

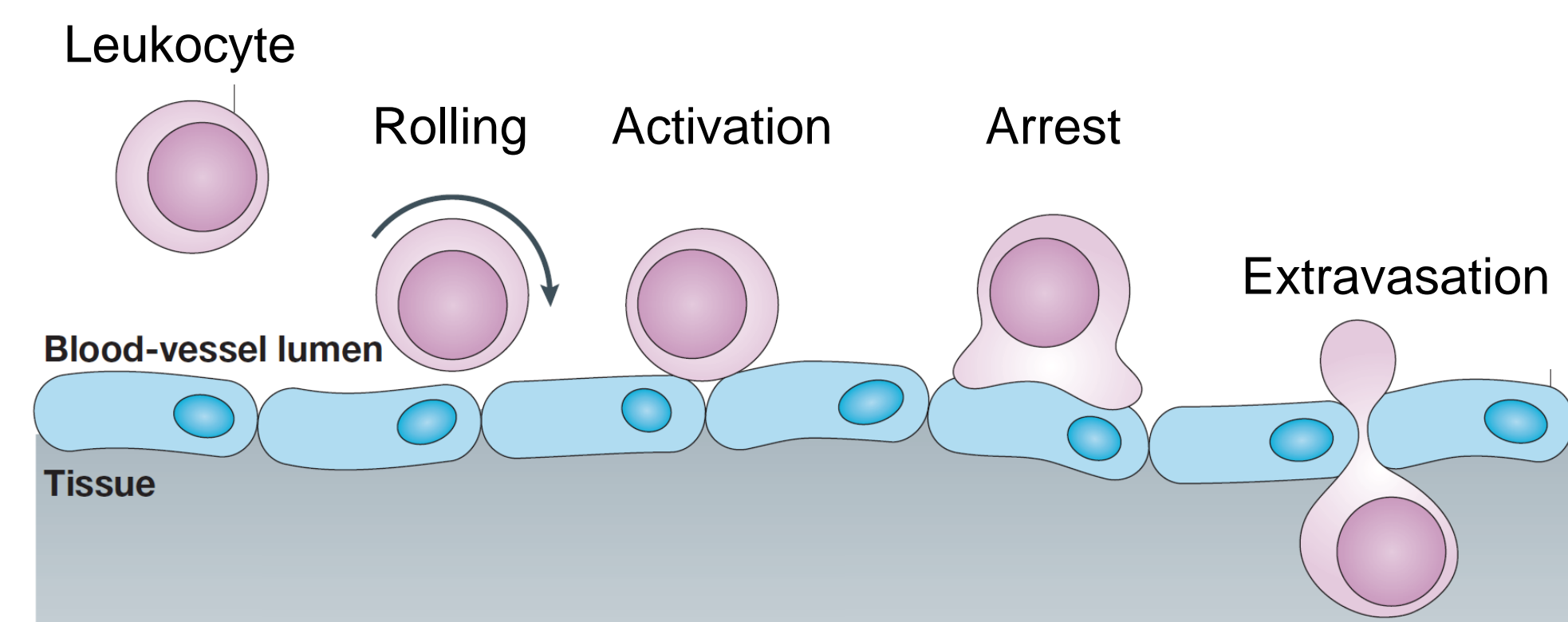
Human Neutrophil Haptokinesis and Chemokinesis on Microcontact Printed Fibronectin

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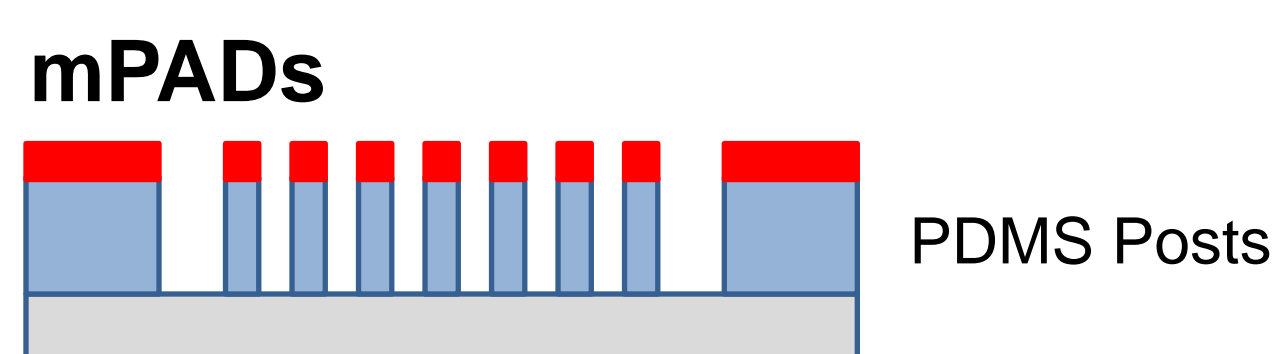
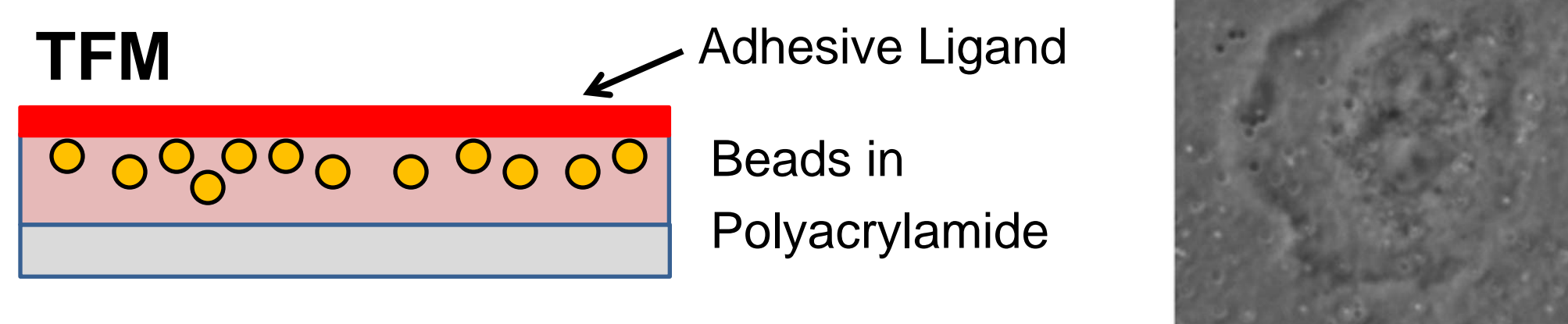
Motivations

