Fetuin-A adsorption to polydimethylsiloxane with varying elastic modulus

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Introduction

An early biological response to the introduction of biomaterials, **protein adsorption** is a precursor to cellular adhesion, thrombus formation, and inflammation

 Total amount and orientation of adsorbed protein influenced by a number of material properties



Polydimethylsiloxane (PDMS)

 Tunable elastic modulus that covers a range of biologically relevant stiffnesses¹

| PDMS Mix | Elastic Modulus |
|-----------------------|-----------------|
| Sylgard 184 | 4.20 MPa |
| 5:1 (Syl 184:Syl 527) | 3.97 MPa |
| 1:1 (Syl 184:Syl 527) | 2.06 MPa |
| 1:5 (Syl 184:Syl 527) | 0.35 MPa |
| 0.50% PDMS-PEG | 3.11 MPa |

Fetuin-A

- Globular plasma protein present in all major organs during fetal development²
- Shown to adsorb to biomaterials in significant amounts affecting cell response³

Hypothesized functions include:

- Regulation of mineralization²
- Pro- and anti- inflammatory responses⁴

Research Aims

Altering the **surface modulus** of an underlying biomaterial has an impact on the subsequent adsorbed protein layer^{5,6} Our aims are to:

- Characterize PDMS samples
- Investigate the amount and orientation of adsorbed Fetuin-A

Methods

PDMS Sample Preparation

- PDMS formulations mixed from Sylgard 184 were prepared in a 10:1 ratio of base to curing agent, and Sylgard 527 was prepared in a 1:1 ratio of part A and B.
- The Sylgard 184 and 527 were then mixed in ratios of 5:1, 1:1 & 1:5.
- Mixed PDMS was poured into petri dishes and placed in a vacuum desiccator for 30mins then left to cure for 48 hours

Protein Adsorption, Elution, Measurement



o.15mg/mL protein + PBS x15 solution for 2hrs

Eluted in 2% sodium dodecyl sulfate (SDS) solution overnight Total protein adsorption quantified by Bicinchoninic Acid (BCA) Assay

Make 1% PDMS

in toluene

solutions

QCM-D Sensor Spin Coating

Deposit 50uL of

solution on

sensor surface

Surface Characterization Surface Roughness

| Water Contact Angle | | | |
|---------------------------|-------------|----|--|
| Sample | Average (°) | SD | |
| Sylgard 184 | 107 | 3 | |
| 5:1 | 108 | 3 | |
| 1:1 | 109 | 3 | |
| 1:5 | 105 | 3 | |
| 0.5% PDMS- PEG | 52 | 2 | |
| Data are mean ± SD, n = 7 | | | |

Similar angles across all but the PDMS-PEG formulation

 No change in wetting with increasing Sylgard 527

Syl 184 5:1 1:1 1:5 Data are mean ± SD, n = 2

RMS Roughness < 1nm across all samples

 No noticeable trend though experiment should be repeated with new AFM tips

XPS

| At % | PDMS | 0.5% PDMS- PEG |
|---------|-------|-------------------|
| С | 44.61 | 44.10 |
| 0 | 31.23 | 31.40 |
| Si | 24.17 | 24.50 |

Spin at 2000rpm

for 45s

 No significant difference suggest the PDMS-PEG was not at the surface during the XPS measurement

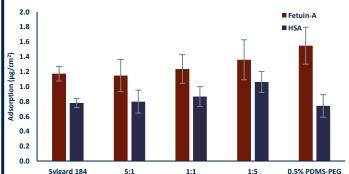
Conclusions

- Varying PDMS formulation has no measurable impact on surface wetting properties, as determined by water contact angle
- The addition of PDMS-PEG copolymer to Sylgard 184 increases the hydrophilicity of the surface
- Surface roughness does not seems to increase with the addition of Sylgard 527
- Increased fetuin-A adsorption over BSA consistent on all Sylgard 184 + PDMS-PEG samples
- Current preparation of 0.5% PDMS-PEG samples is not suitable for the expression of PEG chains at all times

Future Work

- Quartz Crystal Microbalance with Dissipation measurements in progress to determine total adsorption and indicate the adsorption state of the protein
- Adsorption from plasma to determine how fetuin-A adsorbs in competitive environments
- Macrophage studies to determine the pro- or antiinflammatory effect of fetuin-A

Protein Adsorption



- Overall greater adsorption of Fetuin-A over Human Serum Albumin
- No demonstration that changing surface modulus impacts total protein adsorption amounts
- Do not see the expected impact of 0.5% PDMS-PEG samples

Data are mean ± SD, n=5

References

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