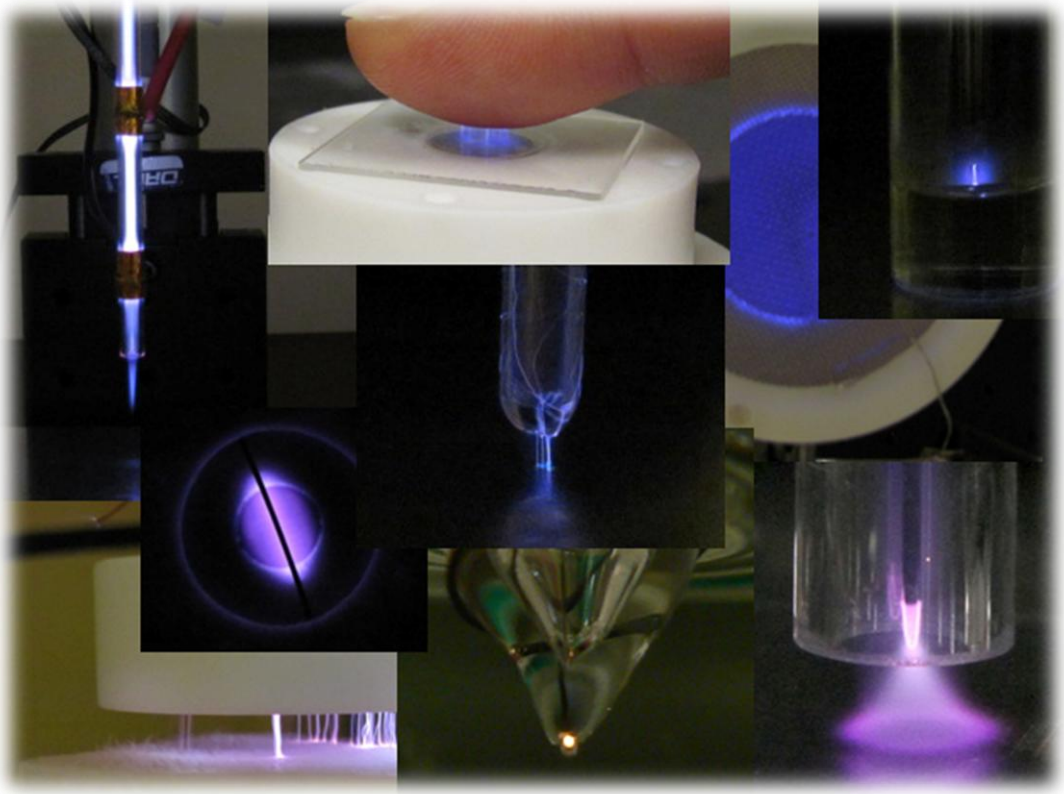

Effect of Discharge Parameters on Ambient-Gas Plasma Disinfection

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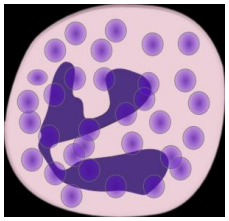
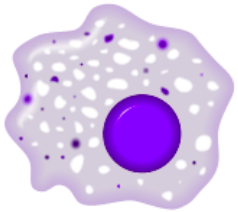
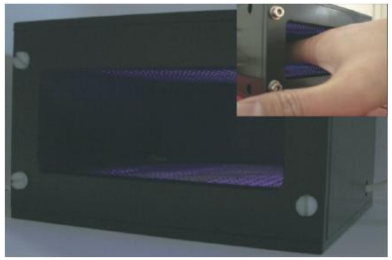
Overview of Ambient-Gas Plasma Disinfection

- Room-temperature, atmospheric-pressure air plasma
- Parallels innate-immune ROS/RNS
- Brief history:





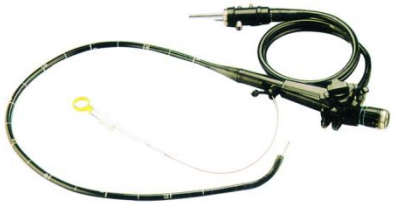
  - Early 20th century: *thermal* plasma for wound treatment
  - 1996: *E. coli* disinfection in solution (M. Laroussi)
  - 1998: *E. coli* disinfection on surfaces (J. R. Roth)
  - 2007: Use of an “indirect” discharge (J. Engemann)
  - 2009: Hand disinfection prototype (G. Morfill)



