A Model of Arterial Plaque Cap Development and Degradation

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Introduction

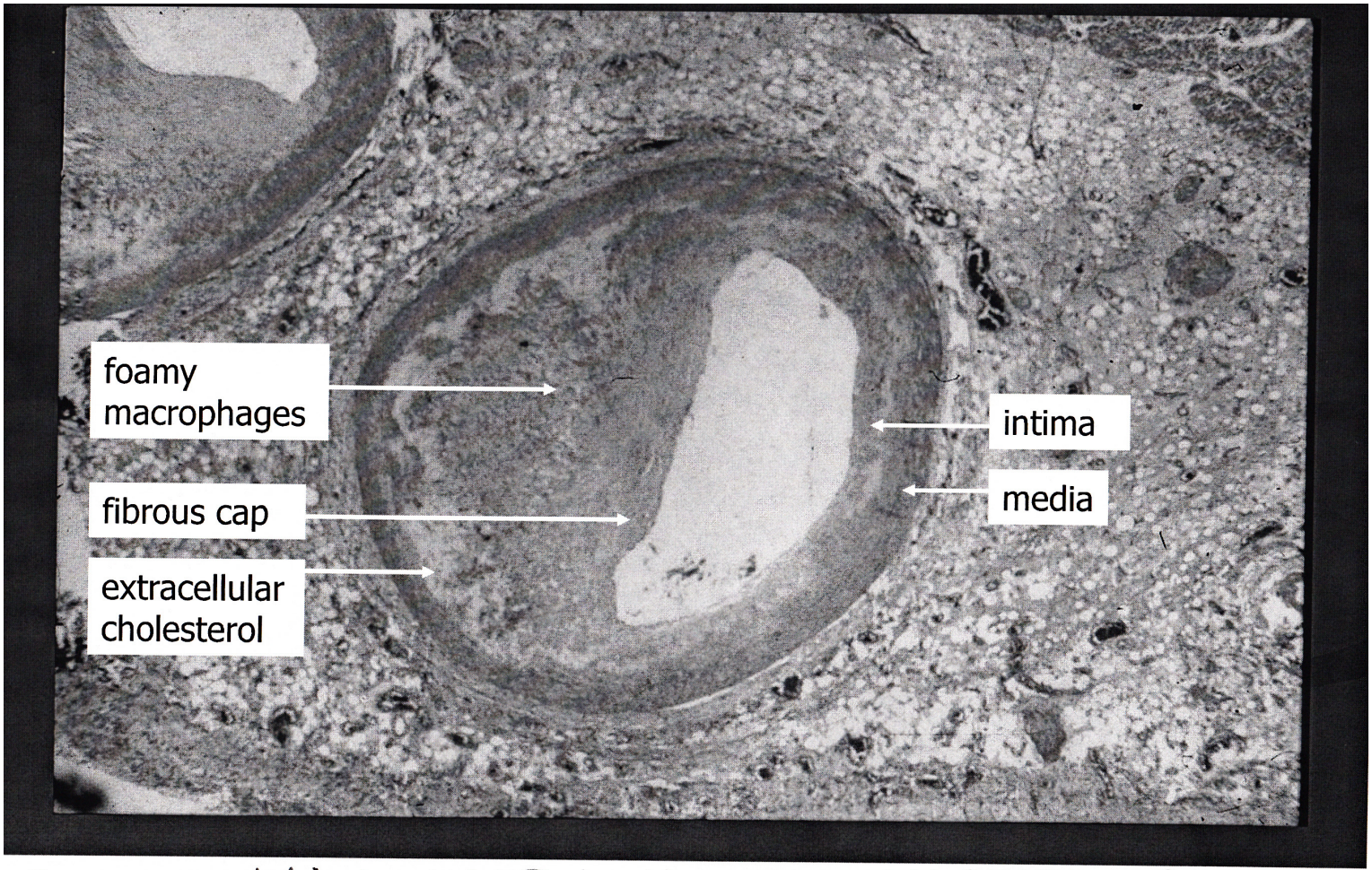
- Cardiovascular disease affects 80 million Americans (2006 data) 2200 Americans die of cardiovascular disease every day (2008) Coronary heart disease was responsible for 1 out of 6 deaths in US
- A common form of cardiovascular disease is atherosclerosis
- Atherosclerosis is an inflammatory disease of large and medium arteries due to fatty lesions containing cholesterol and cell debris in the arterial wall
- Doctors now believe that rupture of certain plaques (vulnerable plaques) are responsible for most deaths
- In one study , 73% of all deaths examined from heart attack were caused by plaque rupture

Arterial Plaques

- A plaque is a lesion that develops in the arterial wall layer (intima)
- It is made up of immune cells, cell debris, lipids (cholesterol, fatty acids,), fibrous connective tissue, etc
- Arterial plaque formation and growth involves complex chemical, hemodynamic, and biomechanical processes
- Arterial plaques have lipid cores separated from the blood flow by an endothelial cell layer and a fibrous cap
- There are basically two types of plaques: stable plaques and unstable plaques (vulnerable plaques (VP), high-risk plaques, thin-cap fibroatheromas (TCFAs))

Plaque Cross Section

section.jpg



From a presentation by Robin Poston, John McGregor, Sophia Collot-Teixera, Saliya Tilmaz
Cardiovascular Division, King's College, London

Characteristics of Vulnerable Plaques

- Large lipid core: more than 40% of the plaque volume
- Thin fibrous cap with little collagen fibers, cap thickness $< 65\mu m$
- Ratio of plaque area occupied by lipid components (macrophages and extracellular lipids) versus fibromuscular components (smooth muscle cells and collagen) is large

Other Characteristics

- Large number of inflammatory cells, macrophages, foam cells, T-lymphocytes
- Inward (negative) remodeling causing stenosis (partial blood flow blockage), and hence variable shear stress on endothelial layer and cap

Main Players in Our Story

Monocytes and macrophages; foam cells; smooth muscle cells; endothelial cells; low density lipoproteins (LDLs); oxidized LDLs; extracellular matrix (ECM) material; matrix metalloproteinases (mmps); various cytokines (TGF-β, TNF-α, IL-1, PDGF, etc.)

