Plasma Medicine:

Physical and Biological Mechanisms of Plasma Interaction with Living Tissue

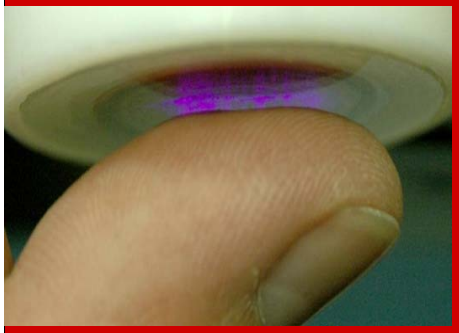
1. Plasma Medicine & Sterilization
2. Discharges in Plasma Medicine
3. Bio-Active Plasma Components
4. Effects on Cell Membrane
5. Intracellular Biochemistry
6. Selectivity of Plasma Treatment
7. Plasma Wound Healing
8. Blood Coagulation
9. Skin Diseases
10. Cancer Treatment
11. Gastrointestinal Diseases

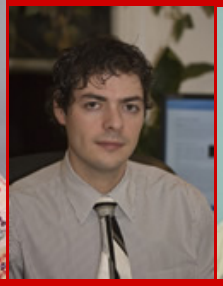
1. Plasma Fuel Conversion

3. Plasma in Aerospace and Material Engineering

3. Plasma Medicine and Biology

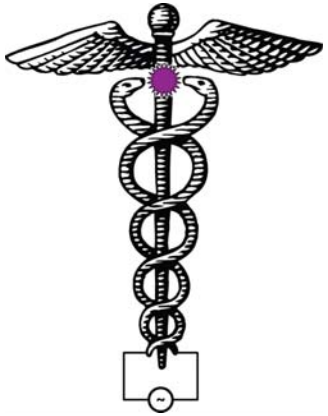
DREXEL PLASMA INSTITUTE

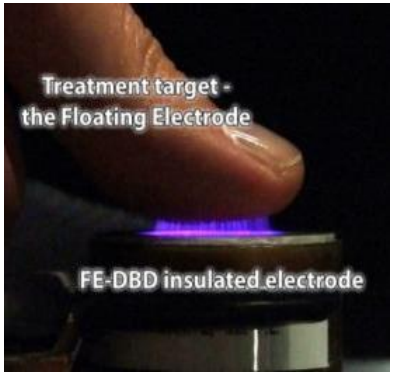




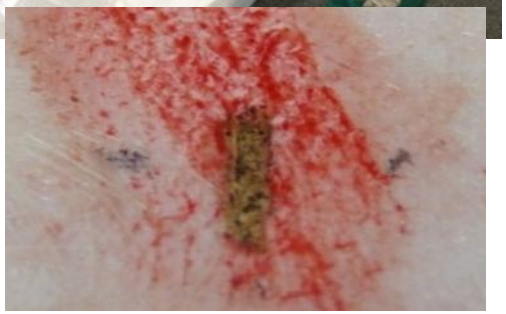
- Critical Mass of People >25 faculty, 6 colleges
- Critical Mass of Projects (>$2.5 M annual budget)
- Critical Mass of Equipment

Plasma Medicine

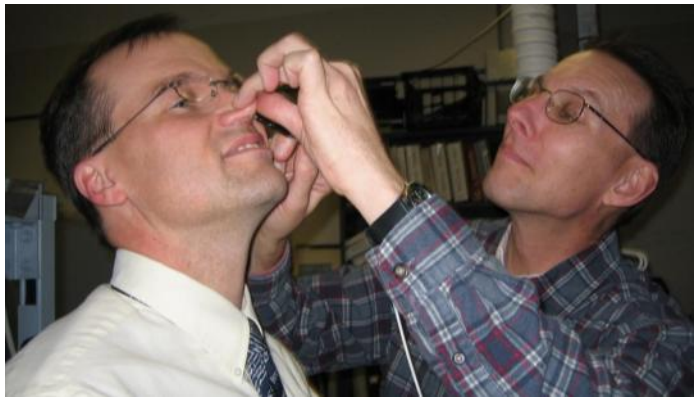


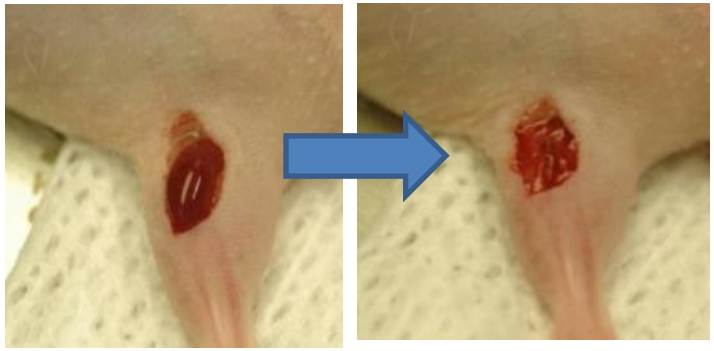


Wound & skin sterilization



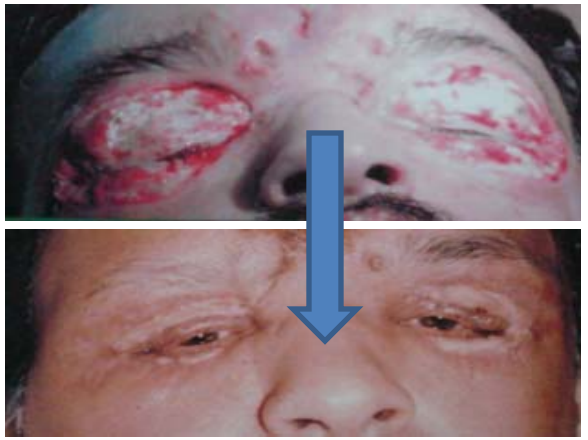
Blood coagulation

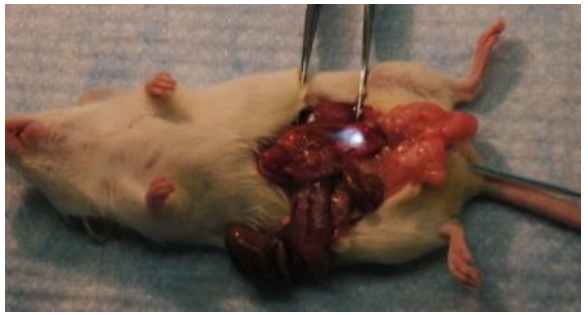




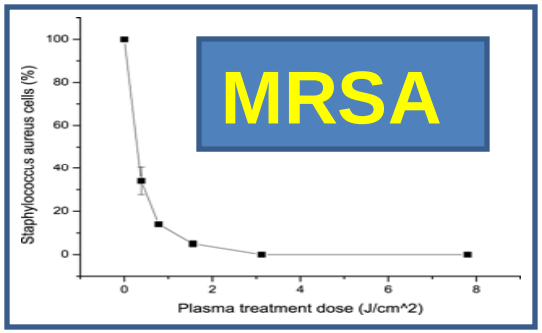
Healing of wounds & diseases

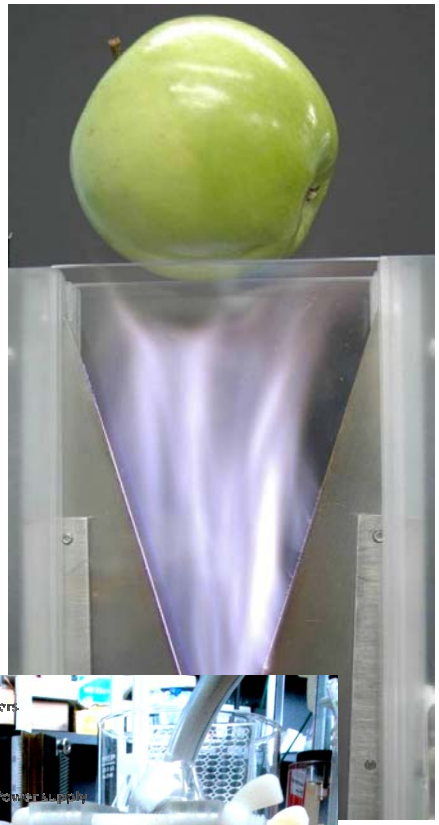
Skin infections and ulcers, gastrointestinal diseases (colitis, etc), cancer, Leishmaniasis

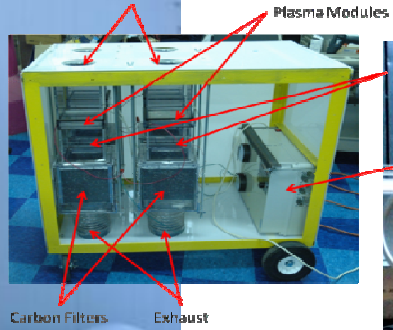


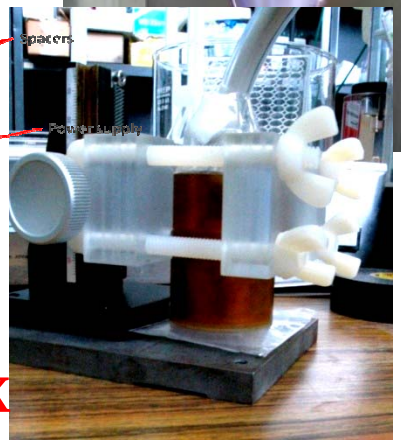


Plasma Sterilization

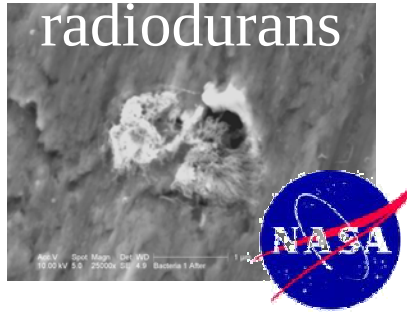






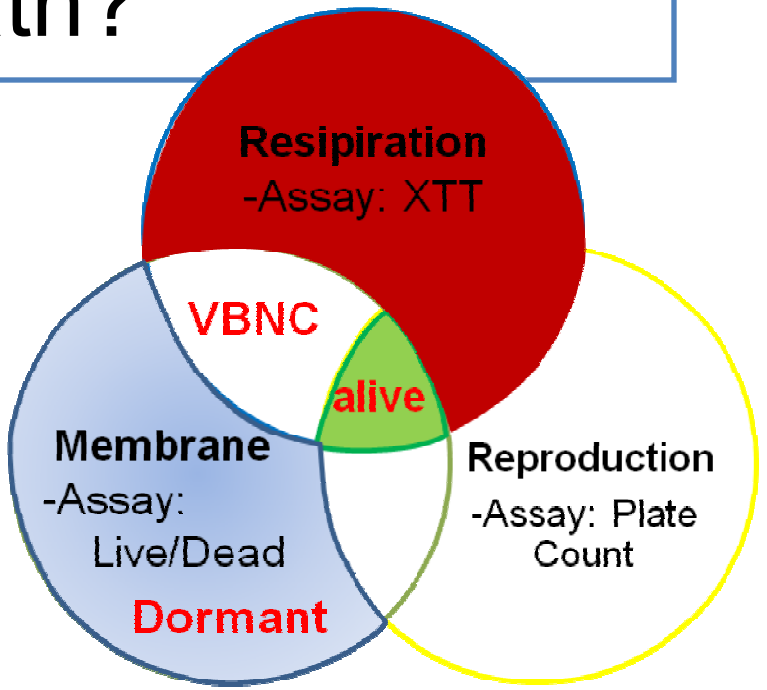


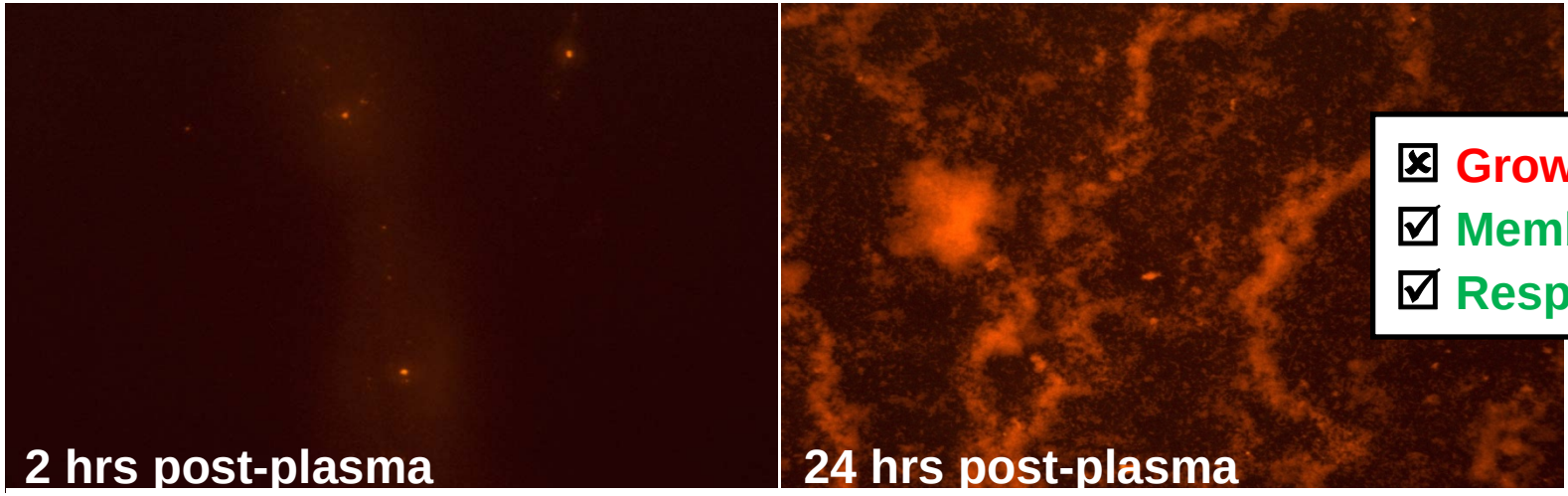
Anthrax



What is Death?

