Modeling Biofilms Using Complex Fluid Models

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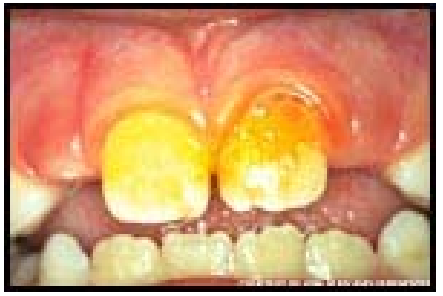
Outline

- Introduction
- Phase field models for biofilms
- Linear stability analysis of constant steady states
- Numerical methods and results for 1-D systems
- Numerical methods and results for 2-D systems
- A kinetic model (optional).
- Conclusion

Introduction to Biofilms

- Biofilms are ubiquitous in nature and manmade materials. Biofilm forms when bacteria adhere to surfaces in moist environments by excreting a slimy, glue-like substance. Sites for biofilm formation include all kinds of surfaces: natural materials above and below ground, metals, plastics, medical implant materials—even plant and body tissue. Wherever you find a combination of moisture, nutrients and a surface, you are likely to find biofilm.

In a pipe



Plaque on teeth

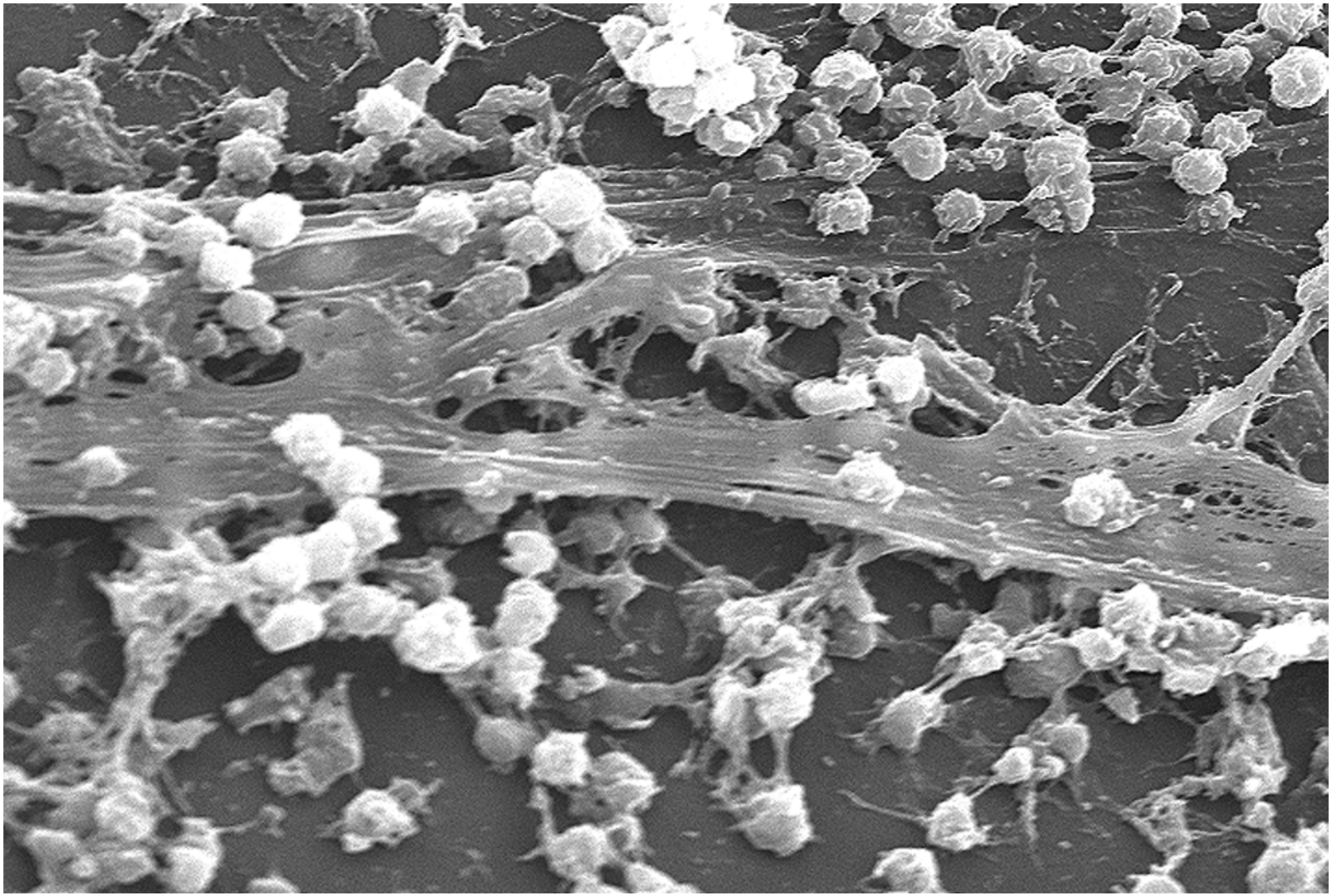
In a creek.

In a membrane

Where do biofilms grow?

- Biofilms grow virtually everywhere, in almost any environment where there is **a combination of moisture, nutrients, and a surface.**
- This streambed in Yellowstone National Park is coated with biofilm that is several inches thick in places. The warm, nutrient-rich water provides an ideal home for this biofilm, which is heavily populated by green algae. The microbes colonizing thermal pools and springs in the Park give them their distinctive and unusual colors.

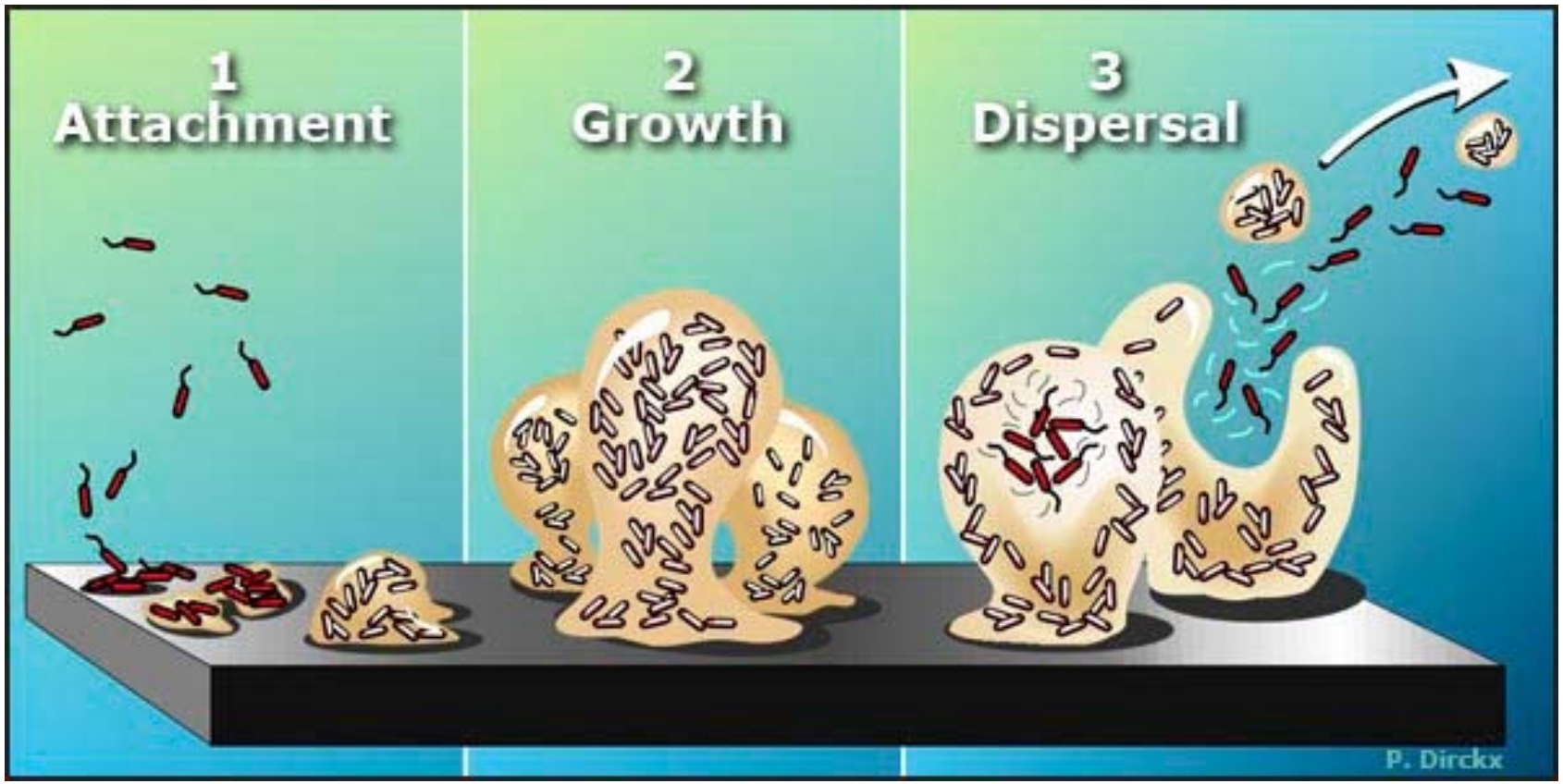
Staph Infection (Staphylococcus aureus biofilm) of the surface of a catheter (CDC)



Bacteria mats near Grand Prismatic Spring in Yellowstone
(Daniel Mayer)

