

Dynamics of the septum during cell division

Dual Degree Project Stage-II

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Indian Institute of Technology Bombay

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Outline

- 1 Motivation
 - The Basic Problem of Septum Dynamics
- 2 Progress towards the solution
 - Image analysis
 - Application of PIV to reveal septum dynamics
 - Theoretical Models
- 3 Fast Relaxation of the septum post laser ablation
 - Theoretical Approach
 - Image analysis
- 4 Simulation using phase field formalism
- 5 Summary and Future scope



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What is septum?

Septum Dynamics

- Visco elastic material
- Activity due to myosin molecular motors
- Diameter of the septum $\sim 10\mu\text{m}$. Dynamics at micro scale.



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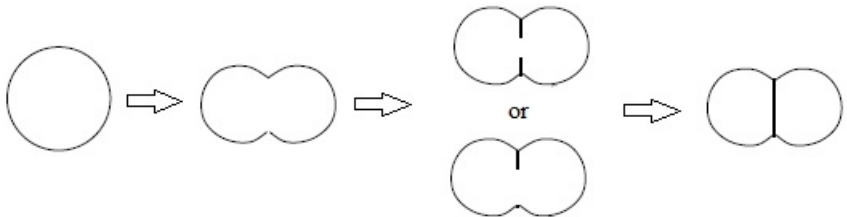
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Symmetric and Asymmetric Closure

Septum Dynamics

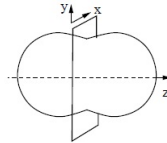


Figure: Septum Plane during cell division



Symmetric and Asymmetric Closure

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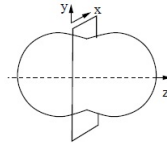


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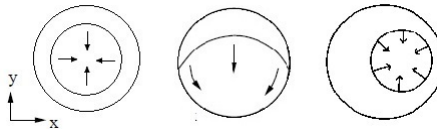


Figure: Cross-section of septum plane: Symmetric and Asymmetric Septum closure



Asymmetric Closure experimentally

Septum Dynamics

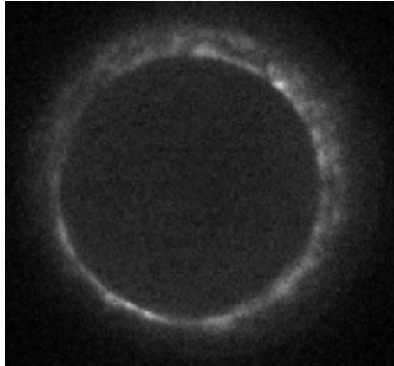


Figure: Experimental movie (obtained from Stephan Grill's lab, CBG Dresden) showing asymmetric septum closure



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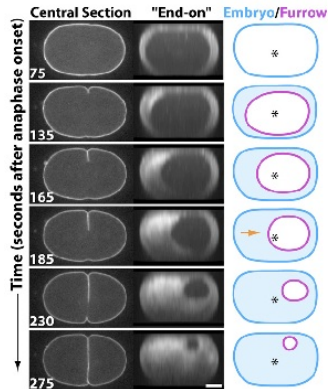


Figure: Time evolution of Septum closure



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Image extraction methods

Image analysis

- To analyze the video, extract a time series of snapshots and analyze them.
- Method 1
 - Using mmreader and avireader in Matlab
 - Difference in number of rows, columns and colour channels
 - Inconsequential. Abandoned.
- Method 2
 - Using snapshot tool in vlc media player
 - Advantages: Septum visibility
 - Disadvantages: Manual, Inaccurate



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Particle Image Velocimetry (PIV) analysis

Image analysis

- PIV Basics

- Open source: GUI and command line versions in Matlab
- Analyze velocity profiles in a dynamic flow
- Role of Interrogation Windows



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Particle Image Velocimetry analysis