An Application of AGP Disinfection: Hospital Infection Control

- Nosocomial infections: staff as vectors, compromised immune systems, enclosed space
- 5-10% of patients; $12,000 and 5-7 day increase per patient; 100,000 deaths/year
- Sensitive devices, chronic wounds, antibiotic-resistant bacteria

+ Scalable
+ Painless
+ No tissue damage?
+ Cheap
+ Convenient
+ Broad activity
+ Circumvents evolved resistance



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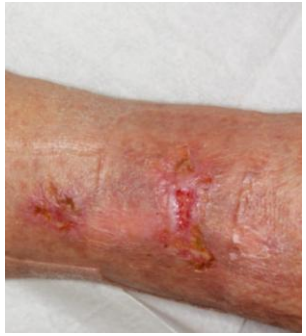

Isbary, *Br. J. Dermatol.* (2010)

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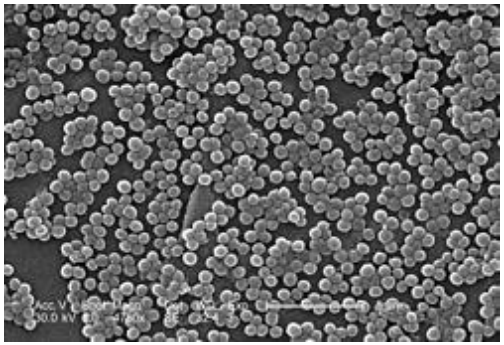
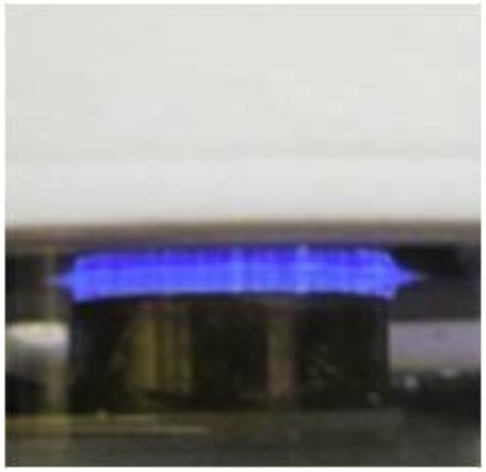


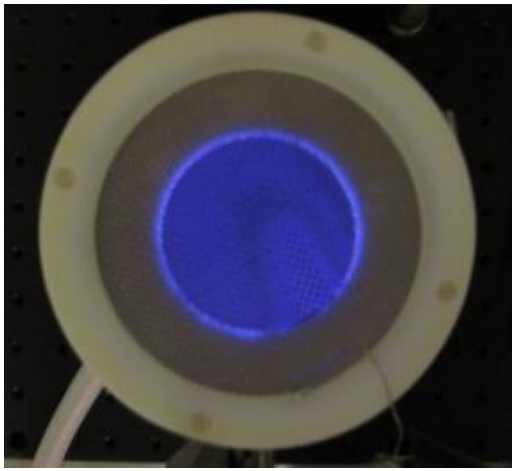
Centers for Disease Control and Prevention
Public Health Image Library

Ambient-Gas Plasma Generation: DBD Device



Direct mode

-Neutrals, ions
-Electric fields
-Filamentous discharge



Indirect mode

-Neutrals only
-No direct electrical effect
-Diffuse discharge

Parameter	Operating Range
Voltage, V	3.3 – 10 kV
Frequency, *f*	4 – 10 kHz
Power density, P	0.5 – 1.5 W/cm^2
Gap distance	1 mm
Treatment time	30 – 300 s

- How do the discharge mode and parameters affect the antimicrobial activity?
- Which conditions are most promising for clinical use?

Surface Disinfection Experimental Setup

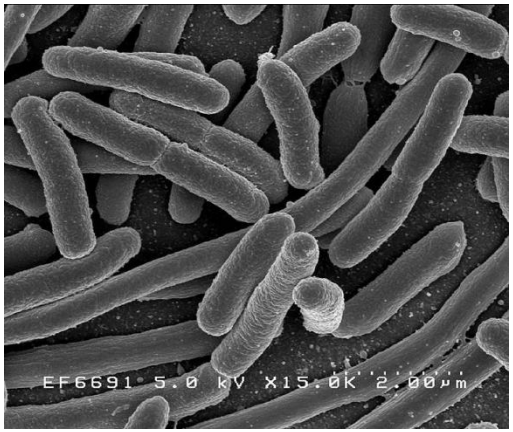
Procedure

1. Contaminate a surface with bacteria
2. Treat the surface with plasma
3. Remove the remaining bacteria by vortexing
4. Dilute, plate, and count colonies

