Modeling Wound Healing and Mass Transfer Effects in Low Temperature Plasma-Liquid Interactions

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Wound healing modeling: investigating ambient gas plasma treatment efficacy

Marat Orazov, Yukinori Sakiyama and David B Graves

\[
\frac{\partial c}{\partial t} = D_c \nabla^2 c - \left( \lambda_1 e + \lambda_2 \left( \frac{P(t)}{1 + \lambda_p e} \right) \right) \frac{c}{\lambda_3 + c} - \lambda_4 bc + \lambda_5 b.
\]
Coupling plasma and wound

But how to couple plasma model with tissue/wound model??
**Hypothesis:** Plasma speeds healing by killing bacteria and increasing $O_2$ availability

- 6-species PDEs in 1-D Cartesian coordinates
- modified parameters and additional terms for plasma treatment

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**Major pathways for wound healing**

- **bacteria**
- **oxygen**
- **chemo-attractants**
- **capillary tips**
- **blood vessels**
- **fibroblasts**
- **ECM**
Wound healing: governing equations (1)

- **Oxygen: \( c \)**

\[
\frac{\partial c}{\partial t} + \nabla \cdot (-D_c \nabla c) = \left( \frac{k_1}{1 + k_b e} + k_2 e \right) \frac{c}{k_3 + c} - k_4 b c + k_5 b
\]

consumption by bacteria

- **Chemoattractants : \( a \)**

\[
\frac{\partial a}{\partial t} + \nabla \cdot (-D_a \nabla a) = -k_6 a b - k_7 a + \frac{k_8 H(c - c_L) H(c_H - c)}{1 + e}
\]

production
Wound healing: governing equations (2)

- **Capillary tips:** $n$

\[ \frac{\partial n}{\partial t} + \nabla \cdot (-D_n \nabla n) = \nabla \cdot \left( \frac{-\kappa_n e n}{(1+e^2)(1+a)^2} \nabla a \right) + a(k_9 b + k_{10} n) - n(k_{11} n + k_{12} b) \]

- **Fibroblasts:** $f$

\[ \frac{\partial f}{\partial t} + \nabla \cdot (-D_f \nabla f) = \nabla \cdot \left( \frac{-\kappa_f f}{(1+a)^2} \nabla a \right) + \frac{k_{16} f c}{1+c} - \frac{k_{17} f^2}{(1+c)(1+e)} \]
Wound healing: governing equations (3)

- **Blood vessels:** $b$
  \[
  \frac{\partial b}{\partial t} = -\frac{\kappa n}{(1 + e^2)(1 + a)^2} \nabla a + k_{13} b(k_{14} e + k_{15} f - b)
  \]
  production by capillary tips

- **ECM:** $e$
  \[
  \frac{\partial e}{\partial t} = k_{18} f c(k_{19} c - e)
  \]
  deposition
Wound healing: infected, untreated wound

\[ t = 0.0 \text{ [week]} \]
Wound healing: effects of gas plasmas

Twice/day plasma treatment
• 99% direct reduction ($R$)
• 90 min doubling time ($k_p$)

Time dependent bacterial load

Oxygen: $c$

$$
\frac{\partial c}{\partial t} + \nabla \cdot (-D_c \nabla c) = -\left( \frac{k_1}{1 + k_b e} P_n + k_2 e \right) \frac{c}{k_3 + c} - k_4 b c + k_5 b
$$

$$
P_n = \frac{R P_{n-1} \exp(k_p t)}{1 + R P_{n-1} \{\exp(k_p t) - 1\}}
$$
Wound healing: *with plasma treatment*

- oxygen
- chemoattractants
- capillary tips
- blood vessels
- fibroblasts
- ECM

$t = 0.0$ [week]
Model prediction of plasma effect on wound healing for infected wound

Plasma effects on bacteria:
• 99% direct reduction
• 90 min doubling time
Wound Healing Model Remarks

1. Wound healing model is obviously relatively crude:
   - e.g. quantities treated as constant parameters are no doubt in reality varying with treatment
   - only a small fraction of relevant processes included in model

2. Effort was made to get consistent parameters from literature

3. Bacterial regrowth kinetics important

4. RONS from plasma directly kill wound surface bacteria but other effects probably important too:
   - protein/lipid reactions; gene expression
   - macrophages & inflammation processes…others…