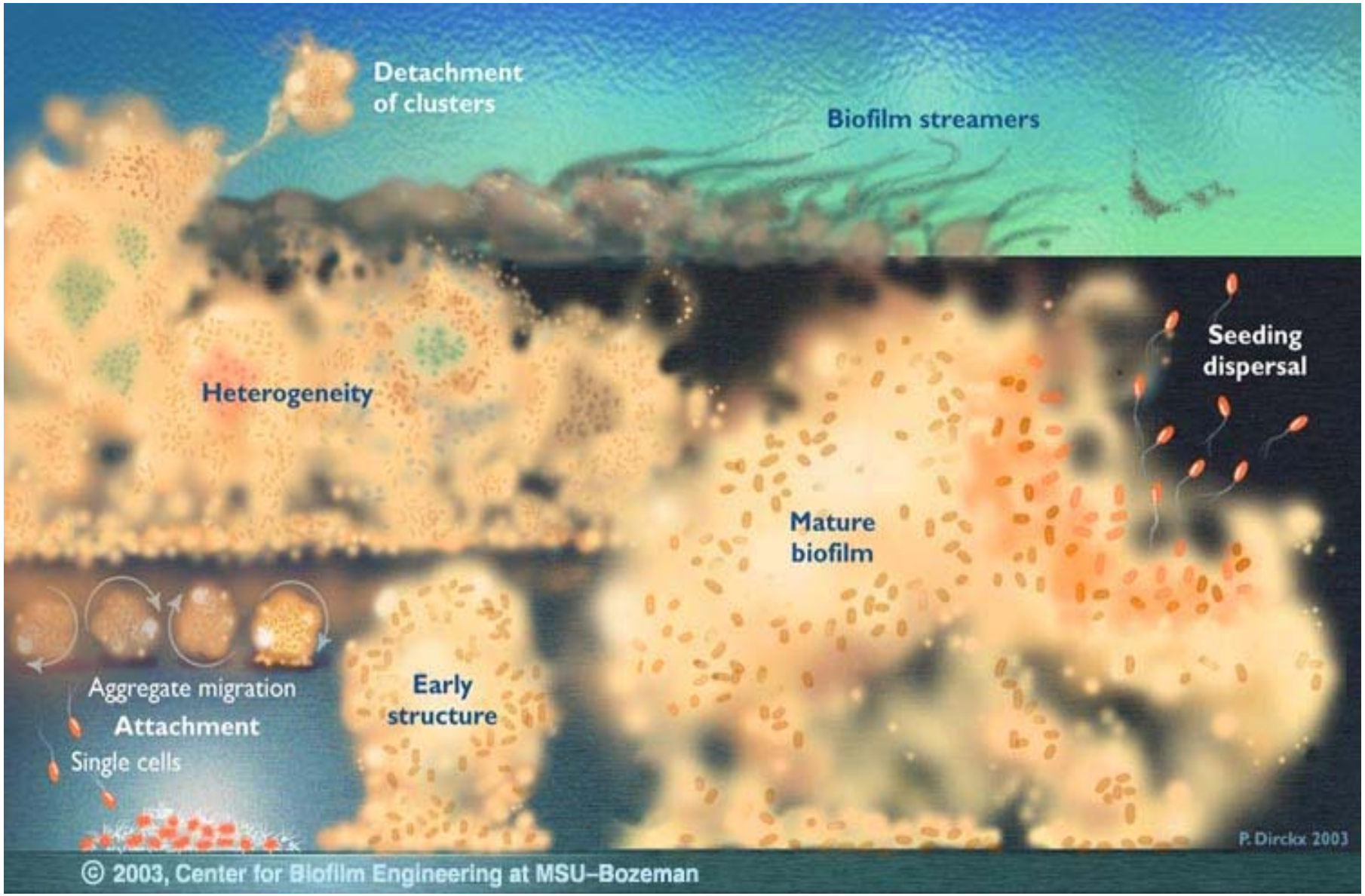
Properties of biofilms

- A biofilm community can be formed by a single bacterial species, but in nature biofilms almost always consist of rich mixtures of many species of bacteria, as well as fungi, algae, yeasts, protozoa, other microorganisms, debris and corrosion products.
- Biofilms are held together by sugary molecular strands, collectively termed "**extracellular polymeric substances" or "EPS**." The cells produce EPS and are held together by these strands, allowing them to develop complex, three-dimensional, resilient, attached communities.
- Biofilms cost the U.S. literally billions of dollars every year in energy losses, equipment damage, product contamination and medical infections.
- But biofilms also offer huge potential for bioremediating hazardous waste sites, biofiltering municipal and industrial water and wastewater, and forming biobarriers to protect soil and groundwater from contamination



1) Free-floating, or *planktonic*, bacteria encounter a submerged surface and within minutes can become attached. They begin to produce slimy extracellular polymeric substances (EPS) and to colonize the surface. 2) EPS production allows the emerging biofilm community to develop a complex, three-dimensional structure that is influenced by a variety of environmental factors. Biofilm communities can develop within hours. 3) Biofilms can propagate through detachment of small or large clumps of cells, or by a type of "seeding dispersal" that releases individual cells. Either type of detachment allows bacteria to attach to a surface or to a biofilm downstream of the original community.

Biofilm growth process



Problems caused by biofilms

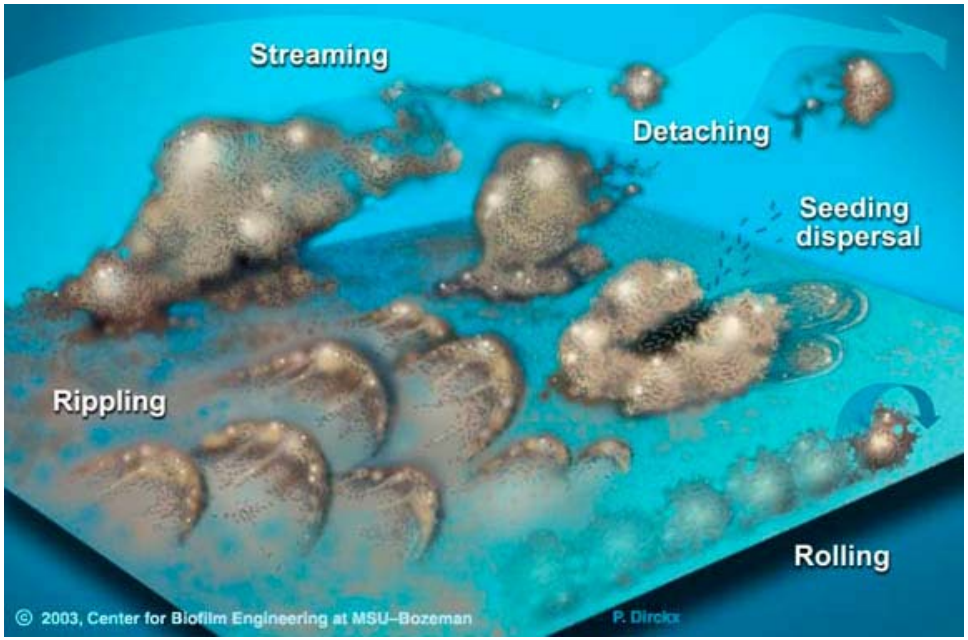
- **In natural environments.** "Microbes can negatively impact environments on a global level including producing and consuming atmospheric gases that affect climate; mobilizing toxic elements such as mercury, arsenic and selenium; and producing toxic algal blooms and creating oxygen depletion zones in lakes, rivers and coastal environments (eutrophication). Furthermore, the incidence of microbial diseases such as plague, cholera, Lyme disease, and West Nile Virus are linked to global change.“
- **In industrial environments.** Biofouling, biocorrosion, equipment damage and product contamination are constant and expensive problems in industry. Biofilm contamination and fouling occur in nearly every industrial water-based process, including water treatment and distribution, pulp and paper manufacturing and the operation of cooling towers.
- **In human health**. The human body is heavily colonized by microbes that have found it a great place to live. chronic, low-grade infections are related to the biofilm mode of growth.

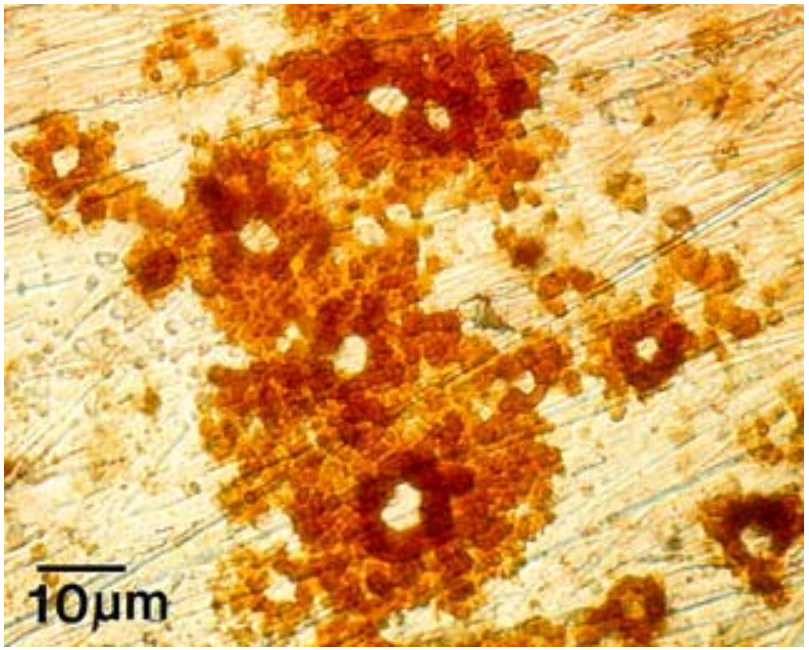
Beneficial effects

- While some bacteria produce effects that are detrimental to surrounding organisms or hosts, most bacteria are harmless or even beneficial.
- Engineers have taken advantage of natural biofilm environmental activity in developing water-cleaning systems.
- When toxic organic contaminants (i.e. gasoline, fuel oil, chlorinated solvents) are accidentally released underground, the native soil bacterial population will, to the degree possible, adjust their ecological composition in order to use the organic contaminants as a food source. This process is commonly referred to as “bioremediation” and if successful, potentially has the ability to render initially toxic organic material into harmless by-products.
- Heap leaching is the most common process used to microbially extract copper and other minerals from spent ore. The process consists of organizing the spent ore fragments into a packed bed configuration which allows water to be trickled through. To initiate the process, acidified water (pH = 1.5 to 3.0) is sprayed over the porous ore bed. Acidophilic bacteria, such as *Thiobacillus ferrooxidans,* actively oxidize the soluble ferrous iron and attack the sulfide minerals, releasing the soluble cupric ion that can then be recovered from aqueous solution. This oxidation process is similar in concept to corrosion of metal surfaces.
- **Other biofilm technologies with promise:** Microbial fuel cells, Biofilm "traps“, Microbial "canaries"

Characteristics of Biofilms

- **Biofilms are complex, dynamic structures**

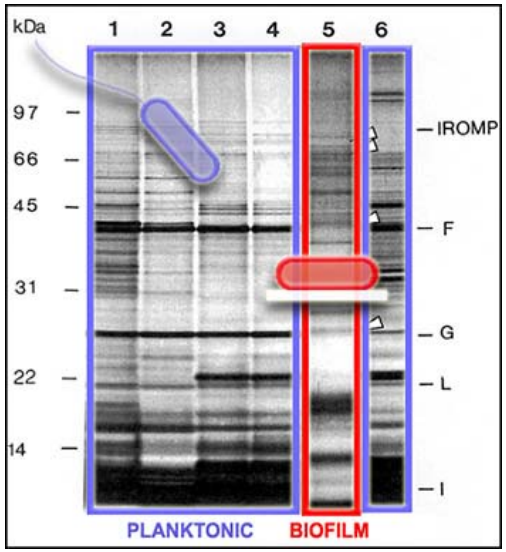


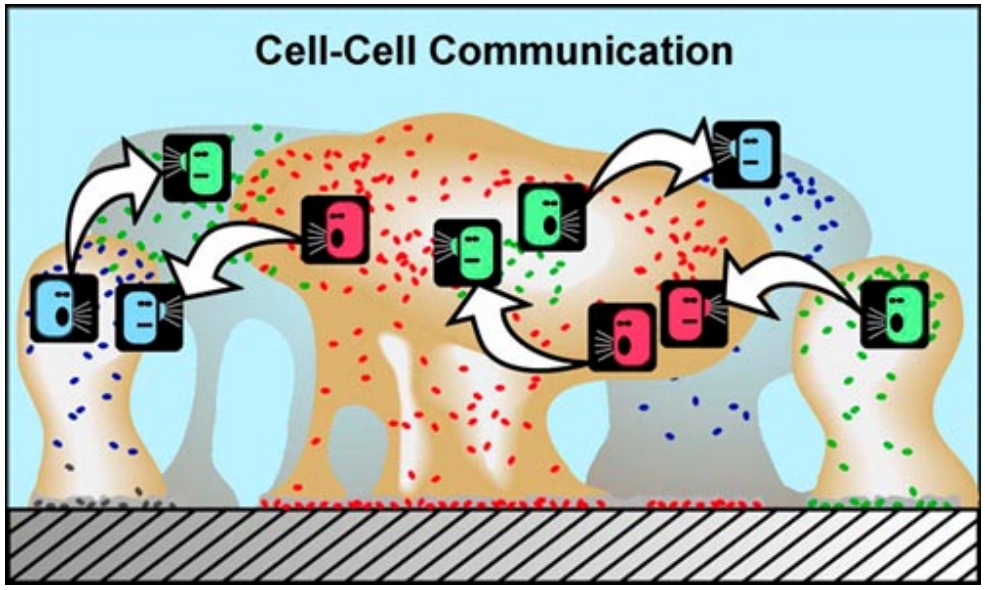


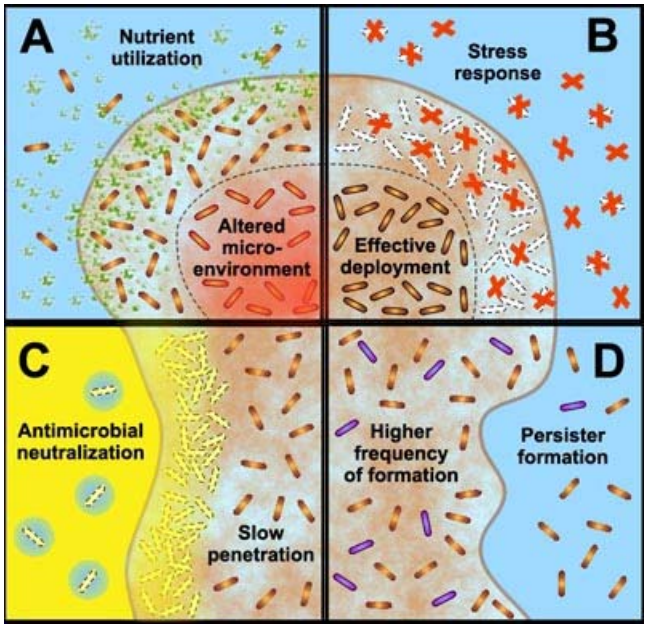
Pitting corrosion on 316S stainless steel, an example of microbially influenced corrosion. *Image, courtesy of Z. Lewandowski and W. Dickinson, MSU-CBE*