Variables

- Discharge mode
- Plasma parameters (P, V, *f*)
- Type of surface (steel, rubber, skin)
- Treatment time

Model organism



Escherichia coli K12

NIAID, Biodefense Image Library

Metric: Log reductions

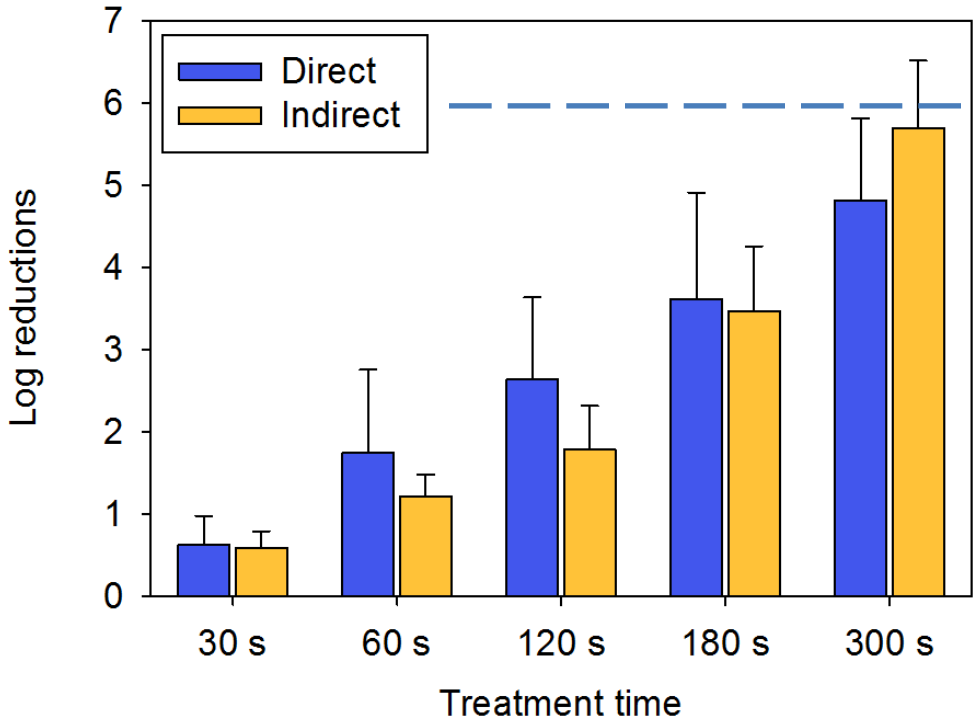
$$\log\left(\frac{N_0}{N}\right)$$

N_0: Number of viable colonies in an untreated sample

N: Number of surviving colonies in a treated sample

If a hand sanitizer kills "99.9%" of bacteria, that's a 3-log reduction

Indirect Mode Has Practical Advantages Over Direct Mode

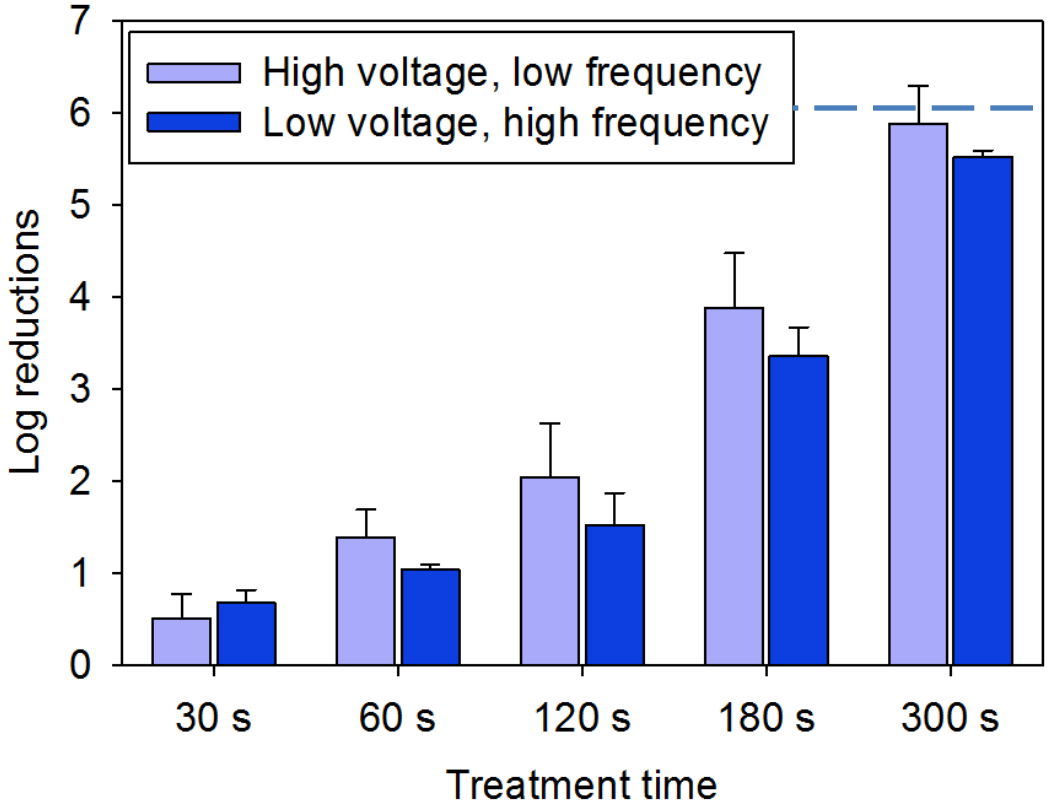


