Bacteria-Triggered Antimicrobial Release from Complexation-Loaded Polyelectrolyte Microgels

Xixi Xiao and Matthew Libera

Dept of Chemical Engineering & Materials Science, Stevens Institute of Technology, Hoboken, NJ, 07030

Statement of Purpose: Self-defensive biomaterial surfaces offer a compelling possible solution for biomedical-device-associated infections (X. Xiao et al., Colloids Surf. B, 2020.110989). In contrast to delivery systems that continuously elute antimicrobials, a self-defensive approach releases an antimicrobial in response to a bacterial trigger only when and where on a surface that challenge occurs (Figure 1). Hence, a self-defensive approach does not unnecessarily promote the development of resistant bacterial strains.

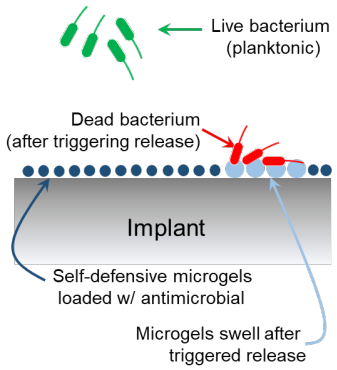

Figure 1. A microgel-coated self-defensive surface responds to a bacterial challenge by locally releasing antimicrobial (light blue). Antimicrobial elsewhere in the coating remains sequestered (dark blue).

Prior work (J. Liang et al., Biomaterials, 2019. 204: p. 25-35) shows that poly(acrylic acid) (PAA) microgels can load and sequester colistin (polymyxin E), an FDA-approved antibiotic, in low-ionic-strength (0.01 M) buffer. Colistin release is triggered by *E.coli* contact. However, under physiological conditions (pH 7.4, ionic strength 0.14 M), the colistin is fully released in a rapid burst because the additional Na^+ interferes with the complexation between the PAA and the colistin. In contrast, two antimicrobial peptides, L5 (+6, 2274 Da) and Sub5 (+7, 1723 Da), show enhanced complexation strength with PAA microgels and exhibit good self-defensive behavior. With the purpose of creating a self-defensive surface using FDA-approved antibiotics, we are investigating the complexation strength between the cationic antibiotics and anionic microgels to ensure long-term sequestration and bacteria-triggered release.

Methods: Poly(styrene sulfonate) (PSS) microgels were prepared by thermo-initiated free-radical polymerization of styrene sulfonate in a water-in-oil suspension. PAA microgels were synthesized by photo-initiated free-radical polymerization of acrylic acid. The antimicrobial loading, sequestration, and release were studied by *in situ* optical microscopy in a continuous flow chamber with control over pH, ionic strength, and antibiotic concentration.

Results: The results show that PSS microgels can load and sequester colistin under physiological conditions. In contrast to PAA where [Na+] = 0.136 M will fully release the complexed colistin, results show that PSS only release part of its payload colistin (Figure 2). Importantly, the colistin/PSS complexation is not fully shielded by salt and. However, as manifested by the change in microgel diameter, that complexation is disrupted by bringing *E. coli* into the flowing medium.

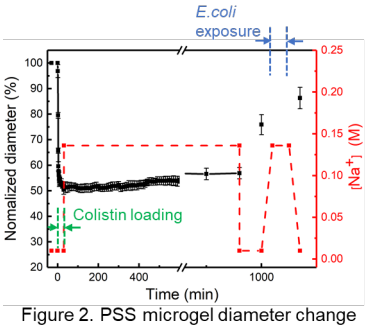

Figure 2. PSS microgel diameter change during loading (1 mg/ml colistin in 0.01 M phosphate buffer) and soaking in phosphate buffer with [Na+] = 0.136 M and adding 10^8 CFU/ml *E.coli*

The chemical structure of polymyxin B is very similar that of colistin except for the substitution of a phenyl ring for a dimethyl group. We studied the threshold [Na^+] and [Antimicrobial] ([Am]) for breaking and maintaining the complexation to demonstrate the complexation strength, both results show an enhanced complexation for polymyxin B with PSS.

Conclusion: We have studied the complexation of FDA-approved antibiotics with PAA and PSS microgels. Both sets of microgels can complex with polymyxin antibiotics. When that complexation is disrupted, colistin is released and the microgels swell. PSS shows stronger complexation with colistin than PAA, and we attribute this to the additional interactions, beyond electrostatics, introduced by aromaticity in both the antibiotic and in the microgel. Importantly, PSS/colistin complex was disrupted when *E.coli* was added into flowing buffer. The negatively charged bacteria envelop is in close physical proximity with antimicrobial-sequestered microgel, provides an energetically favorable environment for cationic antimicrobial, favors the release, and transfer the antimicrobial to the bacterial.