Atmospheric pressure gas plasmas for biomedical applications

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1. Overview of Plasma medicine
   • brief review of “Plasma Medicine”
   • plasma sources for biomedicine
   • ROS/RNS in gas plasmas and biomedicine

2. Experimental projects
   • surface sterilization
   • plasma activated water (PAW)

3. Modeling projects
   • surface DBD
   • plasma needle and ring-shaped killing pattern

4. Concluding remarks
Overview: what is plasma?

• The fourth state of matter (solid, liquid, gas, and plasmas…)
• Ionized gas containing negative and positive charged particles
Overview: history of gas plasmas in biomedicine

1893 A. d’Arsonval: compatibility of HF with nerve and muscle

1926 Bovie knife: the first clinical use of an electrosurgical device

1940 Hyfrecator (Birtcher Co): low power and no ground pad

1995 APC (ERBE GmbH): Ar plasma for endoscopic surgery

Coblation (Arthrocare Co): discharge in saline solution

1996 M. Laroussi: E. coli sterilization (He DBD)

2003 E. Stoffles: non-destructive cell handling (He plasma needle)

2007 G. Fridman: in vitro cancer cell treatment (Air DBD)

2010 G. Isbary: clinical trial for wound healing (MW Ar plasma)
Overview: potential applications

- antibacterial resistance
- nosocomial infection
- chronic wound
- pandemic flu
- dental cavity
- hand hygiene
- cancer
- drug delivery
Overview: cancer treatment

**In vitro study**

**In vivo animal study**
Overview: dental applications

**Tooth bleaching**


**Root canal treatment**

Overview: wound healing (1)

R. A. Bryant, et al., *Acute and Chronic Wounds* (Mosby, Missouri, 2006).

**Inflammatory phase**
- ~48 hours
- bacteria sterilization/debris removal
- blood coagulation

**Proliferative phase**
- 2~10 days
- blood vessels generation
- collagen deposition from fibroblasts

**Remodeling phase**
- 1 year
- tissue reorganization/realignment
- apoptosis of unnecessary cells
Overview: wound healing (2)

- **Plasma health care project**
- lead by G. Morfill at Max-Planck Institute
- 19 PhDs, 11 MDs
- Germany, UK, Russia, Japan, USA

**Phase-I clinical study**

J. Heinlin, *JDDG* 8 (2010) 968

**Microwave Ar plasma torch**

Before treatment

After 11 treatments
Overview: complex nature of “plasma medicine”

prokaryotes (bacteria)

Gram positive: e.g. *B. subtilis*
(vegetative cell or spore)
Gram negative: e.g. *E. coli*

eukaryotes (animal or plant)

planktonic
biofilm
*in vivo*
Devices: three major categories

**Plasma jet/needle**
- rare gas flow
- localized (<1cm²)

**Air DBD**
- static air
- large area possible (>100cm²)

**Solution plasma**
- saline solution, water, etc.
- localized (~1mm)
Devices: rare gas jet/needle

**LF He plasma jet (bullet)**


- **Condition:** 1~5 kV, 1-10 kHz, ~1 W/cm², rare gas, near/at room temperature
- **Agents:** electron, ion, E-field, neutral, photon
- **Property:** remote access localized treatment

**RF He plasma needle**


- **He flow**
- **Tungsten needle**
- **Glass tube**
- **Metal plate**
- **Sample**
Devices: direct/indirect air DBD

**Direct DBD (volume DBD)**

- Metal plate → HV
- Dielectric
- Sample

**Indirect DBD (surface DBD)**

- Metal plate → HV
- Dielectric
- SS mesh
- Sample

<table>
<thead>
<tr>
<th>conditions</th>
<th>~5 kV, 1-10 kHz, ~1 W/cm², air, near/at room temperature</th>
</tr>
</thead>
<tbody>
<tr>
<td>agents</td>
<td>electron, ion, E-field, neutral, photon</td>
</tr>
<tr>
<td>property</td>
<td>direct contact to sample</td>
</tr>
<tr>
<td></td>
<td>neutral, photon</td>
</tr>
<tr>
<td></td>
<td>stability and scalability</td>
</tr>
</tbody>
</table>
Devices: solution plasmas