Stainless steel (similar for all surfaces)

Measurement	Units	Indirect mode	Direct mode
Power range	W/cm^2	0.74 – 0.76	0.43 – 0.97
Power standard deviation	W/cm^2	0.01	0.18
Antimicrobial standard deviation	log reduction	0.52	0.93

- Similar killing in both modes
- Greater reproducibility, uniformity, and accuracy for indirect mode
- Indirect mode is more attractive for therapeutic applications

Plasma Parameters Have Little Effect on Antimicrobial Activity



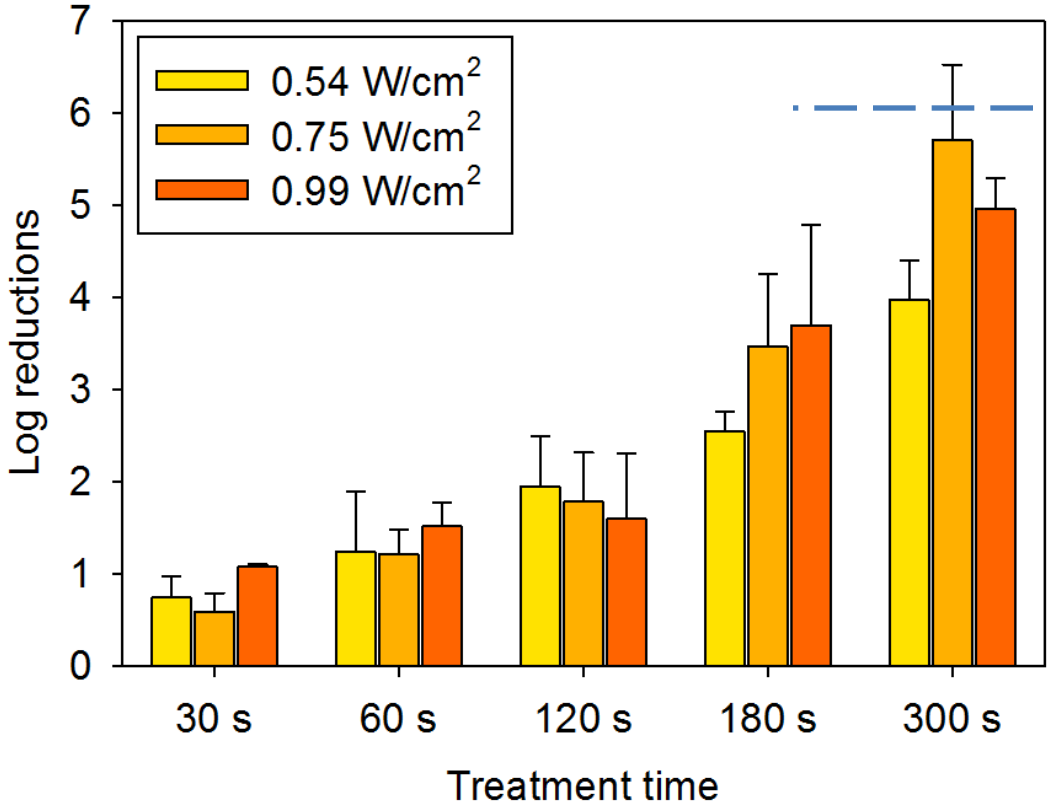
Stainless steel
(similar for all surfaces)