Other Players

T-cells, antigen-presenting cells, HDLs, adhesion molecules, calcified macrophages, ...

Plaque Development

- Some injury to endothelial layer (EL) causing inflammatory response, perhaps triggered by low-density lipoprotein (LDL) excess
- Once in intima, LDL is rapidly oxidized by free radicals, producing ox-LDLs. Free radicals are oxidative agents released by ongoing chemical reactions within cells
- Endothelial cells (ECs) display adhesion molecules on lumen side latching onto monocytes and other immune cells. Secreted chemoattractants lure monocytes into intima that quickly mature into macrophages. Macrophages have scavenger receptors that recognize ox-LDLs, allowing macrophages to ingest them

Plaque Development continued

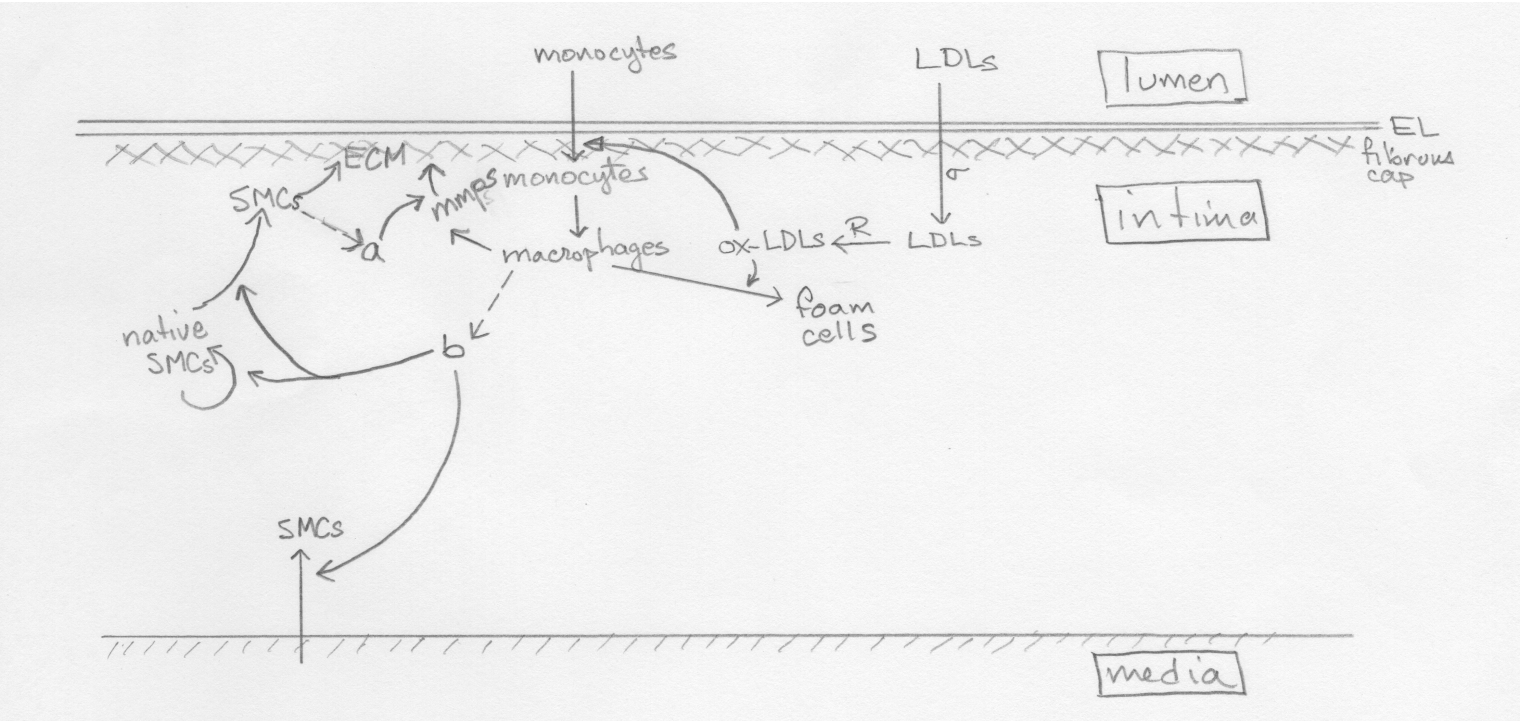
- The result is that macrophages turn into lipid-rich foam cells
- The action of ECs, ox-LDLs, and macrophages release cytokines that cause smooth muscle cell (SMCs) proliferation and migration into plaque (from media). They also move up a chemical gradient toward the EL, and with producing extracellular matrix material (ECM, mostly collagen), a fibrous cap forms behind the EL
- Accumulation of foam cells and extracellular lipid cause the plaque to grow and cause arterial remodeling. Inward remodeling (thickening) impinges on the blood flow (stenosis), changing the distribution of shear stress on the EL and plaque
- Decreased shear stress and production of matrix metalloproteinases (mmps), from macrophages, negatively affect the structure and strength of the cap, and determine the stability of the plaque.