Characteristics of Biofilms

- **Genetic expression is different in biofilm bacteria when compared to planktonic (free floating) bacteria.**
- **Biofilm cells can coordinate behavior via intercellular "communication" using biochemical signaling molecules.**
- **Biofilms are less susceptible to antimicrobial agents .**







Modeling Challenges

- Mathematical models for the growth of the biofilm colony that accounts for the properties of the EPS, nutrient distribution/transport, bacterial dynamics, solvent interactions.
- Intercellular communication, signaling pathways.
- Drug interaction with the biofilm components, especially, the bacterial microbes.
- Studies of complex live biofilms provide an interdisciplinary challenge for researchers across the disciplinary boundaries.
- Past approaches: discrete models and/or mainly multi-fluid continuum models. Disadvantages: how to imposed initial and boundary conditions for each velocity in multi-fluid models?
- Our approach: one fluid, multi-component modeling, systematically to include more components. Advantages: an averaged velocity is used and the material is treated as incompressible.

Phase field models for biofilms

We model the biofilm material as incompressible and denote the following variables:

- $\mathbf{v}$ is the average velocity,
- ϕ_n is the polymer network volume fraction and ϕ_s the volume fraction of the effective solvent, $\phi_n + \phi_s = 1$; ϕ_n is naturally viewed as a phase variable,
- p is the pressure and τ the extra stress tensor,
- c is the nutrient substrate concentration,
- ρ is the density of the mixture.

Constitutive equations

$\tau_n = 2\eta_n \mathbf{D}, \tau_s = 2\eta_s \mathbf{D},$ VA-model

$\tau_n = 2\eta_n \mathbf{D}_n, \tau_s = 2\eta_s \mathbf{D}_s,$ VN-model

$\frac{d\tau_n}{dt} - \mathbf{W} \cdot \tau_n + \tau_n \cdot \mathbf{W} - a[\mathbf{D} \cdot \tau_n + \tau_n \cdot \mathbf{D}] + \frac{\tau_n}{\lambda_1} = \frac{2\eta_p}{\lambda_1}\mathbf{D},$

$\tau_s = 2\eta_s \mathbf{D},$ JSA-model

$\frac{d\tau_n}{dt} - \mathbf{W}_n \cdot \tau_n + \tau_n \cdot \mathbf{W}_n - a[\mathbf{D}_n \cdot \tau_n + \tau_n \cdot \mathbf{D}_n] + \frac{\tau_n}{\lambda_1} = \frac{2\eta_p}{\lambda_1}\mathbf{D}_n,$

$\tau_s = 2\eta_s \mathbf{D}_s,$ JSN-model

where the infinite relaxation time limit $\lambda_1 \to \infty$ yields the rubber elastic theory.

Boundary conditions

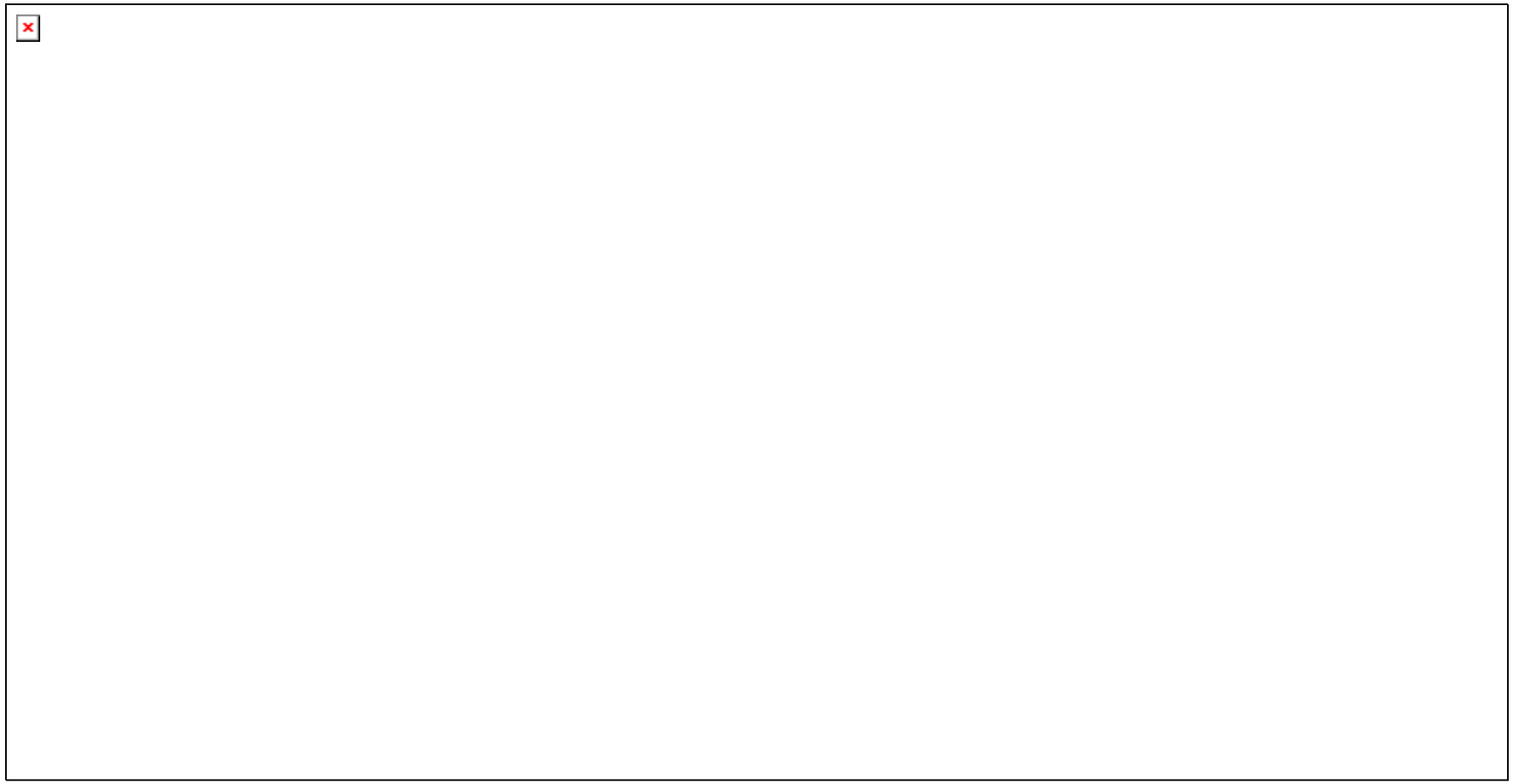
Let I be the domain occupied by the biofilm. The boundary conditions for the governing system of equations are

$$\mathbf{v}|_{\partial I} = 0, [\phi_n \mathbf{n} \cdot \nabla \frac{\delta f}{\delta \phi_n}]_{\partial I} = 0, [\mathbf{n} \cdot \nabla \phi_n]_{\partial I} = 0, [\phi_s \mathbf{n} \cdot \nabla c]_{\partial I} = 0,$$

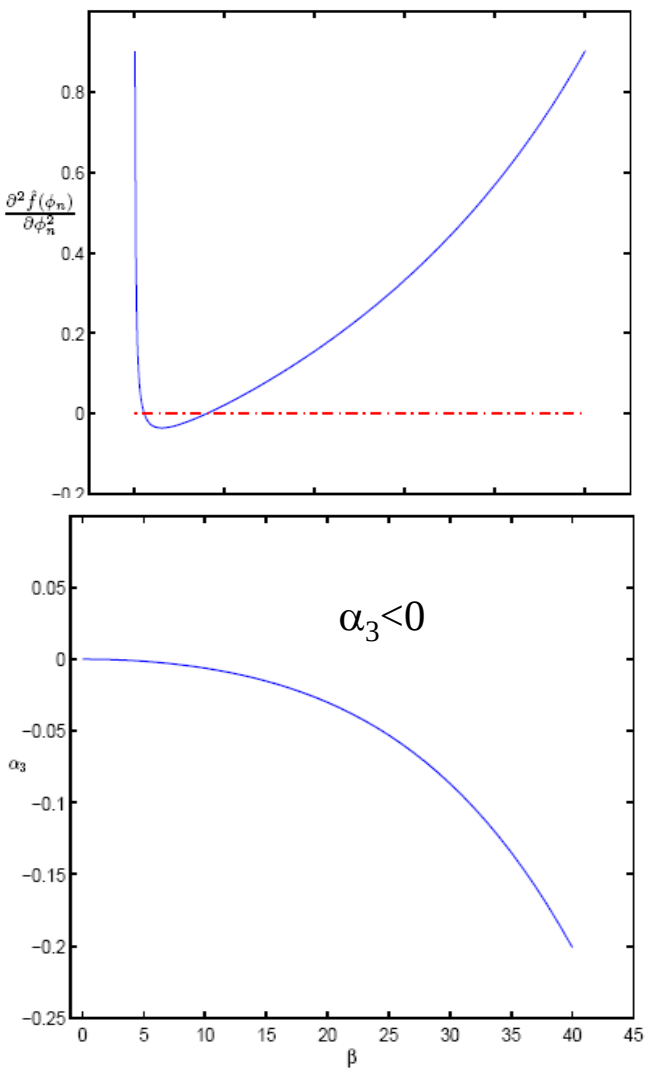
where $\mathbf{n}$ is the unit external normal at the boundary of the domain I. In the simulation presented below, a Dirichlet boundary condition for the nutrient substrate concentration is prescribed at one boundary to mimic an open boundary of the mixture to a large reservior, i.e.,