- A microorganism is traditionally “dead” when it does not exhibit ALL the following: homeostasis; response to stimuli; metabolism; growth; and reproduction.
- Viable but nonculturable (VBNC): “A cell which is metabolically active, while being incapable of undergoing the cellular division required for growth in or on a medium normally supporting growth of that cell”.
- The **VBNC** state is dangerous for pathogenic bacteria as stressed bacteria are **more virulent** than well-fed bacteria.

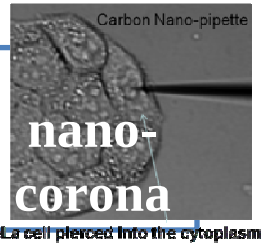




Respiration from few initial survivors (left) increase respiration after 24 hours (right) but remain non-culturable

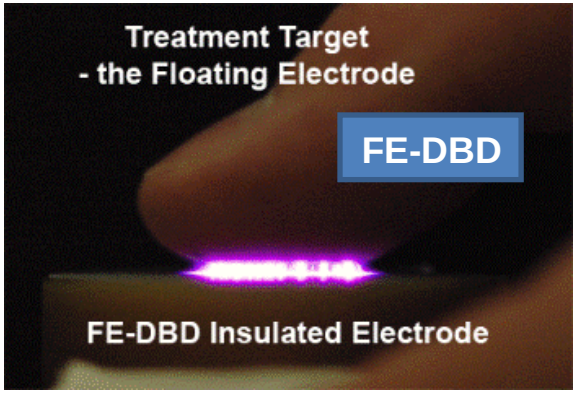

Treatment Target
- the Floating Electrode
FE-DBD Insulated Electrode
Plasma Medicine:
Healing vs. Killing

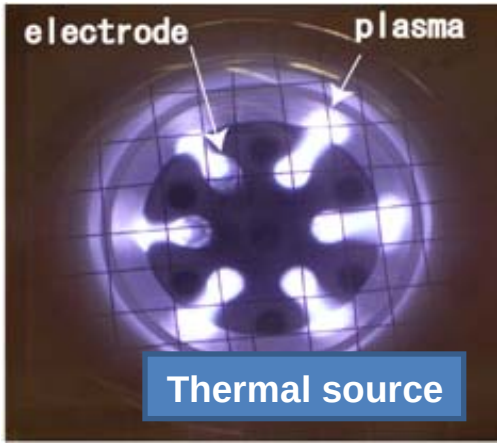
Bio-Active Plasma Components:
Classification of Discharges

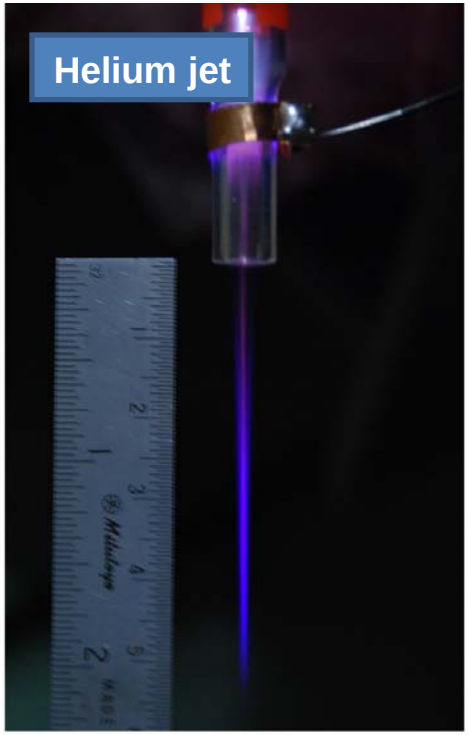


$$e + N_2 \rightarrow N_2^+ + 2e$$
$$e + O_2 + M \rightarrow O_2^- + M$$
$$e + O_2 \rightarrow 2O + O_2 + e$$
$$O + O_2 + M \rightarrow O_3 + M$$
$$e + O_2 \rightarrow O_2(^1\Delta_g) + e$$
$$N_2^+ + H_2O \rightarrow H_2O^+ + N_2$$
$$H_2O^+ + H_2O \rightarrow H_3O^+ + OH$$
$$OH + OH + M \rightarrow H_2O_2 + M$$

$$[NO] \rightsquigarrow UV$$
$$e + N_2 \rightarrow N + N + e$$
$$N + O_2 \rightarrow NO + O$$
$$N + NO \rightarrow N_2 + O$$

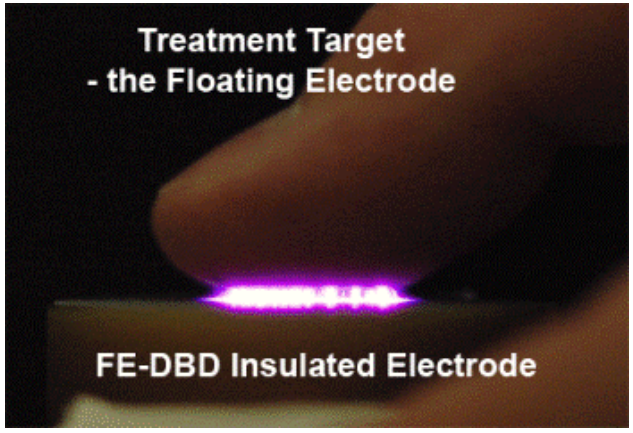


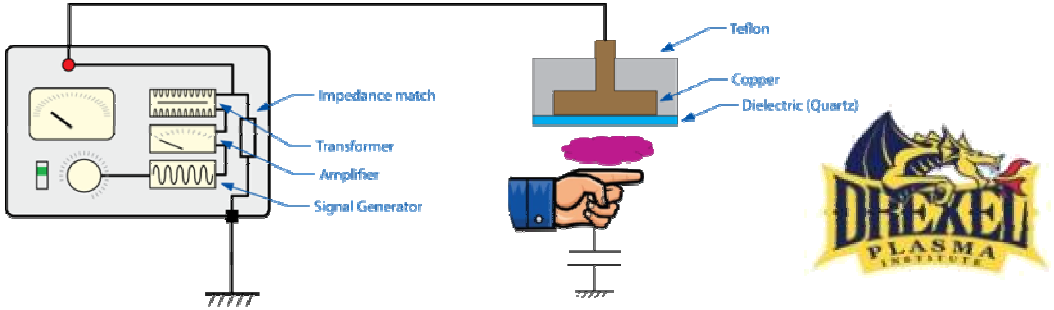




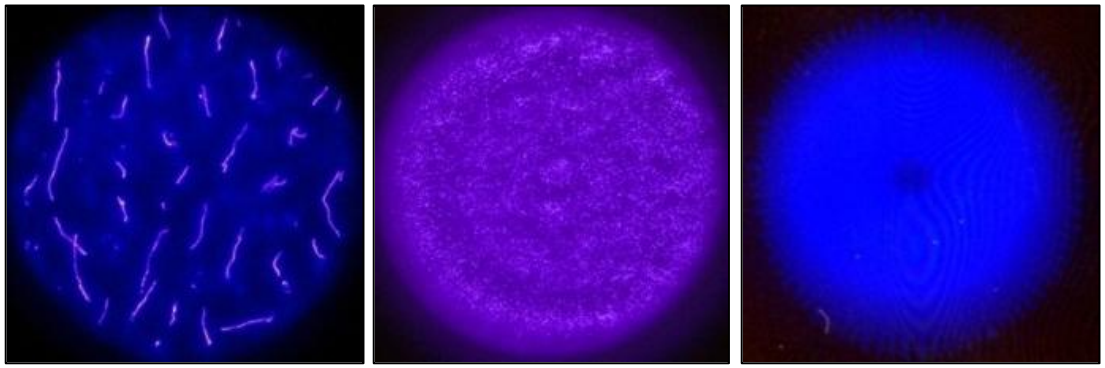
Bio-Active Plasma Components: **FE-DBD**

$$e + N_2 \rightarrow N_2^+ + 2e$$
$$e + O_2 + M \rightarrow O_2^- + M$$
$$e + O_2 \rightarrow 2O + O_2 + e$$
$$O + O_2 + M \rightarrow O_3 + M$$
$$e + O_2 \rightarrow O_2(^1\Delta_g) + e$$
$$N_2^+ + H_2O \rightarrow H_2O^+ + N_2$$
$$H_2O^+ + H_2O \rightarrow H_3O^+ + OH$$
$$OH + OH + M \rightarrow H_2O_2 + M$$





$$[NO] \rightsquigarrow UV$$
$$e + N_2 \rightarrow N + N + e$$
$$N + O_2 \rightarrow NO + O$$
$$N + NO \rightarrow N_2 + O$$



Continuous wave | Microsecond pulse | Nanosecond pulse

Bio-Active Plasma Components: **PHD**

$$e + N_2 \rightarrow N_2^+ + 2e$$
$$e + O_2 + M \rightarrow O_2^- + M$$
$$e + O_2 \rightarrow 2O + O_2 + e$$
$$O + O_2 + M \rightarrow O_3 + M$$
$$e + O_2 \rightarrow O_2(^1\Delta_g) + e$$
$$N_2^+ + H_2O \rightarrow H_2O^+ + N_2$$
$$H_2O^+ + H_2O \rightarrow H_3O^+ + OH$$
$$OH + OH + M \rightarrow H_2O_2 + M$$

$$[NO] \rightsquigarrow UV$$
$$e + N_2 \rightarrow N + N + e$$
$$N + O_2 \rightarrow NO + O$$
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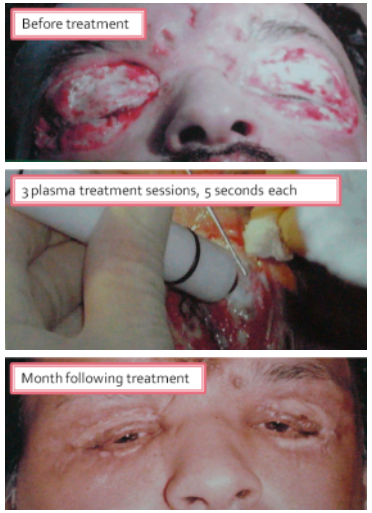
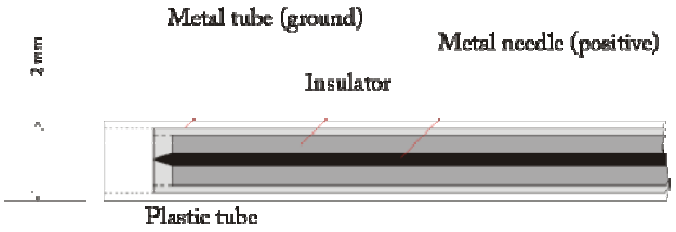
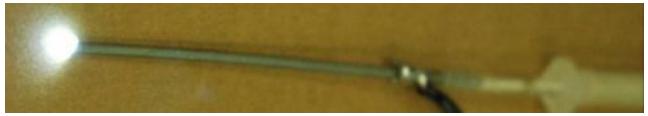
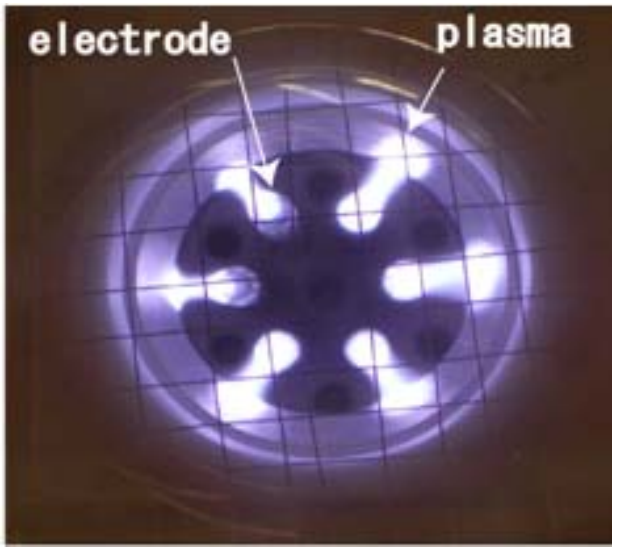