Expanding a Canonical Cascade



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

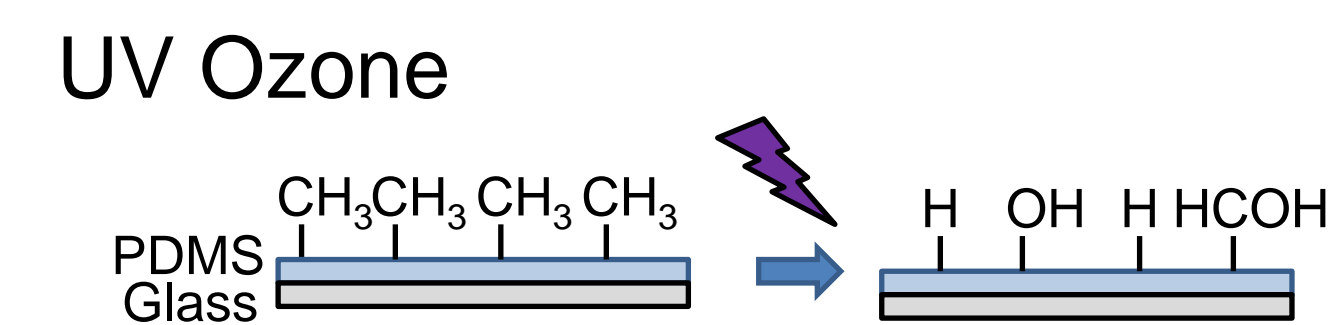
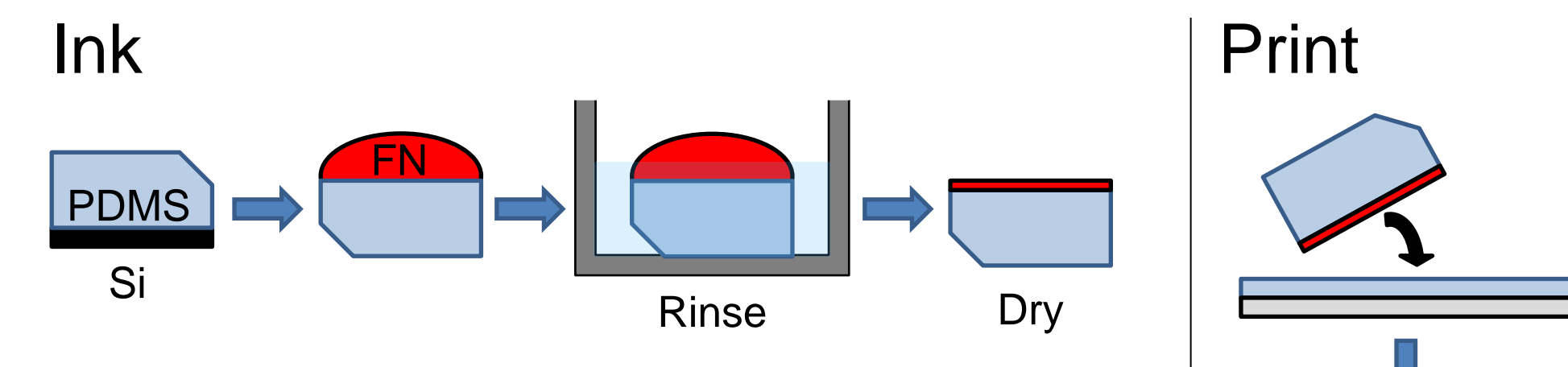
Comparing Traction Assays