PIV basics; Image analysis

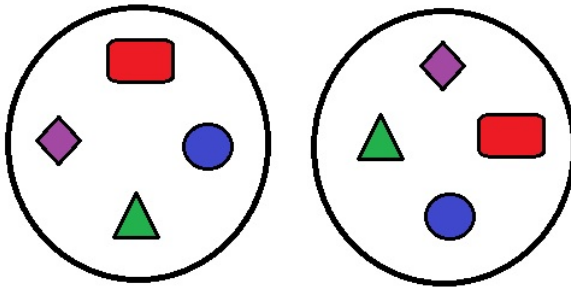


Figure: Two images as input



Particle Image Velocimetry analysis

PIV basics; Image analysis

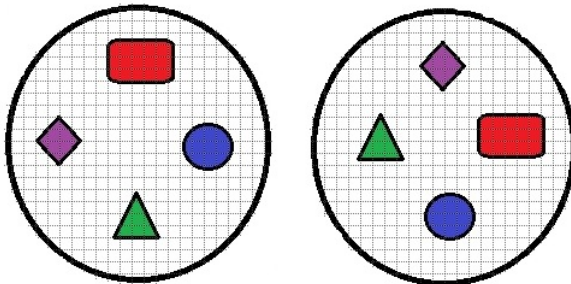


Figure: PIV interrogation regions



Particle Image Velocimetry analysis

PIV basics; Image analysis

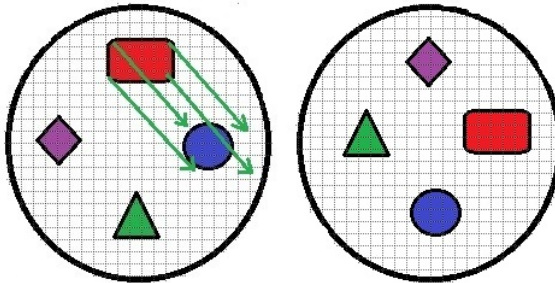


Figure: Mapping using arrows



Particle Image Velocimetry analysis

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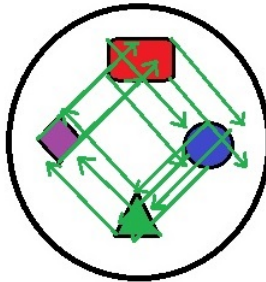


Figure: PIV Output



Parameters on PIV GUI

PIV basics; Image analysis

- Scaling the vectors
- Outlier Filter
- Jump Value
- ROI tool



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Asymmetric closure experimentally

Septum dynamics

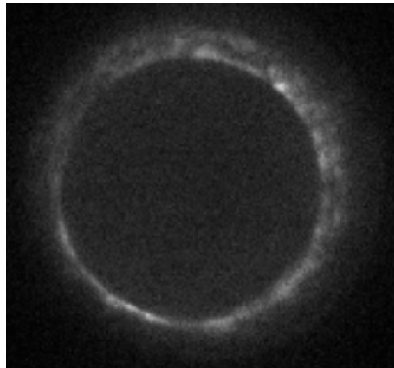
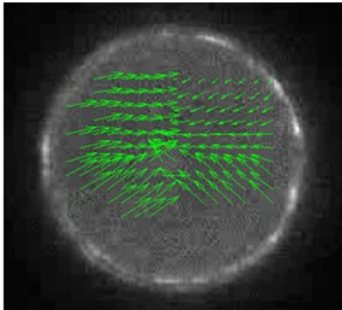


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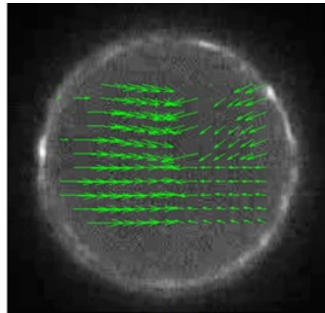