Direct experimental measurements in vivo are crucial for future progress
Understanding how complex phenotypes arise from individual molecules and their interactions is a primary challenge in biology that computational approaches are poised to tackle. We report a whole-cell computational model of the life cycle of the human pathogen *Mycoplasma genitalium* that includes all of its molecular components and their interactions. An integrative approach to modeling that combines diverse mathematics enabled the simultaneous inclusion of fundamentally different cellular processes and experimental measurements. Our whole-cell model accounts for all annotated gene functions and was validated against a broad range of data. The model provides insights into many previously unobserved cellular behaviors, including in vivo rates of protein-DNA association and an inverse relationship between the durations of DNA replication initiation and replication. In addition, experimental analysis directed by model predictions identified previously undetected kinetic parameters and biological functions. We conclude that comprehensive whole-cell models can be used to facilitate biological discovery.
Figure 1. *M. genitalium* Whole-Cell Model Integrates 28 Submodels of Diverse Cellular Processes

(A) Diagram schematically depicts the 28 submodels as colored words—grouped by category as metabolic (orange), RNA (green), protein (blue), and DNA (red)—in the context of a single *M. genitalium* cell with its characteristic flask-like shape. Submodels are connected through common metabolites, RNA, protein, and the chromosome, which are depicted as orange, green, blue, and red arrows, respectively.
(B) The model integrates cellular function submodels through 16 cell variables. First, simulations are randomly initialized to the beginning of the cell cycle (left gray arrow). Next, for each 1 s time step (dark black arrows), the submodels retrieve the current values of the cellular variables, calculate their contributions to the temporal evolution of the cell variables, and update the values of the cellular variables. This is repeated thousands of times during the course of each simulation. For clarity, cell functions and variables are grouped into five physiologic categories: DNA (red), RNA (green), protein (blue), metabolite (orange), and other (black). Colored lines between the variables and submodels indicate the cell variables predicted by each submodel. The number of genes associated with each submodel is indicated in parentheses. Finally, simulations are terminated upon cell division when the septum diameter equals zero (right gray arrow).
What is our *Conceptual Model* for Plasma-Therapy?

RONS gas species: transfer into liquid *and* surface cells

(Liquid: water, salt, proteins, lipids; RONS react to form products)

Postulate: RONS-macromolecule products must induce RONS/redox stress in surface cells, followed by cell-cell communication.
Bystander effects as manifestation of intercellular communication of DNA damage and of the cellular oxidative status

Holger Klammer, Emil Mladenov, Fanghua Li, George Iliakis*

“Oxy-nitroso burst stress model”
Ozone Correlates with Antibacterial Effects from Indirect Air Dielectric Barrier Discharge Treatment of Water

Matthew J Pavlovich, Hung-Wen Chang, Yukinori Sakiyama, Douglas S Clark, David B Graves

![Diagram of dielectric barrier discharge treatment of water]

**Graphs:**
- **(a)**: Concentration of $\text{H}_2\text{O}_2$ vs. power density (W/cm$^2$)
- **(b)**: Concentration of $\text{NO}_2^-$, $\text{NO}_3^-$ vs. power density (W/cm$^2$)

**Chemical Compounds:**
- $\text{H}_2\text{O}_2$
- $\text{NO}_2^-$
- $\text{NO}_3^-$
- $\text{O}_3$
**E coli Suspension tests**

- 5 second contact time

**Buffered and unbuffered:** pH not important

**Power density (W/cm²):**
- 0.4 W/cm²
- 0.05 W/cm²

**Log reductions vs. Exposure time (s):**

- Vortexed
- NOT vortexed

**O₃ concentration (mg/l) vs. Power density (W/cm²):**
Nitric oxide delivery system for biological media

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\[
4 \text{NO} + \text{O}_2 + 2 \text{H}_2\text{O} \xrightarrow{k_1} 4 \text{NO}_2^- + 4 \text{H}^+
\]

\[
\frac{d\bar{C}_{\text{NO}}}{dt} = \frac{\chi_{\text{NO}} A_m k_m^{\text{NO}}}{V} \left( \alpha_{\text{NO} P_{m, \text{NO}}} - \bar{C}_{\text{NO}} \right) - \frac{A_t k_t^{\text{NO}}}{V} \bar{C}_{\text{NO}} - 4 \gamma k_1 \bar{C}_{\text{NO}}^2 \bar{C}_{\text{O}_2},
\]

\[
\frac{d\bar{C}_{\text{O}_2}}{dt} = \frac{\alpha_{\text{O}_2 P_{t, \text{O}_2}} - \bar{C}_{\text{O}_2}}{V} - \frac{\chi_{\text{O}_2} A_m k_m^{\text{O}_2}}{V} \bar{C}_{\text{O}_2} - \gamma k_1 \bar{C}_{\text{NO}}^2 \bar{C}_{\text{O}_2},
\]

\[
\frac{d\bar{C}_{\text{NO}_2^-}}{dt} = 4 \gamma k_1 \bar{C}_{\text{NO}}^2 \bar{C}_{\text{O}_2}.
\]