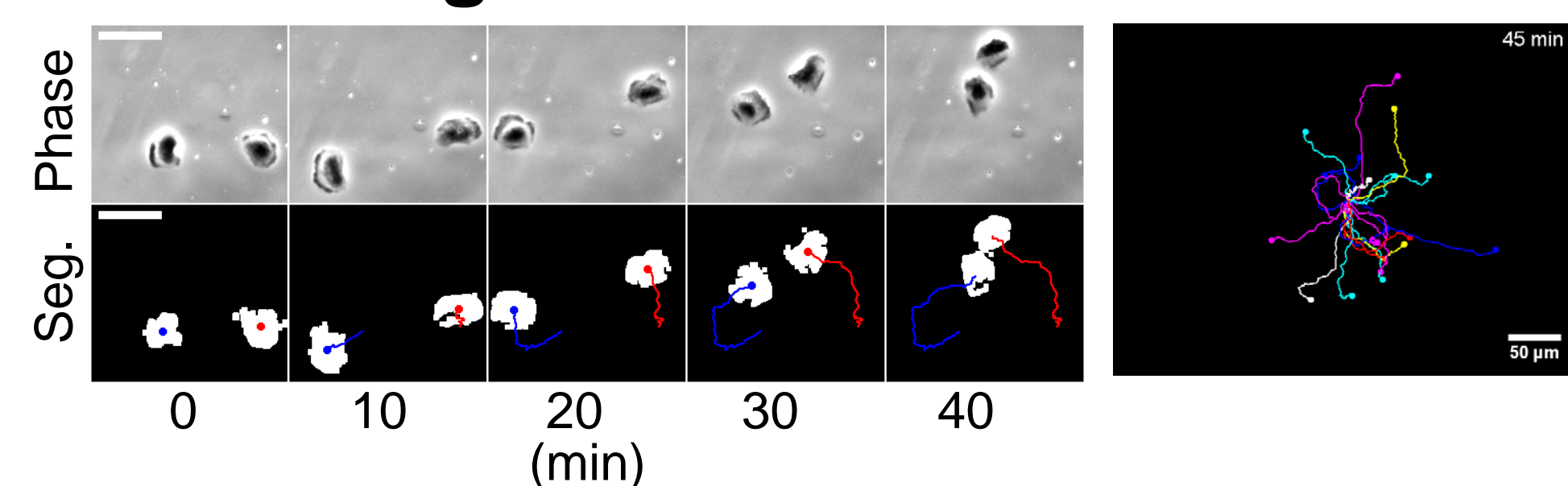
Neutrophils are an ideal cell type for the comparison of Traction Force Microscopy (TFM) and microfabricated-Post-Array-Detectors (mPADs). Presently, all traction maps of motile neutrophils are via TFM. Map adapted from (2).

Approach

Microcontact Printing



Cell Tracking

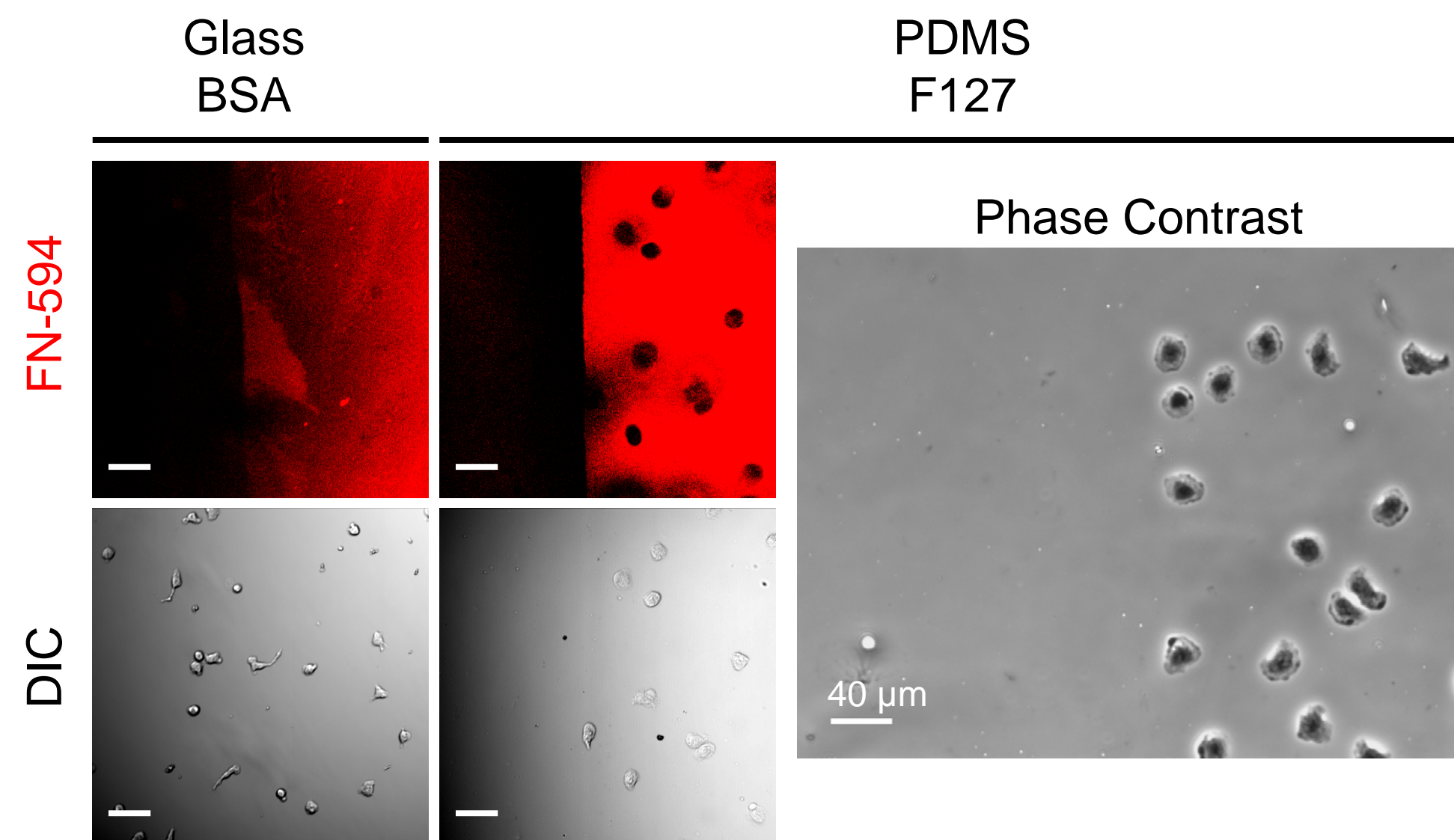


References:

- (1) Tatsuo Kinashi. 2005. Nat Rev Immunol. 5(7):564
- (2) Jannat, R et al. 2011. Biophys J. 101(3):575
- (3) Kishimoto, TK et al. 1989. Science. 245(4923):1238

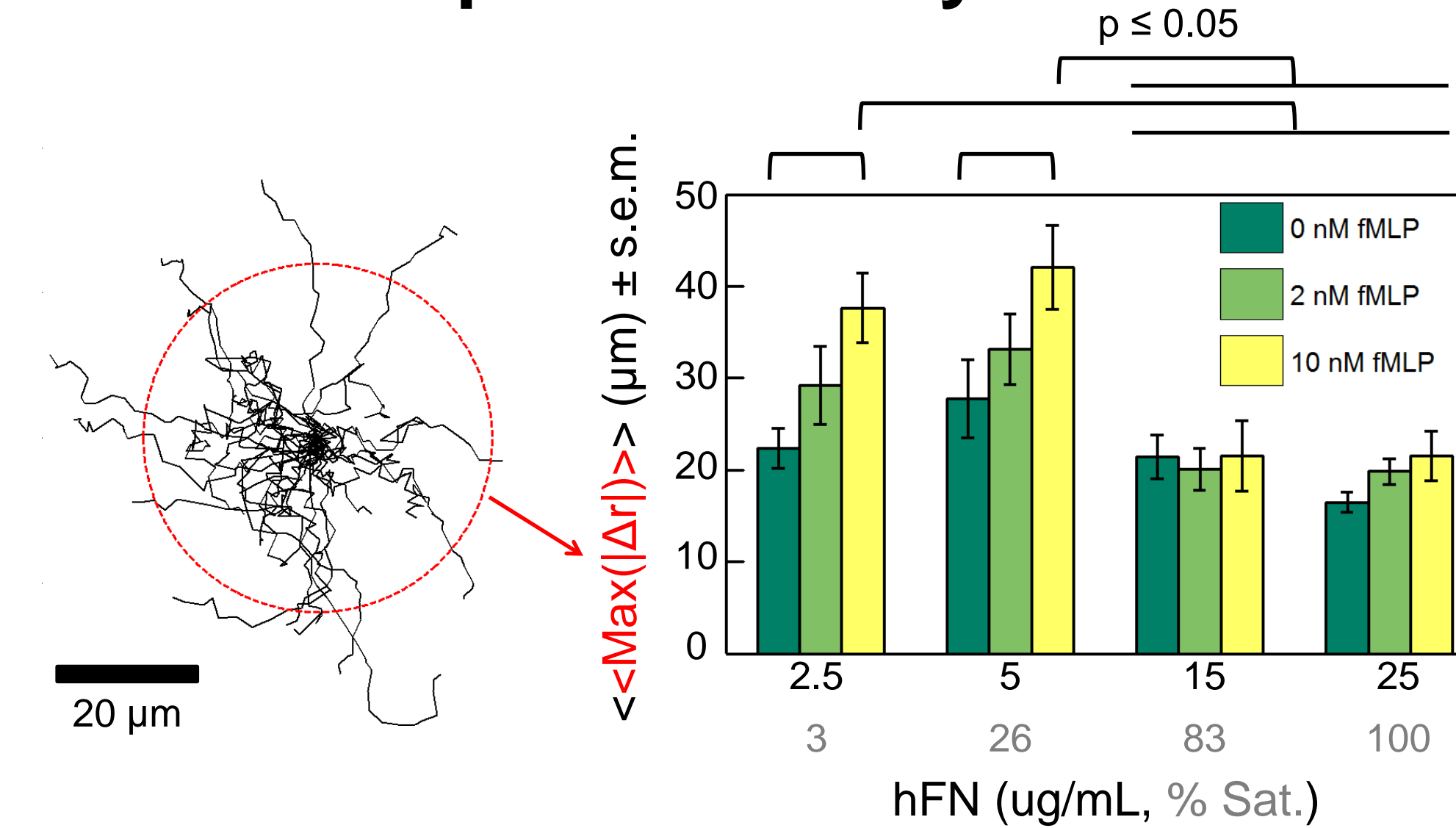
Results

Exquisite cell-FN interaction



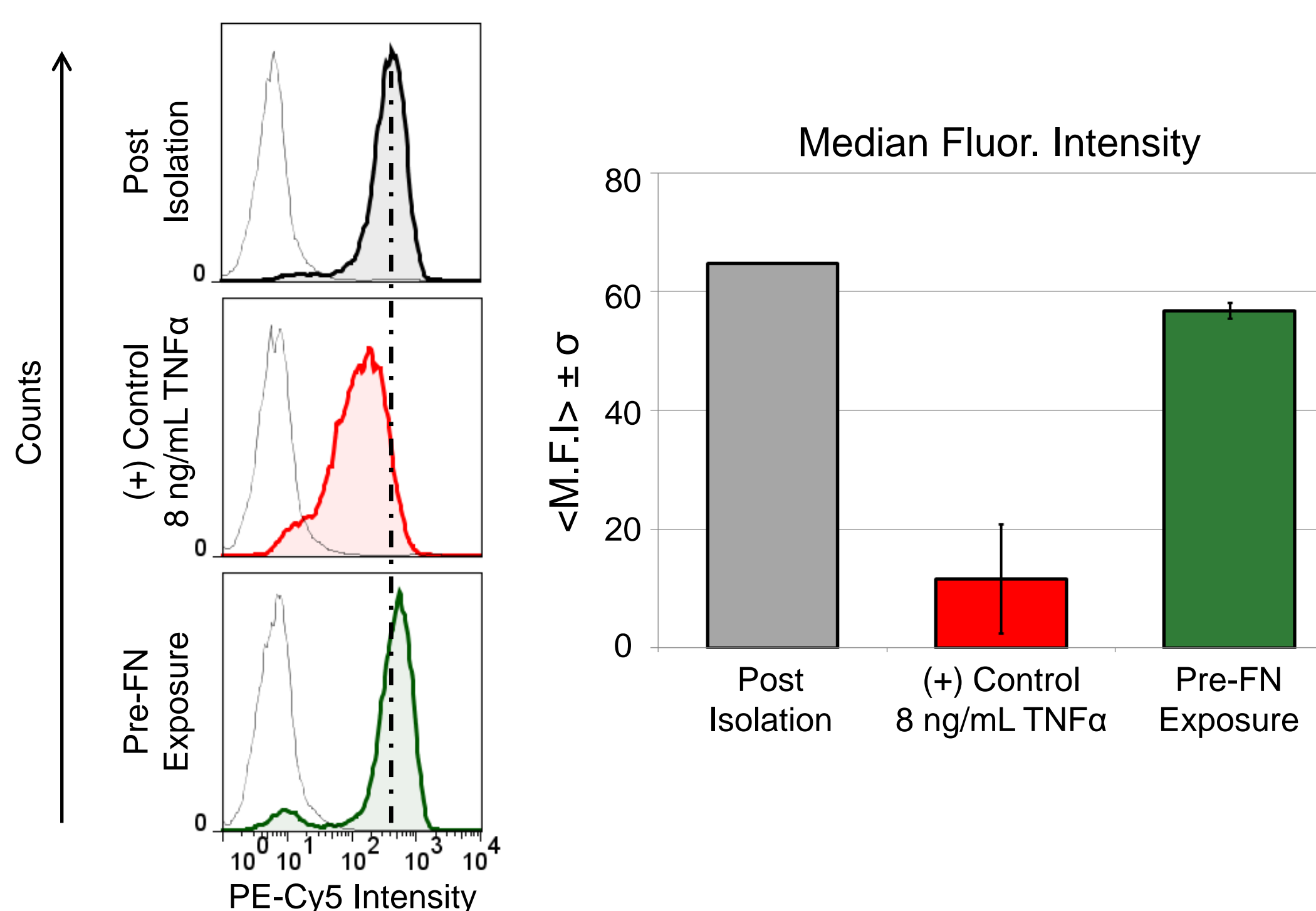