Applications of PIV

PIV on snapshots of asymmetric septum closure



(a) 10-20

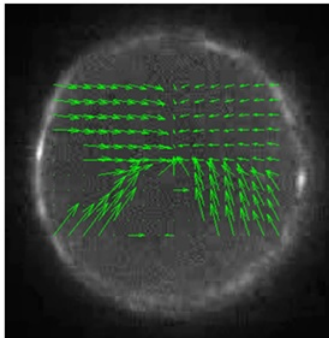


(b) 20-30

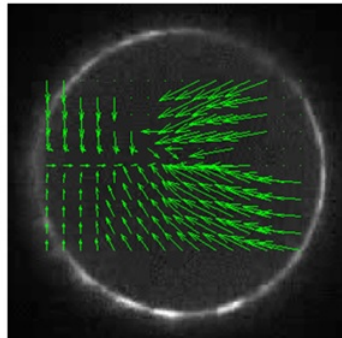


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(c) 30-40

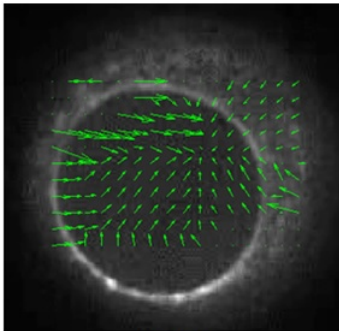


(d) 40-50

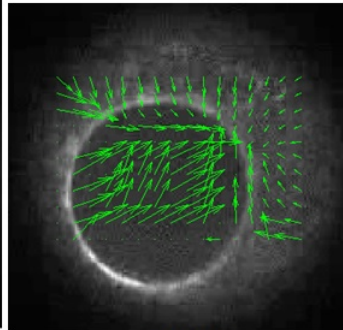


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(g) 70-80

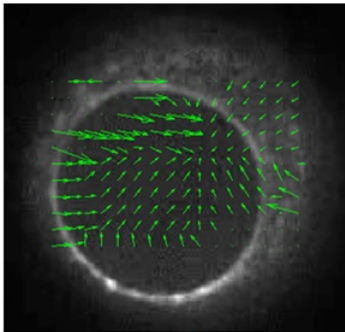


(h) 80-85

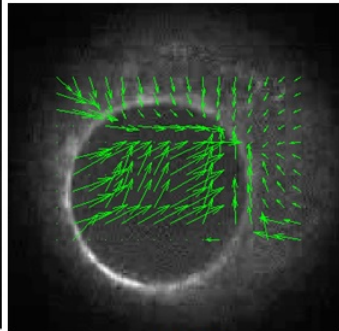


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Inference from PIV maps

PIV on snapshots of asymmetric septum closure

- Rotation of the acto-myosin ring is evident as its radius decreases.
- v_ϕ must depend on r .



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Defining the system

Septum dynamics

- Navier-Stokes Equation:



Defining the system

Septum dynamics

- Navier-Stokes Equation:

$$\rho \left[\frac{\partial \mathbf{v}}{\partial t} + (\mathbf{v} \cdot \nabla) \mathbf{v} \right] = -\nabla p + \eta \Delta \mathbf{v} + \left(\zeta + \frac{1}{3} \eta \right) \nabla (\nabla \cdot \mathbf{v})$$



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$\nabla \cdot \mathbf{v} = 0$ which becomes

$$\frac{1}{r} \frac{\partial (rv_r)}{\partial r} + \frac{1}{r} \frac{\partial (v_\phi)}{\partial \phi} + \frac{\partial v_z}{\partial z} = 0$$



Defining the system

Basic equations; Septum dynamics

In cylindrical co-ordinates:



Defining the system

Basic equations; Septum dynamics

In cylindrical co-ordinates:

- $$\frac{\partial v_r}{\partial t} + (v \cdot \nabla) v_r - \frac{v_\phi^2}{r} = -\frac{1}{\rho} \frac{\partial p}{\partial r} + \nu \left(\nabla^2 v_r - \frac{2}{r^2} \frac{\partial v_\phi}{\partial \phi} - \frac{v_r}{r^2} \right)$$



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Basic equations; Septum dynamics

Stress-velocity relations in cylindrical co-ordinates:

- $\sigma_{rr} = -p + 2\eta \frac{\partial v_r}{\partial r}$
- $\sigma_{r\phi} = \eta \left(\frac{1}{r} \frac{\partial v_r}{\partial \phi} + \frac{\partial v_\phi}{\partial r} - \frac{v_\phi}{r} \right)$
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Boundary conditions

Septum dynamics

At outer periphery the velocity boundary conditions are:

- $v_r(R_1) = -v_0$
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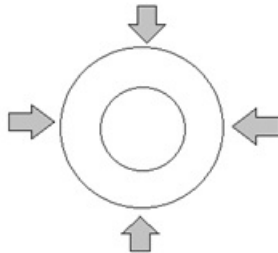


Figure: ϕ symmetric vesicle addition through the outer rim

Boundary conditions

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- At inner periphery the stress boundary condition is:

$$\sigma_{nn}(R_2) = \frac{\Sigma}{R_2}$$

- Radius of curvature changes at every point on the inner periphery in the asymmetric case



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Different approaches that didn't work

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- Case 1: $v_r = v_r(r)$, $v_\phi = 0$
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Streamfunction formalism

Different approaches that didn't work; Septum dynamics

- Streamfunction ϕ^S obeys $\mathbf{v} = \nabla\phi^S \times \hat{\mathbf{z}}$

- In terms of ϕ^S

$$\mathbf{v} = v_r \hat{\mathbf{r}} + v_\phi \hat{\phi} = \frac{1}{r} \frac{\partial \phi^S}{\partial \phi} \hat{\mathbf{r}} - \frac{\partial \phi^S}{\partial r} \hat{\phi}$$

