



Proteinaceous Surface Appendage Contribution to *Pseudomonas aeruginosa* PAO1 Surface Properties and Adhesive Ability

J. B. Morrow♣, B.F.Smets¥, and D.GrassoEUR

♣Biochemical Science Division, National Institute of Standards and Technology, Gaithersburg, MD

¥Environment and Resources DTU, Technical University of Denmark, DK-2800 Lyngby, Denmark

EUR College of Engineering and Mathematical Sciences, The University of Vermont, Burlington, VT

Bacterial transport and attachment to surfaces is of considerable importance to engineered and natural systems. Bacterial surface association and subsequent biofilm formation is governed by the initial attachment step. Initial attachment of pathogenic bacteria to environmentally and medically relevant substrata is governed by cell and substratum surface features, including nanoscale roughness, charge and surface chemistry, and the chemistry of the intervening fluid. This work focused on the role of specific proteinaceous appendages in altering *Pseudomonas aeruginosa* PAO1 cell surface properties and macro and micro-scale adhesive ability to glass. Mutants for various chromosomal encoded surface appendages, flagella, (*fliM*-), type IV pili (*pilA*-), both type IV pili and flagella (*pilA*- and *fliM*-) were used to determine appendage contributions to PAO1 adhesion. Type IV pili had the most pronounced effect on cell surface properties and adhesion to glass in batch adhesion studies (batch liquid and Total Internal Reflectance Fluorescence, TIRF, microscopy observations). Flagella had a larger impact in column studies represented by larger blocking factor values $\beta = 44$ and $\beta = 21$ for WT and *pilA*-, respectively, compared to $\beta = 7$ for both *fliM*- and *fliM-pilA*- mutants. Adhesive behavior determined in macro-scale, batch adhesion studies to glass slides (quantified by surface coverage, \tilde{A} , sgg after 30 min exposure) was larger for appendage laden wild type (WT) cells ($\tilde{A} = 48 \times 10^{11}$ cells/m²) than for mutants ($\tilde{A} = 0.92 - 19.6 \times 10^{11}$ cells/m²). Macro-scale adhesion behavior was at-

tributed to cell surface interaction due to appendage initiated attachment. Micro-scale measurements collected with atomic force microscopy (AFM) indicated that all PAO1 strains exhibited large pull-off forces. Larger micro-scale adhesive forces observed for PilA- strains were attributed to increased polymer/probe interaction for type IV deficient cells. Micro and macro-scale measurements suggest proteinaceous appendages initiate contact with the substratum and moderate adhesive strength.