No off-FN adhesion is observed on printed PDMS, blocked with Pluronic F127.

Model-independent analysis



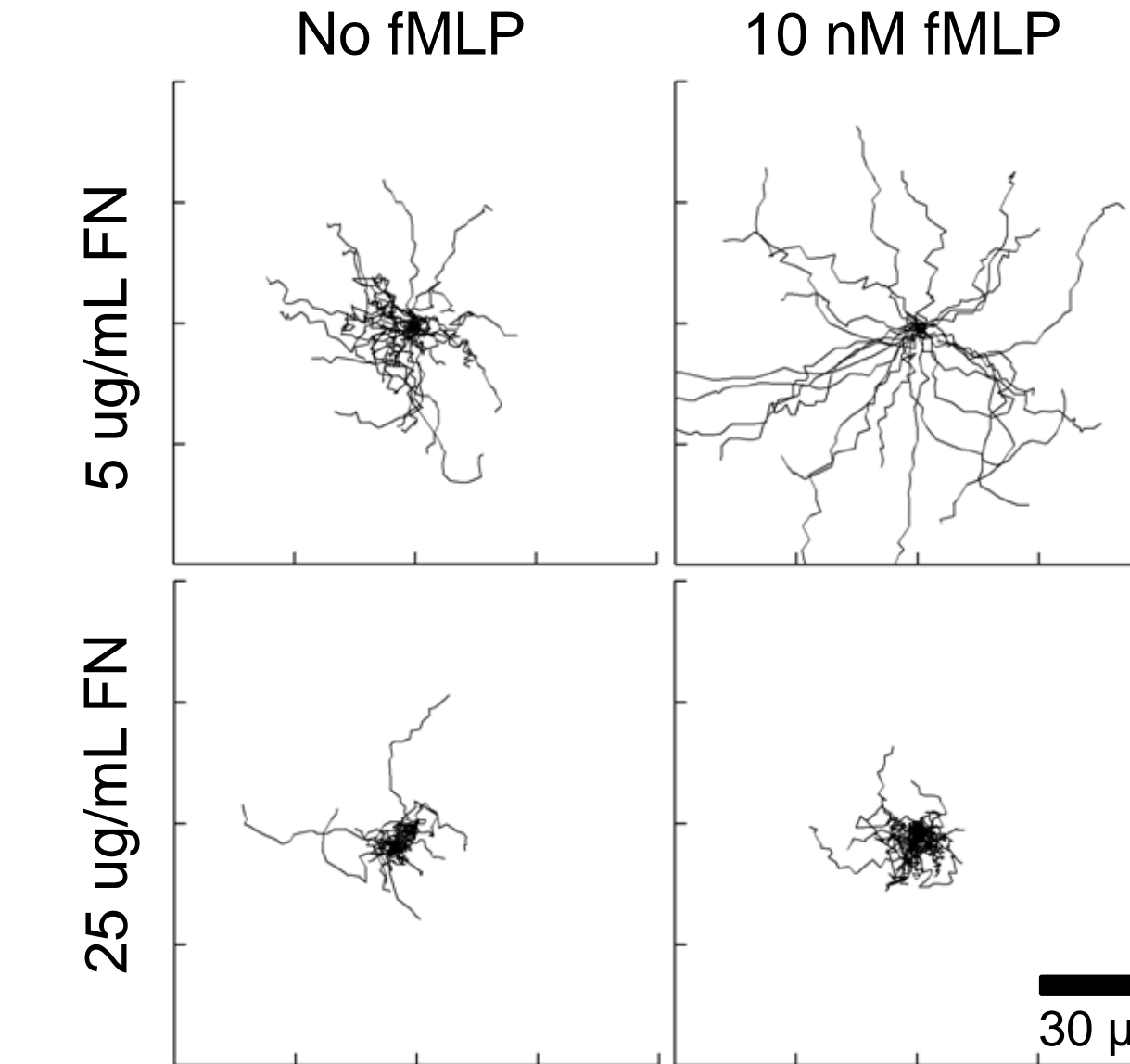
At low adhesiveness, fMLP increases extent of motility, but beyond an adhesive threshold fMLP sensitivity is attenuated.