**Coblation (ArthroCare, Corp)**

http://www.arthrocare.com


**PEAK (PEAK surgical, Inc)**

http://www.peaksurgical.com/

e.g. A. Vankov, *J. Appl. Phys.* **101** (2007) 124701

<table>
<thead>
<tr>
<th>medium conditions</th>
<th>normal saline</th>
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<tbody>
<tr>
<td>purpose</td>
<td></td>
</tr>
<tr>
<td>~200 $V_{rms}$, 100 kHz</td>
<td>10-100 ns, 4-6 kV, ~0.5 mJ/pulse</td>
</tr>
<tr>
<td>coagulation and ablation</td>
<td>localized incision</td>
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</table>
## ROS/RNS: possible biomedical effects

<table>
<thead>
<tr>
<th></th>
<th>physics</th>
<th>chemistry</th>
<th>electrostatic</th>
</tr>
</thead>
<tbody>
<tr>
<td>electronsions</td>
<td>1-10 eV</td>
<td>~ 1 eV</td>
<td>negative</td>
</tr>
<tr>
<td>radicals</td>
<td>~ 1 eV</td>
<td>ROS/RNS</td>
<td>positive/negative</td>
</tr>
<tr>
<td>photons</td>
<td>4-12 eV (UVC)</td>
<td>ROS/RNS</td>
<td></td>
</tr>
<tr>
<td>electric field</td>
<td></td>
<td>ROS/RNS</td>
<td>10^6-10^7 V/m</td>
</tr>
<tr>
<td>effects</td>
<td>• bond breaking (e.g., DNA) • sputtering</td>
<td>• oxidation (e.g., O, OH) • signaling (e.g., NO)</td>
<td>• membrane disruption (~10^9 V/m) • stimulation</td>
</tr>
</tbody>
</table>
**ROS/RNS: widely recognized agents**

- **ROS** (reactive oxygen species): O, O$_2^*$, O$_3$, O$_2^-$, OH, H$_2$O$_2$
- **RNS** (reactive nitrogen species): NO, NO$_2$, ONOO$^-$

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Abbas, *Cellular and Molecular Immunology* (Elsevier, 2005).
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Surface sterilization: experimental setup

- **Frequency**: 10 kHz
- **Voltage**: 10 kV $V_{p-k}$
- **Power consumption**: ~5 W
- **Distance to sample**: 1 mm
- **Plasma on**: 0.5-5 min

- **Diameter**: 50 mm
- **Bacteria**: E. coli K12
- **Material**: SS, silicone, pig skin

**Procedure**

1. Rinse
2. Diluted specimen
3. Over night
4. Count #CFU

**Materials**

- Power supply
- Copper electrode
- SS mesh electrode
- Teflon block
- Glass plate
- E. coli
- Agar plate
- Material: SS, silicone, pig skin
Surface sterilization: different surfaces

log reduction = \log_{10} \left( \frac{\text{initial number}}{\text{number of survivors}} \right)
Surface sterilization: direct and indirect

- Direct
- Indirect

Bar chart showing log reduction vs. exposure time for direct and indirect sterilization methods. The chart includes data points at 30, 60, 120, 180, and 300 seconds, with error bars indicating variability.
PAW = plasma activated water


- **frequency**: 10 kHz
- **voltage**: 10 kV $V_{pkpk}$
- **power consumption**: ~5 W
- **distance to sample**: ~40 mm
- **plasma on**: 20 min
- **volume**: 10 ml
- **medium**: distilled water
- **storage period**: 0-7 days
PAW: chemical properties

**Chemical Properties**

- **pH**: Initial pH: 6.2
- **Chemical Compounds**:
  - Hydrogen peroxide ($H_2O_2$)
  - Nitrite ($HNO_2$)
  - Nitrate ($HNO_3$)

**Graphs**:

- **Left Graph**:
  - pH vs. time [hour]
  - pH remains relatively stable with initial pH: 6.2

- **Right Graph**:
  - Nitrite/nitrate vs. hydrogen peroxide [mM]
  - Nitrite and Nitrate concentrations over time [hour]
PAW: antimicrobial activity (1)

\[
\text{log reduction} = \log_{10} \left( \frac{\text{initial number}}{\text{number of survivors}} \right)
\]

short-lived species?
PAW: antimicrobial activity

log reduction = \log_{10} \left( \frac{\text{initial number}}{\text{number of survivors}} \right)

