Human Neutrophil Haptokinesis and Chemokinesis on Microcontact Printed Fibronectin

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Motivations
Expanding a Canonical Cascade

Quantifying motility and traction on post-extravasation ligands (e.g. fibronectin) will help extend the present model of leukocyte recruitment.

Comparing Traction Assays
TFM
Adhesive Ligand
Beads in Polyacrylamide
mPADs
PDMS Posts

Neutrophils are an ideal cell type for the comparison of Traction Force Microscopy (TFM) and microfabricated-Post-Array-Detectors (mPADs). Presently, all traction maps of motile neutrophils are via TFM. Map adapted from (2).

Approach
Microcontact Printing
Ink
PDMS
Si
UV Ozone
Dry

UV Ozone

Cell Tracking

References:

Results

Exquisite cell-FN interaction

No off-FN adhesion is observed on printed PDMS, blocked with Pluronic F127.

Model-independent analysis

At low adhesiveness, fMLP increases extent of motility, but beyond an adhesive threshold fMLP sensitivity is attenuated.

L-selectin as activation marker

L-selectin is a sensitive marker of activation state (3). An active phenotype is not found before FN stimulation suggesting activation is FN-induced via an outside-in pathway.

Summary

• fMLP potentiates human neutrophil motility on µCP FN (chemokinesis)
• Quantified baseline motility metrics on functionalized half-spaces
• FN haptokinesis is not result of pre-FN activated phenotype suggesting model of outside-in activation
• Adhesiveness alone can potentiate motility suggesting haptotactic potential of FN

Future Directions

Immobilizing adhesive gradients

Applying a power-law model preserves trend revealed previously. Across all conditions tested, initial motility (30 min) is superdiffusive (α ~ 1.5).

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