$$c|_{\partial I +} = c^*.$$

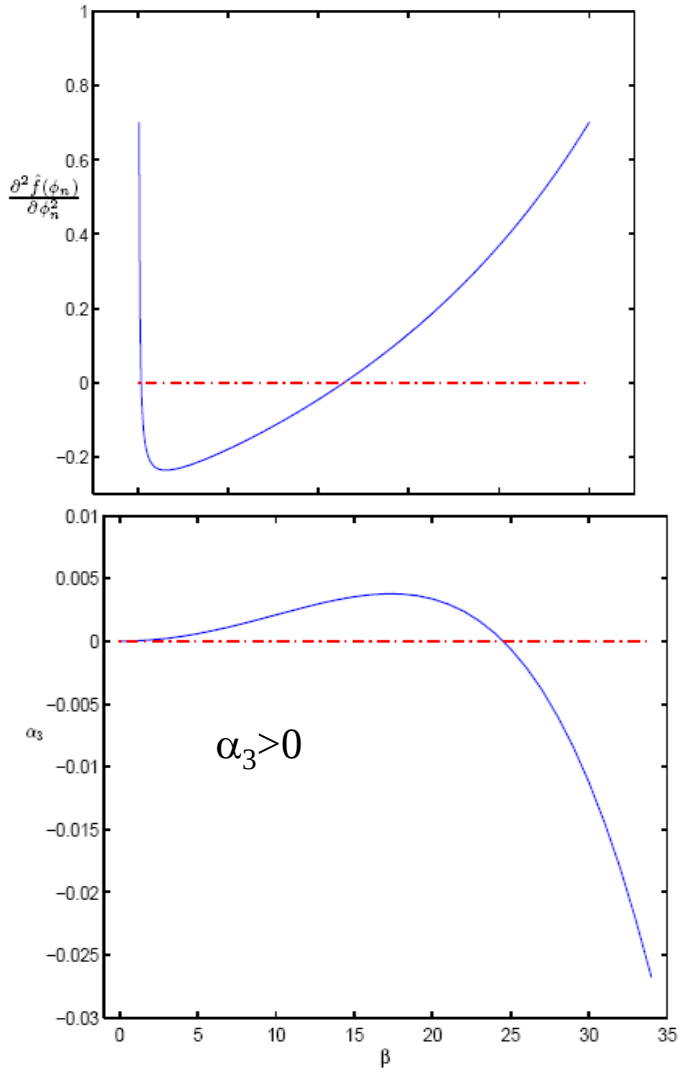
Stability of constant steady states



Info for the mixing energy and growth rates

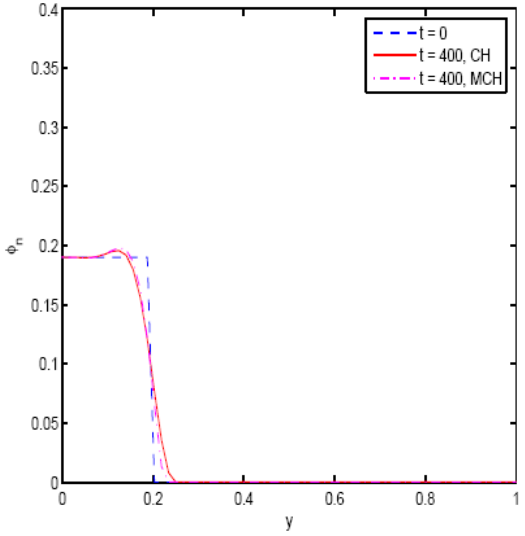


(a) Steady state 1, $\phi_n = \phi_0 = 0.19, c = 0$, and $\chi = 0.55$

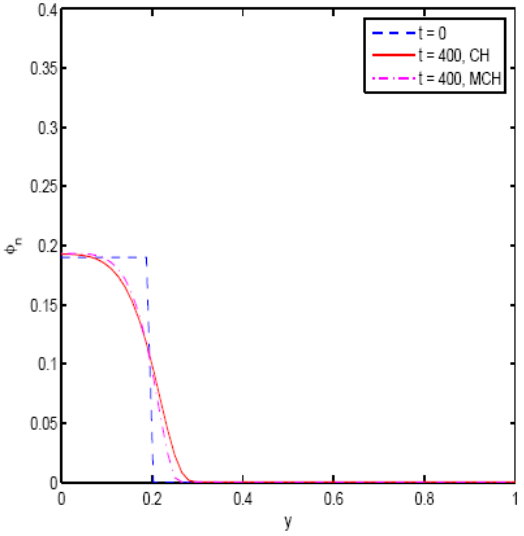


(b) Steady state 1, $\phi_n = \phi_0 = 0.19, c = 0$, and $\chi = 0.65$

1-D simulations: growth of a step distribution without eps production



(a) $\lambda = 10^{-5}$



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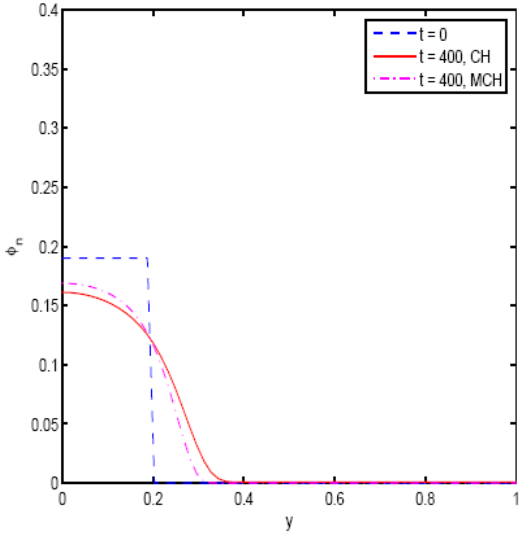

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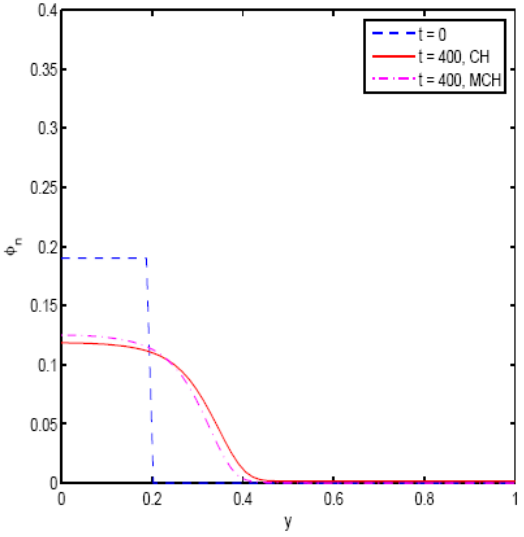
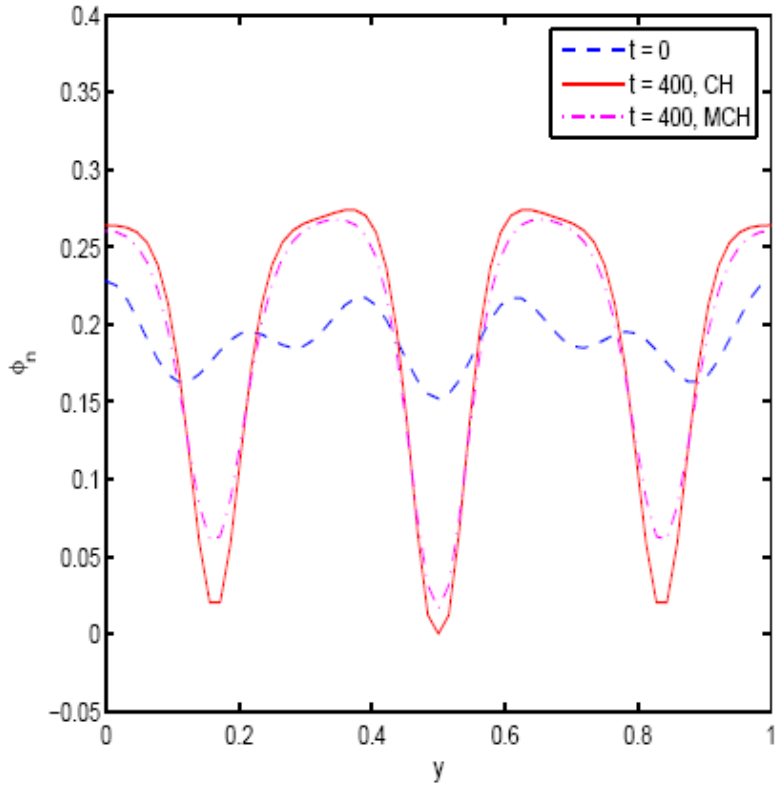
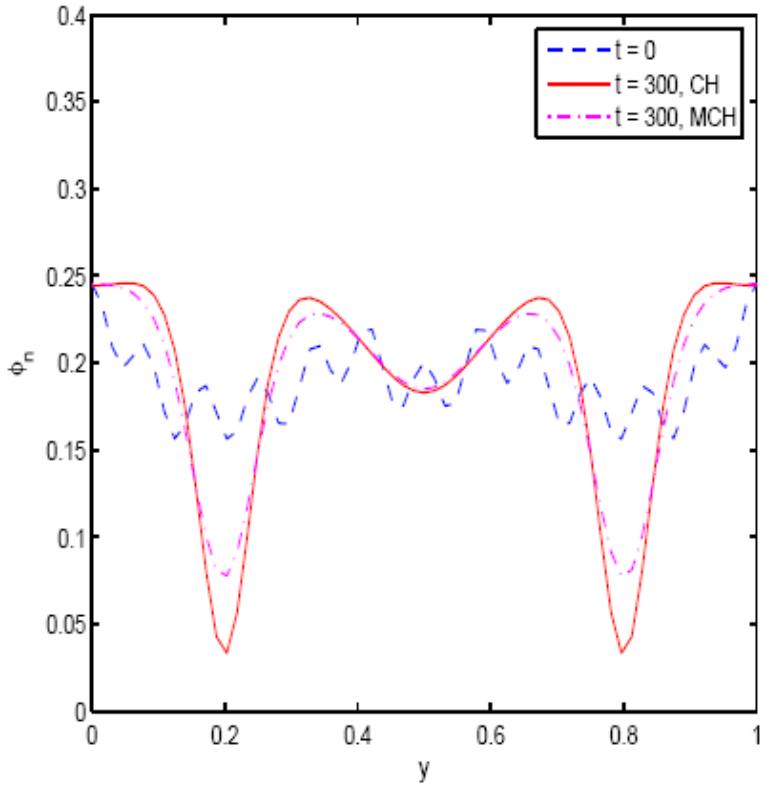


(d) $\lambda = 10^{-2}$

Coarsening of the biofilm profile without eps production

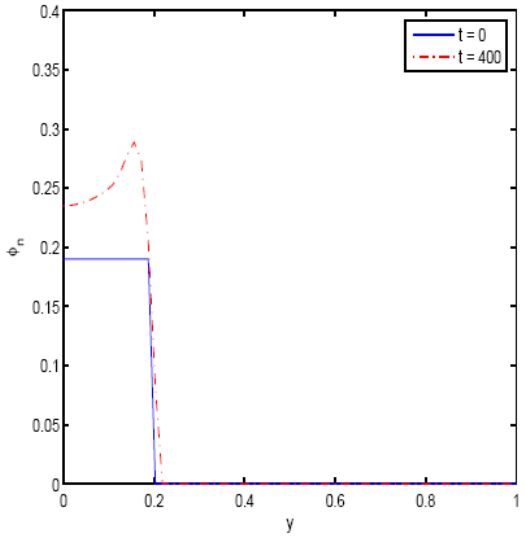


(a) 2 modes



(b) 4 modes

Biofilm growth with eps production (MCH)



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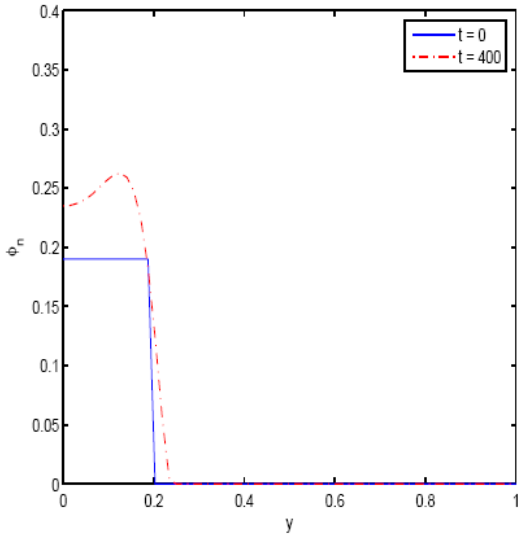

