BP Manchester Summer Intern Programme 2016

Project Title: The Growth of Bacterial Biofilms on Graphene

Student(s): Justinas Rumbutis (in collaboration with Jack Hart)

Academic Supervisor(s): Dr Thomas Andrew Waigh

Project Description

Bacterial biofilms cause serious problems in oil and gas industry by damaging metal pipes and producing marine biofouling on sensors and the hulls of ships. This topic has received much attention of researchers and a lot of investigation has been done in understanding the adhesion process, the first stage of biofilm formation [1]. It is anticipated that graphene surfaces can reduce the bacterial adhesion and this nanomaterial can be used in making anti-biofouling paints. However this effect has not been fully explored yet to draw final conclusions [2].

In this project an experiment of bacterial biofilm growth on a graphene oxide surface was performed and a simulation of bacteria adhesion to the surface process was developed. Several theoretical models were used and by comparing results of deposition kinetics obtained by these models to the actual experimental data the dominant effects of the process were identified.

Project Objectives / Deliverables

The main objective of this project was to develop a simulation capable of predicting experimental results. Several methods were used and several versions of the simulation were developed. A real and a simulated image of bacterial deposition are shown in Fig. 1 and Fig. 2 respectively.

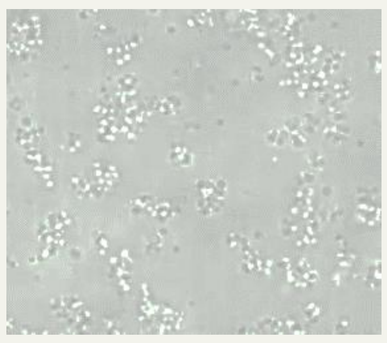

Figure 1. Staphylococcus aureus (~1μm in size) adsorption measured using a bright field microscope, ultrafast CMOS camera and microfluidic flow cell.

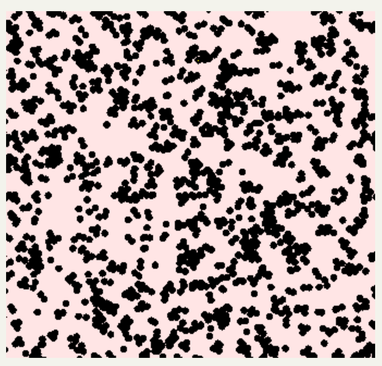

Figure 2. Deposition simulation of polydisperse particles, moving in Brownian motion under gravity with a sticking probability of 5%.

References

[1] Mazza, M. G., The Physics of Biofilms—an Introduction, *Journal of Physics D: Applied Physics*, 49,20, 2016.

[2] Yoshida, N., Miyata, Y., Doi, K., Goto, Y., Nagao, Y., Tero, R., Hiraishi, A., Graphene Oxide-Dependent Growth and Self-Aggregation Into a Hydrogel Complex of Exoelectrogenic Bacteria, *Scientific Reports*, 6, 21867, 2016.

[3] Privman, V., Recent Theoretical Results of Nonequilibrium Deposition of Submicron Partcles, *The Journal of Adhesion,* 74, 421-440, 2000.

[4] Carla, P., Schxccmittbuhla, M., Schaafb P., Stoltzc, J., Voegela, J., Senger, B., Experimental and Modeled Deposition Kinetics of Large Colloidal Particles, *Physica A: Statistical Mechanics and its Applications*, 298, 1-2, p. 198-228, 2001.

[5] Bybee, M. D., *Hydrodynamic simulations of colloidal gels: Microstructure, dynamics, and rheology*, PhD thesis, University of Illinois at Urbana-Champaign, 2009.

[6] Delong, S., Usabiaga, F. B., Donev, A., Brownian dynamics of confined rigid bodies, *J. Chem. Phys.,* 143, 144107, 2015.

[7] Waigh, T. A., *The Physics of Living Processes: A Mesoscopic Approach*, Wiley, First Edition, 2014, p. 69-78.

[8] Bolintineanu, D. S., Grest, G. S., Lechman, J. B., Pierce, F., Plimpton, S. J., Schunk, P. R., Particle dynamics modeling methods for colloid suspensions, *Computational Particle Mechanics*, 1,3, p. 321–356, 2014.

[9] Redon, S., Kheddar, A., Coquillart, S., Fast Continuous Collision Detection between Rigid Bodies, *Eurographics*, 21, 3, 2002.

[10] Baraff D., Fast Contact Force Computation for Nonpenetrating Rigid Bodies, *Proceedings of the 21st Annual Conference on Computer Graphics and Interactive Techniques - SIGGRAPH '94*, p. 23–34, 1994.

