A model for murine layer growth and cell shape during cell division in Caulobacter

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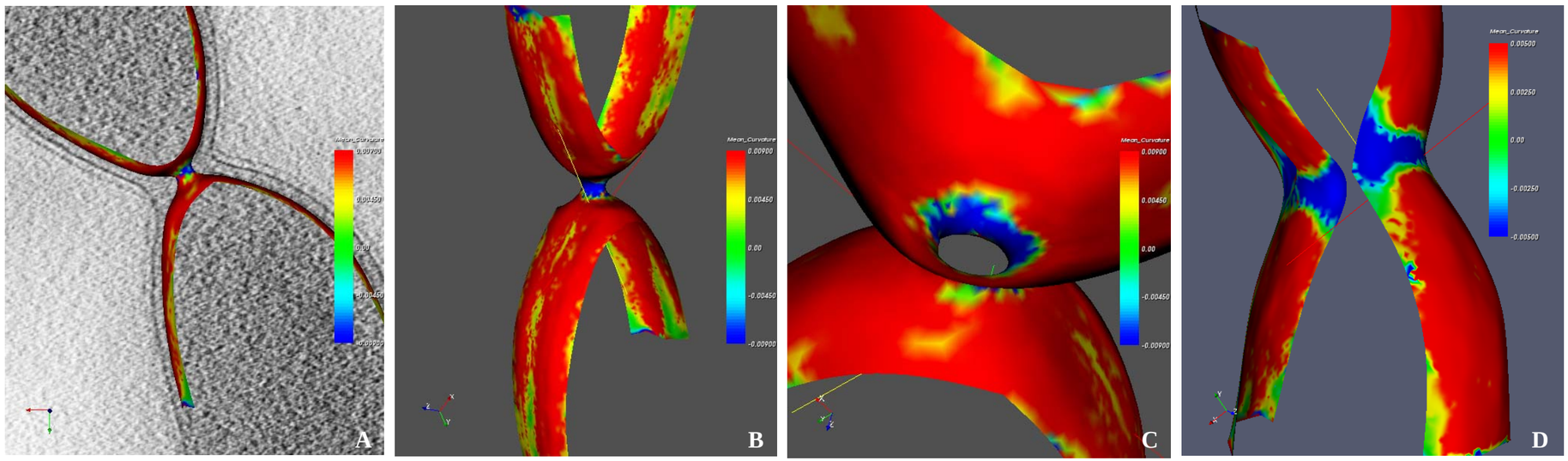
Abstract

The purpose of this study is to understand the distinct shape profile of the dividing Caulobacter crescentus (Cc) cell near the division midplane at various stages of cell division, visible in new high-resolution Cryo EM tomographic images. These images of Cc during cell division provide the needed resolution for understanding cell wall dynamics under the influence of the contractile ring at the division midplane (FtsZ ring). The bacterial cell wall is a peptidoglycan mesh that acts like a fabric that can support shear as well as distinct normal forces, as opposed to the lipid membranes which are usually assumed not to support shear or any differences in normal forces. New tomographic images clearly show the S-layer, inner, outer membranes and peptidoglycan mesh.

The shape of the cell wall during division and the deflection at the division plane is a function of the material properties of the cell wall, the growth rate, and the force due to the FtsZ ring. Our hypothesis is that cell growth and cell contraction occur on similar timescales, and that the contractile force at the division midplane is minimal, serving to direct the growth of the peptidoglycan mesh very near the division plane so as to eventually pinch and isolate the two halves of the dividing cell. In this model the potential energy stored in the peptidoglycan mesh under the strain induced by the FtsZ ring is always minimal and the insertion of new cell wall material near the division midplane at the point of inflection in the dividing cell is essential. Using this model we can predict the force due to the contractile ring given specific elastic parameters and growth rate.

Cryo EM Image Modeling. (A) A slice from a tomographic reconstruction and partial surface rendering of the inner membrane obtained from the reconstruction. (B),(C), (D) Models of Inner and outer membranes for different stages of cell division. Color map: local curvature of the surface.