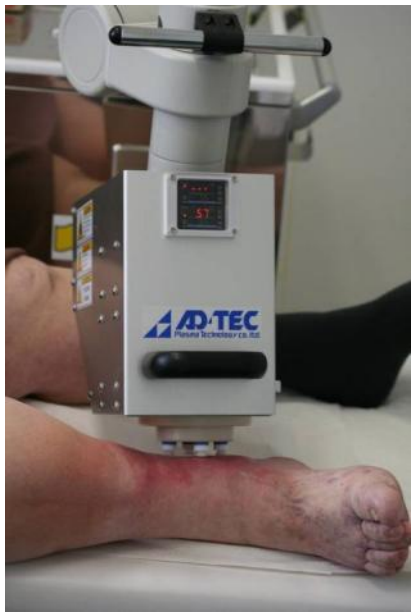


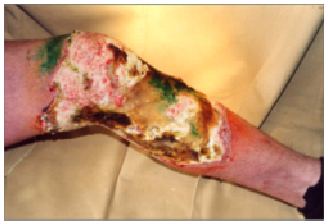


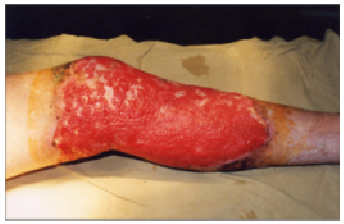
Bio-Active Plasma Components: **Plasma NO**

$$e + N_2 \rightarrow N_2^+ + 2e$$
$$e + O_2 + M \rightarrow O_2^- + M$$
$$e + O_2 \rightarrow 2O + O_2 + e$$
$$O + O_2 + M \rightarrow O_3 + M$$
$$e + O_2 \rightarrow O_2(^1\Delta_g) + e$$
$$N_2^+ + H_2O \rightarrow H_2O^+ + N_2$$
$$H_2O^+ + H_2O \rightarrow H_3O^+ + OH$$
$$OH + OH + M \rightarrow H_2O_2 + M$$









Before treatment

After 7 days of NO-therapy (5 sessions)

$$[NO] \rightsquigarrow UV$$
$$e + N_2 \rightarrow N + N + e$$
$$N + O_2 \rightarrow NO + O$$
$$N + NO \rightarrow N_2 + O$$

Plasma Technologies, Inc.

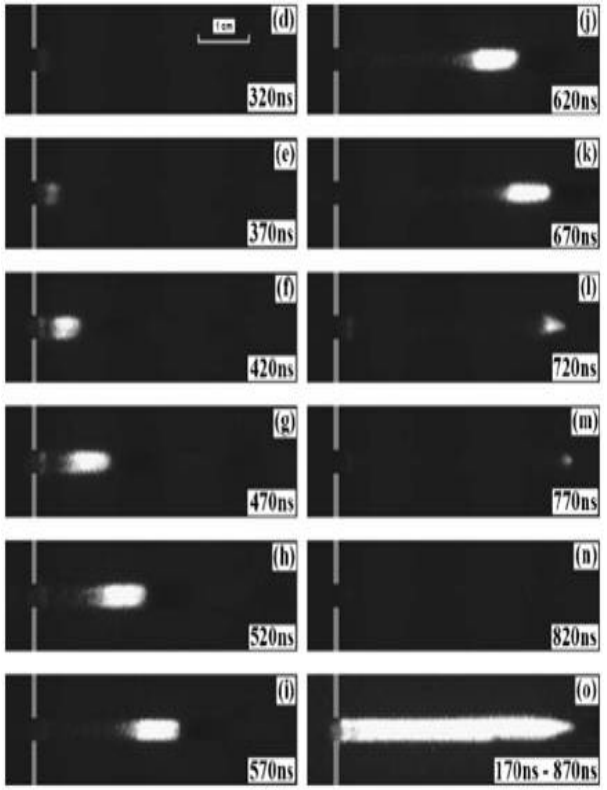
MAX-PLANCK-GESELLSCHAFT

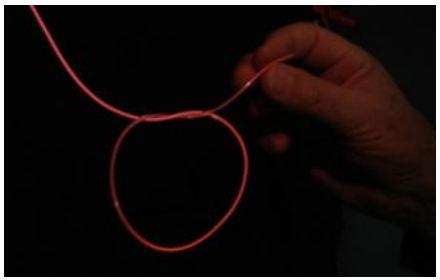
städtisches Klinikum München

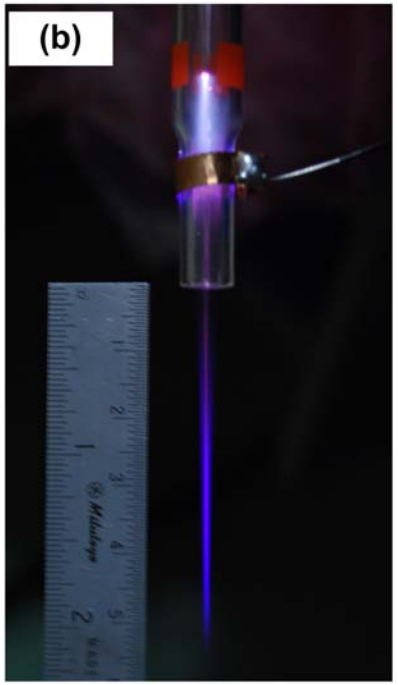
Bio-Active Plasma Components: **Helium Jets, Bullets**

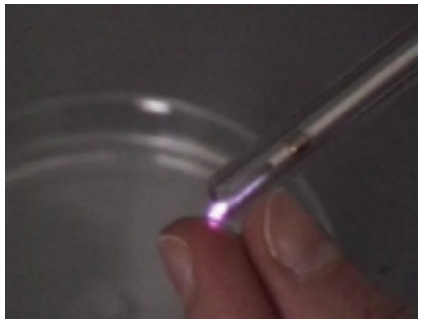
$$e + N_2 \rightarrow N_2^+ + 2e$$
$$e + O_2 + M \rightarrow O_2^- + M$$
$$e + O_2 \rightarrow 2O + O_2 + e$$
$$O + O_2 + M \rightarrow O_3 + M$$
$$e + O_2 \rightarrow O_2(^1\Delta_g) + e$$
$$N_2^+ + H_2O \rightarrow H_2O^+ + N_2$$
$$H_2O^+ + H_2O \rightarrow H_3O^+ + OH$$
$$OH + OH + M \rightarrow H_2O_2 + M$$

$$[NO] \rightsquigarrow UV$$
$$e + N_2 \rightarrow N + N + e$$
$$N + O_2 \rightarrow NO + O$$
$$N + NO \rightarrow N_2 + O$$









OLD DOMINION UNIVERSITY

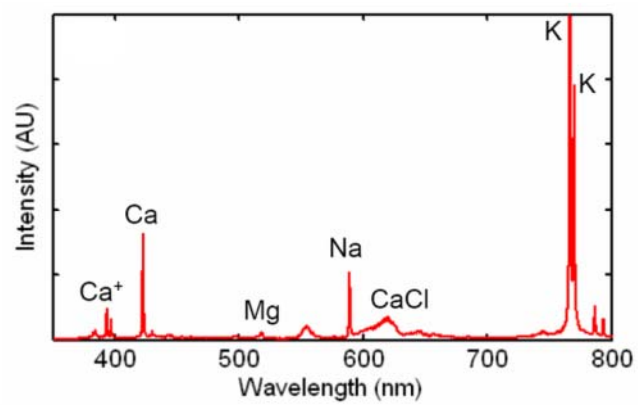
TU/e

Bio-Active Plasma Components:
Surface Plasma / Plasma Blanket

$$e + N_2 \rightarrow N_2^+ + 2e$$
$$e + O_2 + M \rightarrow O_2^- + M$$
$$e + O_2 \rightarrow 2O + O_2 + e$$
$$O + O_2 + M \rightarrow O_3 + M$$
$$e + O_2 \rightarrow O_2(^1\Delta_g) + e$$
$$N_2^+ + H_2O \rightarrow H_2O^+ + N_2$$
$$H_2O^+ + H_2O \rightarrow H_3O^+ + OH$$
$$OH + OH + M \rightarrow H_2O_2 + M$$

$$[NO] \rightsquigarrow UV$$
$$e + N_2 \rightarrow N + N + e$$
$$N + O_2 \rightarrow NO + O$$
$$N + NO \rightarrow N_2 + O$$

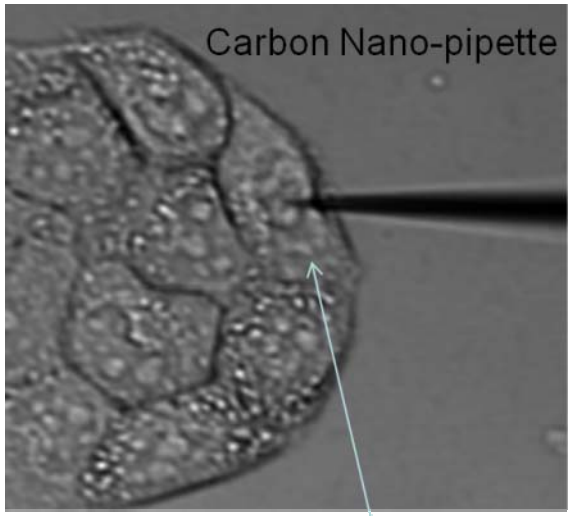
Nano-scale nano-pulse plasma inside liquids



Detected Species:

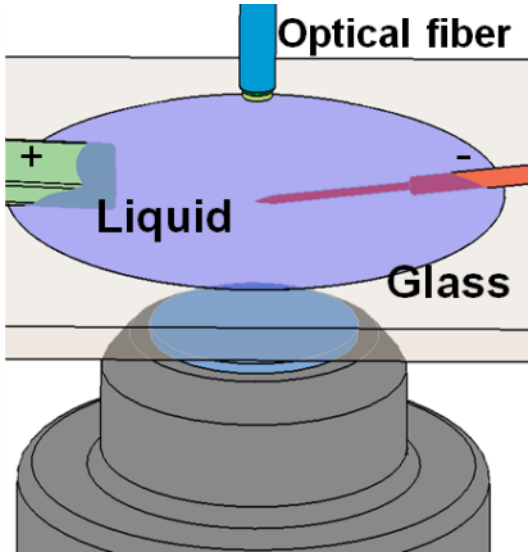
K. Na. Ca. Mg. Ca^+ Pb. Ga. Zn. W. P

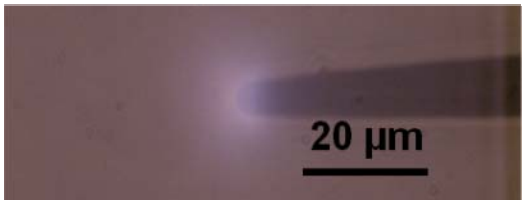
O. H. OH C_2. CH



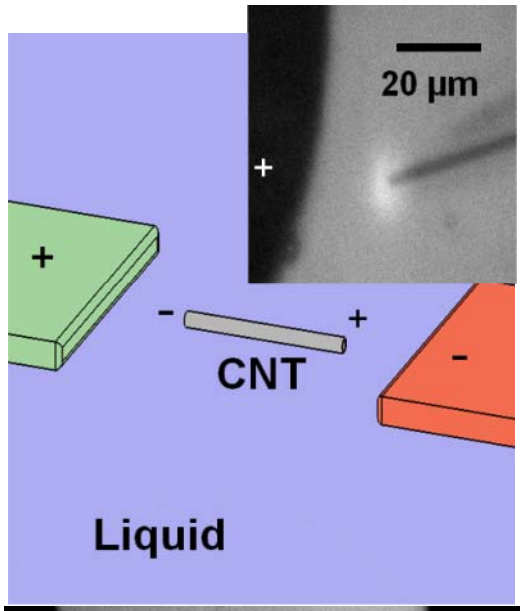
HeLa cell pierced into the cytoplasm

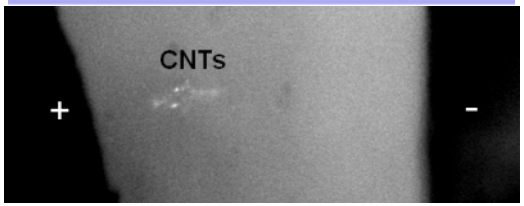
Direct Discharges





Remote Discharges



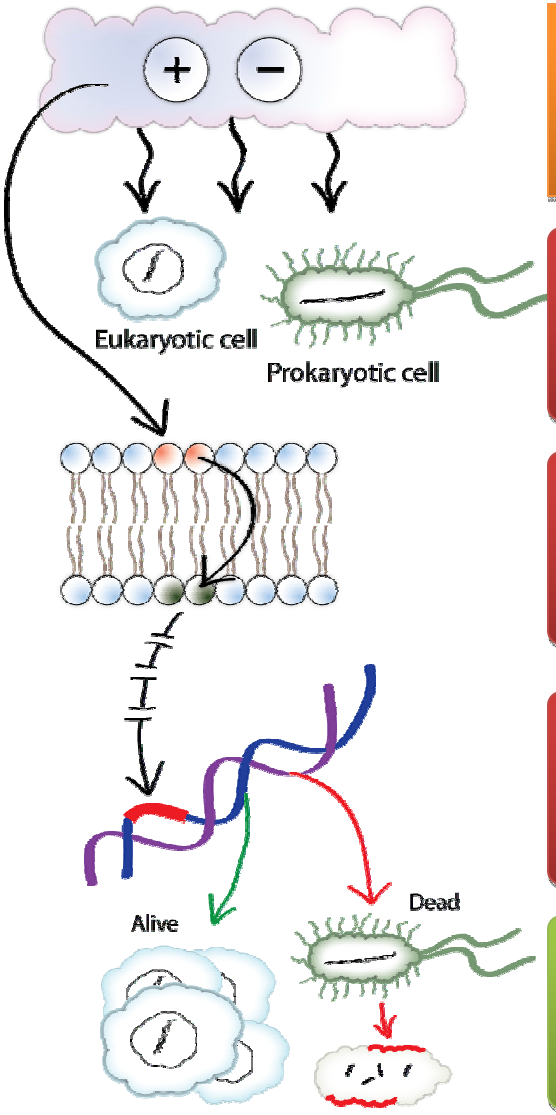


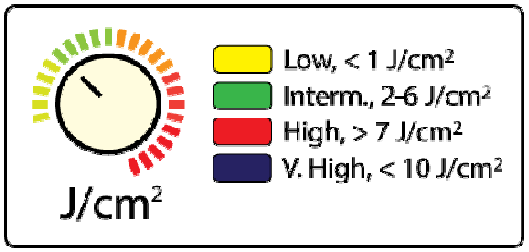
Aditya Chitre, David Staack, Z. Orynbayeva, J. Clifford, Y. Gogotsi, A. Fridman, G. Friedman

