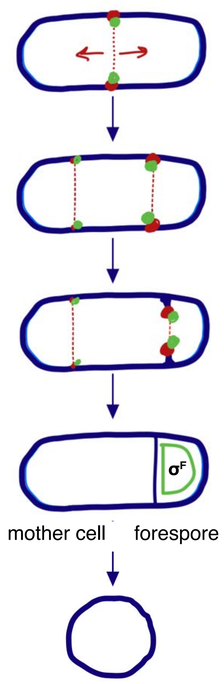
SpoIIE alters the dynamic polymer network of the division machinery to break symmetry

Alexis Ryan[1,2] and Niels Bradshaw[2]

Molecular and Cell Biology Program and Department of Biochemistry, Brandeis University

SpoIIE promotes asymmetric division and activates σ^F in the forespore



I. Medial **FtsZ rings** composed of the tubulin homolog FtsZ scaffolded on the actin homolog FtsA are redirected to the cell poles.

II. Polar **FtsZ rings** form

SpoIIE is inactive as a phosphatase

III. One Z-ring constricts as the new cell wall is built

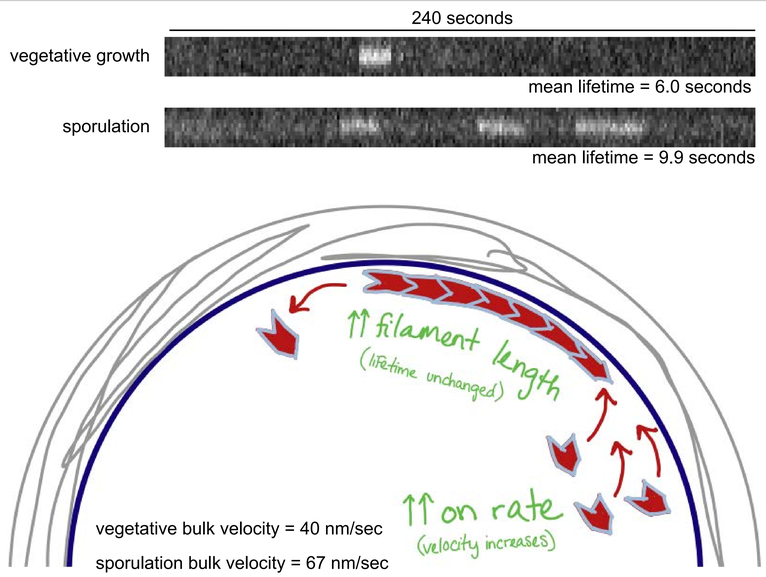
Inactive SpoIIE constricts with Z rings

IV. Z-rings are no longer present.

SpoIIE dephosphorylates its substrate (AA) in the forespore, leading to σ^F activation.

V. Sporulation continues under σ^F-dependent programming to form a stable endospore.

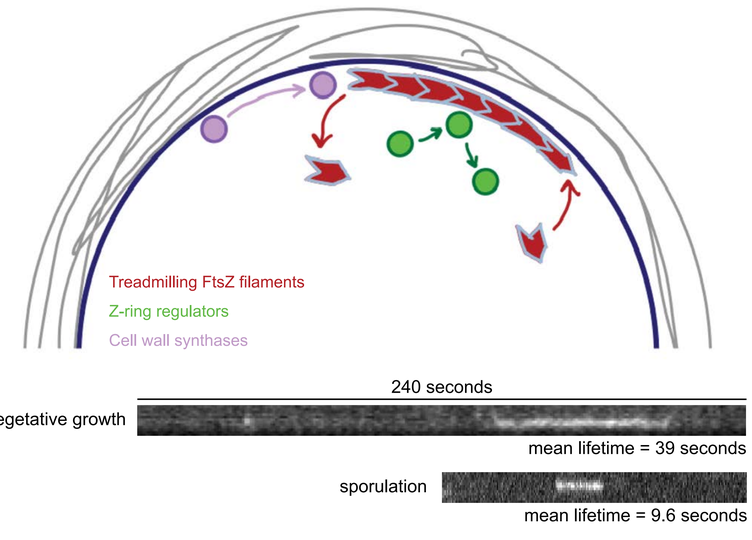
Treadmilling dynamics of the tubulin FtsZ change during sporulation



The mean lifetime of FtsZ molecules is higher during sporulation

Since bulk treadmilling velocity also increases, this indicates an increase in filament length and an increase in on rate during sporulation

SpoIIE single molecule behavior indicates a role as an FtsZ regulator

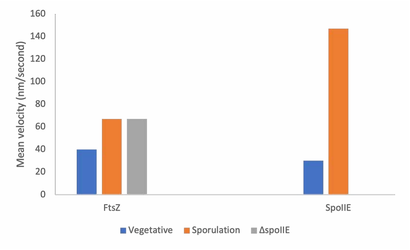


The FtsZ-ring regulators bind to stationary monomers within a treadmilling FtsZ filament. These factors are stationary at a single molecule level.