Indirect mode

P = 0.75 W/cm^2 ± 5%

- Similar killing at different V, *f* for a given P
- Similar killing for P within ± 30%
- Contact time (and therefore biological interaction) is important

Plasma Parameters Have Little Effect on Antimicrobial Activity



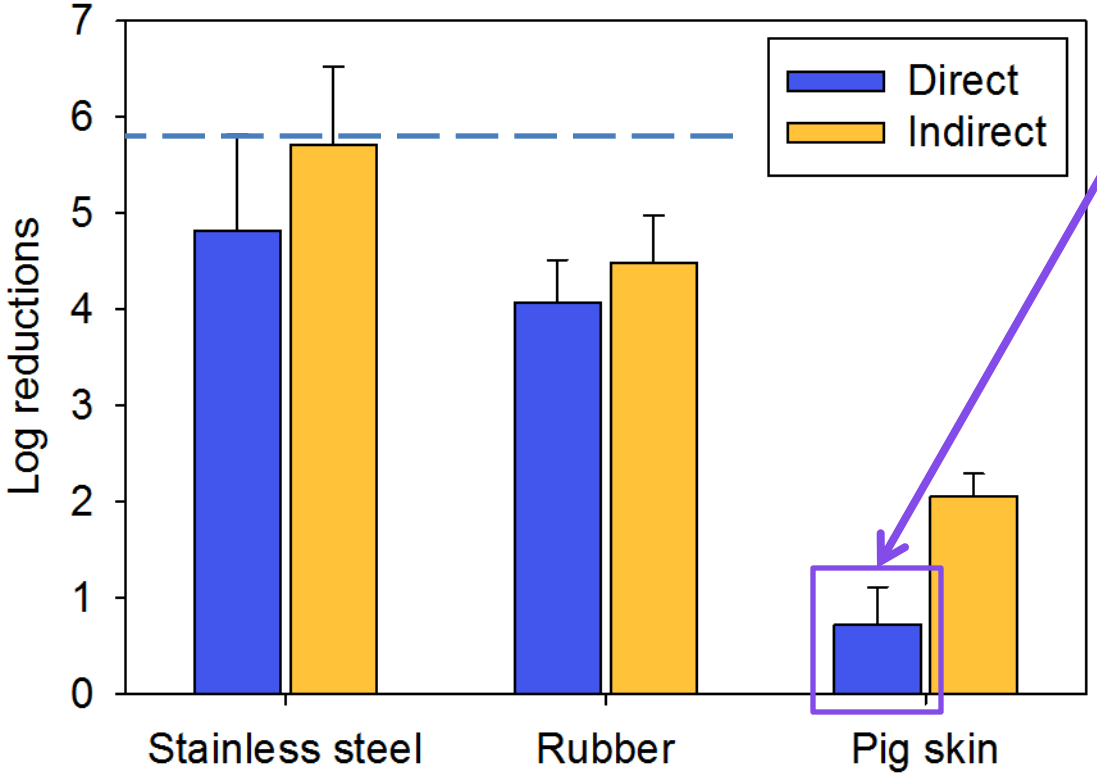
Stainless steel
(similar for all surfaces)

Indirect mode

Manipulated V, *f* to change P

- Similar killing at different V, *f* for a given P
- Similar killing for P within ± 30%
- Contact time (and therefore biological interaction) is important

Plasma Disinfects Non-Biological Surfaces Better Than Biological Surfaces



Skin burns due to discharge nonuniformity in direct mode

P = 0.80 W/cm^2 ± 10%

300 s treatment time

- Similar performance on non-biological surfaces
- Significantly worse on pig skin
- Why? (Porosity, roughness, layers?)

Conclusions

- Ambient-gas plasma presents a novel and potentially useful strategy for infection control
- Indirect-mode discharges have practical advantages over direct-mode
- Some surfaces are more amenable to disinfection than others
- Contact time is the most important controllable parameter for determining the antimicrobial effect

Remaining Questions and Next Steps

- What properties of a surface determine its susceptibility to plasma disinfection?
  - Can we correlate roughness, porosity, etc. to antimicrobial effect?
- What is plasma treatment actually doing to kill bacteria?
  - Can we detect biomolecule modifications with redox proteomic/lipidomic techniques?
  - Can we measure gene expression with microarrays?

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