[11] MATLAB, Version R2015, The Math Works Inc, Natick, Mass 01760.

Approach / Methodology Employed Experiments, Models, Designs Used / Developed

The experiment was performed using Staphylococcus aureus bacteria that are spherical in shape and do not have any motility mechanism. The deposition process was recorded using high frame rate camera and poly particle tracker for finding positions of bacteria and obtaining the surface coverage, the number of bacteria adsorbed to the surface as a function of time.

The first stage in developing the simulation was to implement two basic models of particle deposition and compare results with theoretical predictions. The first model, random sequential adsorption (RSA), describes particle adsorption in the case where thermal motion dominates over gravity. If a particle directly hits the surface without colliding with other particles, its position is fixed and hence it is adsorbed. Otherwise the particle is removed from the system [3]. The second model, ballistic deposition (BD), describes the deposition process governed by gravity and considers particles rolling over pre-adsorbed ones. A particle moving in a path of steepest descent can reach the surface and be adsorbed or it can stop at a position of stable equilibrium on top of deposit particles. These two models are the two limiting cases of the deposition process [4]. They were then extended to an agent-based simulation which included hydrodynamic interactions and electrostatic forces between simulated particles. The time step of updating particle positions was much larger than the particle momentum relaxation time, so a non-inertial version of Langevin equation was used. It can be written as:

$$\boldsymbol{F}^{H} + \boldsymbol{F}^{B} + \boldsymbol{F}^{C} = \boldsymbol{0}, \qquad (1)$$

where $\boldsymbol{F}^{H}, \boldsymbol{F}^{B}$ and $\boldsymbol{F}^{C}$ are $6N$ (N is a number of particles) dimensional vectors containing all hydrodynamic, Brownian and conservative forces and torques respectively. Using the linearity of Stokes equations that describe fluid motion, the hydrodynamic force was expressed as a linear function of all particle velocities:

$$\boldsymbol{F}^{H} = \boldsymbol{R} \cdot \boldsymbol{U}, \qquad (2)$$

where $\boldsymbol{R}$ is the resistance tensor which was obtained using fast lubricant dynamics (FLD) technique [5], $\boldsymbol{U}$ is a vector containing all velocities/angular velocities of the particles with respect to the fluid. The Brownian forces were generated using random vectors obeying the fluctuation-dissipation theorem [6]. The conservative forces consisted of gravity, electrostatic double layer force and dipole – dipole (dispersion) interactions. The potential of the last two forces, predicted by DLVO theory is shown in Fig. 3.

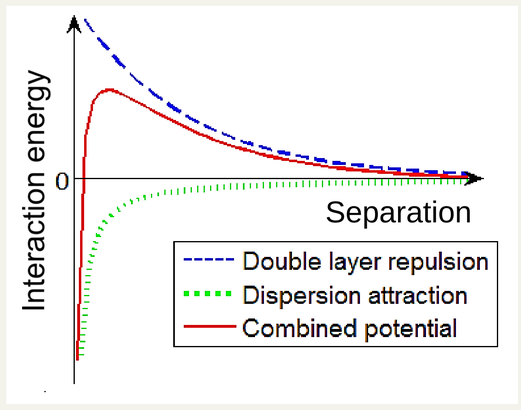

Figure 3. Electrostatic double layer and dipole-dipole interaction potentials. The double layer potential arises from electrostatic interactions between charged surfaces of particles and ions in the solvent. The dispersion attraction is caused by interactions of induced electric dipoles in molecules of the particles [7].

Using velocities obtained from Eq. (1) and (2) the motion of particles was simulated using the mid-point time stepping scheme [8], the continuous collision detection [9] and the fast contact force [10] algorithms. From the experimental data it was noticed that the bacteria tend to aggregate. This behaviour was modelled by introducing a probability that during a collision two particles stick together and also by simulating rigid bodies made of several spheres.

Results

The results obtained from BD and RSA simulations were found to be consistent with theoretical predictions. The surface coverage as a function of time for these two models is given in Fig. 4 and Fig 5. The final surface coverages were found to be 0.6108 ± 0.0003 and 0.5500 ± 0.0001, while theory predicts 0.6105 and 0.55, for BD [4] and RSA [3] models respectively.