TEXAS A&M UNIVERSITY

DREXEL PLASMA INSTITUTE

Direct Plasma - Cell Interactions for both, sterilization and healing





A

Plasma ions play key role

Role of positive and negative ions is related to catalysis of peroxidation processes in presence of reactive oxygen species.

B

Primary target is cell membrane

Peroxidation of phospholipids and polysaccharides is catalyzed by charged species. Water is required and effect of surrounding medium is important.

C

Intracellular biochemistry

Lipid layer peroxidation leads to intracellular pathway activation (i.e. malondialdehyde, MDA formation) which alters DNA structure.

D

Selectivity

Tissue sterilization without visible or microscopic (histological) damage; selective development of apoptosis in cancer cells; difference in metabolism of peroxidation products; etc.

E

Effect of dose

Low doses: sterilization, blood coagulation.
Intermediate doses: cell proliferation, growth factor release, apoptosis in cancers, etc. *High doses*: normal cell death.
Very high doses: necrosis.

A

Plasma ions play key role

Role of positive and negative ions is related to catalysis of peroxidation processes in presence of reactive oxygen species.

For mammalian and bacterial cells; for healing and for sterilization.

Air Plasma Major Bio-Active Components

$$e + N_2 \rightarrow N_2^+ + 2e$$
$$e + O_2 + M \rightarrow O_2^- + M$$
$$e + O_2 \rightarrow 2O + O_2 + e$$
$$O + O_2 + M \rightarrow O_3 + M$$
$$e + O_2 \rightarrow O_2(^1\Delta_g) + e$$
$$N_2^+ + H_2O \rightarrow H_2O^+ + N_2$$
$$H_2O^+ + H_2O \rightarrow H_3O^+ + OH$$
$$OH + OH + M \rightarrow H_2O_2 + M$$

$$[NO] \rightsquigarrow UV$$
$$e + N_2 \rightarrow N + N + e$$
$$N + O_2 \rightarrow NO + O$$
$$N + NO \rightarrow N_2 + O$$

Air Plasma Parameters

Voltage: 5 – 35+ kV (p2p)

Frequency: 0.1 kHz – 100 kHz

Rise times: 1 – 3,000 V/ns

Gas temperature: ~ 300 K

[e^-] conc.: 10^8 – 10^{11} cm^{-3}

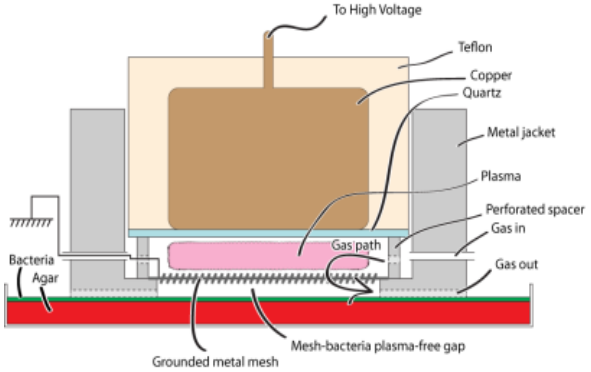
[e^-] temp.: 1 – 2 eV

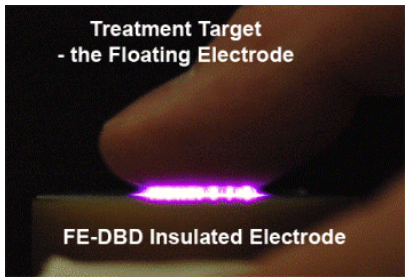
Ion conc.: 10^9 – 10^{12} cm^{-3}

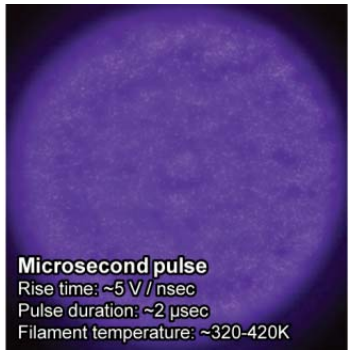
Ion temp.: 1/40 eV (~300 K)

UV (180-300 nm) & VUV (110-180 nm)

10^{-9}-10^{-8} W/cm^2 total UV surface power







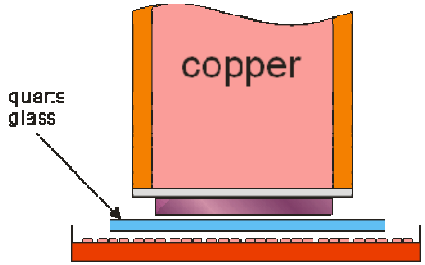
A

Plasma ions play key role

Role of positive and negative ions is related to catalysis of peroxidation processes in presence of reactive oxygen species.

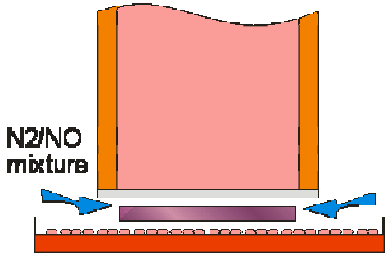
No significant effect of UV, Ozone, H_2O_2.

Quartz glass to leave only UV

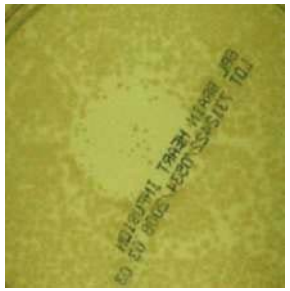


No sterilization

N_2/NO mixture to destroy Ozone



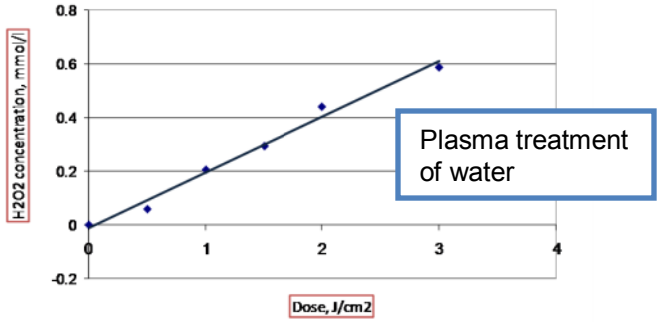
Air



N2/NO

Sterilization efficiency does not depend on presence of ozone

H_2O_2



Concentration of commercially available H2O2 used:

%	mol/L	Result
50	20	100% sterile
5	2	
0.5	200 mmol/L	Some (~ 50%)
$5*10^{-2}$	20 mmol/L	No sterilization
$5*10^{-3}$	2 mmol/L	
$5*10^{-4}$	0.2 mmol/L	

Plasma produces 0.2 to 0.8 mM solutions of H2O2 in water, which is not effective for sterilization (>200 mM is required)

E. coli, skin flora: strep, staph, yeast

~10^7 CFU/ml, 700 µl – on agar, dry for ~ 30 min;

Treatment: up to 3 J/cm^2

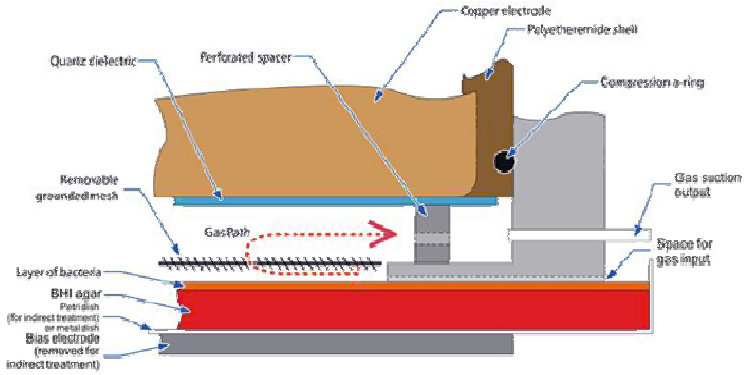
Dobrynin, D., et al., 48th ICAAC & 46th IDSA, October 25-28, 2008, Washington, DC, USA.

A

Plasma ions play key role

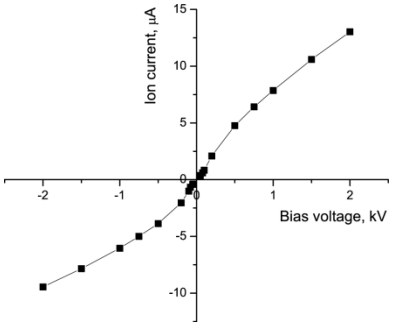
Role of **positive and negative ions** is related to catalysis of peroxidation processes in presence of reactive oxygen species.

Effect is chemical.

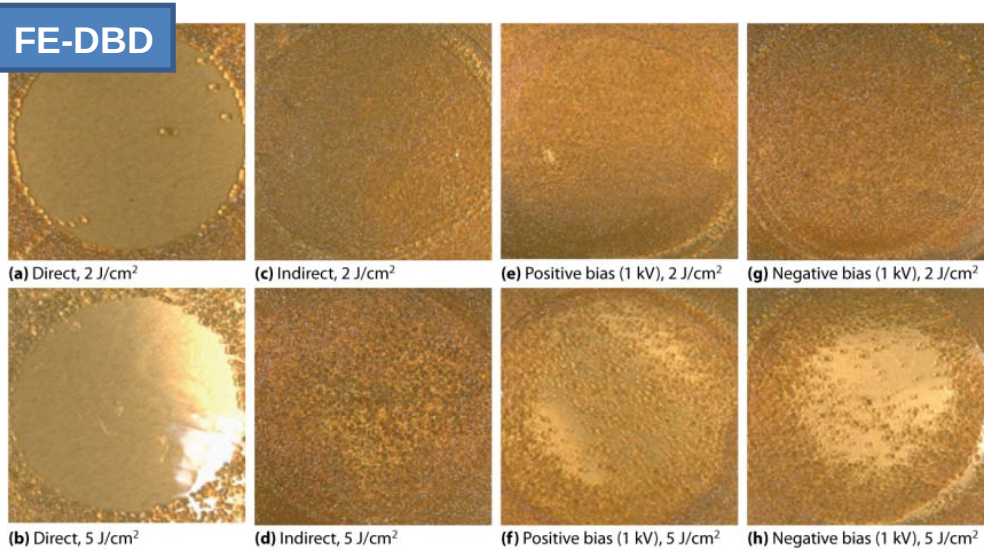


Ion current: ~ 1 – 10 μA

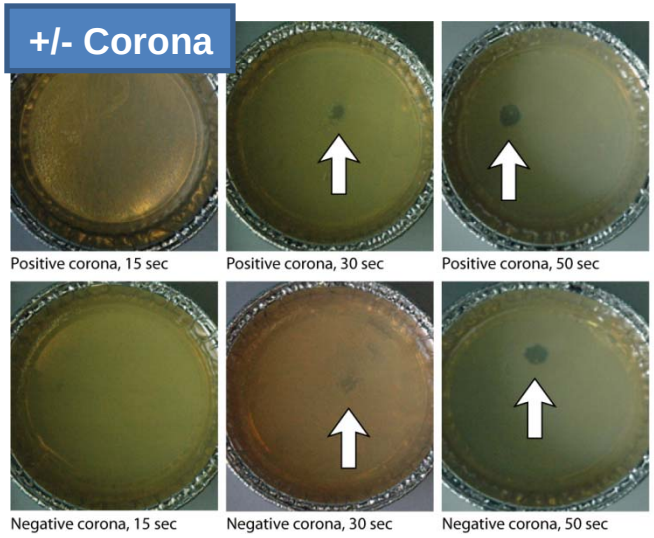
Both positive and negative ions are effective in sterilization