Model Assumptions

- One space dimension: intima defined by interval $0 < x < 1$.
- Once inflammation begins, LDLs $\Rightarrow$ oxidized-LDLs fast; so dynamics is collapsed into ox-LDL flux and diffusion
- SMCs, both native to intima, and imported, are the source of ECM building material for the cap. Rather than follow dynamically the ECM concentration, we assume it is proportional to SMC concentration.
- Macrophages produce mmps and other substances that tend to degrade the cap. Rather than follow dynamically the mmp concentration, we assume it is proportional to macrophage concentration
- Chemoattractant activity is important. Production of macrophage chemoattractants, like chemotactic-peptide-1 (MCP-1), from SMCs, and production of growth factors, like PDGF-B, from macrophages, that play a role in SMC migration and proliferation

Diagram of Model Interaction in Intima Layer

Main model variables: $\mathbf{O}$ = ox-LDL conc.; $\mathbf{M}$ = macrophage conc.; $\mathbf{N}$ = smooth muscle cell conc.; $\mathbf{a}, \mathbf{b}$ = two chemoattractants conc.;



Model Equations

The system is defined on $(0,1) \times (0,T)$:

$$\begin{aligned} O_t &= D_O O_{xx} - \mu_O O \\ M_t &= D_M M_{xx} - \alpha_1 (B_1(M) a_x)_x - \mu_M M \\ N_t &= D_N N_{xx} - \alpha_2 (B_2(N) b_x)_x + \rho(b) f(N) \\ a_t &= D_a a_{xx} + g_1(O) N - \mu_a a \\ b_t &= D_b b_{xx} + g_2 M - \mu_b b \end{aligned} \tag{1}$$

along with non-negative initial conditions, and boundary conditions

$$\begin{aligned} &\text{At } x = 0 : O_x = M_x = a_x = b_x = 0, -N_x = g(b(0,t)) \\ &\text{At } x = 1 : a_x = b_x = N_x = 0, M_x = h(O(1,t)), O_x = \sigma \end{aligned} \tag{2}$$

Question: With this limited use of chemoattractants, can we get N, hence ECM, and M, hence $mmps$, in sufficient concentrations near the EL boundary?

Standard Chemotaxis

Origin: Keller-Segel JTB 1970, 1971 (aggregation of slime mould)
u = cell concentration , v = chemical concentration

$$\begin{cases} u_t = D\triangle u - \chi\nabla(u\nabla v) \\ v_t = \triangle v + u - v \end{cases}$$

1D case: global existence (Osaki, 2001)
Models:

- Cell mobility in embryonic development
- Worm C. elegans motility in response to external chemical signals
- sperm cells attracted to chemical releases from eggs
- Immune cells migrating to sites of inflammation

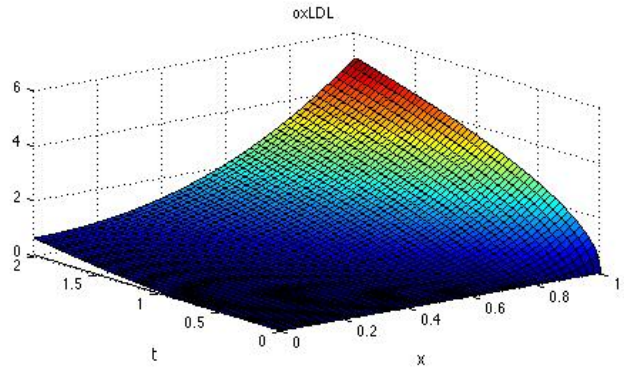
Standard Chemotaxis: Higher Dimensions nD

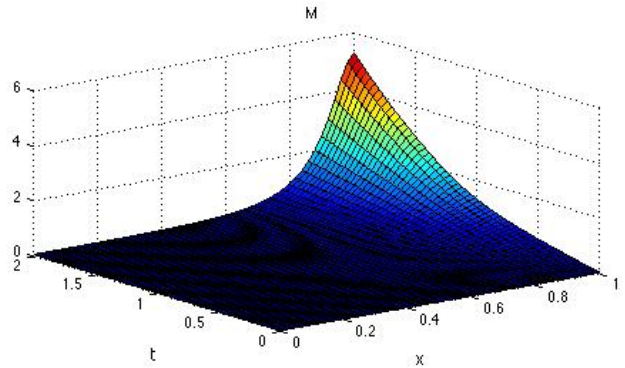
For $n = 2$: $(u_0 \doteq u(x, 0))$

1. $m \doteq \int_0^1 u_0 < 4\pi$, all sol's are bounded (Gajewski, Zacharias, 1998)
2. For any $\varepsilon > 0$, there exist unbounded sol's with $m > 4\pi + \varepsilon$ (Horstmann, Wang, 2001)
   no knowledge whether blow-up time is finite or infinite!
3. For radial symmetric case, $m > 8\pi$, sol's can be constructed to blow-up in finite time (Herrero, Velazquez, 1997)