PAW spray?
Outline

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SDBD: model description (1)

- Computational domain
  - $d_{pls}$
  - $d_{gas}$
  - $\Gamma_{pg}$

- SMD
  - Zero flux

- Neutral reactor
  - Zero flux
  - Treated surface
  - e.g. metal surface with diluted bacteria

- Measured current properties

- Mesh electrode (grounded)
- Insulator
- Copper electrode (powered)

- Microdischarges

- Electrons
- Ions
- Neutrals

- $E$
- 10 ns
SDBD: model description (2)

For charged particles:
\[ \Gamma_{pg} = 0 \]

For neutrals:
\[ \Gamma_{pg} = \frac{D_{gas}(n_{pls} - n_{gas})}{d_{gas}} \]

- **SMD Neutral reactor**
  - Computational domain
  - \( d_{pls} \) and \( d_{gas} \)
  - \( \Gamma_{pg} \)

- **Neutral reactor**
  - \( \frac{\partial n_{pls}}{\partial t} = \sum_j R_j - \frac{1}{d_{pls}} \Gamma_{pg} \)

- **Treated surface**
  - \( \frac{\partial n_{gas}}{\partial t} = \sum_j R_j + \frac{1}{d_{gas}} \Gamma_{pg} \)

- **Example surface**
  - E.g. metal surface with diluted bacteria
  - \( \Gamma_{pg} \)

- **Particle Types**
  - Electrons
  - Ions
  - Neutrons

- **Time Scale**
  - \( E \) and 10 ns
SDBD: model description (3)

- **Computational domain**
  - SMD
  - Neutral reactor
  - e.g. metal surface with diluted bacteria

- **Zero flux**
  - $d_{pls}$
  - $d_{gas}$
  - $\Gamma_{pg}$

- **Electrical field**
  - $E$
  - 10 ns

**Negative particles**: $e^-, O^-, O_2^-, O_3^-, O_4^-, H^-, OH^-, NO^-, N_2O^-, NO_2^-, NO_3^-

**Positive particles**: $N^+, N_2^+, N_3^+, N_4^+, O^+, O_2^+, O_4^+, NO^+, N_2O^+, NO_2^+, H^+$, $H_2^+, H_3^+, OH^+, H_2O^+, H_3O^+$

**Neutrals**: $N$, $N^*$, $N_2$, $N_2^*$, $N_2^{**}$, $O$, $O^*$, $O_2$, $O_2^*$, $O_3$, $NO$, $N_2O$, $NO_2$, $NO_3$, $N_2O_5$, $H$, $H_2$, $OH$, $H_2O$, $HO_2$, $H_2O_2$, $HNO$, $HNO_2$, $HNO_3$
SDBD: multiple time-scale phenomena

- Electron impact reactions
- Charge transfer, ion recombination
- Neutral reactions
- Applied voltage period
- Gas diffusion
- Exposure time

Simulation procedure:

- SMD (electrons, ions, neutrals)
- SMD (neutrals)
- Cycle-averaged reaction rates
- SMD (neutrals)
- Neutral reactor (neutrals)
SDBD: dynamics of charged particles

- Power density: 0.1 W/cm²
- Frequency: 10 kHz
- Gap distance: 1 mm
- Humidity: 0% (dry)

**Positive ions**
- $N_x^+$
- $O_x^+$
- $N_xO_y^+$

**Negative ions**
- $N_xO_y^-$
- $O_x^-$

**Time vs. Density**
- Density [m⁻³]
- Time [s]

**Graphs**
- Positive ions graph
- Negative ions graph
SDBD: cycle-averaged neutral density

Power density: 0.1 W/cm²
Frequency: 10 kHz
Gap distance: 1 mm
Humidity: 0% (dry)
SDBD: comparison between dry and humid air

- Power density: 0.1 W/cm²
- Frequency: 10 kHz
- Gap distance: 1 mm
- Humidity: 0% (dry), 30% (humid)

Graph showing the concentration of various chemical species such as hydrogen (H₂), oxygen (O₂), nitrogen oxide (NO), ozone (O₃), nitrous oxide (N₂O), nitric oxide (NO₂), nitric acid (HNO₃), and others, with different colors representing dry and 30% humidity conditions.
RF plasma needle: ring-shaped killing profile