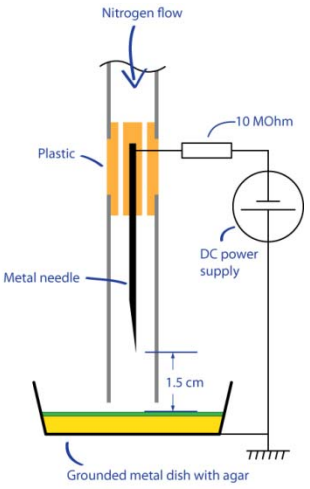


FE-DBD



+/- Corona





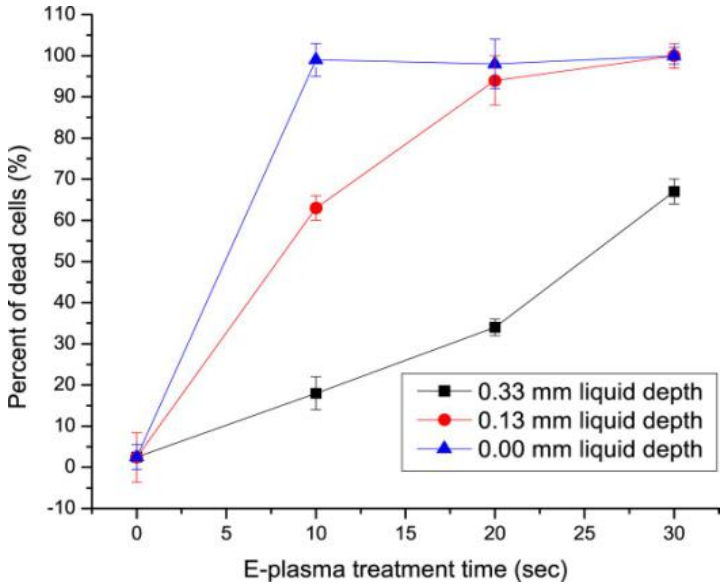
Dobrynin, D., et al., 48th ICAAC & 46th IDSA, October 25-28, 2008, Washington, DC, USA.

A

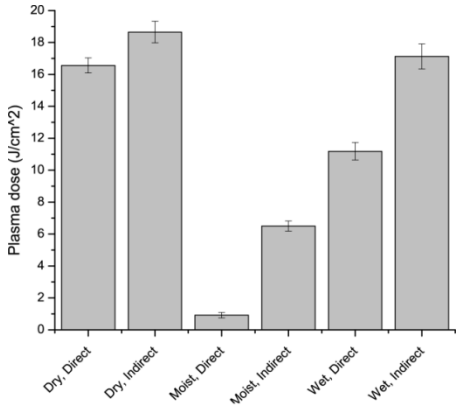
Plasma ions play key role

Role of positive and negative ions is related to catalysis of peroxidation processes **in presence of reactive oxygen species**.

Effect of O_2 and water.



gas \ method	Jet, 1 lpm	Jet, 10 lpm	Direct
O_2	10^5 reduction 120-140 s	10^5 reduction 120-140 s	$>10^6$ reduction 15 s
air	10^5 reduction 120-140 s	10^5 reduction 120-140 s	$>10^6$ reduction 15 s
N_2	little	little	little
Argon	little	little	little
He	little	little	little
N_2/NO	little	little	little



Plasma treatment penetrated liquid but the effect is significantly reduced for cells and bacteria. Best effect is achieved on moist surfaces where there is no liquid water.

Oxygen enhances direct plasma effect.

Absence or overabundance H_2O suppresses the effect, optimal water layer is ~10 μm

G. Fridman, et al., *Plasma Process. Polym., 2007, 4, 370-375*.

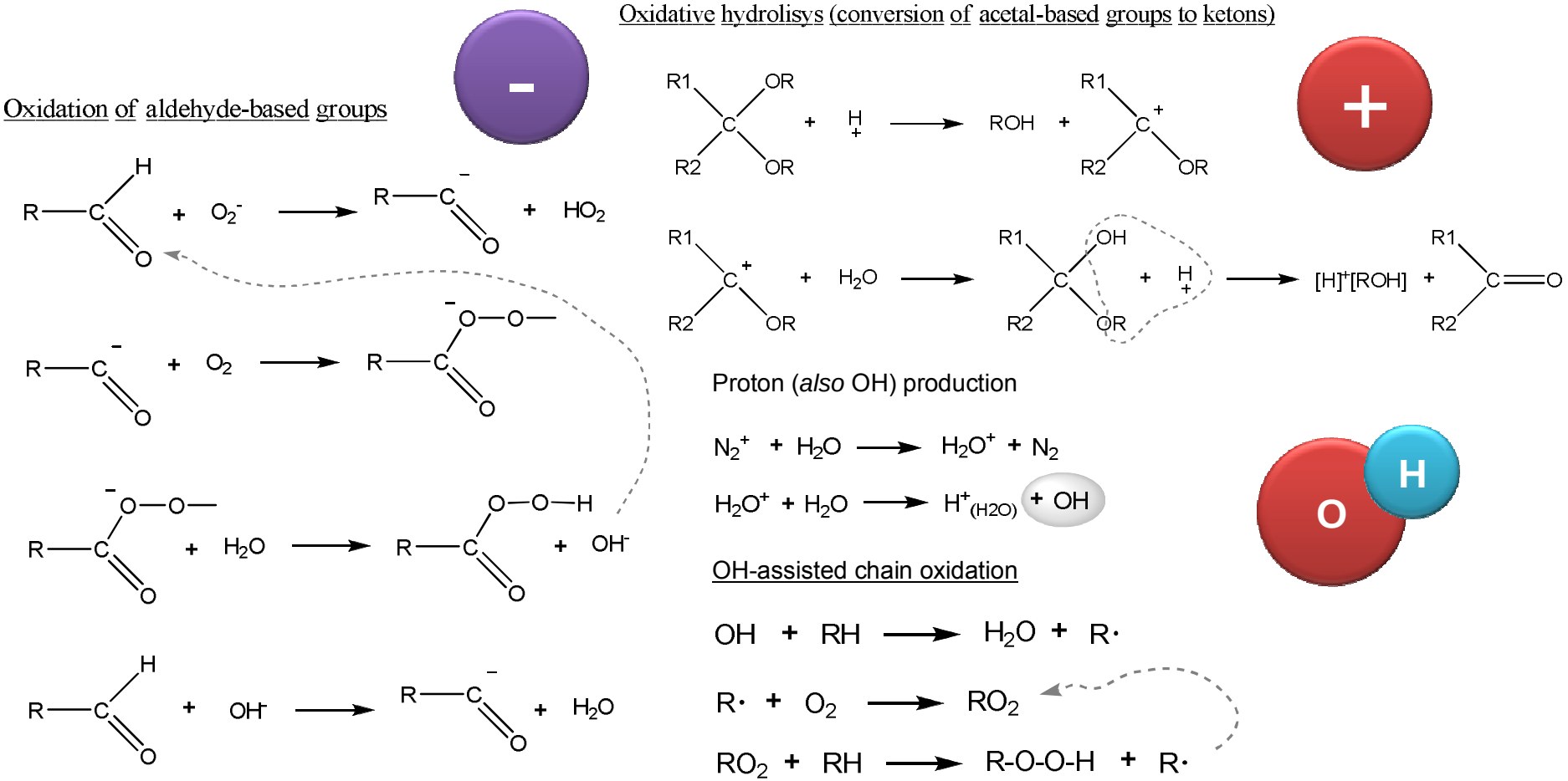
G. Fridman, et al., *Plasma Chemistry and Plasma Processing, 27, 2, 163-176 (2007)*.

A

Plasma ions play key role

Role of positive and negative ions is related to **catalysis of peroxidation** processes in presence of reactive oxygen species.

Ions create long chains (netrals, too, but much shorter).



Ion oxidation chain length: 10,000 to 100,000

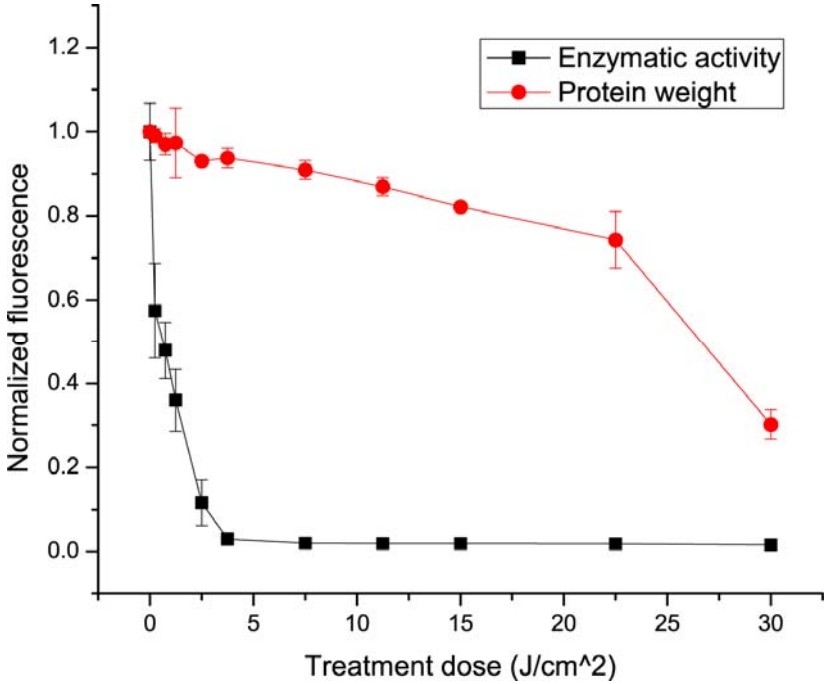
OH oxidation (i.e. phospholopid peroxidation) chain length: 3 to 10

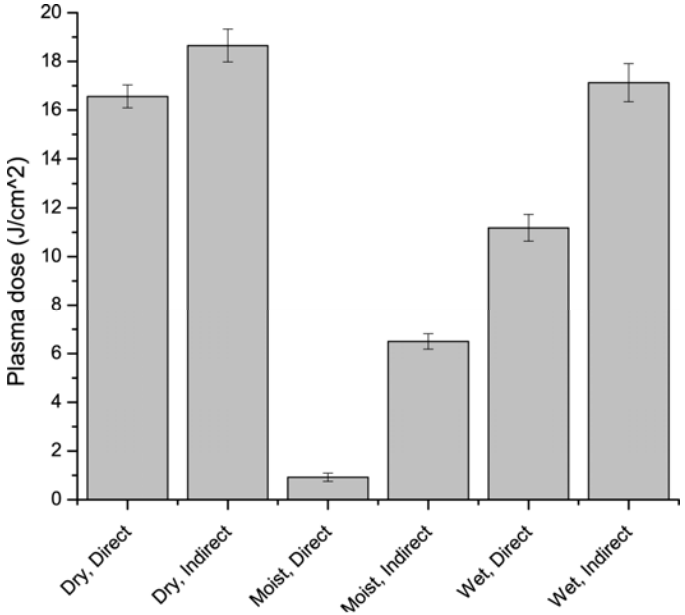
B

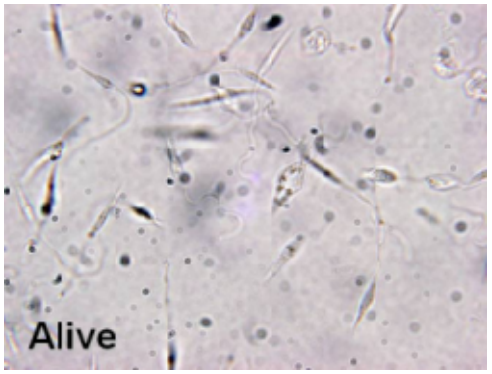
Primary target is cell membrane

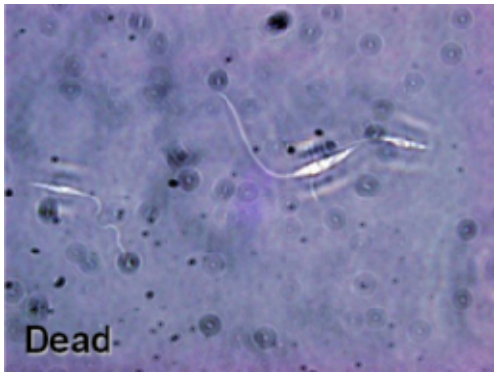
Peroxidation of phospholipids and polysaccharides is catalyzed by charged species. Water is required and effect of surrounding medium is important.