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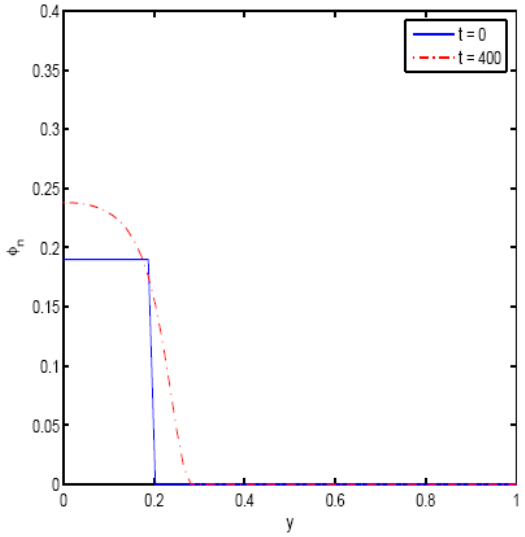

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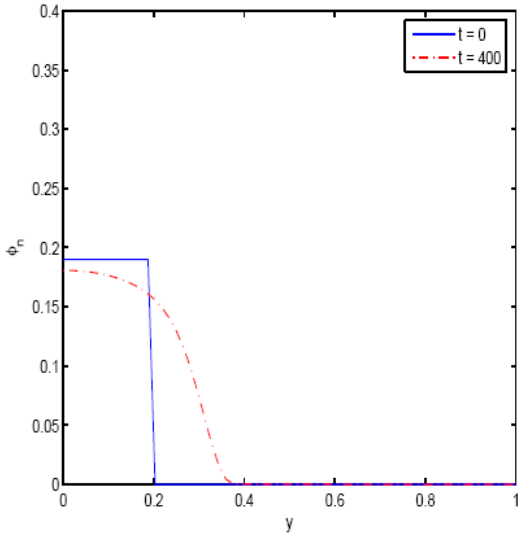

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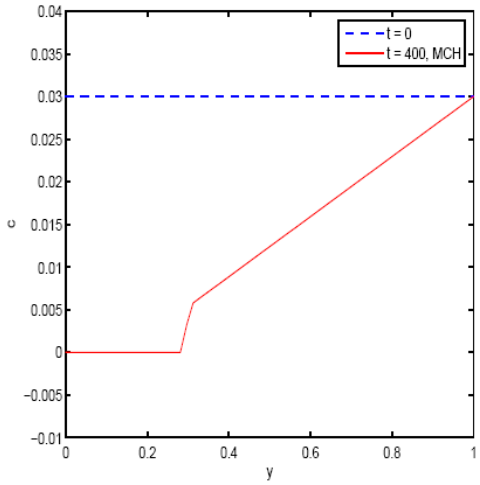
Nutrient substrate concentration



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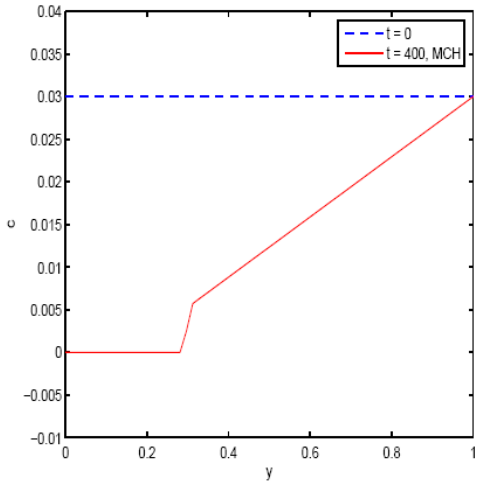

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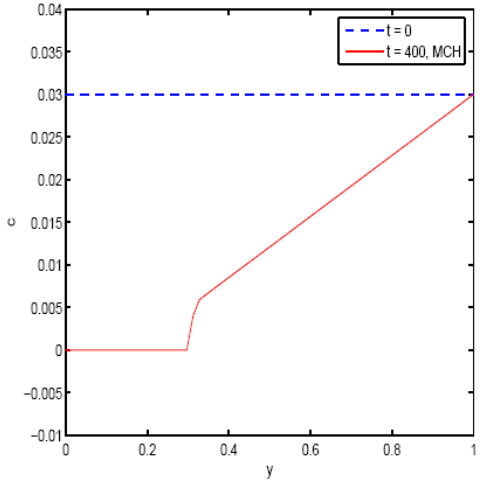

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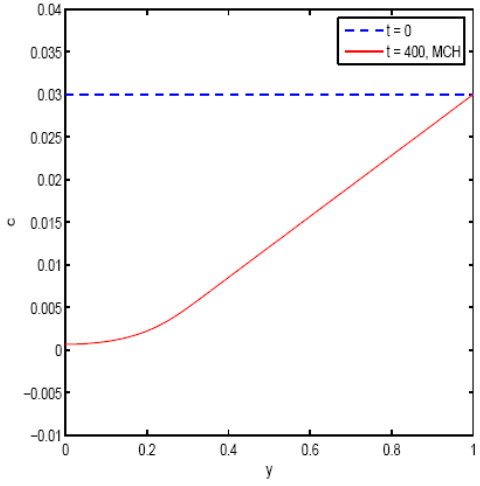

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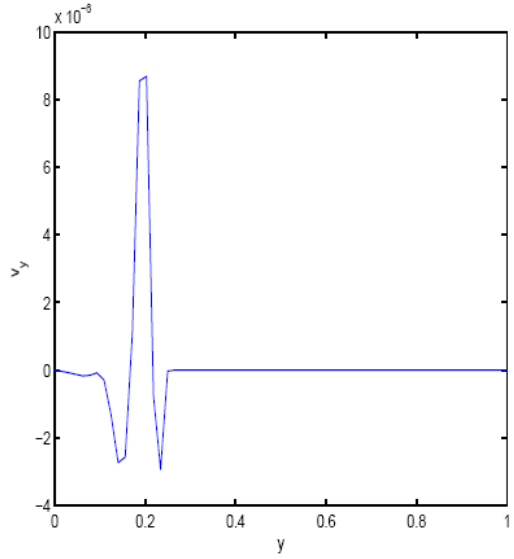
Velocity distribution



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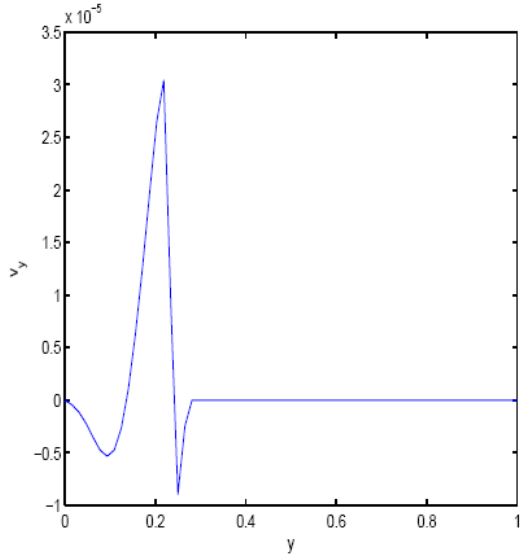

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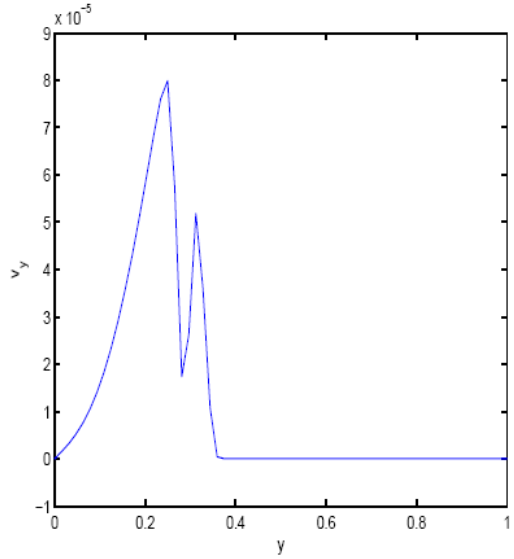

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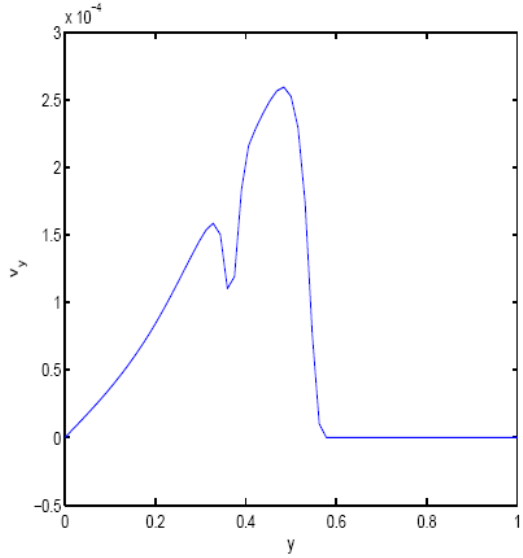
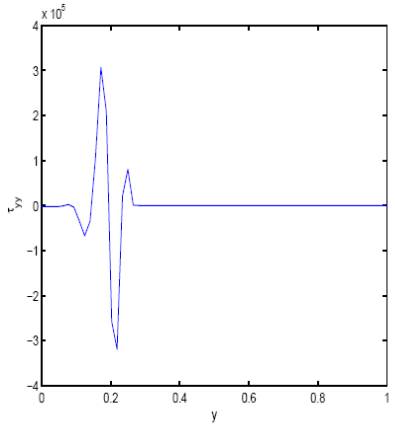


(d) $\lambda = 10^{-2}$

Pressure and normal stress distribution τ_{yy}



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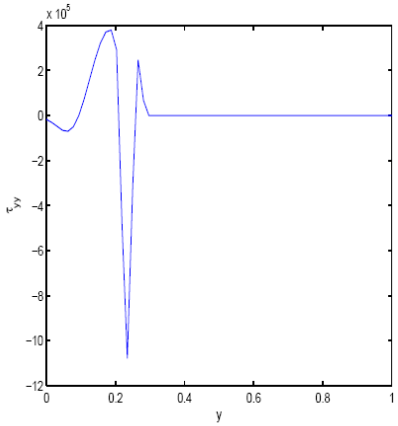

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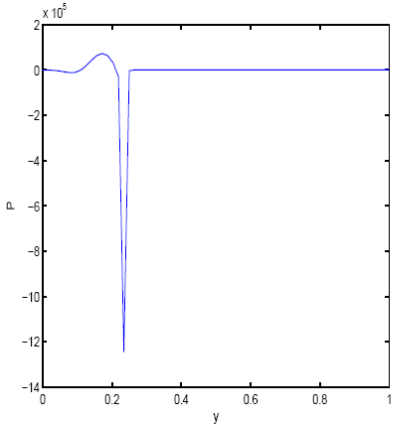

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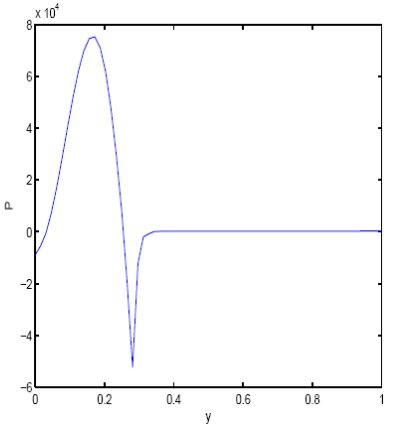

