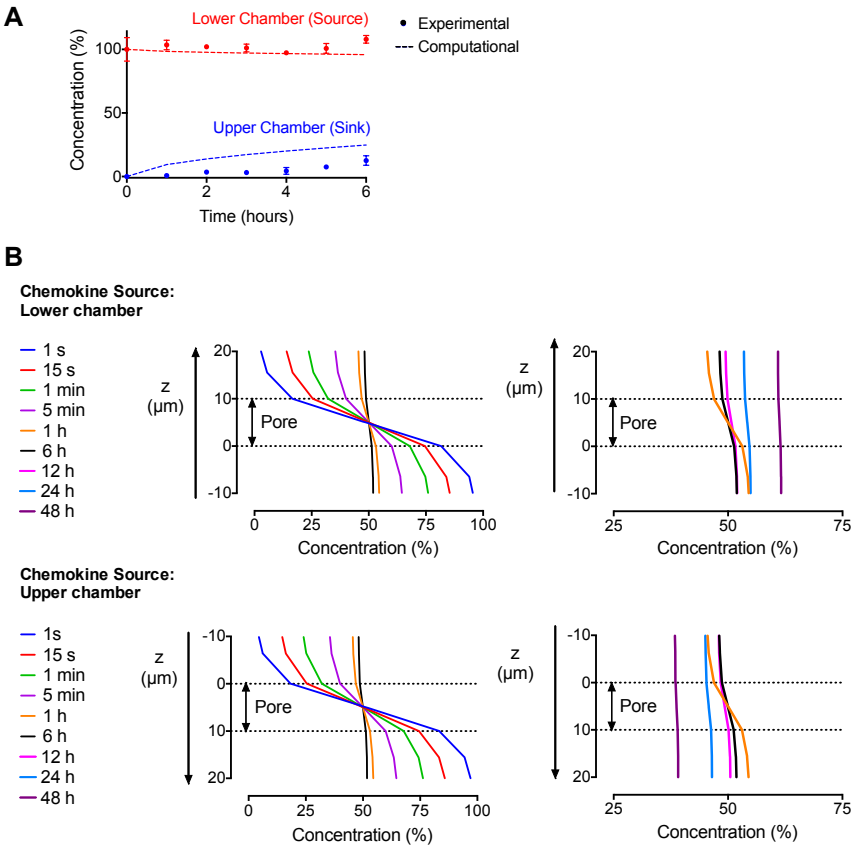
Electronic Supplementary Material (ESI) for Integrative Biology.

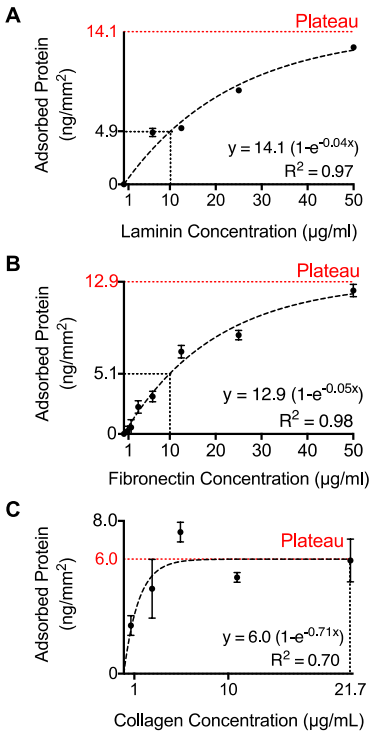
Supplemental Information
Figures



Supplemental Fig. 1 Western blot analysis of extracts from myoblasts treated with bFGF. Myoblasts were treated for 0 (no treatment), 1, 5, 10, 15, or 30 min before lysing and probing for the presence of phosphorylated FGFR1 (p-FGFR1). Beta-tubulin was used to normalize for protein volume.



Supplemental Fig. 2 Solute concentrations in the upper and lower chambers of the Transwell do not accurately represent concentrations experienced by cells traversing the porous membrane. (A) Measurements of the bulk concentrations in the lower and upper chambers over a 6-h time period were obtained using both experimental and computational methods. (B) FEM simulations of the concentration gradients across the pore of a Transwell membrane over time (1 s – 6 h, left and 1 h – 48 h, right). The top panel shows concentration gradients when the lower chamber is used as the chemokine source, and the lower panel shows how the concentration gradient is changed when the setup is reversed and the upper chamber is used as the source. The y-axis is reversed in the lower panel for easier comparison.



Supplemental Fig. 3 The microfluidic device is amenable to quantification of ECM protein adsorption. Adsorption isotherms for laminin (A), fibronectin (B), and collagen (C) were created to determine the amount of protein (ng/mm^2) adsorbed to the glass surface given the concentration (µg/mL) of the solution used to coat the surface. For these experiments, the substrate concentrations were determined to be 4.9 ng/mm^2 laminin, 5.1 ng/mm^2 fibronectin, and 6.0 ng/mm^2 Type I collagen.

Supplemental Information
Materials and Methods

Protein adsorption assay

Adsorbed protein substrates were prepared by incubating glass coverslips (Bellco Glass, 1943-10015) in fibronectin (Sigma, F2006), laminin (Life Technologies, 23017-015), or Type I collagen (Sigma, C-8919) solutions overnight at 37°C. For fibronectin and laminin the coating concentrations were 0.78, 1.56, 3.13, 6.25, 12.5, 25, and 50 μg/ml, whereas the concentrations for collagen were 0.68, 2.71, 5.42, 10.85, and 21.7 μg/mL. Coverslips were rinsed three times in warm PBS, placed in 24-well plates, and covered with 250 μl PBS and 250 μL of bicinchoninic acid (BCA) assay working reagent (Sigma, QPBCA). Simultaneously, a set of standard solutions of known fibronectin (0, 0.63, 1.25, 2.5, 5, and 10 μg/ml), laminin (0, 0.63, 1.25, 2.5, 5, and 10 μg/ml), and collagen (0, 0.67, 1.36, 2.71, 5.42, 10.85, and 21.7 μg/mL) were prepared in PBS buffer and mixed with an equal volume of BCA working reagent. Plates were sealed with parafilm and incubated for 1 hour at 60°C. Plates were allowed to cool for 30 minutes before transferring contents to a black, 96-well, clear-bottom plate (Costar, 3631) and measuring absorbance at 562 nm. Data for standard concentrations were fit to a Langmuir adsorption isotherm.