- Cell lifecycle changes without membrane damage (no cell lysis);
- Biochemical effects persist long after treatment;
- Composition and amount of medium are important;
- DNA damage is secondary.

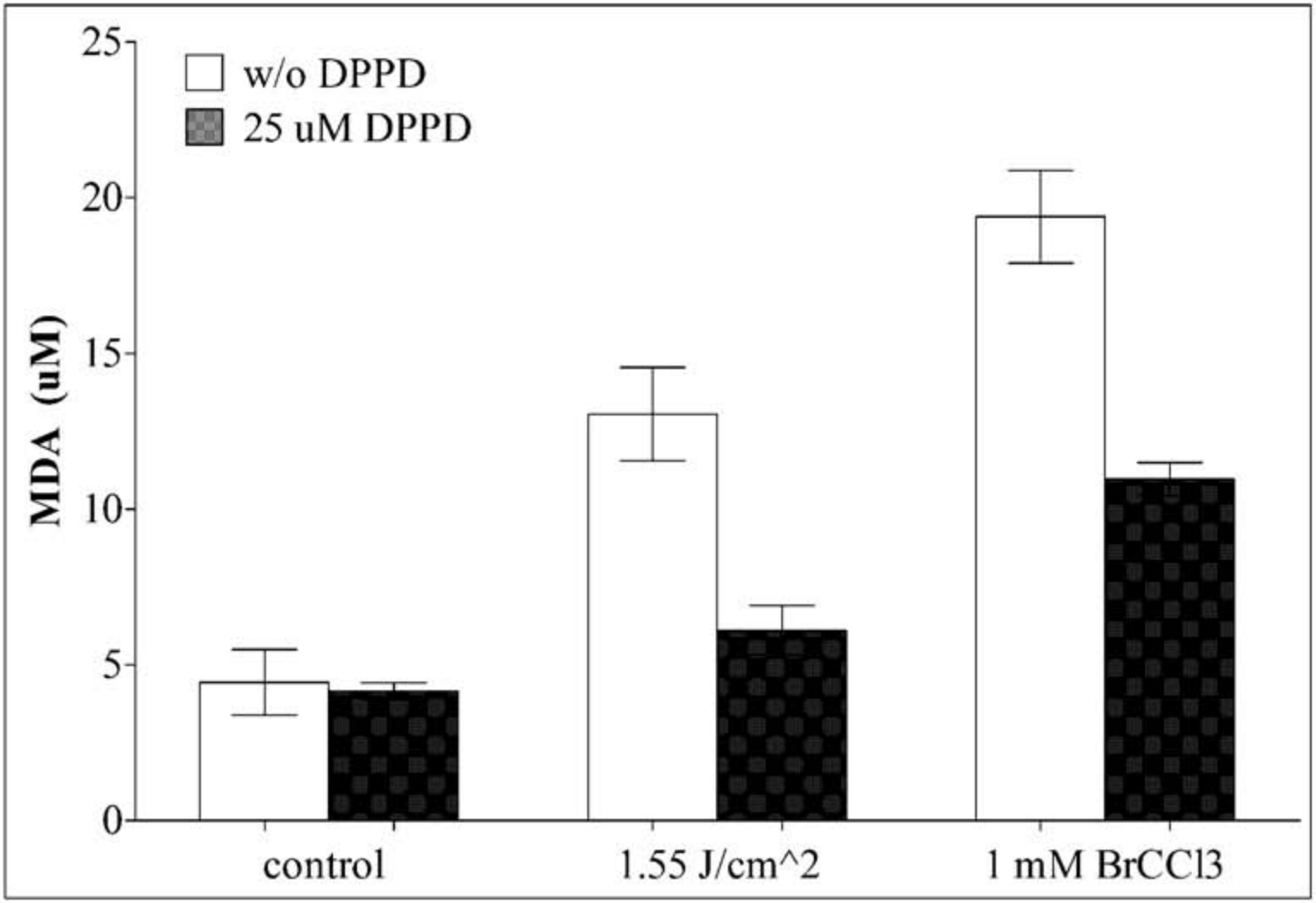








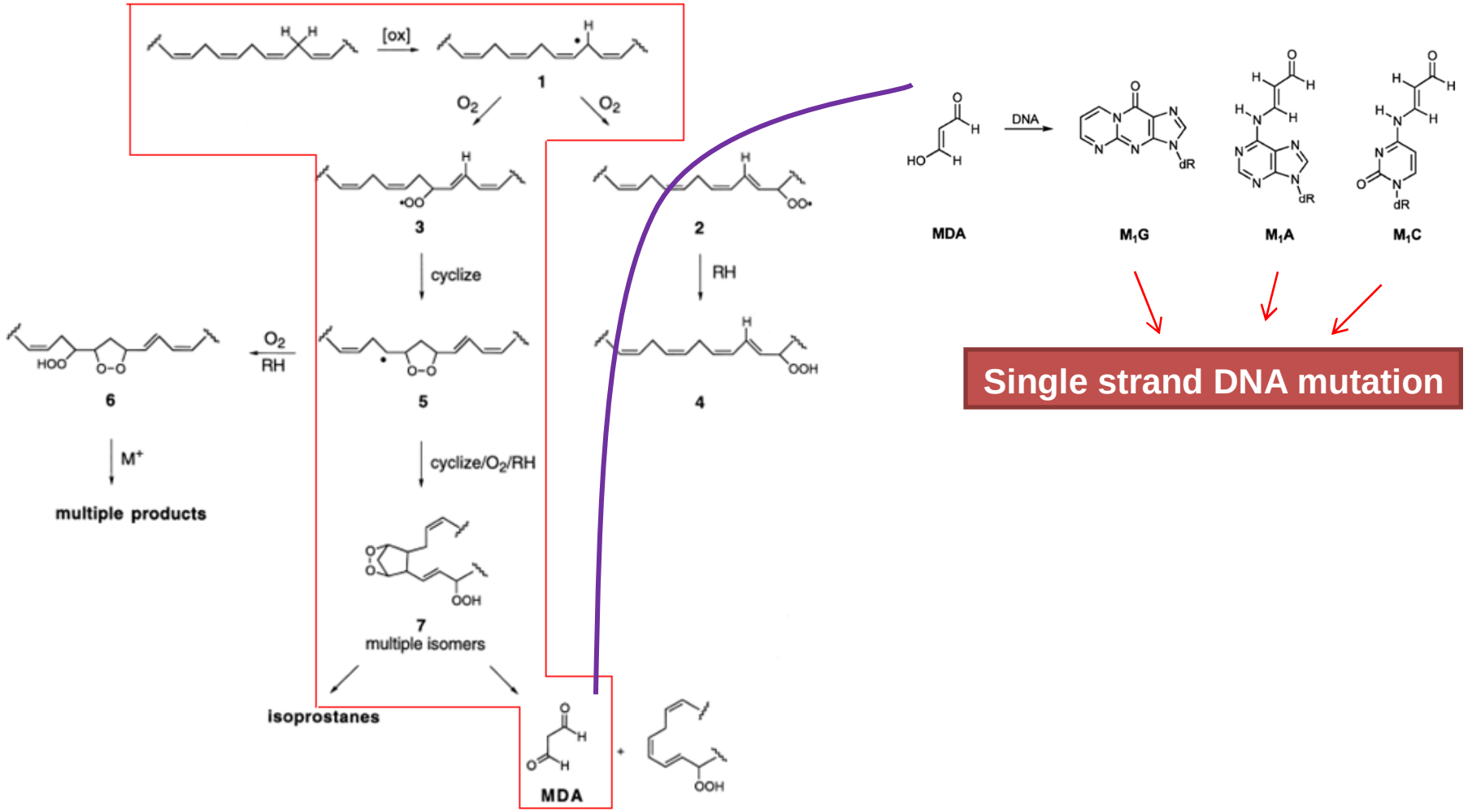
Lipid peroxidation leads to and is measured by MDA release



C

Intracellular biochemistry

Lipid layer peroxidation leads to intracellular pathway activation (i.e. malondialdehyde, MDA formation) which alters DNA structure.



Marnett, Lawrence J. "Lipid Peroxidation - DNA Damage by Malondialdehyde." Mutation Research (1999): 83-95.

Biological Mechanisms:

DNA double strand breaks (DSBs) in mammalian cells

REACTIVE OXYGEN SPECIES (ROS) INDUCES DSBs:

- Superoxide generation:
  $e_{aq} + O_2(H_2O) \rightarrow O_2^-(H_2O)$
- Superoxide dismutation into hydrogen peroxide (catalyzed by Super Oxide Dismutase)
  $2\ O_2^- + 2\ H^+ [+SOD] \rightarrow H_2O_2 + O_2 [+SOD]$
- OH generation (Fenton mechanism):
  $H_2O_2 + Fe^{2+} \rightarrow OH + OH^- + Fe^{3+}$
  $Fe^{3+} + O_2^- \rightarrow Fe^{2+} + O_2$
  $M^+ + H_2O \rightarrow M + H_2O^+$
  $H_2O^+ + H_2O \rightarrow H_3O^+ + OH$

- Hydroxyl chain oxidation:
  $OH + RH \rightarrow R + H_2O$
  $R + O_2 \rightarrow RO_2$
  $RO_2 + RH \rightarrow RO_2H + R$

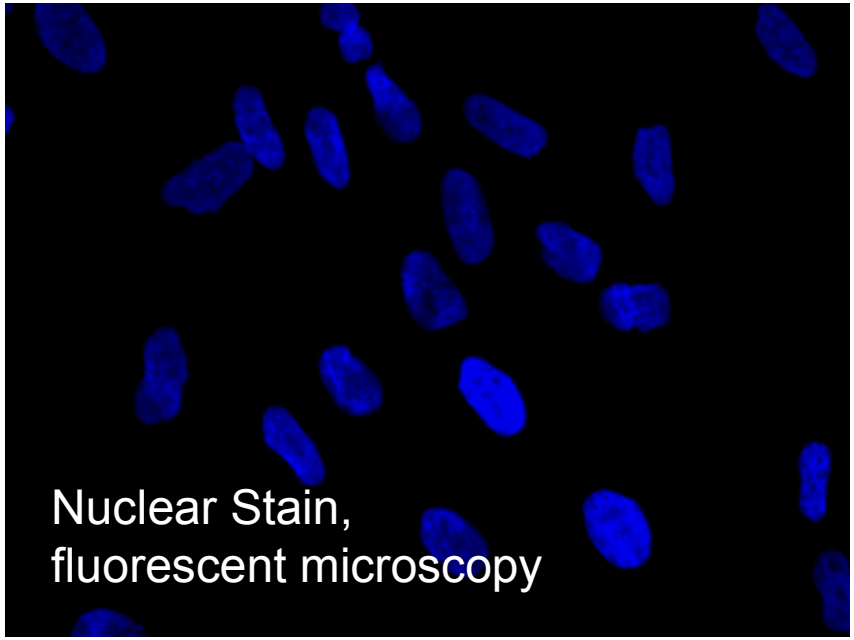
Both in radiation and plasma DNA double strand breaks are created by ion-related ROS (not by direct radiation).

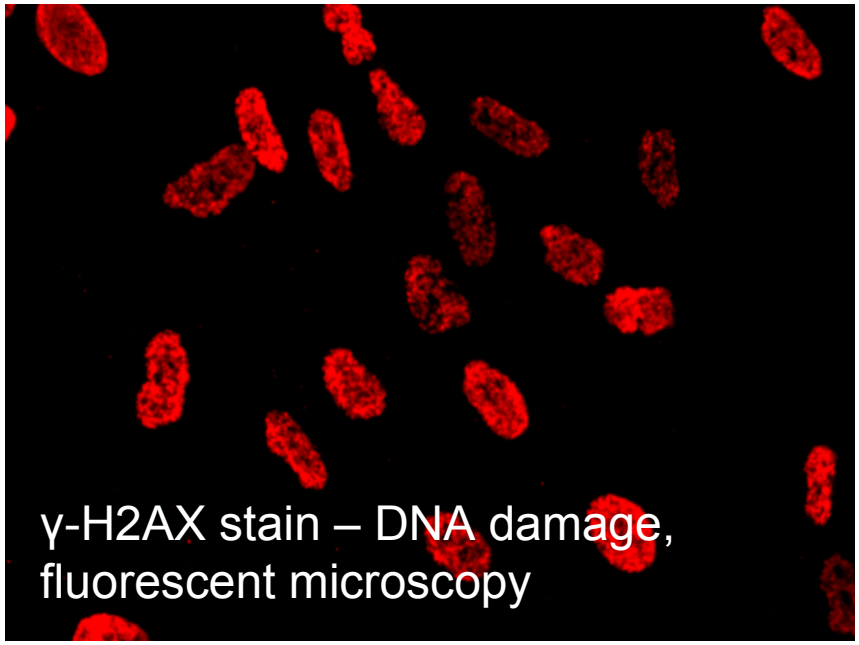
Plasma produces much more ROS than radiation biology, however transport into the cell is limiting DSBs

Samuni, A., J. Aronovitch, D. Godinget, M. Chevion, and G. Czapski, European Journal of Biochemistry, 1983. 137(1-2): p. 119-124.
McClune, G.J. and J.A. Fee, FEBS Letters, 1976. 67(3): p. 294-298.

