Beyond Elastic Modulus: The Role of Interfacial Mechanics in Cell Behavior

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Statement of Purpose: It has been well established that mechanical properties of the cellular microenvironment influence cell function, including proliferation, adhesion, migration, and viability[1]. Many common models have been established to evaluate cell response to mechanics, including hydrogel and thin film substrates that can be manufactured with a range of mechanical properties spanning a few kPa to the GPa range. However, this work has primarily focused on measurement of the Young's or Elastic modulus of the material. Recently, we have shown that interfacial properties that occur at the interface between the thin film and its support can strongly influence cell response. Here, we extend upon this work to show that unsupported materials, which have the same elastic modulus, but different tension, can exhibit altered mechanical properties that influence cell behavior.

Methods: Two materials were examined, Matrigel hydrogels and poly(caprolactone) electrospun fiber mats (EFMs). These materials were synthesized as described previously[2, 3]. For Matrigel, OSU-2 cells obtained from glioma patients under written consent and IRB protocol 2005C0075 were prelabeled with Cell Tracker Green CMFDA (Invitrogen) and cultured[2] at 3000 cells/80 μL Matrigel hydrogel (40wt%) supported on a glass substrate. Cell morphology and migration were evaluated as a function of depth from the gel-glass interface using confocal fluorescence microscopy. Finite element modeling (FEM) using ABAQUS CAE 6.8-1 (Dassault Systèmes Simulia Corporation) was performed to evaluate mechanical properties as a function of depth from the gel-glass interface. Also, PCL EFMs were examined in three configurations: supported on cylindrical polydimethylsiloxane (PDMS) substrates (E, PDMS ~ 1.7 MPa vs. 0.8 MPa for EFMs), supported on donut-shaped PDMS substrates, resulting in an unsupported interior region, and supported on PDMS substrates but after severing attachments at the fiber mat edge ("tension-released"). Cell morphology of OSU-2 cells described above and U87 and U251 glioma lines obtained from ATCC and cultured as per manufacturer's instructions was examined as a function of mat thickness (50-200 μm) on supported EFMs and as a function of tension (donut and tension-released systems).

Results: In the Matrigel model, cells exhibited differences in both morphology and migration as a function of distance from the glass interface. Specifically, cells closer to the more rigid glass interface (> 100,000 Pa vs. ~450 Pa for Matrigel) exhibited a higher degree of spreading, polarization, and migration, consistent with *in vivo* like behaviors. At a depth of ~ 50 μm from the gel-glass interface, cells became rounded with little or no observable migration. Additionally, cells closest to the interface exhibited distinct behaviors from those cultured on the glass interface alone, including reduced cell area but increased aspect ratio, indicating the role of the 3D Matrigel substrate in influencing cell response.

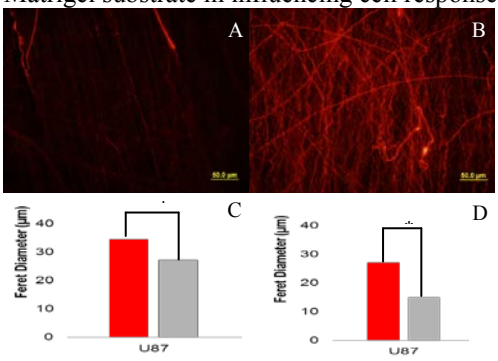

Figure 1. (A) EFM under tension (B) Tension-Released EFM. Cell spreading on (C) supported (red) vs. unsupported (gray) EFMs and (D) supported (red) vs. tension released (gray) EFMs.

For EFMs, cell spreading (feret diameter) in all three systems decreased with increasing mat thickness. Additionally, a dependence on tension was observed, with a statistically significant decline in feret diameter in unsupported systems for U87 cells, but increases in feret diameter for U251 cells. This suggests differential response to mechanical stimuli by cell type and possible involvement of differing chemical mechanisms. The moduli of these systems were also modeled using FEM, which predicted declining modulus with decreasing support.

Conclusions: Here, we demonstrate the importance of interfacial mechanics in modulating cell behavior, which should be considered in experimental design. Measurement of Elastic modulus is not sufficient to fully characterize material behaviors and must be coupled with consideration of the support material (if any) and the tension of the underlying materials as well.

References:

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