L-selectin as activation marker



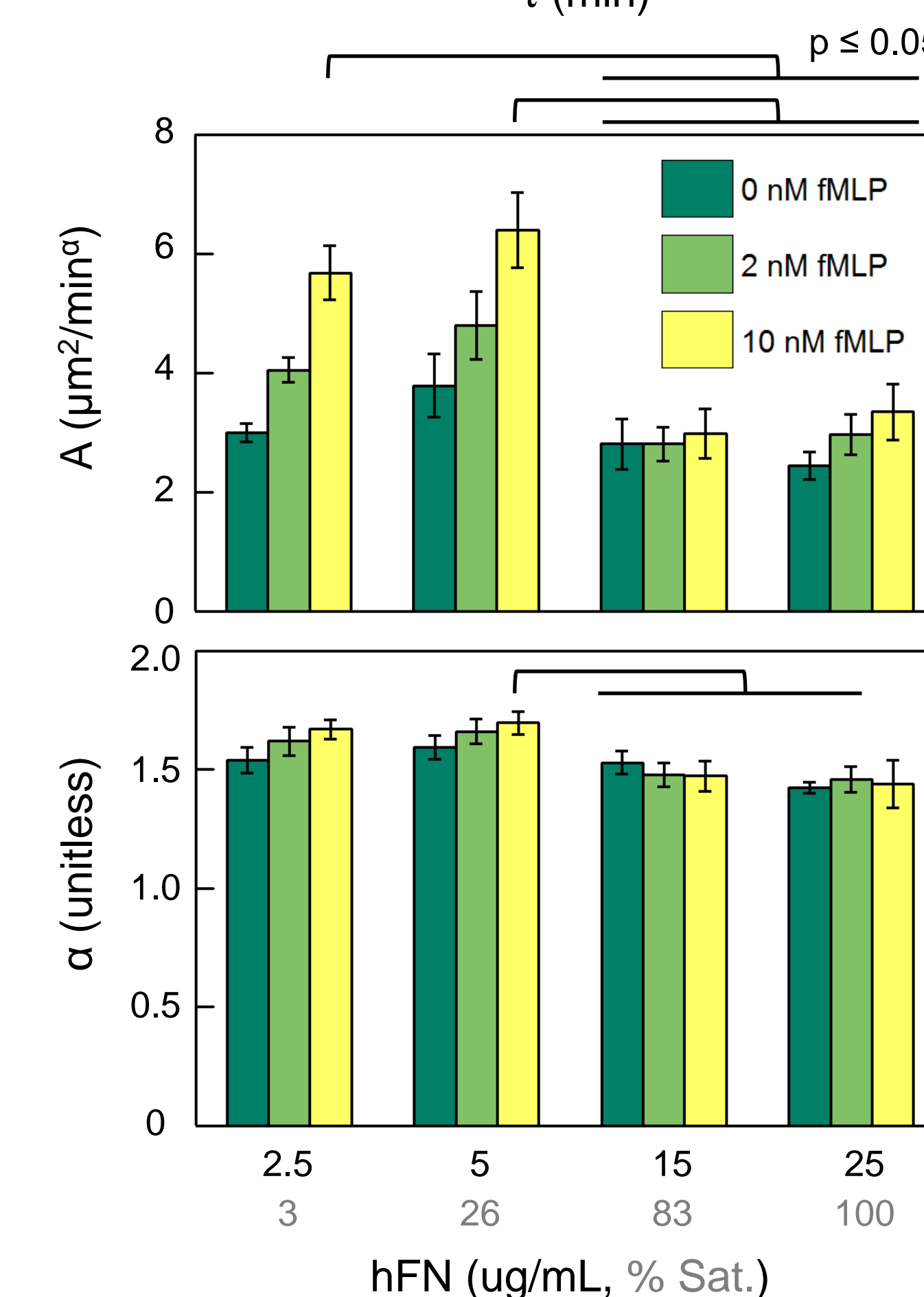
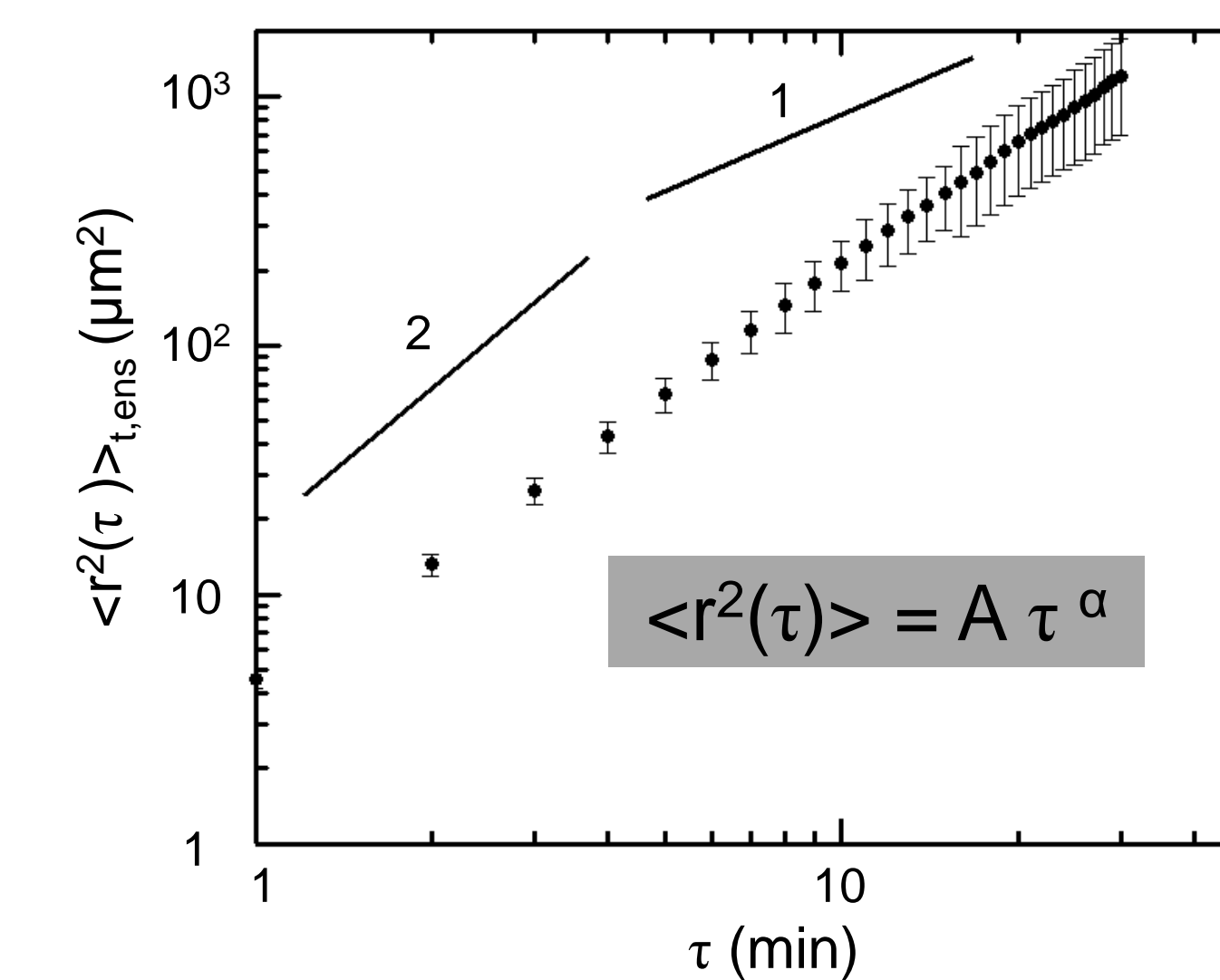
L-selectin is a sensitive marker of activation state (3). An active phenotype is **not** found before FN stimulation suggesting activation is FN-induced via an outside-in pathway.

Tuning motility



Single donor trajectories. Increasing adhesiveness decreases fMLP potentiation.