For $n \geq 3$: no example of a sol. that undergoes finite time blow-up

Simulation


oxLDL
6
4
2
0
2
1.5
1
0.5
0
t
0
0.2
0.4
0.6
0.8
1
x


M
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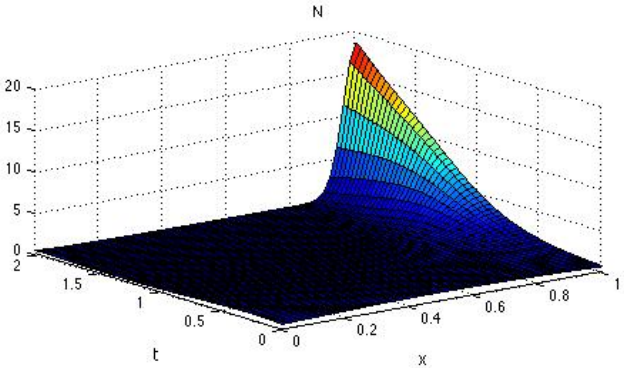
N
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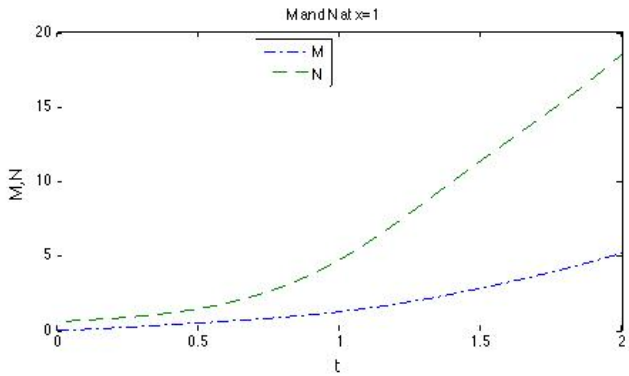

M and N at x=1
M
N
20
15
10
5
0
M,N
0
0.5
1
1.5
2
t

Prototype Cross-Chemotaxis Model

$$
\begin{aligned}
u_t &= D_1 u_{xx} - \alpha_1 (u a_x)_x - \mu u \\
v_t &= D_2 v_{xx} - \alpha_2 (v b_x)_x + \rho(b) f(N) \\
a_t &= D_3 a_{xx} + g_1 v - \mu_a a \\
b_t &= D_4 b_{xx} + g_2 u - \mu_b b
\end{aligned}
\tag{3}
$$

with non-negative initial conditions and the boundary conditions

$$
\begin{cases}
\text{At } x = 0 : a_x = b_x = u_x = 0, -v_x = h(b(0,t)) \\
\text{At } x = 1 : a_x = b_x = v_x = 0, u_x = g(t)
\end{cases}
\tag{4}
$$

Local existence

Theorem 1: There exists a local (in time) weak solution of the (u, v, a, b) system on $[0, T]$ for some $T > 0$ with the following regularity properties:

$$u, v, a, b \in C([0, T]; L^2) \cap L^2([0, T]; H^1) \quad \text{and}$$

$$a, b \in L^\infty([0, T]; H^1) \cap L^2([0, T]; H^2)$$

Proof is through a contraction mapping argument
(paper submitted; co-author: Animikh Biswas, UMBC)
(See arXiv.org/abs/1511.02304)

Positivity and Global Solvability

Positivity of solutions: Let $u_0(x) = u(x, 0)$, etc.
$u_0, v_0, a_0, b_0 \geq 0$ on $[0, 1]$ implies $u, v, a, b \geq 0$ on $(0, 1) \times (0, T)$

Global bounds: Among other bounds we show, for every $T > 0$,

$$\sup_{t\in[0,T]} \max\{||u(t)||_{L^1}, ||v(t)||_{L^1}, ||a(t)||_{L^1}, ||b(t)||_{L^1}\} \leq C^T < \infty$$

$$\sup_{t\in[0,T]} \max\{||a_x(t)||_{L^1}, ||b_x(t)||_{L^1}\} \leq \widetilde{C}^T < \infty$$

$C^T, \widetilde{C}^T$ may depend on initial data, parameters, and T, but are independent of the solution. We also make use of the diffusion semigroup with Neumann b.c.s.

Basic Idea of Positivity Proof

Proofs of solvability, regularity are quite technical.

$w_- = \min\{w, 0\}$, $w = u, v, a, b$

Take inner product of the w equation with w, do estimates, use Young's inequality, etc. $\Rightarrow$ (by Gronwall inequality)

$$\frac{d}{dt}||w_-||^2 \leq \text{stuff}||w_-||^2 \quad \rightarrow \quad ||w_-||^2 \leq ||w_{0-}||^2 e^{C\int_0^t \text{stuff}}$$

$$\text{but } w_0 \geq 0, \text{ so } ||w_{0-}|| = 0, \text{ implying } w \geq 0$$

Dynamics of Average Concentrations

Let $\bar{u}(t) \doteq \int_0^1 u(x,t)dx$, etc. so $\bar{u}_0 = \int_0^1 u(x,0)dx$, etc.
Then, for every $T > 0$,

$$\bar{u}(t) \leq \bar{u}_0 + \frac{D_1}{\sqrt{2\mu}}||g||_{L^2(0,T)} \quad , \quad \bar{u}(t) \to \bar{u}_c \text{ for some } 0 \leq \bar{u}_c < \infty$$

and this gives $\sup_{t\geq 0} \bar{b}(t) < \infty$, and $\bar{b}(t) \to \bar{b}_c$ for some $0 \leq \bar{b}_c < \infty$. Now if
$h(b) \leq h_M, \rho(b) \leq \rho_M, f_M = \sup_v |f(v)| < \infty$, then

$$\bar{v}(t) \leq \bar{v}_0 + (D_2 h_M + \rho_M f_M)t \quad , \quad t \in (0,T)$$

$$\bar{a}(t) \leq \bar{a}_0 + g_1 \bar{v}_0 t + \tfrac{g_1}{2}(D_2 h_M + \rho_M f_M)t^2$$

Examples of Multi-Chemotactic Activity

- Macrophage chemoattractants, like MCP-1, from smooth muscle cells, and PDGF-B, from macrophages, plays a role in smooth muscle cell migration and proliferation.
  HB-Macrophages also produce HB-EGF that plays a role in smooth muscle cell migration
- Activated endothelial cells produce PDGF-BB and insulin-like growth factor 1 (IGF-1). Other chemicals that have chemoattractant properties include macrophage colony-stimulating factor, endothelin-1, etc.