The cell wall synthesis machinery follows FtsZ filaments, and single molecules move directionally at a similar rate to bulk-labeled FtsZ (between 30-40 nm/sec).

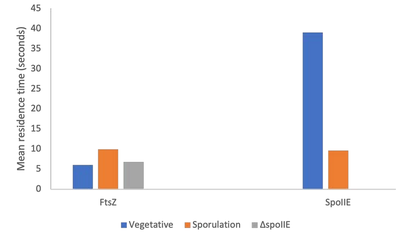
The stationary behavior of SpoIIE both during vegetative growth and sporulation are consistent with a role as an FtsZ structural regulator

Sporulation bulk velocities support increases in filament length and on rate



Based on increased FtsZ velocity during sporulation, the on rate of FtsZ monomers is faster.

SpoIIE does not affect FtsZ velocity. SpoIIE velocity increases 5-fold during sporulation.



Since FtsZ monomer residence time is modestly increased, FtsZ filaments are longer during sporulation.

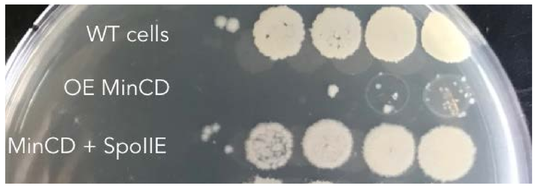
SpoIIE increases FtsZ filament length.

SpoIIE residence times are 5-fold shorter during sporulation, indicating a similar timescale for binding to FtsZ.

The steady state dynamics of FtsZ appear to be different during sporulation. However, SpoIIE dynamics are not proportionally altered. This could be a result of increased FtsZ expression, factors that are present in higher concentrations at polar sites, or sporulation specific factors that further alter dynamics of FtsZ-ring binding proteins.

SpoIIE opposes the midcell placement machinery that debundles FtsZ rings

The Min complex forms a gradient that inhibits FtsZ-ring assembly near the cell poles. Overexpressing MinCD kills cells.



SpoIIE opposes the Min complex and rescues cell division when expressed during vegetative growth. .

Future Directions: Incorporation with MRSEC IRG2

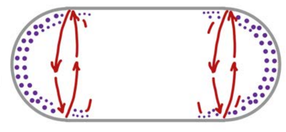
The FtsZ network exhibits unique properties that could translate into active matters with different uses from matters assembled from eukaryotic proteins. Both the actin (FtsA) and tubulin (FtsZ) homologues in this system treadmill, meaning they may be useful in forming new types of active materials. Unlike eukaryotic actin and tubulin, FtsA and FtsZ can polymerize together along the membrane to form curved filaments. Though FtsA/Z filaments exhibit slightly different properties of dynamic behavior, they can be manipulated through similar mechanisms of distortion such as proteins that control bundling (SepF, EzrA, and ZapA) and polymerization (MciZ and MinC). Ultimately, these bacterial polymers could be copolymerized on supportive lipid bilayers to produce active materials with different structural outcomes.

Our goals for understanding how SpoIIE promotes FtsZ-ring asymmetry and changes in force constriction that influence cell division strongly align with IRG2 goals of modeling biological systms to better understand large scale cellular outcomes. We plan to collaborate with the MRSEC to model and explain how these organisms generate emergent spatial properties and consequent force to divide and to define a role for SpoIIE in these sporulation-specific morphological changes.

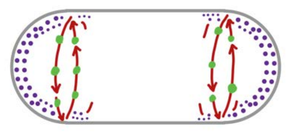
Model: SpoIIE stabilizes FtsZ filaments to overcome polar debundling

During vegetative growth, the Min complex restricts FtsZ treadmilling to the midcell. At the point of lowest Min concentration, FtsZ-rings can form larger bundles.

During sporulation, FtsZ rings must form near the poles where high concentrations of Min prevent bundling.
A thinner FtsZ-ring and increased expression of FtsZ during sporulation could explain how FtsZ velocity increases.



SpoIIE could bind to FtsZ-rings and stabilize filaments (preventing GTP hydrolysis, affecting lateral interactions between filaments). This would explain the increase in FtsZ dwell time, while the polar Min maintains FtsZ velocity.



Funding

Brandeis MRSEC
NIH/NIGMS