Caulobacter cell wall

- Cc is a small (700 nm x 3000 nm at cell division) gram negative rod shaped bacterium with a very thin (~3 nm) stretchy peptidoglycan cell wall. The cell wall is anisotropic, with a higher Young's modulus in the hoop direction, the direction of the glycan strands, than in the longitudinal direction, the direction of the peptide bonds.
- Using atomic force microscope measurements [1] of cell wall deflection and by utilizing a simple model for the peptidoglycan mesh as a network of stiff glycan bonds and soft peptide bonds, Boulbitch et al. made an estimate for the longitudinal Young's modulus of 3 x 10^7 [2].
- The cell is pressurized at 2-3 atmospheres. The cell is not perfectly axially symmetric but has a mild crescent shape. The crescent shape of Cc may be due to internal structural trans membrane proteins that effect cell shape [5].
- During cell division the cell begins to constrict at a point near the middle of the cell. This constriction is due to forces exerted by motor proteins whose combined action results in a radially symmetric inward force on the peptidoglycan mesh at the division midplane. At some point near the climax of cell division the inner membranes transitions to a configuration whereby the two halves of the cell compartmentalize. At a later time the peptidoglycan mesh separates, the outer membrane and S layers separate, and the newly divided cells then break away from each other and go on to lead separate lives [3].
- The shape of the balloon-like pressurized Cc cell is due to the mechanical properties of the anisotropic peptidoglycan mesh, combined with the influence of underlying cytoskeletal structures. We will consider the mechanical properties of the regularly arrayed but anisotropic peptidoglycan mesh to have first order effects on the cell shape, while additional shape determining protein structures are second order effects[4-6].

Deflections due to FtsZ ring

How is the deflection of the dividing cell wall and its shape during division related to the force, or pressure, that the contractile ring is exerting on the cell wall, and the elastic parameters of the cell wall?

Under any hypothesis, the shape of the cell wall during division and the deflection at the division plane must be functions of the material properties of the cell wall at a given point in the division process and the force due to the FtsZ ring.

Our hypothesis is that cell growth and cell contraction occur on similar timescales, and that the contractile force at the division midplane is minimal and serves only to direct the growth of the peptidoglycan mesh very near the division plane in ever decreasing concentric circles so as to eventually pinch and isolate the two halves of the dividing cell.

In this model the potential energy stored in the peptidoglycan mesh under the strain induced by the FtsZ ring is always minimal. In this "directed growth" model, the insertion of new cell wall material near the division midplane, at the point of inflection in the dividing cell, is essential. It is only at the division midplane that new, shorter glycan strands and relaxed peptide bonds can be inserted into the mesh at a radius dictated by the deflection induced by the FtsZ ring. Using this model we can make a prediction about the rate at which the cell diameter closes off assuming a constant turn over of cell wall components (constant overall growth rate).

Alternative models relying only on the contraction of the FtsZ ring are found by both analytic and FEM analysis to require unrealistically large contractile pressures (FIGURE 1)

Dynamic Directed Growth

In this model the cell adds rings of glycoprotein at constant rate and at specific growth regions. The average time required to synthesize an entire circumferential ring or band is t_o. The width of this newly synthesized band is w_o. The nominal cell radius is r_o. If the cell is under the influence of a circumferential contractile ring, the nominal deflection angle caused by the pressure of the contractile ring is θ_o. We ask what the deflection $d(t)$ of the cell wall as a function of time will be. We derive:

$$d(t) = \left[\frac{Log\left[\frac{t}{t_o}(\alpha-1)+1\right]-1}{Log[\alpha]} \right] w_o \sin(\theta_o).$$

This formula gives us deflection as a function of time. Using this formula and the similar one for the lengthening of the cell in time allows us to plot the cell deflection and lengthening as a function of time, and to make a parametric plot of the cell shape as a function of time. A linear parameterization of the change in the deflection angle as a function of time, $\alpha = 1 - w_o \sin(at+b)/r_o$, yields a function for the deflection and lengthening of the cell and a realistic looking profile for the dividing cell (FIGURE 2). A more accurate parameterization for the change in the deflection angle as a function of time can be obtained from the curvature parameters of the cell itself, or by doing a new FEM calculation to obtain the new deflection under constant pressure for each growth ring added to the cell wall. In any case, this simple model shows the correct behavior of the cell wall in time and shows that cell wall growth coupled with constant contractile ring pressure allows the cell to change shape and "pinch off" without requiring ever increasing pressures. For a particular set of model parameters a reasonable fit can be achieved with the linear parameterization of the change in the deflection angle as a function of time.

Results

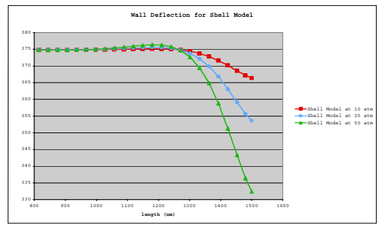


Figure 1. Finite Element Method Shell model for static cell wall under internal pressure and contractile forces due to the FtsZ ring. A Static model requires unrealistically large contractile pressures to pinch off dividing cell.

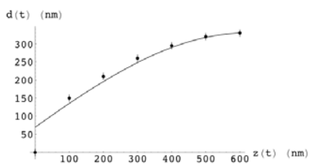


Figure 2. Dynamic Directed Growth Model reproduces cell wall shape with only a small contractile force due to the FtsZ ring. Shape data (points with error bars) is taken from high resolution Cryo EM images.

References

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