RF (13.56 MHz) power supply

- Glass tube (Ø 4 mm)
- Needle (Ø 0.5 mm)

0.3 m/s light intensity 1.0 m/s

5 mm

Alive

Inactivated
RF plasma needle: fully coupled plasma-flow model

Neutral Gas flow (He, Air)

\[ \nabla \cdot (\rho \mathbf{u}) = 0, \quad \nabla \cdot (\rho \omega_{\text{air}} \mathbf{u} - \rho D \nabla \omega_{\text{air}}) = 0 \] (mass conservation)

\[ \nabla \cdot (\rho \mathbf{u} \mathbf{u}_i) = -\nabla p - \nabla \cdot \mathbf{\tau} + \sum q_i n_i \mathbf{E} \] (momentum conservation)

\[ \nabla \cdot \left( -\lambda \nabla T + u c_p T \right) = \Phi + \sum q_i \Gamma_i \mathbf{E} + Q_{el} \] (energy conservation)

Plasma dynamics

\[ \frac{\partial n_i}{\partial t} + \nabla \cdot \mathbf{\Gamma}_i = S_i \] (mass conservation)

\[ \mathbf{\Gamma}_i = \text{sgn}(q_i) n_i \mu_i \mathbf{E} - D_i \nabla n_i + n_i \mathbf{u} \] (drift-diffusion)

\[ \frac{\partial (n_e \varepsilon)}{\partial t} + \nabla \cdot \left( \frac{5}{3} \varepsilon \mathbf{\Gamma}_e - \frac{5}{3} n_e D_e \nabla \varepsilon \right) = -\mathbf{\Gamma}_e \cdot \mathbf{E} - Q \] (electron energy)

\[ \varepsilon_0 \nabla \cdot \mathbf{E} = \sum q_i n_i \] (Poisson’s equation)

RF plasma needle: off-axis ionization peak

Y. Sakyiama et al, Plasma Sources Sci. Technol. 18 (2009) 025022
RF plasma needle: off-axis ionization peak (2)

Predicted emission pattern

CCD image with optical filters

dark  bright

He+, He2+  O2+, N2+

He, He+, O2, N2

He* 706 nm

N2+ 397 nm
RF plasma needle: from emission to killing pattern

Experimental observation

emission profile

Killing pattern

alive inactivated

alive inactivated

Modeling approach

Plasma chemistry simulation
- 46 species
- 214 reactions
- Flux distribution of species

RF plasma needle: flux on treated surface

on-axis

off-axis

ROs/RNS

$\text{flux} \left[ 10^{19} \text{ m}^{-2} \text{ s}^{-1} \right]$

$\text{flux} \left[ 10^{19} \text{ m}^{-2} \text{ s}^{-1} \right]$

$\text{He}_2^+$ $\text{N}_2^+$ $\text{O}_2^+$

$\text{H}_2\text{O}^+$ $\text{N}_2^+$ $\text{O}_2^+$ $\text{H}$ $\text{N}$ $\text{OH}$ $\text{NO}^+$ $\text{O}_2^+$
RF plasma needle: TALIF measurement of O atom

TALIF (two photon absorbed laser induced fluorescence)
- Collaboration with Ruhr-Universitat Bochum (Germany)
- Absolute density of O atom

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Concluding Remarks

1. Plasma-medicine is promising field and has huge potentials and impact.

2. Plasma-generated ROS/RNS would play significant roles in plasma medicine.

3. Neutral gas flow and air chemistry are keys to control plasma-generated ROS/RNS.
Acknowledgements

Dr. M. Traylor (Graves group)
M. Pavlovich, S. Karim, and Z. Chen (Graves group)

Prof. D. Clark (UC Berkeley, US)
Prof. H. Nikaido (UC Berkeley, US)

Dr. T. Shimizu and Prof. G. Morfill (Max-Planck Institute, DE)
Dr. V. Schulz-von der Gathen (Ruhr University Bochum, DE)
Dr. J. Jarrige and Prof. M. Laroussi (Old Dominion University, US)
Prof. J. Goree (University of Iowa, US)
Dr. E. Stoffels
Related publications

Reviews for plasma medicine


Plasma-biomaterial interaction in Graves group