- Advantages:

- Equation of continuity automatically satisfied
- Everything gets reduced in terms of scalar function ϕ^S

- Doesn't work due to the conflict b/w

- Periodicity constraint in $\hat{\phi}$ and
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- Advantages:
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Different approaches that didn't work; Septum dynamics

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Working theoretical model

Lubrication approximation; Septum dynamics

- Proceed with Case 3 above i.e. assume
 $v_r = v_r(r), v_\phi = v_\phi(r)$
- **Lubrication approximation** says that the pressure is zero.
- Forget incompressibility in 2D; z direction vesicle addition allowed



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Lubrication approximation; Septum dynamics

Under these conditions stress velocity relations reduce as

- $\sigma_{rr} = 2\eta \frac{\partial v_r}{\partial r}$
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- $\frac{\partial \sigma_{rr}}{\partial r} + \frac{1}{r} \frac{\partial \sigma_{r\phi}}{\partial \phi} + \frac{1}{r} (\sigma_{rr} - \sigma_{\phi\phi}) = 0$
- $\frac{\partial \sigma_{r\phi}}{\partial r} + \frac{1}{r} \frac{\partial \sigma_{\phi\phi}}{\partial \phi} + \frac{2}{r} (\sigma_{r\phi}) = 0$



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Working theoretical model

Lubrication approximation; Septum dynamics

- Substituting, we get

- $r^2 \frac{\partial^2 v_r}{\partial r^2} + r \frac{\partial v_r}{\partial r} - v_r$
- $r^2 \frac{\partial^2 v_\phi}{\partial r^2} + r \frac{\partial v_\phi}{\partial r} - v_\phi$

- Putting $v_r = r^m$ and $v_\phi = r^n$ we get $m, n = \pm 1$. Thus v_r and v_ϕ take the same form

- $v_r = Ar + \frac{B}{r}$
- $v_\phi = Ar + \frac{B}{r}$



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Working theoretical model

Lubrication approximation; Septum dynamics

- Applying the boundary conditions at outer periphery

- $v_r(R_1) = -v_0,$

- $v_\phi(R_1) = 0,$

- $v_r(r) = Ar - \frac{AR_1^2}{r} - \frac{v_0 R_1}{r}$

- $v_\phi(r) = Cr - \frac{CR_1^2}{r}$

- Applying the boundary condition at inner periphery

$\sigma_{rr}(R_2) = \frac{\Sigma}{R_2}$ in symmetric case, we get

$$A = \left[\frac{1}{R_1^2 + R_2^2} \right] \left[\frac{\Sigma R_2}{2\eta} - v_0 R_1 \right]$$



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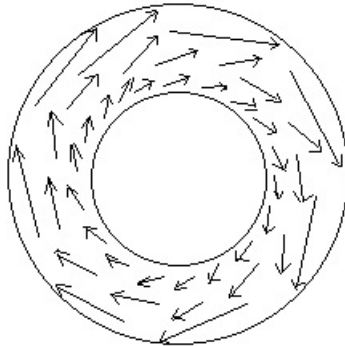


Figure: Schematic representation of the flows under lubrication approximation model showing radial dependence of v_r and v_ϕ



Outline

- 1 Motivation
 - The Basic Problem of Septum Dynamics
- 2 Progress towards the solution
 - Image analysis
 - Application of PIV to reveal septum dynamics
 - Theoretical Models
- 3 Fast Relaxation of the septum post laser ablation
 - Theoretical Approach
 - Image analysis
- 4 Simulation using phase field formalism
- 5 Summary and Future scope



Revisiting the system

Septum Dynamics

- 2D viscous tensor components inside septum:

$$\sigma_{rr} = \sigma_0 + 2\eta \frac{\partial v_r}{\partial r}, \sigma_{\phi\phi} = \sigma_0 + 2\eta \frac{v_r}{r}$$

- Force balance Equation:

$$\partial_r \sigma_{rr} + \frac{1}{r} (\sigma_{rr} - \sigma_{\phi\phi}) = 0$$

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$$\sigma_{rr} = \sigma_0 + \frac{\eta}{A} \frac{dA}{dt} \left(1 + \frac{r_0^2}{r^2} \right)$$



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Laser ablation on cortex

Ablation response

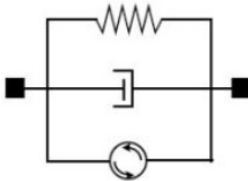
- Cortex assumed as an active visco-elastic solid modelled by Kelvin Voigt Material