Generalized Cross-Chemotactic System

Cell concentrations: u_i, $i = 1..N$
chemical concentrations: c_j, $j = 1..M$
For each i, u_i produces $k(i)$ chemicals $c_{i_1}, \ldots, c_{i_{k(i)}}$
u_i is "influenced" by $s(i)$ chemicals $c_{i_{-1}}, \ldots, c_{i_{-s(i)}}$

$$\begin{cases} u_{it} = d_i u_{ixx} - \sum_{1 \le l \le s(i)} \alpha_{i_l} (u_i c_{i_{-l}x})_x + f_i(u_i) \ , \ i = 1..N \\ \\ c_{i_j t} = D_{i_j} c_{i_j xx} + g_{i_j} u_i - m_{i_j} c_{i_j} \ , \ j = 1..k(i) \end{cases}$$

with non-negative i.c.s $\{u_i(x,0), c_{i_j}(x,0)\} = \{u_{i0}(x), c_{i_j 0}(x)\}$ and homogeneous Neumann b.c.s.

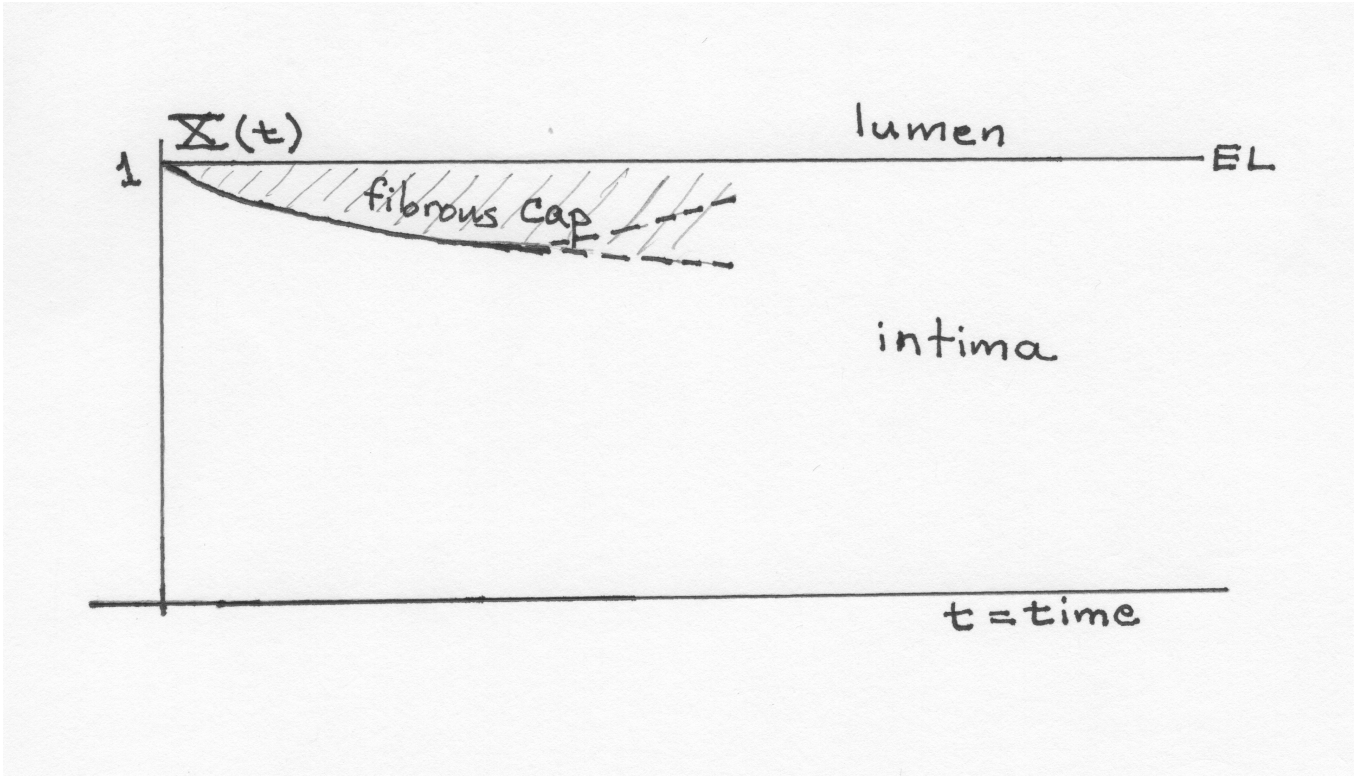
Fibrous Cap (Thickness) Model

Assumptions:

- No inward remodeling, so EL remains fixed at $x = 1$. Cap region: $X(t) < x < 1$, so cap thickness$=1 - X(t)$.
- The cap dynamics is considered a competition between SMCs (depositing ECM building material), and macrophages (releasing destructive mmps). So,

$$\frac{dX}{dt} = -\varepsilon\{F_1(N(X(t),t)) - F_2(M(X(t),t))\}\ ,\ X(0) = 1\ , \quad (5)$$

Fibrous Cap (Thickness) Model



Question: What conditions lead to the eventual rise of X(t) (weakening of the cap: the vulnerable plaque case), versus continued decrease of X(t) (cap thickening: the stable plaque case)?