**Fig. 1.** Schematic of NO delivery system.
Simulated and observed NO and $O_2$ concentrations and NO rise times for simultaneous NO and $O_2$ delivery at various $O_2$ concentrations

<table>
<thead>
<tr>
<th>$P_{O_2}$ (mm Hg)</th>
<th>%O$_2$</th>
<th>%NO</th>
<th>$L$ (cm)</th>
<th>Predicted $\bar{C}_{NO}$ (µM)</th>
<th>Predicted $\bar{C}_{O_2}$ (µM)</th>
<th>Predicted $\tau_{NO}$ (min)</th>
<th>Observed $\bar{C}_{NO}$ (µM)</th>
<th>Observed $\bar{C}_{O_2}$ (µM)</th>
<th>Observed $\tau_{NO}$ (min)</th>
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<td>30%</td>
<td>10%</td>
<td>8.0</td>
<td>2.2</td>
<td>270</td>
<td>1.6</td>
<td>2.2</td>
<td>260</td>
<td>1.7</td>
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<tr>
<td>160</td>
<td>35%</td>
<td>10%</td>
<td>5.1</td>
<td>2.2</td>
<td>270</td>
<td>1.6</td>
<td>2.3</td>
<td>260</td>
<td>1.6</td>
</tr>
<tr>
<td>160</td>
<td>30%$^a$</td>
<td>1%</td>
<td>6.3</td>
<td>0.69</td>
<td>260</td>
<td>5.2</td>
<td>0.67</td>
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<td>1%</td>
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<td>0.86</td>
<td>160</td>
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<td>160</td>
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<td>1%</td>
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<td>13</td>
<td>1.3</td>
<td>43</td>
<td>11</td>
</tr>
</tbody>
</table>

$^a$ Indicates a specific concentration level.
A nitrogen dioxide delivery system for biological media

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\[
\begin{align*}
Q, \tilde{C}_{j,f} & \quad Q, \tilde{C}_{j} \\
\text{Headspace} & \quad \tilde{C}_{j} \\
\text{Liquid Film} & \quad C_{j}(x) \quad x = 0 \\
\text{Stirred Liquid} & \quad C_{j}(\delta) \quad x = \delta
\end{align*}
\]

\[
D_{NO_2} \frac{d^2 C_{NO_2}}{dx^2} = 2R_1 + k_3 C_{NO_2} C_{ABTS},
\]

\[
-D_{NO_2} \frac{dC_{NO_2}}{dx}(0) = \tilde{h}_{NO_2} \left( \tilde{C}_{NO_2} - \frac{C_{NO_2}(0)}{\Phi_{NO_2}} \right)
\]

\[
Q \left( \tilde{C}_{NO_2,f} - \tilde{C}_{NO_2} \right) = \tilde{h}_{NO_2} A \left( \tilde{C}_{NO_2} - \frac{C_{NO_2}(0)}{\Phi_{NO_2}} \right) + 2\tilde{V}\tilde{R}_1,
\]

\[
2 \text{NO}_2 \xrightarrow{k_1} \text{N}_2\text{O}_4,
\]

\[
\text{N}_2\text{O}_4 + \text{H}_2\text{O} \xrightarrow{k_2} \text{HNO}_2 + \text{HNO}_3,
\]

\[
\text{NO}_2 + \text{ABTS} \xrightarrow{k_3} \text{NO}_2^- + \text{ABTS}^+.
\]

\begin{figure}
\centering
\includegraphics[width=\textwidth]{no2_concentration.png}
\caption{NO\textsubscript{2} Concentration (mM) vs. NO\textsubscript{2} Mixture at 23 \textdegree C and 37 \textdegree C.}
\end{figure}
Prediction of nitric oxide concentrations in melanomas

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Note relatively short diffusion distances associated with tissue and tumors.
Note the short diffusion distances under physiological conditions.
Concluding Remarks:
future of coupled plasma-biological system models

Incorporating such complex models of a cell/tissue to plasma models is probably unrealistic, certainly any time soon.

However, coupling plasma models to *models of lipid bilayers* or to *model biofilms* or *model wounds* seems possible and realistic.

We should think about (model) experimental systems that can be intelligently coupled with plasma models!