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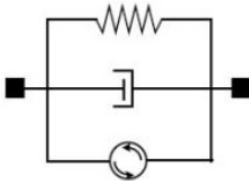
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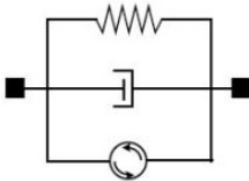
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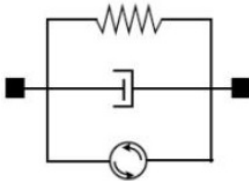
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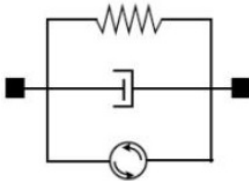
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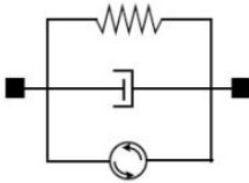
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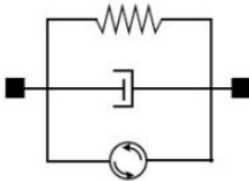
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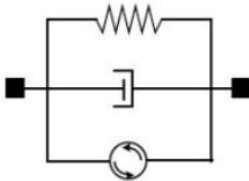
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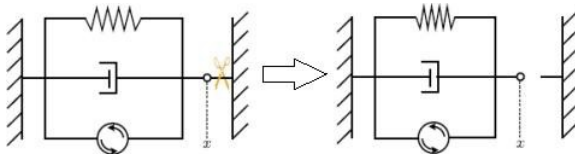
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Cortical dynamics post ablation

Ablation response

- Dynamics of the rapid displacements

$$T - kx - \zeta \dot{x} - C = 0$$

- The initial condition

$$T(t) = T_0(1 - U(t)), \text{ where } U(t) \text{ is the unit step function}$$

- Solving this equation,

$$v = \dot{x} = -\frac{T_0}{\eta} e^{-\frac{t}{\tau}}; t > 0$$

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Stress balance equation

Theoretical approach

- Radial septum stress $\sigma_r = \sigma_0 + \frac{\eta}{A} \frac{dA}{dt} \left(1 + \frac{A_0}{A_0 - A}\right)$
- Two components of transverse septum stress at ring boundary:
 - Active component: $\sigma_t^a = \Sigma_0 + \zeta \frac{u(r)}{r}$
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- In 2D, $\sigma_r = \frac{\sigma_t}{r}$ and thus we obtain
$$\sigma_0 + \frac{\eta}{A} \frac{dA}{dt} \left(1 + \frac{A_0}{A_0 - A}\right) = \frac{1}{r} \left(\Sigma_0 + \zeta \frac{u(r)}{r} + k \frac{u(r)}{r} \right)$$



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Stress balance equation

Theoretical approach

- Radial septum stress $\sigma_r = \sigma_0 + \frac{\eta}{A} \frac{dA}{dt} \left(1 + \frac{A_0}{A_0 - A}\right)$
- Two components of transverse septum stress at ring boundary:
 - Active component: $\sigma_t^a = \Sigma_0 + \zeta \frac{u(r)}{r}$
 - Passive component: $\sigma_t^p = k \frac{u(r)}{r}$
- In 2D, $\sigma_r = \frac{\sigma_t}{r}$ and thus we obtain

$$\sigma_0 + \frac{\eta}{A} \frac{dA}{dt} \left(1 + \frac{A_0}{A_0 - A}\right) = \frac{1}{r} \left(\Sigma_0 + \zeta \frac{u(r)}{r} + k \frac{u(r)}{r} \right)$$



A specific solution

Theoretical approach

- Neglecting the elastic component of the ring,

$$\frac{dA}{dt} = -\frac{\sigma_0 A}{\eta} \frac{A_0 - A}{2A_0 - A}$$

- Solving this we get,

$$A(t) = \frac{1}{2} \left(e^{-\frac{\sigma_0 t}{\eta}} \right) \left[e^c \pm e^{\frac{c}{2}} \sqrt{e^c - 4A_0 e^{\frac{\sigma_0 t}{\eta}}} \right]$$



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Outline

- 1 Motivation
 - The Basic Problem of Septum Dynamics
- 2 Progress towards the solution
 - Image analysis
 - Application of PIV to reveal septum dynamics
 - Theoretical Models
- 3 Fast Relaxation of the septum post laser ablation
 - Theoretical Approach
 - Image analysis
- 4 Simulation using phase field formalism
- 5 Summary and Future scope



