Biomaterial Interfaces

Room 321 - Session BI-MoM

Biofilms, Biofouling, and Biomolecules at Interfaces

Moderators: Sapun Parekh, University of Texas at Austin, **Rong Yang**, Cornell University

10:00am **BI-MoM-1 Harnessing the Photorhabdus Luminescens Biofilm for Continuous Production and Downstream Processing**, ***Gabela Nelson***, *Christoper Green,* U.S. Naval Research Laboratory*; Nathan McDonald,* U.S. Army DEVCOM CBC*; Matthew Yates,* U.S. Naval Research Laboratory

Continuous flow bioreactors could allow for steady operation resulting in consistent production, minimized downtime, and reduced cost when compared to traditional batch reactor systems. Our goal is to determine the spatial and temporal production of small molecules in a fixed bed biofilm reactor to assess any heterogeneities and potential inefficiencies. We are currently testing this technology using *Photorhabdus luminescens*, a biofilm forming bacterial strain that produces small molecules, including anthraquinones and stilbenoids. We assessed reactor performance by measuring oxygen utilization under different operating conditions and resulting small molecule production during each experiment. The reactors from each experiment were evaluated for protein content and small molecule production via extraction. We were able to extract significant amounts of pigmented molecules and total protein (a proxy for biofilm biomass) from the reactors operated under different flow regimes (single pass vs. recirculation). The maximum amount of small molecules extracted was approximately 8 g/L, four times higher than *P. luminescens* grown in shake flasks. Further, protein production was approximately 1.4 g/L. These data indicate that *P. luminescens* biofilms are potentially capable of being employed in continuous biomanufacturing systems. Our results lay a fundamental groundwork toward accomplishing our long-term goal of developing an efficient system to produce small molecules in a biofilm reactor for long periods of time and at scale.

10:15am **BI-MoM-2 Flow Regulates the Fibrin–Enzyme Interface Through Multiscale Structural Remodeling**, *Sajjad Norouzi,* University of Texas at Austin*;* ***Sapun Parekh***, University of Texas Austin

Thrombolysis with tissue Plasminogen Activator (tPA), approved for treating acute ischemic stroke (AIS) within 3–4.5 h of symptom onset, converts Plasminogen (Plg) to Plasmin to degrade fibrin, while fibrin also enhances Plg activation by binding to both tPA and Plg. Therefore, in this study, we investigated whether arterial-like flow, characteristic of AIS, alters fibrin structure and susceptibility to fibrinolysis. Using a newly developed platform for quantitative imaging and spectroscopy, we found that flow generates denser fibrin networks with reduced molecular transport despite reduced protofibril packing within fibrin fibers. Raman spectroscopy revealed an α-helix-to-β-sheet transition, accompanied by reduced Plg and tPA binding, although the reduction in tPA binding emerged only after prolonged flow exposure. Consistently, multi-scale molecular dynamics simulations showed that the Plg binding site destabilized at lower forces than the primary tPA binding site. Together, these multi-scale findings provide unique molecular-level insight to explain the limited efficacy and narrow therapeutic window that underlies thrombolysis treatment.

10:30am **BI-MoM-3 Tailoring the Inert Properties of Zwitterionic Coatings**, *Jana Karthäuser, Lisa Schardt, Katrin Ademmer,* Ruhr University Bochum, Germany*; Jasper Hansen, Andre Laschewsky,* Potsdam University, Germany*;* ***Axel Rosenhahn***, Ruhr University Bochum, Germany

Manmade materials in contact with aqueous environments become rapidly colonized by living matter like proteins, macromolecules, bacteria, diatoms, and other fouling species. Frequently failure of devices and substantial maintenance costs are among the penalties associated with fouling processes. As the use of biocides is heavily restricted in many applications, environmentally friendly low-fouling materials are intensively explored [1]. In the recent years zwitterionic materials have emerged as highly promising building blocks with inherent inert properties. We synthesized different molecular architectures and studied the effect of structural variation on their inert properties [2]. The resistance to protein and microbe attachment was correlated with their anti-polyelectrolyte properties using several analytical in-situ methods including SPR, AFM, and spectroscopic ellipsometry [3,4,5]. Based on the obtained data, fundamental structure-property correlations and the implications for inert zwitterionic coatings will be discussed.

[1] M. Callow, J. Callow, Nature Communications 2011, 2, 244[2] A. Laschewsky, A. Rosenhahn, Langmuir 2018, 35, 1056[3] J.F. Karthäuser, D. Gruhn, A.M. Guajardo, R. Kopecz, N. Babel, U. Stervbo, A. Laschewsky, R. Viebahn, J. Salber, A. RosenhahnFrontiers in Bioengineering and Biotechnology 2024, 12, 1403654[4] L. Schardt, S. Da Vela, A. Martínez Guajardo, M. Jaugstetter, K. Tschulik, A. Laschewsky, A. RosenhahnAdvanced Materials Interfaces 2025, 12, 2400610[5] J. Karthäuser, J. Hansen, A. Smajlji, K.Z. Hunsucker, Y. Tenzin, C. Braga, T. Patschorke, G. Swain, A. Rosenhahn, A. LaschewskyLangmuir 2025, 41, 4545, 10.1021/acs.langmuir.4c04351

10:45am **BI-MoM-4 Graphene Oxide Coatings Enhance Biofilm Formation and Robustness in 3D-Printed Bioreactors**, ***Maryssa Beasley***, *Christopher Green, Megan Pitz,* US Naval Research Laboratory*; K.A. Shiral Fernando, Iman Eizadynejad, Oscar Ruiz,* Air Force Research Laboratory*; Rebecca Mickol,* US Naval Research Laboratory