Biological Mechanisms:

DNA single/double strand breaks in mammalian cells





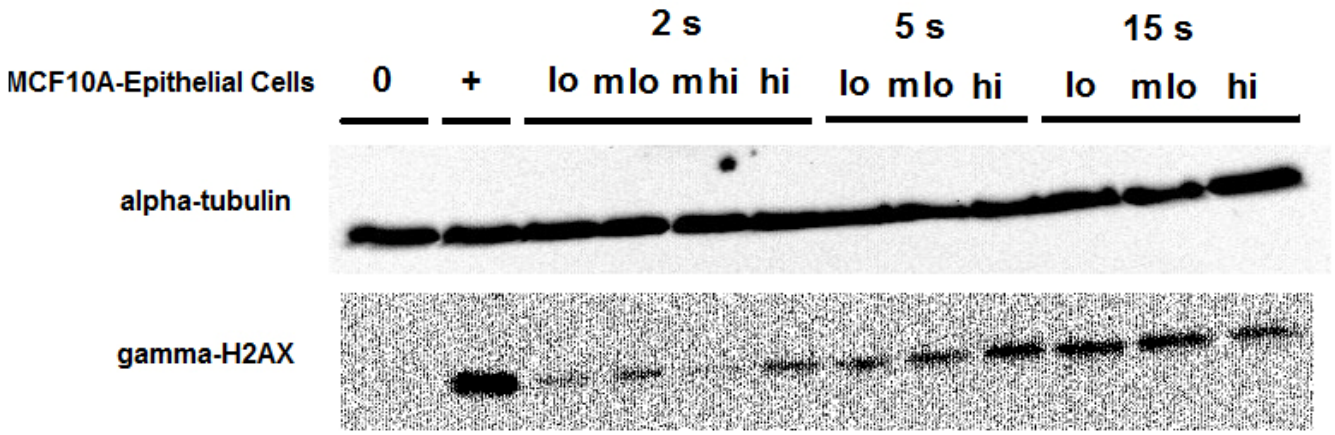
Skin sterilization is effective at seconds of FE-DBD plasma treatment (doses below J/cm^2)

2 - 15 second (>1 J/cm^2) treatment promotes DNA DSB's inside nucleus without damage to the cell itself (cell remains intact).
(Cell line: human Fibroblasts)

S. Kalghatgi, et al., 61st Annual Gaseous Electronics Conference, Oct 13th-Oct 17th, Dallas, Texas, USA.

Western Blot Analysis of DNA DSB/SSBs:

Low dose plasma causes no/few DSB/SSBs



0 no treatment
+ 200 uM H2O2

lo 500 Hz (0.13 W/cm^2)
m lo 800 Hz (0.19 W/cm^2)
m hi 1000 Hz (0.25 W/cm^2)
hi 1500 Hz (0.31 W/cm^2)

Low plasma dose (<1J/cm^2) ➔ No DSB/SSBs
Higher plasma doze (1-10 J/cm2) ➔ *Repairable* DSB/SSBs

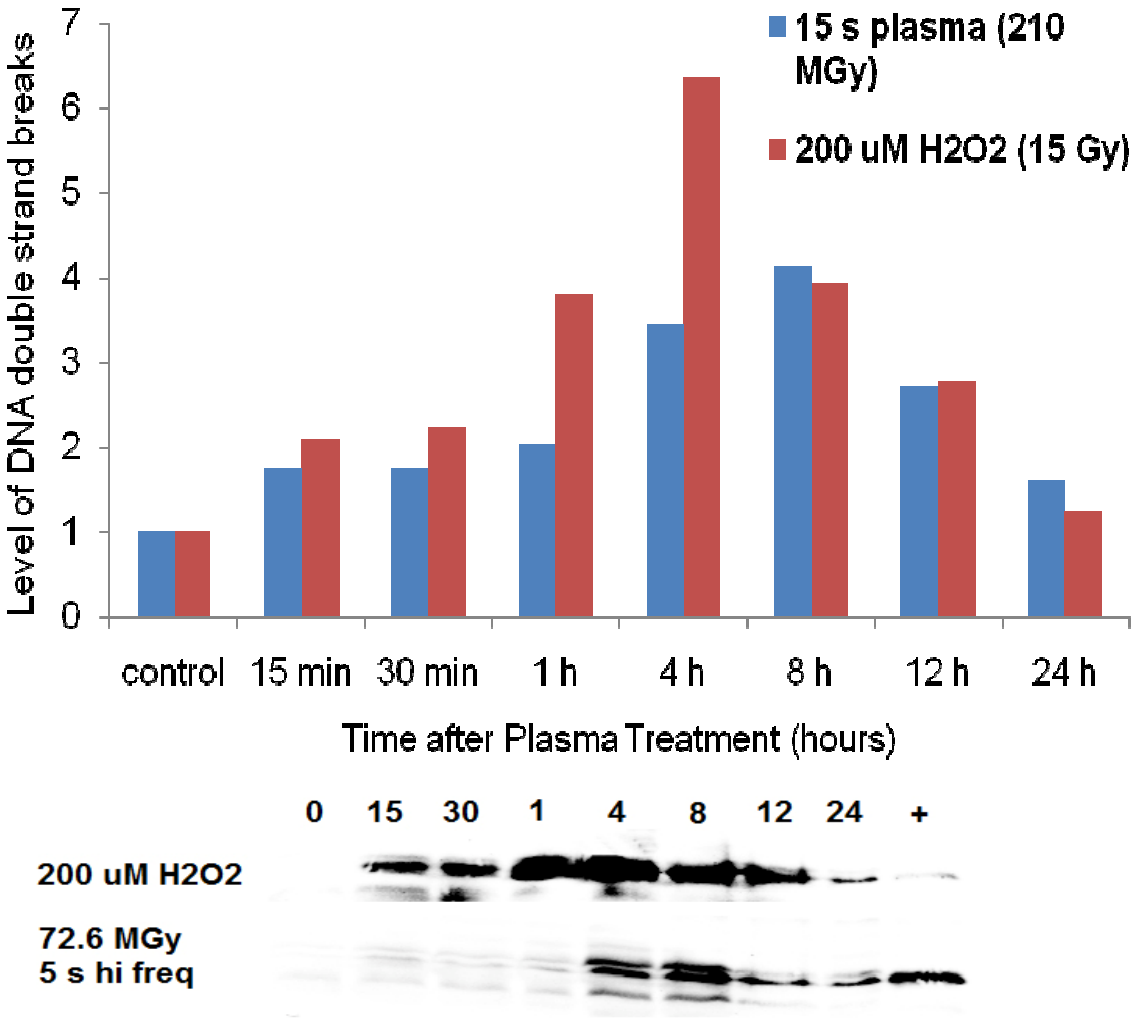
1. Low doses of non-thermal plasma (10-200 MGy) do not induce DNA double strand breaks while, higher doses of non-thermal plasma (>200 MGy) induces DNA double strand breaks which are repaired within 12-18 hours.

2. In the human body a normal cell undergoes 20,000 DNA double strand breaks every single day but the level of DNA double strand breaks produced by non-thermal plasma is much less than these naturally occurring double strand breaks

3. Non-thermal plasma is non-toxic and non-lethal to mammalian cells at low doses

S. Kalghatgi, et al., 61st Annual Gaseous Electronics Conference, Oct 13th-Oct 17th, Dallas, Texas, USA.

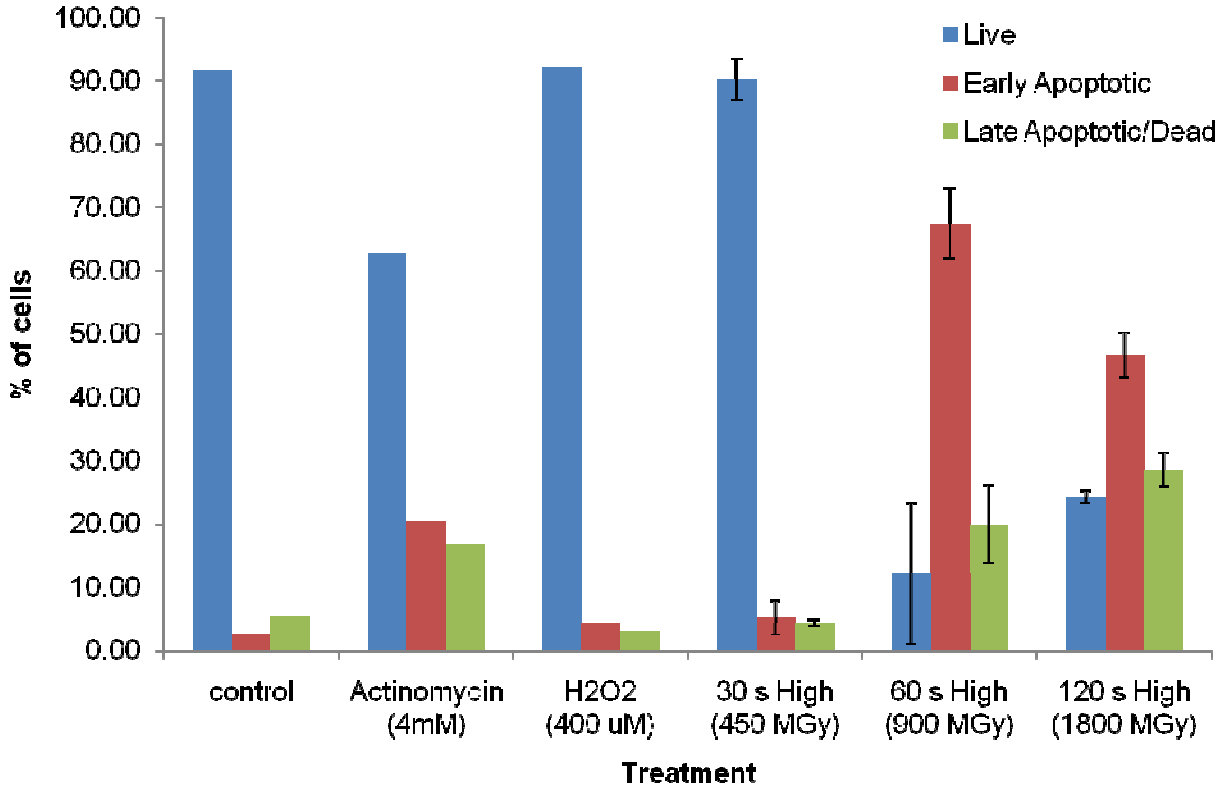
DNA DSB/SSBs are repaired by cell within 24 hrs



DNA damages induced by non-thermal plasma are repaired by the cells naturally within 24 hours

S. Kalghatgi, et al., 61st Annual Gaseous Electronics Conference, Oct 13th-Oct 17th, Dallas, Texas, USA.

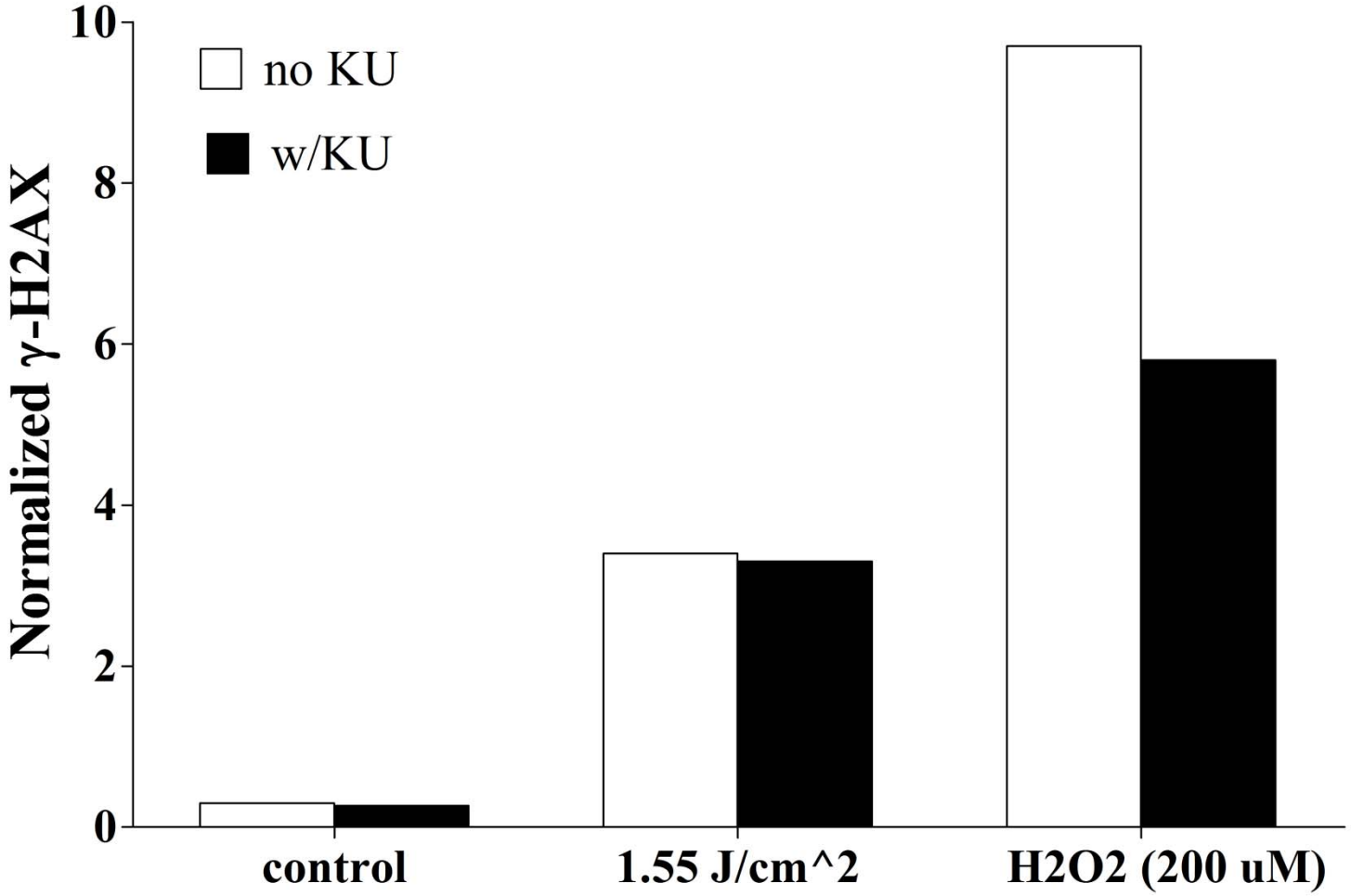
Higher Doses of Plasma induce Apoptosis



Higher doses of Plasma (>10 J/cm^2) induces significant DNA damages which are not repaired and the cells then undergo apoptosis, which prevents mutations in the cells