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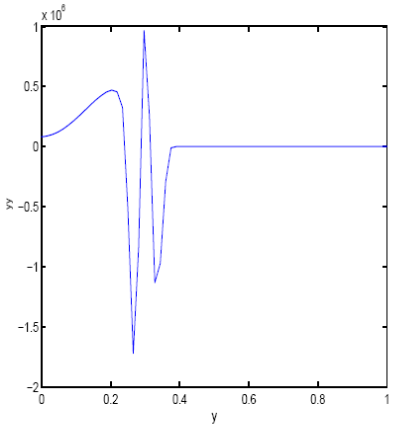

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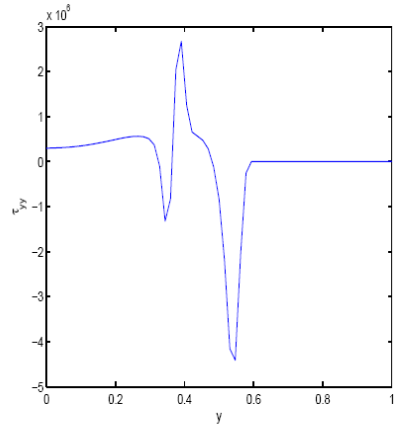

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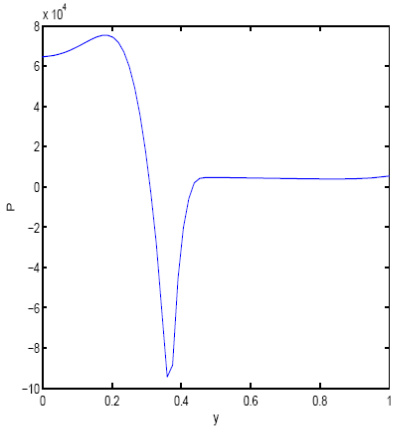

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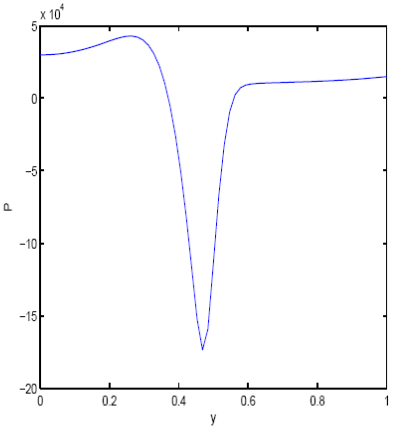
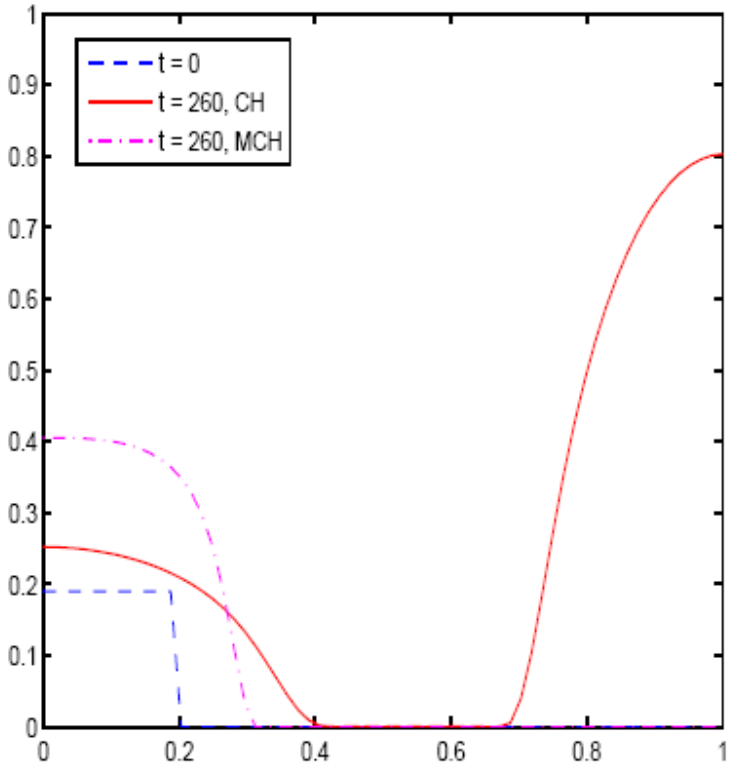
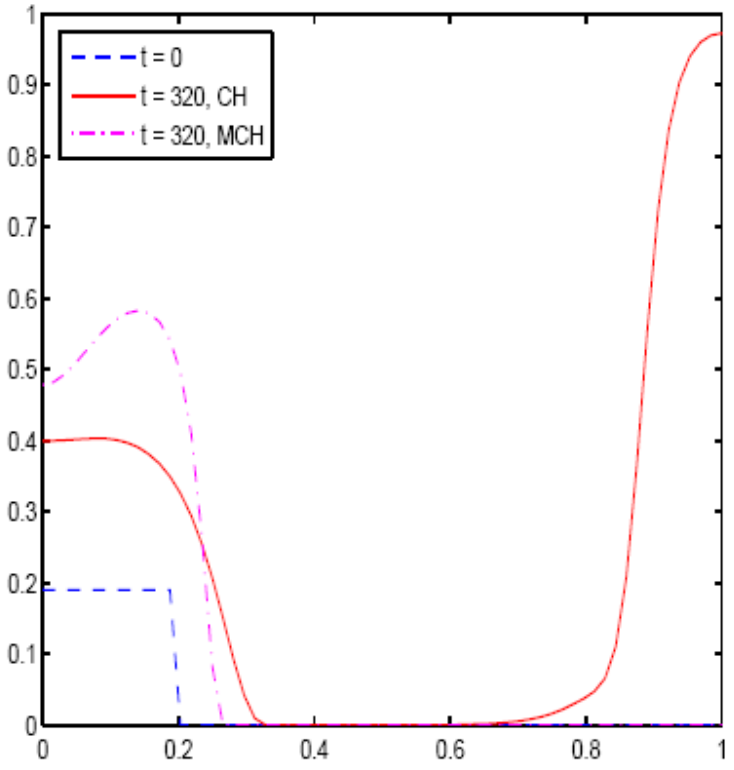


(d) $\lambda = 10^{-2}$

Failure of CH model with eps production vs MCH (Artificial growth near the nutrient rich region)

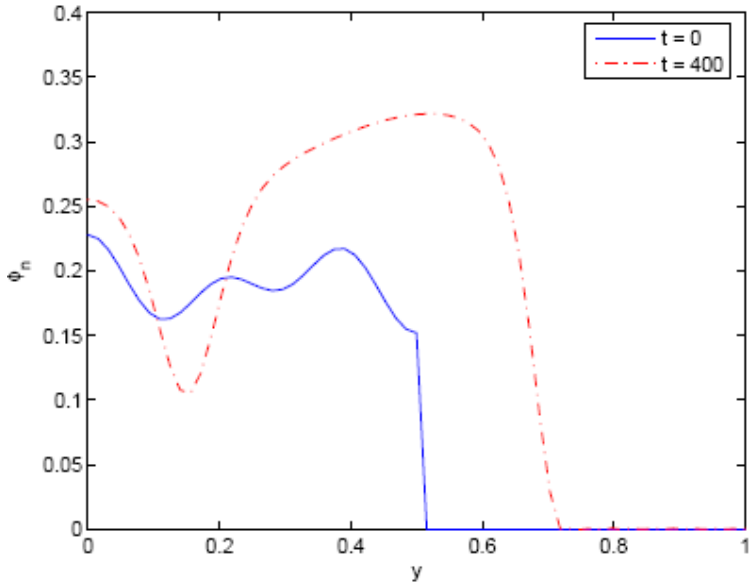


(a) $\lambda = 10^{-3}$

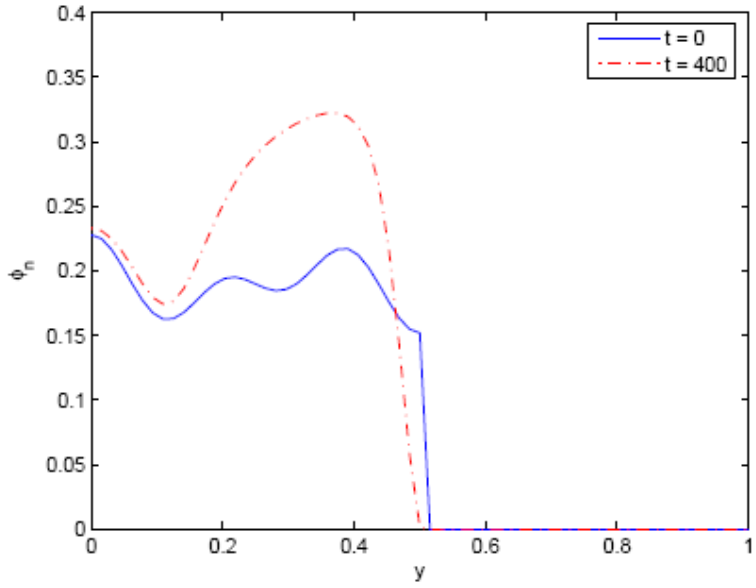


(b) $\lambda = 10^{-4}$

Volume fraction growth of an inhomogeneous step profile

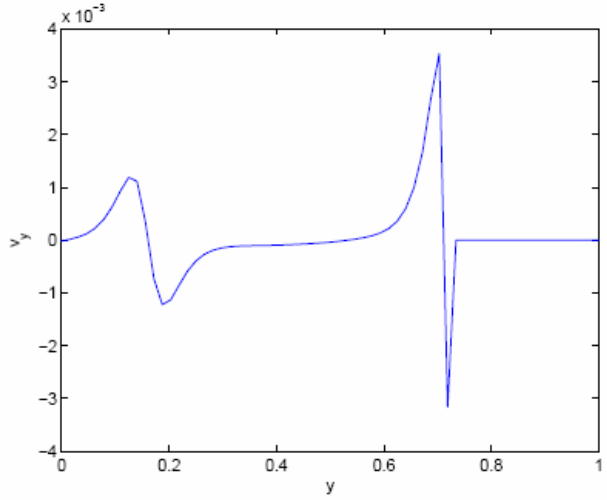


(a) ϕ_n profile,CH.

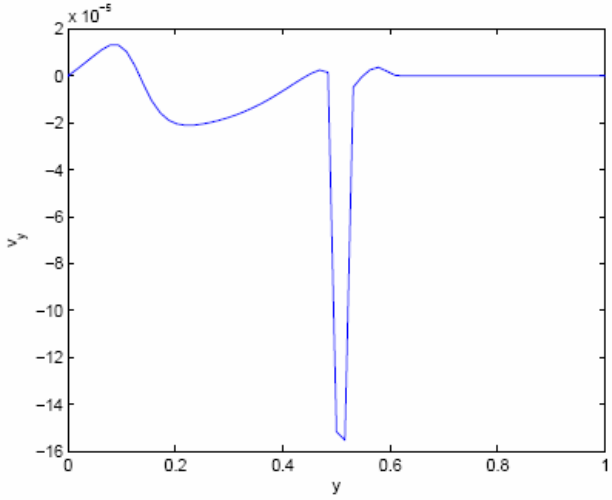


(b) ϕ_n profile, MCH, $\lambda = \lambda/\phi_0$.

