

Computational Analysis and Visualization of Biofilm Development on Implant Materials

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Purpose: Understanding how infection develops following surgery is of paramount importance. This is particularly true in orthopaedics, where arthroplasty and fracture implants may be colonized by opportunistic pathogens. These organisms form biofilms, dense colonies of bacteria largely resistant to antibiotics and the immune system. Research to formulate materials and irrigation methods that reduce or remove biofilm formation on implants is being carried out, but no reproducible platform to quantitate biofilm formation is available. There is a need for a technique to evaluate the efficacy of these therapeutic methods and to understand biofilm development.

Methods: A computational tool was developed in the Python coding language that consistently identifies and measures the presence of bacterial biofilm on implant materials. This tool takes a series of confocal microscopy images corresponding to vertical slices of the biofilm and calculates the amount of protein, extracellular DNA, and exopolysaccharide, which have been stained with Sypro Ruby, TOTO-1, and Concanavalin A dyes. The relative ratio and number of pixels covered by each component is output and a 3-dimensional plot is created depicting biofilm generation in a spacio-temporal manner.

Results: The program has shown to be successful by consistently and accurately producing coverage values and 3-dimensional plots for explant materials and in vitro testing. This tool, in conjunction with an in vitro biofilm platform and hospital-acquired explant materials, has been able to quantify the components of biofilms in as little as 30 seconds. In experiments aimed at showing the efficacy of irrigation agents on pre-established biofilms, this technology demonstrated an 86% decrease in biofilm mass in a 3-dimensional and easy-to-read output.

Conclusion: With this technique, various interventions designed to limit biofilm formation on implant materials can be tested and compared with the goal of discovering the most effective form of treatment. The data acquired via this high-throughput, quantitative, and visual tool will prove critical to the study of biofilms and removal methodologies.