Simulation: stable case

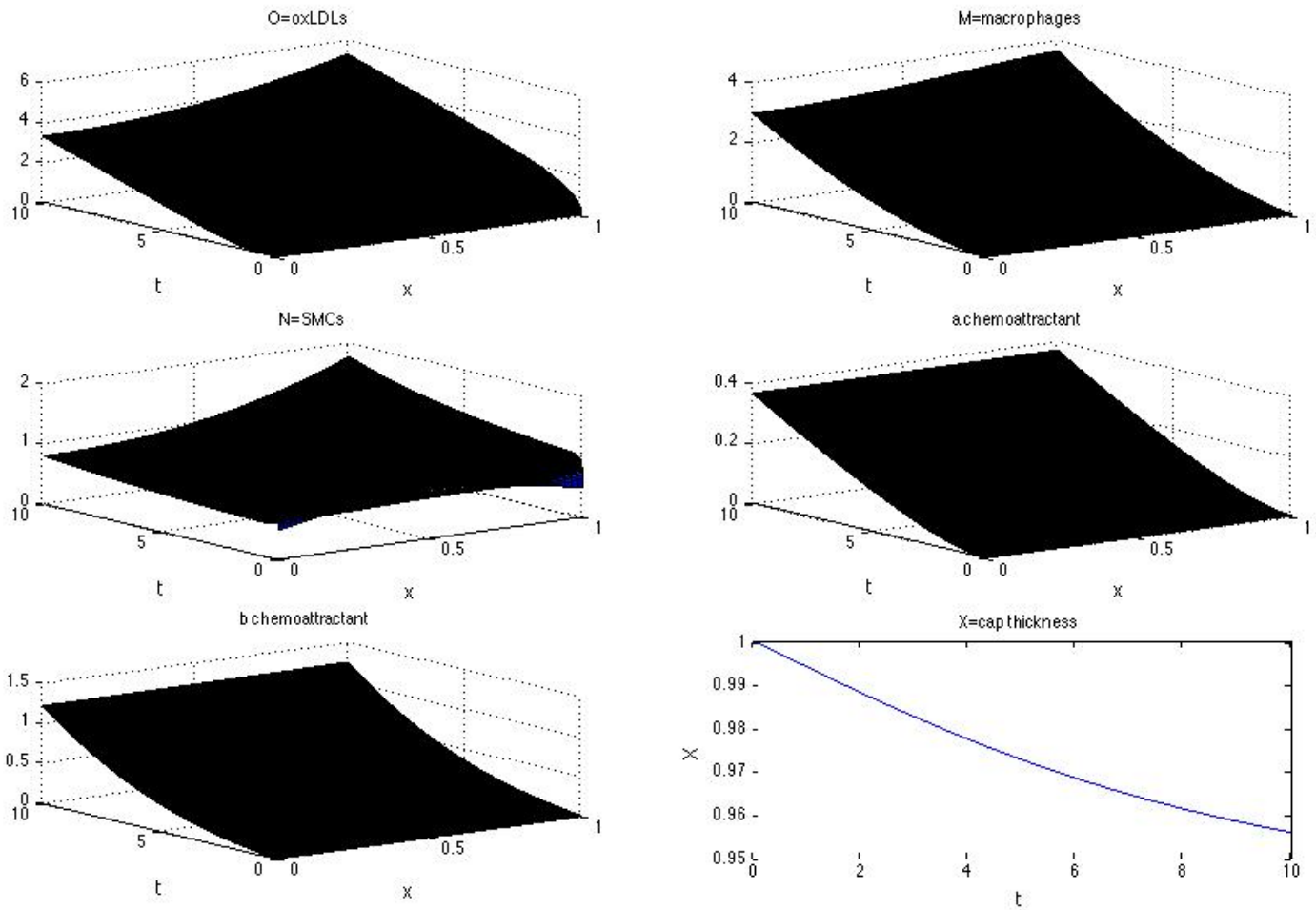


Figure: Stable case with $\sigma = 0.4$, $\sigma_b = 0.04$, $\sigma_a = 0.0$.