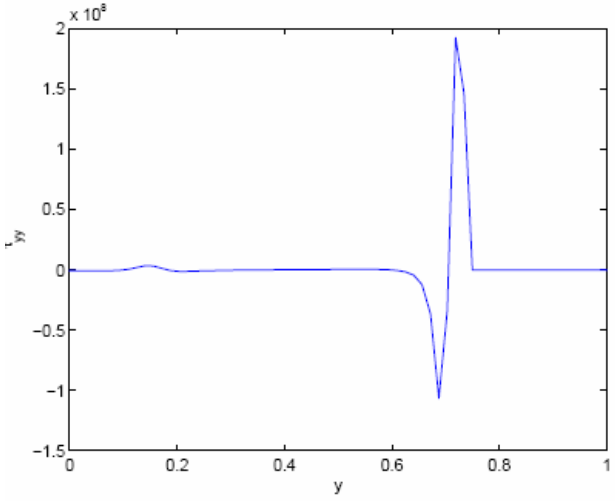
Velocity and stress



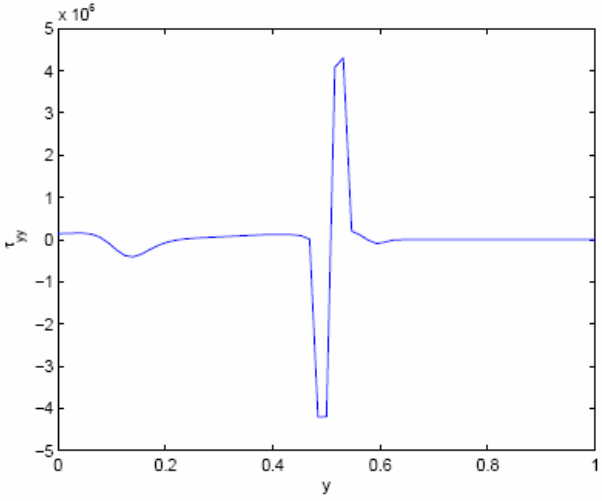
(c) v_y, CH.



(d) v_y, MCH, $\lambda = \lambda/\phi_0$.

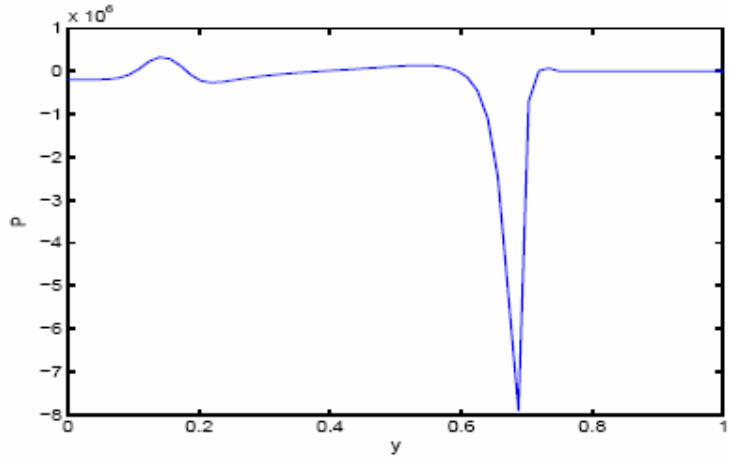


(e) Stress τ_{yy}, CH.

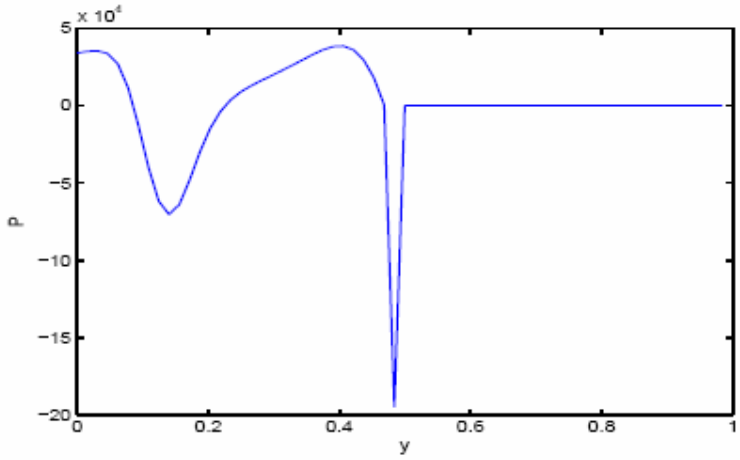


(f) Stress τ_{yy}, MCH, $\lambda = \lambda/\phi_0$.

Pressure



(g) Pressure P,CH.

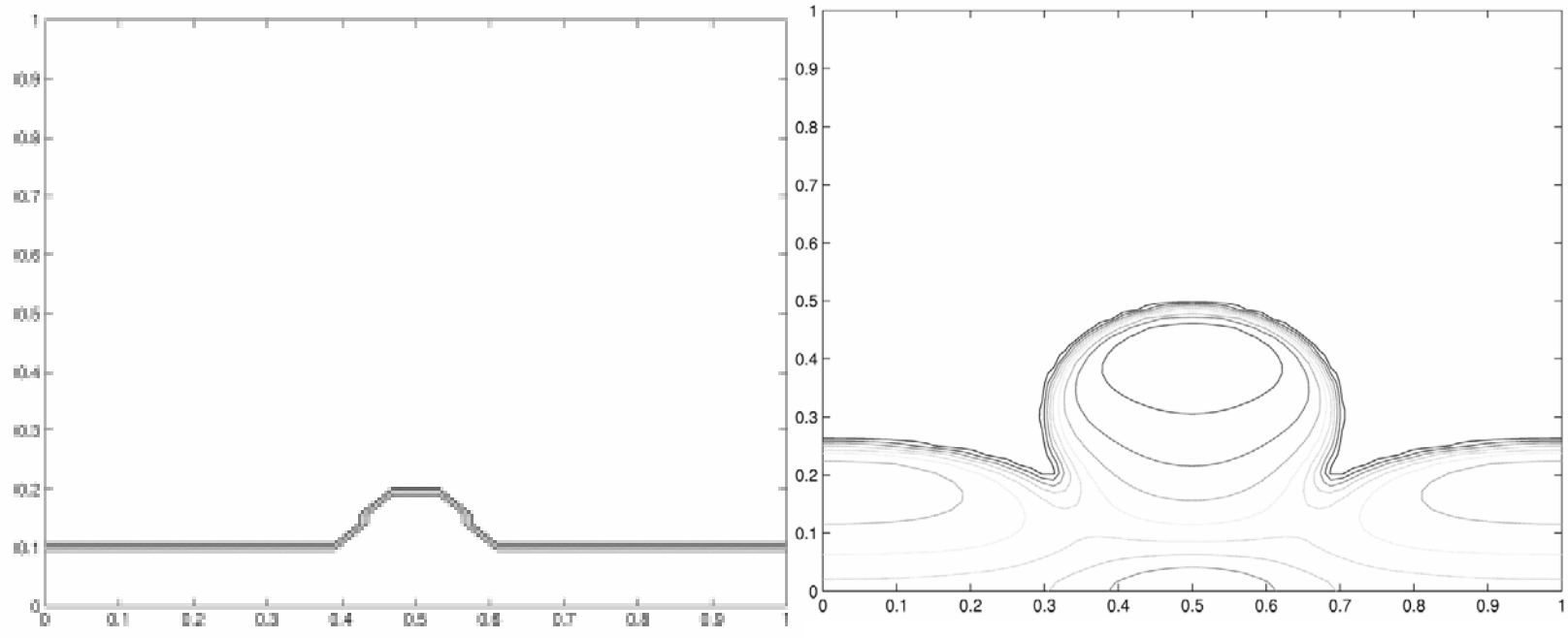


(h) Pressure P, MCH, $\lambda = \lambda/\phi_0$.

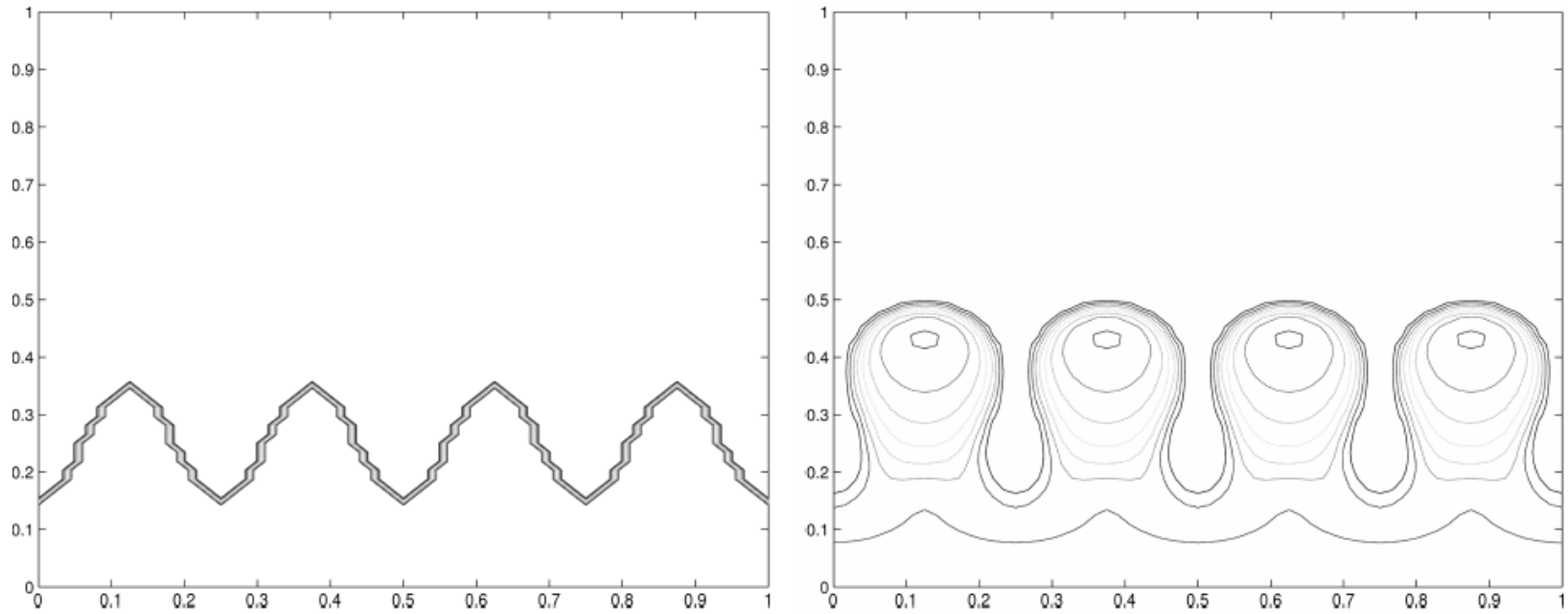
2-D Numerical Methods

- Finite difference of uniform girds, spatially second and temporally second order.
- Velocity corrected projection method for the momentum transport equation (Jie Shen et al., Jianguo Liu et al., etc.).
- Second order discretization for the modified Cahn-Hilliard equation and nutrient substrate transport equation.
- Examples presented below are for the the extended Newtonian model.