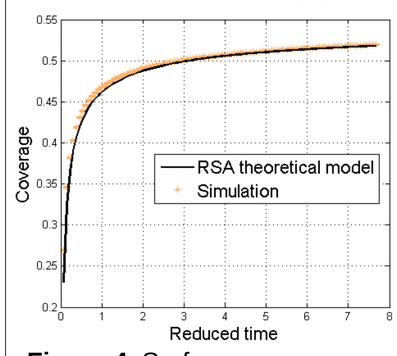

Figure 4. Surface coverage as a function of time of RSA simulation. The dimensionless reduced time is the time weighted by the incoming particle number density.

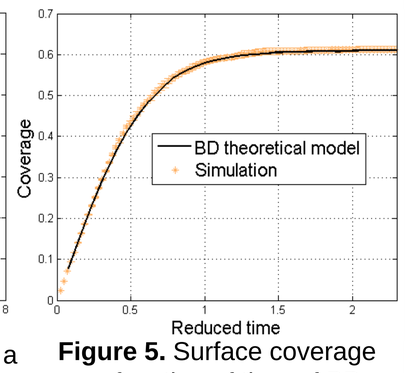

Figure 5. Surface coverage as a function of time of BD simulation.

Results of simulation with/without hydrodynamic and conservative interactions and with polydispersity of particles are shown in Fig. 6 and Fig. 7 respectively. The experimental result (Fig. 8) shows reasonable agreement with a simulation of polydisperse particle deposition.

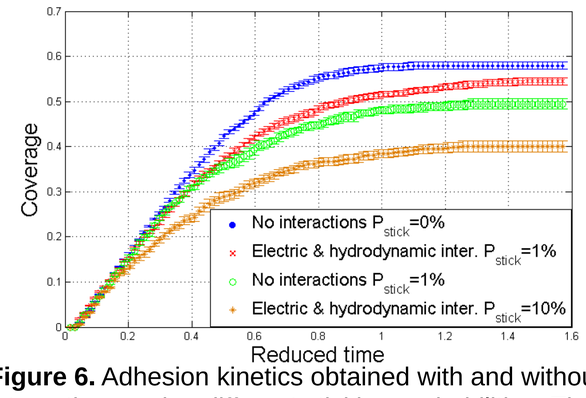

Figure 6. Adhesion kinetics obtained with and without particle interactions, using different sticking probabilities. These effects change the final surface coverage as well as the functional form of the kinetics.

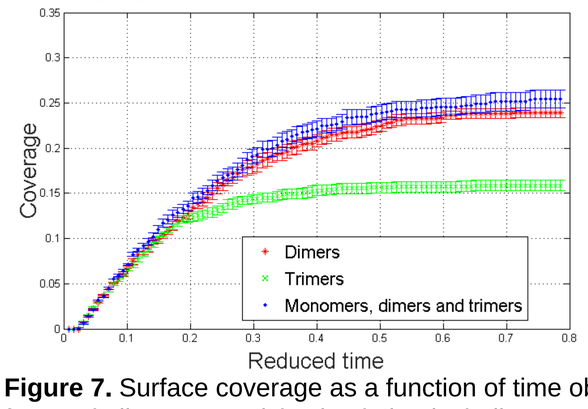

Figure 7. Surface coverage as a function of time obtained from polydisperse particle simulation including monomers (unaggregated spheres), dimers and trimmers.

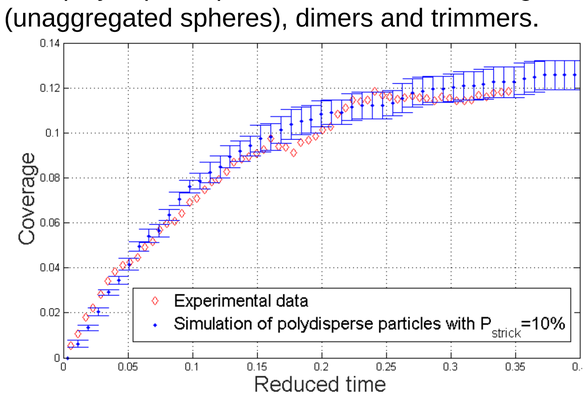

Figure 8. Experimental data of surface coverage as a function of time and results of simulation of polydisperse particles with a sticking probability of 10 %.

Overall Findings / Conclusions

By comparing the experimental data to the simulation results it was found that BD and RSA models of unaggregated spherical particles can not predict kinetics of bacterial deposition when both thermal motion and gravity are equally important. Also, it was observed that other effects, such as conservative and hydrodynamic interactions and bacterial aggregation, are significant in modelling the adsorption process. The closest result to the experimental data was obtained by simulating equal amounts of monomer, dimer and trimer particles with a sticking probability of 10%.