

An Electrochemically-Active Biosensor to Study the Development of Biofilm in Wild-Type and Fimbriae- Deficient *E. coli* Mutants

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I. INTRODUCTION

Biofilms are the clusters of bacterial aggregates, encased in a self-produced matrix of extracellular polymeric substances and adherent to the surface. Biofilms pose significant consequences in medical settings, associated with 80% of microbial and 60% of all infections, originating in a hospital. ¹. A standard technique for biofilm determination is through crystal violet staining. An irreversible assay, it produces inconsistent response in the case of different mutants or antimicrobial agents ². To address this, a novel biosensor has been developed to investigate *E. coli* biofilm life cycle by selecting wild-type (WT) and strains without fimbriae (genes involved in biofilm formation – *fimC* and *fimD*) mutants.

II. RESULTS

WT, *fim C* and *fim D* mutants were grown on an interdigitated electrode and cyclic voltammetry with 3 cycles from -0.3 to 1.0V was performed in every 6 hours within the period of 48 hours to study the full life cycle (Fig. 1).

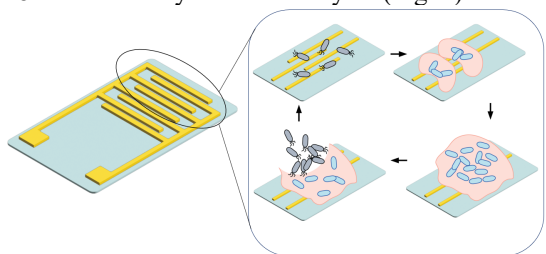


Fig. 1 The schematic view of cell growth on the gold-coated electrodes

The cyclic voltammogram are achieved for different strains. WT, *fim C* and *fim D* mutants show a broad peak at 0.7 V that can be interpreted as adsorption of oxygen for the electrode (Fig. 2 A). Initially, the enhanced electron transfer between bacteria (that are closer to the surface) and the electrode promotes the adhesion of biofilm and increases the current. After biofilm maturation, the electroactive area for electrochemical interaction diminishes due to the coverage of cells on the surface, which decreases the current peak.

Finally, within the period of 42-48 hours, the peak of the current drastically decreases, suggesting the dispersal stage of the biofilm. In comparison, both *fimC* and *fimD* mutants demonstrate a consistent voltammogram within 48 h (Fig. 2 B and C). Type 1 fimbriae are versatile virulence factors that stabilize and mediate the adhesion and required for biofilm formation in *E. coli* ³. Due to the lack of fimbrial adhesins, cells in *fimC* and *fimD* do not adhere to the surface and affect the voltammogram. Therefore, the electroactive area coverage for *fim C* and *fim D* remains constant overtime (Fig. 2D).

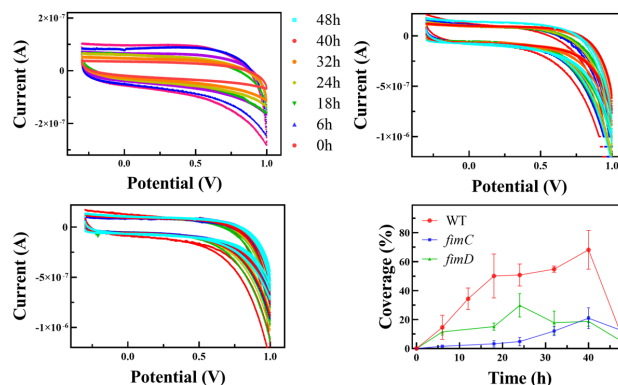


Fig. 2 Cyclic voltammogram of A) wild-type, B) *fimC*-, C) *fimD*-deficient (legend shows the measurement at different time spots), and D) the coverage area of the three strains vs time

The results suggest the developed biosensor can be used to determine the biofilm full life cycle and differentiate between the initial adhesion, maturation, and dispersion of biofilm as well as between biofilm-promoter *E. coli* mutants.

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