Superdiffusive motility



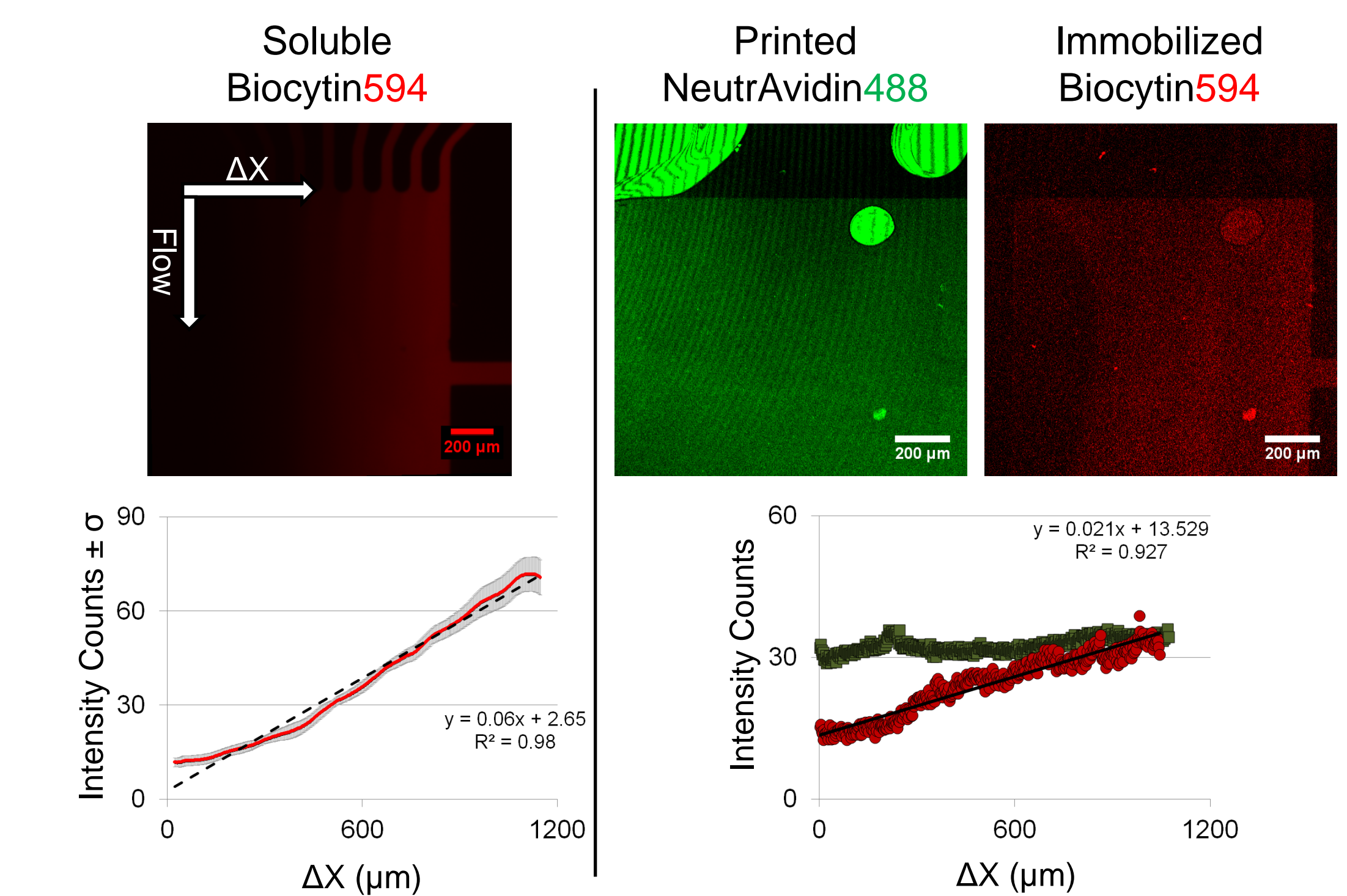
Applying a power-law model preserves trend revealed previously. Across all conditions tested, initial motility (30 min) is superdiffusive ($\alpha \sim 1.5$).

Summary

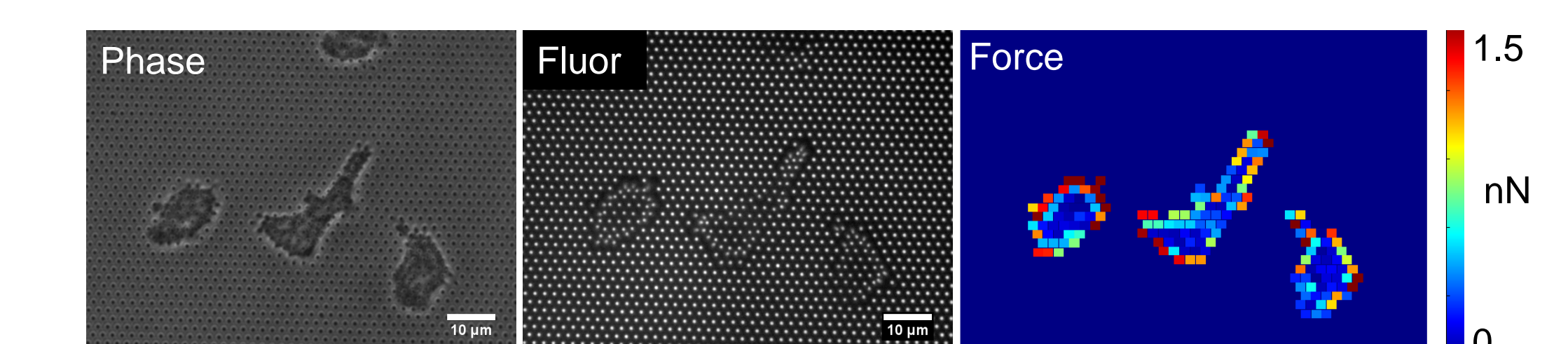
- fMLP potentiates human neutrophil motility on μ CP FN (chemokinesis)
- Quantified baseline motility metrics on functionalized half-spaces
- FN haptokinesis is not result of pre-FN activated phenotype suggesting model of outside-in activation
- Adhesiveness alone can potentiate motility suggesting haptotactic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

Future Directions

Immobilizing adhesive gradients



Traction Mapping via mPADs



Acknowledgements

- Gratitude is expressed to Christopher S. Chen, PhD for sharing μ CP and mPADs expertise.
- Work on gradient immobilization is collaborative endeavor with Neha P. Kamat.
- This material is based upon work supported by the National Science Foundation Graduate Research Fellowship.