Simulation: vulnerable case

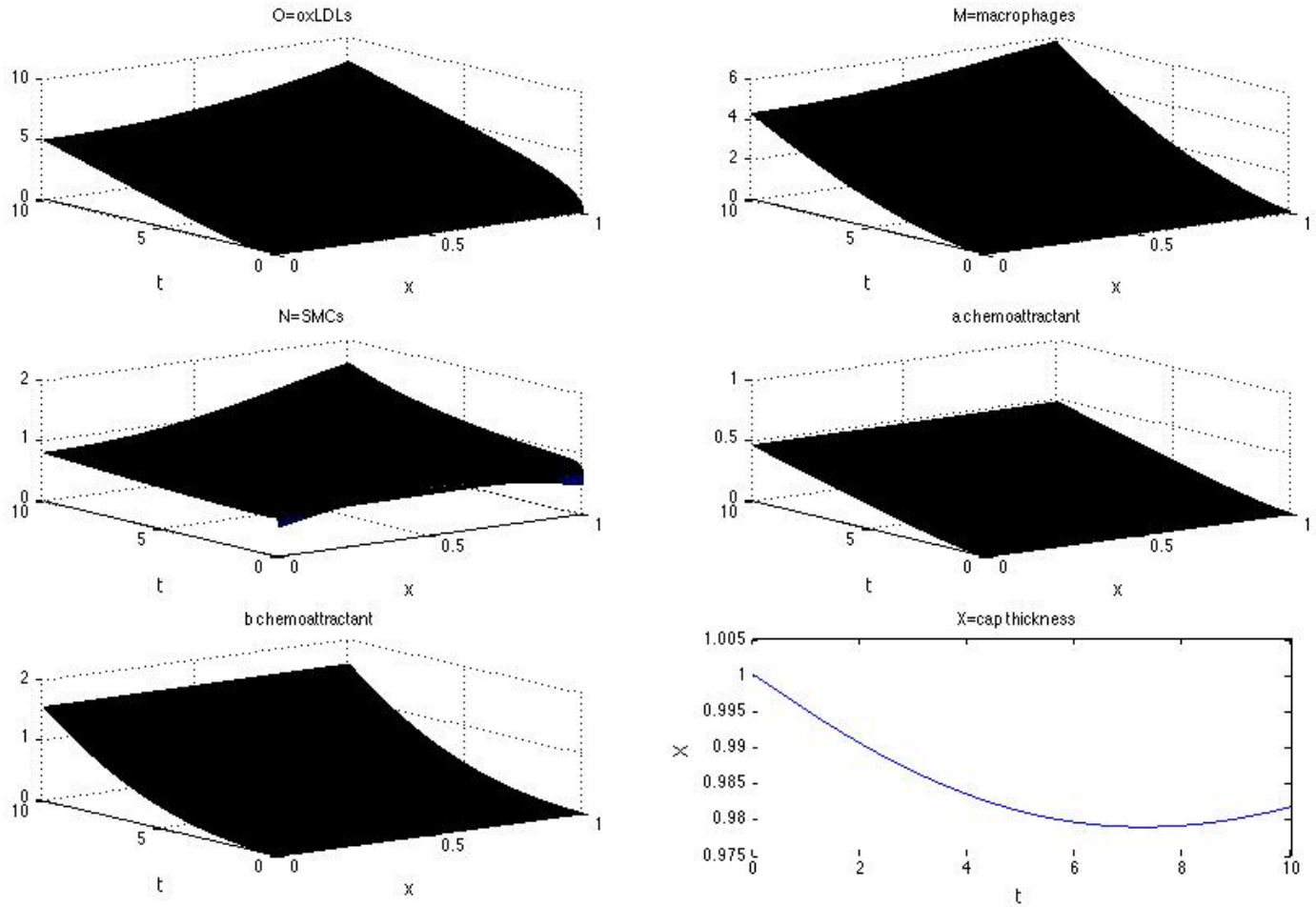


Figure: Unstable case with $\sigma = 0.6$, $\sigma_b = 0.02$, $\sigma_a = 0.01$.

Shear Stress and EC Dynamics

- Atherosclerotic lesions localize preferentially in arterial regions exposed to low shear stress (SS) (places of disturbed flow)
- ECs seem to sense SS as a mechanical signal and transmit it to the cells interior
- Experimental studies indicate various transduction pathways are activated by SS (ion channels, G proteins, adhesion proteins, tyrosine kinase receptors, cytoskeleton, etc.)
- Cytoskeleton constitutes a central mediator of signaling that connects laminal, basal, and junctional cell formations

Shear Stress and EC Dynamics

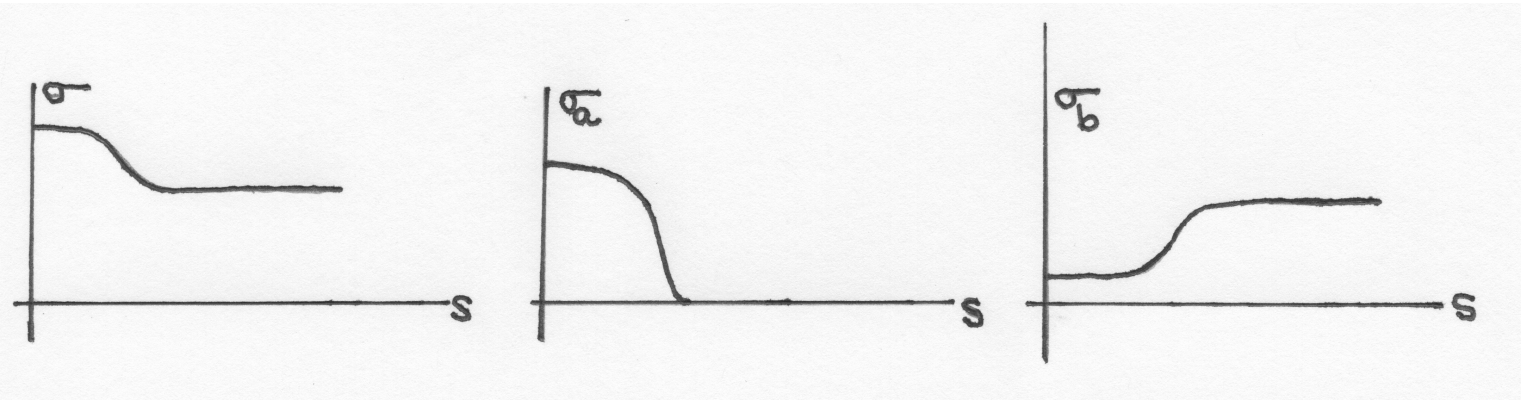
- Pathways lead to phosphorylation of several transcription factors that bind to positive or negative SS response elements
- In regions of non-disturbed flow, ECs express various atheroprotective genes and suppress pro-atherogenic genes
- In regions of low or disturbed flow where low SS occurs, atheroprotective gene expression is suppressed, pro-atherogenic gene expression is up-regulated

This suggests a bifurcation mechanism (measure of SS as a bifurcation parameter).