Here we demonstrate that both graphene oxide (GO)-coated and GO-impregnated bioreactors result in faster growth and more robust biofilm formation than uncoated bioreactors when inoculated with *Marinobacter atlanticus*. GO is a nanomaterial typically used as either an irreversible binding substrate to trap microbial cells or as an antimicrobial coating, often dependent on GO concentration. Here, GO was incorporated either prior to the final resin cure or as a coating to 3D-printed bioreactors in order to increase cellular adhesion, enhance biofilm formation, and ultimately, increase bioproduction capabilities. Initial assays showed dense biofilm formation on bioreactor segments coated 10x with 0.1 wt% GO-in-water, compared to other solutions. In contrast, crystal violet biofilm assays demonstrated maximum biofilm formation on segments coated only 1x, indicating that high GO concentrations transition from biofilm-promoting to antimicrobial. In bioreactor experiments, effluent oxygen concentrations and final protein concentrations demonstrated that *M. atlanticus* biofilms colonized and grew faster in GO-coated or GO-impregnated systems than in uncoated bioreactors. Recovery, defined as the highest effluent oxygen concentration measured immediately following inoculation, reached only 40.5±13.3% oxygen and 55.5±5.7% oxygen in GO-impregnated and GO-coated bioreactors, respectively, compared to uncoated bioreactors (89.6± 4.1% oxygen). Similarly, biofilms within GO-impregnated and GO-coated bioreactors grew to maximum biomass (here, measured as effluent oxygen concentration < 2%) in less time (3.5±0.8 h and 10.5±2.3 h, respectively) than biofilms within uncoated bioreactors (15.2±3.9 h). Additionally, the same total biomass (as measured by protein concentration) was achieved in half the time with GO-impregnated bioreactors than with uncoated bioreactors (50 h vs. 100 h). When *M. atlanticus* was inoculated into bioreactors with medium that did not contain trace minerals, biofilm growth was fastest in GO-coated bioreactors than in GO-impregnated or uncoated bioreactors, with maximum biomass achieved within 13.9±2.5 h. These results suggest that GO can promote microbial growth, potentially through electron transfer, as previously seen with GO-modified electrodes, although more research is necessary. These data indicate a complex relationship between GO concentration and microbial growth, which requires further investigation to improve and optimize GO coatings for enhanced microbial production.

11:00am **BI-MoM-5 Zwitterionic Polymers for Antifouling Surfaces in Soft Devices**, ***Abdon Pena-Francesch***, University of Michigan **INVITED**

Despite recent advances in medical device technology, seamless integration in biological environments remains a challenge. When interfacing with biological environments, devices are prone to the accumulation of fouling proteins, microorganisms, and biofilms that lead to inflammation, infections, foreign body response, and failure or rejection. Thus, there is an urgent need for long-lasting, highly-stable solutions to prevent biofouling to the wide range of materials, designs, and length scales that comprise medical devices. In this talk, we will present the design of zwitterionic polymer coatings as a bioinspired surface engineering approach to avoid the attachment of proteins, bacteria, and other microorganisms to soft devices. We have recently developed surface functionalization methods compatible with medical-grade silicones (which are typically inert and difficult to functionalize), as well as zwitterionic photoresin formulations compatible with 3D-printing technology, that we have implemented in soft anti-biofouling devices. We will demonstrate their application in two case studies: (i) zwitterionic polymer coatings in urinary catheters that reduce >99% bacterial attachment and the formation of biofilms, outperforming state-of-the-art commercial antimicrobial catheter devices, and (ii) 3D-printed microrobots that avoid immune recognition and clearance by macrophages, significantly prolonging the functional lifetime of medical microrobots.

11:30am **BI-MoM-7 Molecular Shape and Peeling-Stretching Mechanics of Collagen Adsorbed on Silicate Surfaces***, **Bruno Zappone,*** Consiglio Nazionale delle Ricerche - Istituto di Nanotechnology (CNR-Nanotec), Italy
INVITED

Collagen is the most abundant protein in mammals and plays a critical role in tissue formation, stability, and mechanics, closely linked to its unique triple-helix structure. We used atomic force microscopy (AFM) and the surface forces apparatus (SFA) to study fibril-forming type I and III human collagen absorbed on atomically smooth mica surfaces. The two-dimensional contours of individual collagen molecules showed non-uniform curvature for both types, which persisted after drying the surface and dehydrating the molecules. This finding suggests that collagen either has an intrinsic three-dimensional curvature in solution, or it acquires a two-dimensional curvature due to a particularly strong interaction with the surface. The former case has direct relevance to the self-assembly and elasticity of collagen fibrils, whereas the latter has potential implications for biomaterial design and tissue engineering.

SFA measurements showed strong adhesion between a collagen-coated and an uncoated mica surface, and repulsion between equally coated surfaces, suggesting that collagen-mica interaction is mainly electrostatic. Moreover, AFM force measurements showed a large adhesive force for a single collagen molecule being peeled by one end from the mica surface. Interestingly, a collagen molecule could be pulled and stretched without failing to a contour length of up to 900 nm—nearly three times its native length—over thousands of stretching cycles until the triple-helix started unravelling. During this process, the alpha-chains slipped progressively, irreversibly, and almost entirely past each other, before being caught by strong physical interactions between overlapping chain ends. These findings indicate that a controlled slippage mechanism at the molecular scale underpins the exceptional toughness of collagen fibrils and collagen-rich tissues such as tendons and skin.

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