Images extracted using method 2

Image extraction; Image analysis

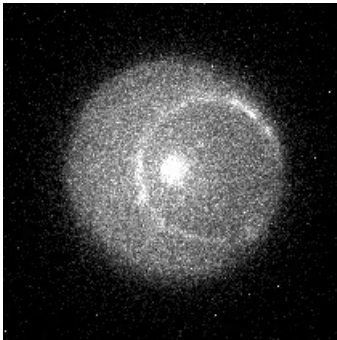


Figure: Movie showing fast relaxation of septum post laser ablation



Images extracted using method 2

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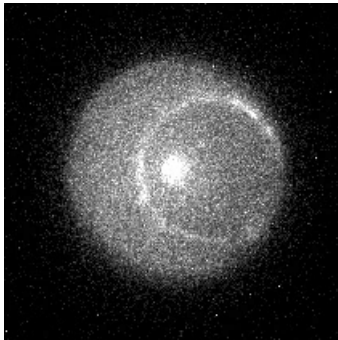
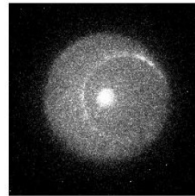
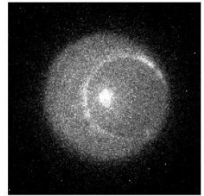


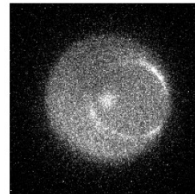
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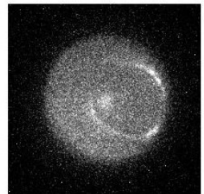
(a)



(b)



(c)



(d)



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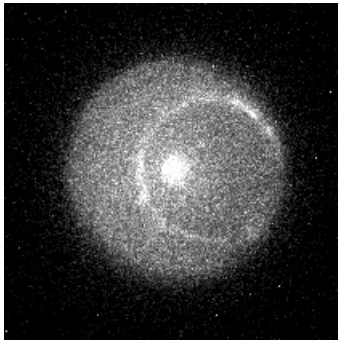
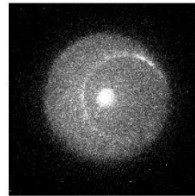
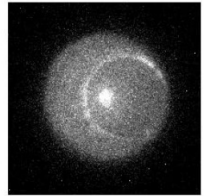


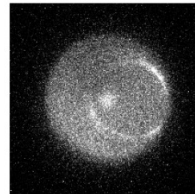
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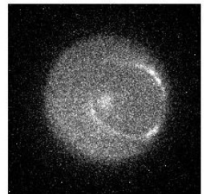
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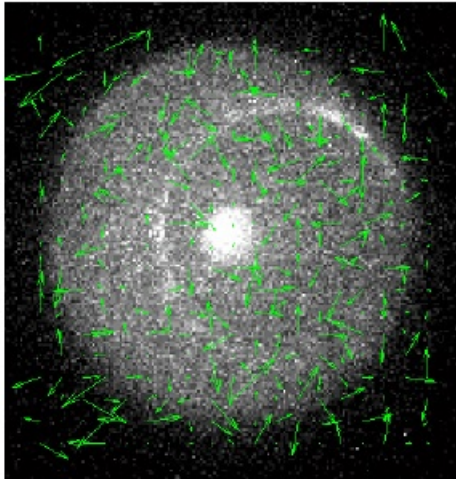


(d)



PIV on extracted images; Inconclusive

PIV application; Image analysis



Phase Field formalism

Septum Dynamics

- The dynamic equation for the phase field is

$$\frac{\partial \phi}{\partial t} = -\mathbf{u} \cdot \nabla \phi + \Gamma(\epsilon \nabla^2 \phi - \frac{G'}{\epsilon})$$

- $G(\phi) = \phi^2(1 - \phi)^2$

- The second module describes the actin network as

$$\nu_0 \nabla \cdot [\phi(\nabla \mathbf{u} + \nabla \mathbf{u}^T)] + \nabla \cdot \sigma_{myo} = 0$$

- ϕ takes care of the boundary conditions.



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Summary and Future scope

- Velocity maps obtained from PIV gave way towards theoretical model under lubrication approximation
- Better video quality; Improved velocity maps; Can help resolving the rotation of the ring.
- Area of the septum decreases with time under fast relaxation.
- Simulation results can provide further insight in the underlying dynamics.



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