Simulation of the mechanical response of encapsulated individual cells during normal force spectroscopy measurements

Katerina Foteinopoulou, Manuel Laso

Institute for Optoelectronics and Microsystems (ISOM) and ETSII, Universidad Politécnica de Madrid, Spain
kfoteinopoulou@etsii.upm.es

Abstract
The atomic force microscope (AFM) [1] is a useful tool for investigating individual cells. Material properties of individual mammalian cells and bacteria have been investigated by using AFM to measure forces, a technique known as force spectroscopy. In a force spectroscopy measurement, cantilever deflection is measured as a function of sample position and is subsequently converted into force using appropriate calibrations giving the force curve (i.e. a plot of force versus sample position). However, interpreting measured force curves can be complicated because forces arising from material deformation (elastic and hydrodynamic forces) may not be distinguishable from other forces (surface forces, steric forces etc). Accurate interpretation relies on understanding the contribution of all invoked elements in the measured force curve.

In the work presented here, the mechanical response, the force-indentation relationship, in normal force spectroscopy measurements carried out on individual polysaccharide encapsulated bacteria is modelled, considering the elastic response of the bacterium and cantilever in combination with a fluid (hydrodynamic) model for the polysaccharide layer. For the hydrodynamic description of the polysaccharide layer a two-dimensional, axisymmetric, fully viscoelastic description is adopted, incorporating multi-modal Phan-Thien-Tanner (PTT) or Giesekus models. For the problem solution, viscoelastic, axisymmetric 2D time-dependent complex flow finite element (FE) calculations were carried out, taking into consideration the viscoelastic character of the polysaccharide liquid incorporating all elements of the system: AFM tip, bacterium stage and the surrounding extracellurar polysaccharide (EP) layer. Figure 1 shows a schematic representation of the problem geometry. The flow FE calculations are coupled with an adaptive remeshing strategy which is based on mapping the physical time-varying domain onto a simpler computational mesh and solving subsequently a set of elliptic partial differential equations for the physical coordinates.

In all studied cases the simulation model rigorously considers the time dependent rheological-mechanical coupling between the elastic and fluid viscoelastic physical components of the experimental setup. Effects of inherent variability in geometrical and material properties of the bacterium and polysaccharide layer on the measurable response are quantified [1]. Calculations are used to predict and explain the mechanical response in an AFM experiment of single encapsulated bacteria which are conducted to provide physical insight into the pathology of certain diseases and the development of novel therapeutics [1]. The FE results are compared with experimentally obtained curves on Staphylococcus aureus without incorporating any adjustable parameters. Rheological data for the surrounding polysaccharide layer (i.e. relaxation time and zero-shear rate viscosity) are considered from steady-state rheological measurements on bacterium polysaccharide extracted from its native environment. It is demonstrated that experimental results can be accurately described by the FE solutions of the viscoelastic fluid model [3]. Furthermore results are presented and compared for the force curve and time-dependent deformation of the physical domain (defined by the bacterium, the cantilever deflection and the EP fluid layers surrounding the bacterium). The viscoelastic fluid parameters invoked were equal to published parameters for advanced multimode constitutive equations of PTT and Giesekus as resulted from fittings of the above models to experimental rheological data of various EP viscoelastic fluids [4]. Representative results are shown in figure 2.
Our numerical method can accurately predict and describe the mechanical response of isolated encapsulated cells during a force spectroscopy measurement without the need of any adjustable parameter since all the intrinsic bacterial and EP layer parameters serve as input parameters of the model. However the EP production is an intrinsically variable property of bacteria with possible variation of the EP inherent properties (layer thickness, viscosity, relaxation time) within a population of cells. Bacterial properties (radius and stiffness) are also subject to significant variations. Therefore achieving high reproducibility in experimental measurements of single encapsulated bacteria is extremely difficult [3]. Supported by model calculations, we also point the way to methods of *in vivo* rheological

characterization of the extracellular polysaccharide as a preferable alternative to characterization after its removal from the native environment.

References

[1] Binnig, G., Quate, C.F., Gerber, C. Phys. Rev. Lett. **56**, (1986) 930.
[2] F. M. Coldren, K. Foteinopoulou, D. L. Carroll, M. Laso, Langmuir **24** (2008) 9575.
[3] F. M. Coldren, K. Foteinopoulou, W. M. H. Verbeeten, D. L. Carroll, M. Laso, Langmuir **24** (2008) 9588.
[4] W.M.H. Verbeeten, J. of Rheology **54** (2010) 447.

Figures

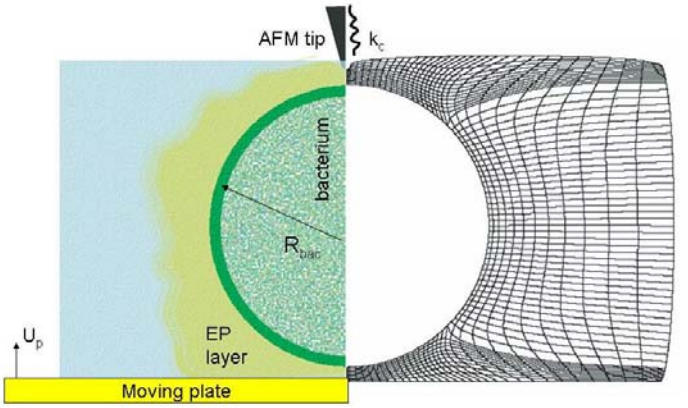

Figure 1 Schematic view of the problem geometry (left) together with the mesh generated for its finite element numerical solution (right)

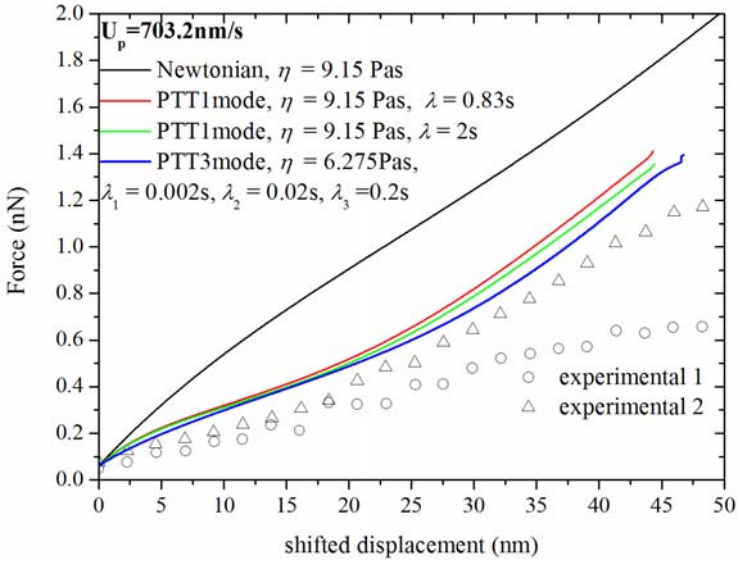

Figure 2 Representative simulation results adopting different models (Newtonian, 1mode PTT, 3mode PTT) for the description of the EP fluid and their comparison the experimental results under the same operative conditions