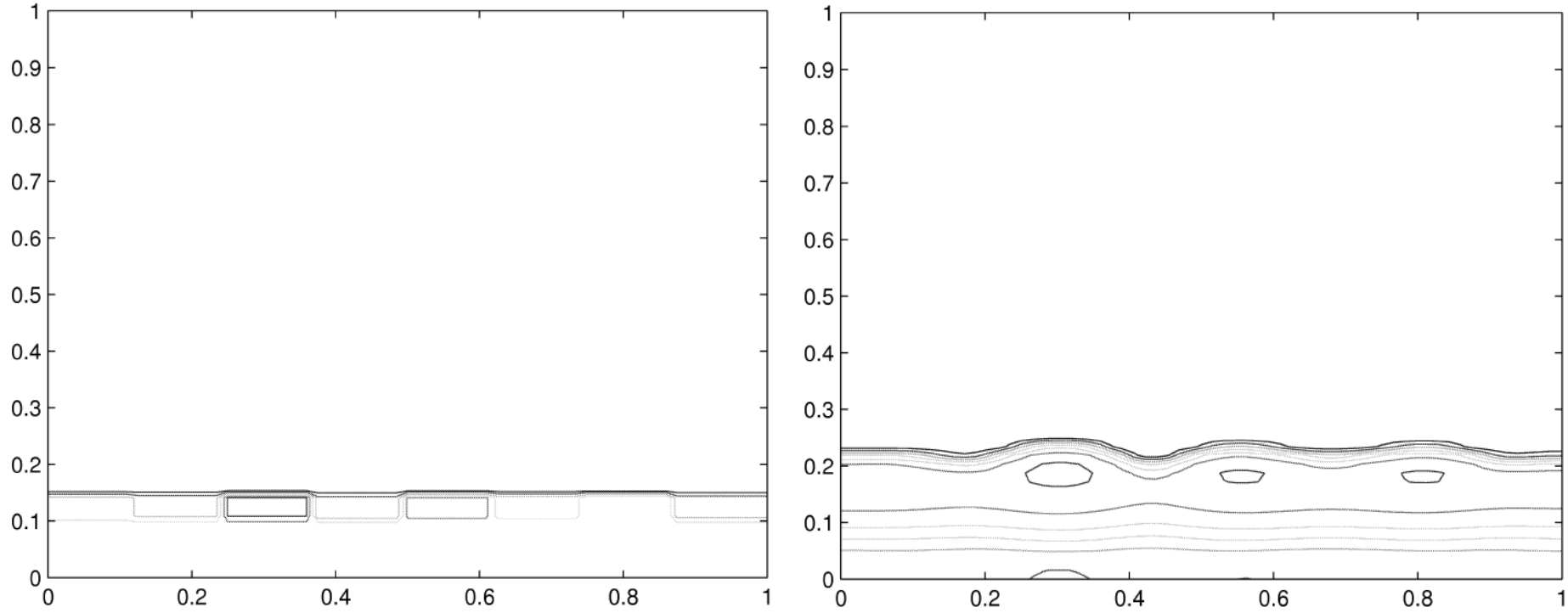
Single bump growth t=320



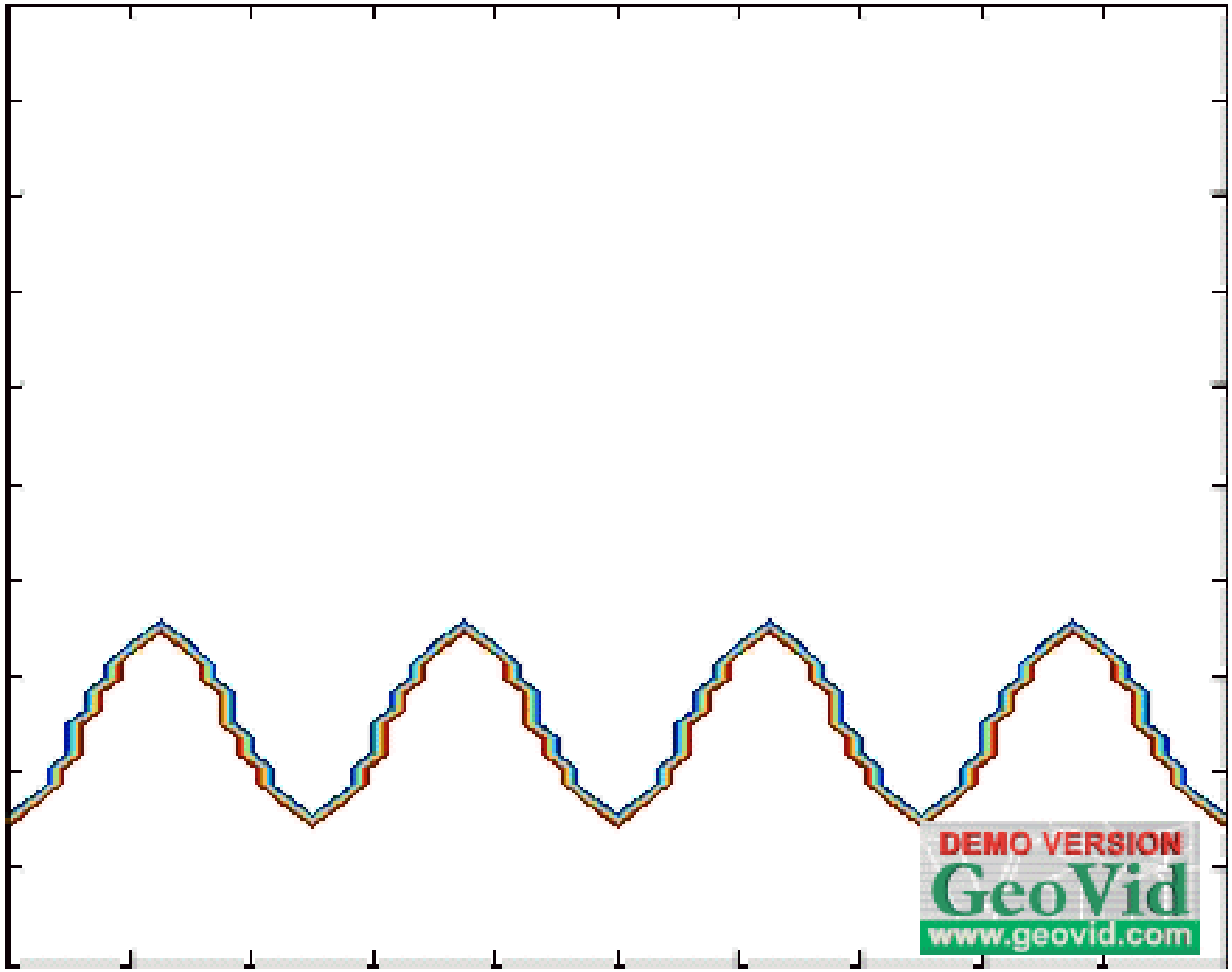
Growth of multiple humps (t=320)



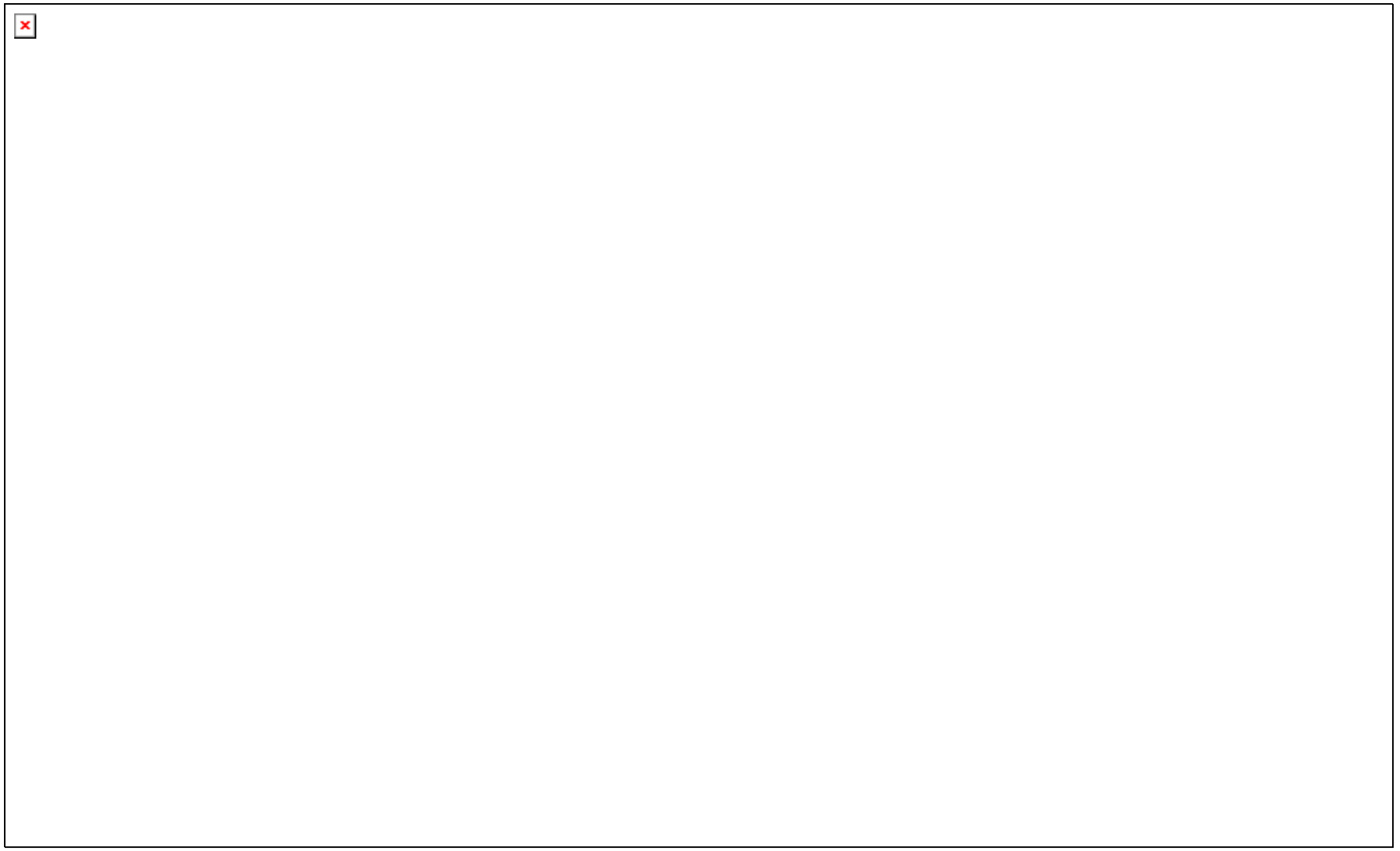
Flat-face inhomogeneous biofilm growth (t=320)



A movie about biofilm growth up to t=320



A kinetic model for biofilms



Conclusion

- We developed a set of one fluid multicomponent phase field models for biofilms accounting for the eps production, polymer network structure and deformation, and nutrient substrate transport.
- The modified Cahn-Hilliard (or singular Cahn-Hilliard) dynamics gives the better resolution for the phases and growth.
- Both gel and temporary network dynamics of eps are incorporated.
- The frame work can be readily extended to account for bacteria as an independent third component and modeled as a viscous fluid.
- Dispersal dynamics and rheological properties of the biofilms can be handled.

Public Health

- Between 1980 and 1992, infectious disease deaths increased by 58% (39% after age adjustment); the major contributors were HIV infection and AIDS, respiratory disease (primarily pneumonia), and bloodstream infection. Infectious diseases are still broadly endemic and maintain a large reservoir of agents that have the potential for rapid and widespread dissemination. Infectious diseases remain the leading cause of death worldwide and the third leading cause of death in the United States. In the United States, each year, approximately 25% of physician visits are attributable to infectious diseases, with direct and indirect costs estimated at more than $120 billion. Because recent research implicates biofilms as reservoirs for pathogenic organisms and sources of disease outbreaks, biotechnology measures are being created to control biofilms and/or sever the routes by which pathogenic organisms are transmitted from biofilms to susceptible people.
- Biofilms are implicated in **otitis media**, the most common acute ear infection in children in the U.S. Other diseases in which biofilms play a role include **bacterial endocarditis** (infection of the inner surface of the heart and its valves), **cystic fibrosis** (a chronic disorder resulting in increased susceptibility to serious lung infection), and **Legionnaire's disease** (an acute respiratory infection resulting from the aspiration of clumps of *Legionnella* biofilms detached from air and water heating/cooling and distribution systems).
- Biofilms may also be responsible for a wide variety of nosocomial (hospital-acquired) infections. Sources of biofilm-related infections can include the surfaces of catheters, medical implants, wound dressings, or other types of medical devices.
- Biofilms avidly colonize many household surfaces, including toilets, sinks, countertops, and cutting boards in the kitchen and bath. Poor disinfection practices and ineffective cleaning products may increase the incidence of illnesses associated with pathogenic organisms in the household environment.