Incorporating Aspects of Shear Stress in Model

Simplest Strategy: $s =$ measure of endothelial shear stress

$$O_x(1,t) = \sigma(s)\ ,\ a_x(1,t) = \sigma_a(s)\ ,\ \ b_x(1,t) = \sigma_b(s)$$



Remark: Sample simulations above used a piecewise constant version of these.

Transegrity Hypothesis

Fluid mechanical stimulus sensed by structures (flow sensors) at the EC surface transmit a signal via cytoskeleton to various sites, including the nucleus, cell-cell adhesion proteins, and focal adhesion sites, where it is a transducer to a biochemical response (Ingber, et al, 1994)

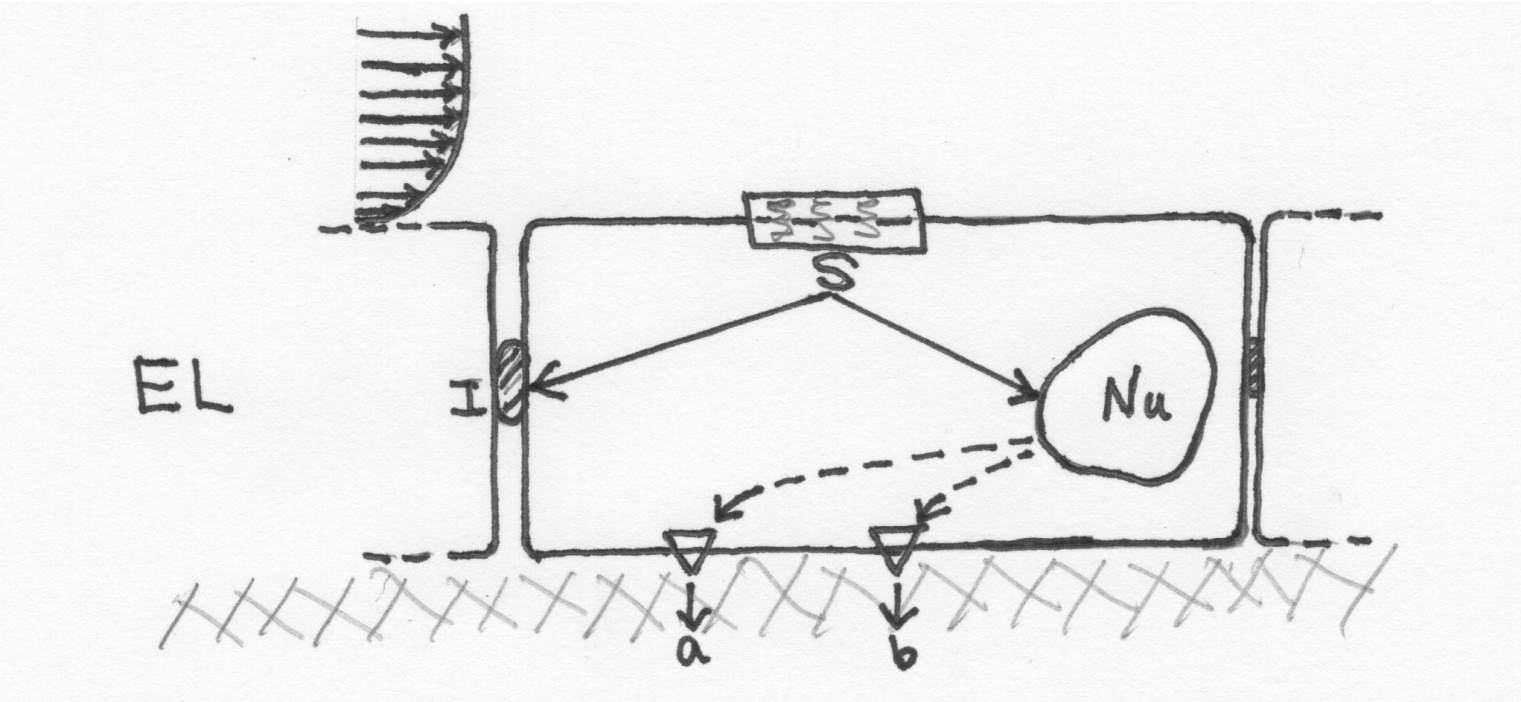
Flow sensors, cell-cell adhesion proteins, etc. can be modeled as a viscoelastic material through coupling Kelvin bodies together (Davies, Barakat, ...)

Another Approach to EL Dynamics

At $x = 1$: $O_x = \sigma(u_I)$, $a_x = \sigma_a(n)$, $b_x = \sigma_b(n)$

$$\nu dn/dt = f_0(n) + f_1(u_{Nu})$$

$f_0(\cdot)$ provides a biological switch (bistable), $f_1(\cdot)$ is a bounded, smooth, monotone increasing function.



Summary

Project more a modeling framework than definitive model: lots of a priori unknown parameters (a detailed parameter investigation hasn't been done), lots of chemistry missing, plaque core mechanics missing, multi-scale issues (temporal, spatial) investigated incompletely, hemodynamics and plaque geometry effects not included, etc.

Though we might determine conditions for cap thinning without EL contributions, simulations suggest EC regulatory dynamics may play a critical role in destabilizing plaque development.

Thank you for your attention

Figure: This is AfterMath, my cruising home