S. Kalghatgi, et al., 61st Annual Gaseous Electronics Conference, Oct 13th-Oct 17th, Dallas, Texas, USA.
S. Kalghatgi, et al., 2nd International Conference on Plasma Medicine (ICPM-2), Mar 16th – Mar 20th 2009, San Antonio, Texas, USA

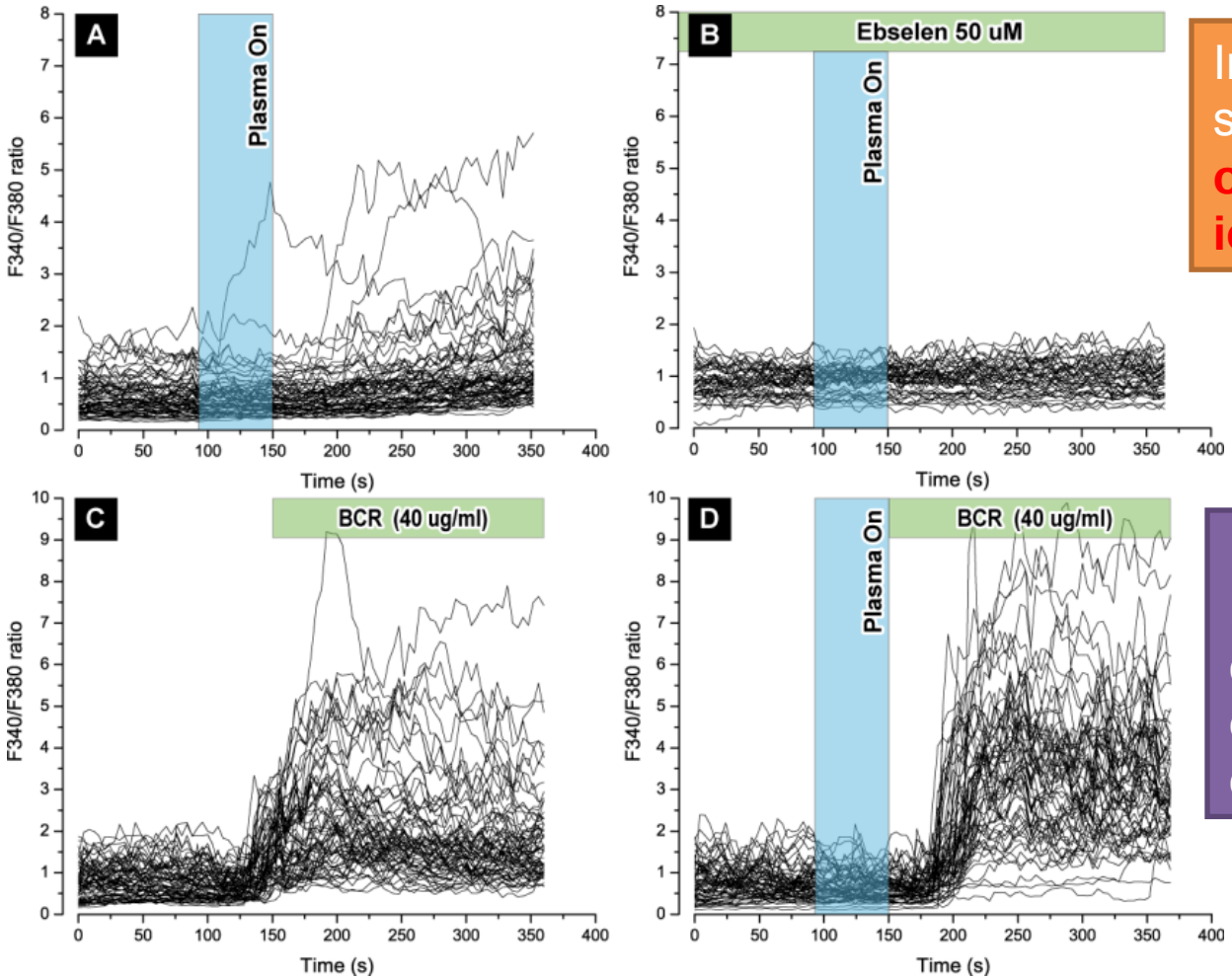
Plasma does not Induce DNA Double Strand Breaks at Low and Intermediate Doses



C

Intracellular biochemistry

Lipid layer peroxidation leads to **intracellular pathway activation** (i.e. malondialdehyde, MDA formation) which alters DNA structure.

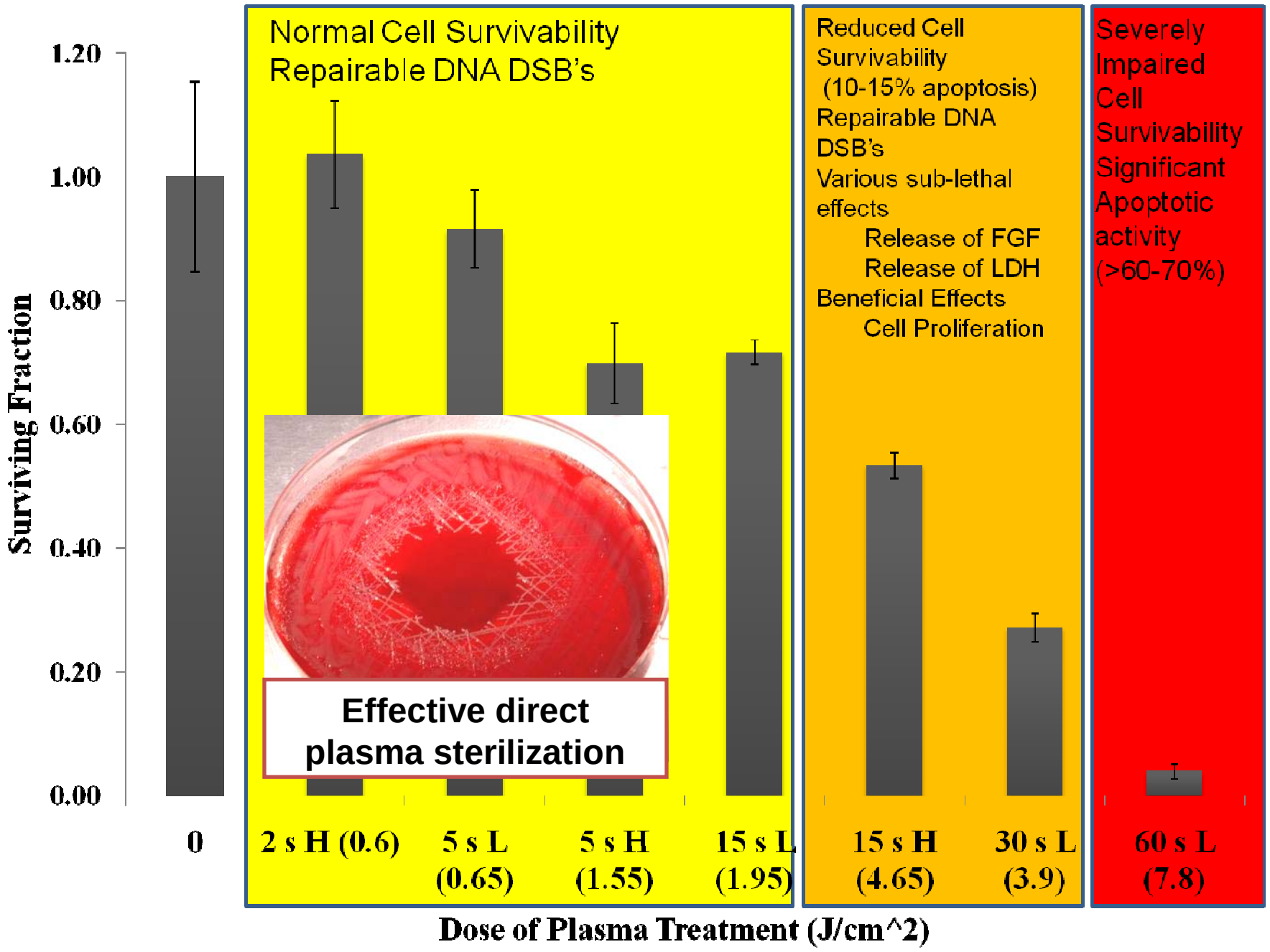


Inhibiting ROS activity suppresses **plasma effect on opening of Calcium ion channels**

Enzymatically promoting ROS activity multiplies the **effect of plasma on opening of Calcium ion channels**

Purified splenic B cells; Plasma 1 min; ebselen 50 μM preincubation 15', BCR 40 μg/ml.

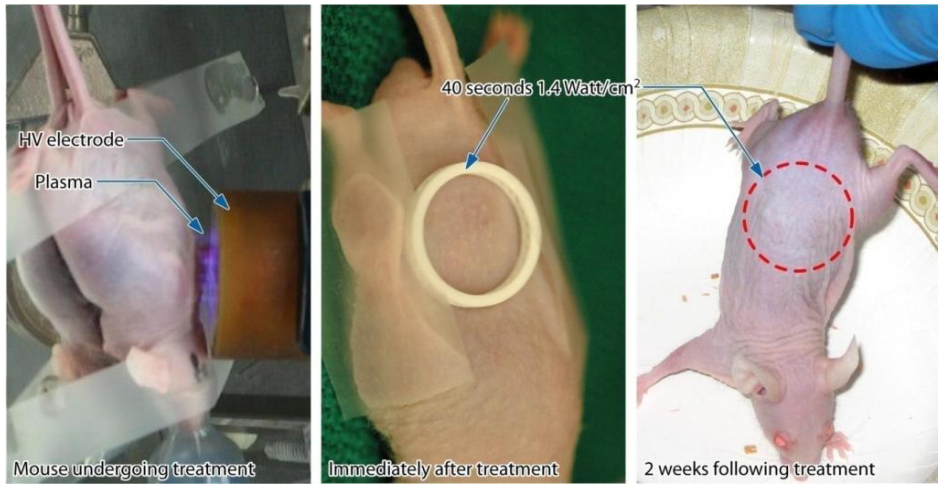
G Fridman, et al., 35th IEEE International Conference on Plasma Science (ICOPS), June 15 - 19, 2008, Congress Center Karlsruhe, Germany.

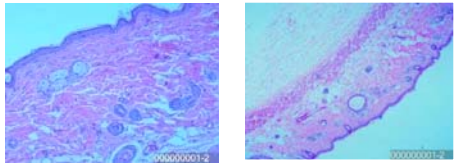

Normal Cell Survivability
Repairable DNA DSB's
Reduced Cell Survivability
(10-15% apoptosis)
Repairable DNA DSB's
Various sub-lethal effects
Release of FGF
Release of LDH
Beneficial Effects
Cell Proliferation
Severely Impaired Cell Survivability
Significant Apoptotic activity
(>60-70%)
Effective direct plasma sterilization
Surviving Fraction
1.20
1.00
0.80
0.60
0.40
0.20
0.00
0
2 s H (0.6)
5 s L (0.65)
5 s H (1.55)
15 s L (1.95)
15 s H (4.65)
30 s L (3.9)
60 s L (7.8)
Dose of Plasma Treatment (J/cm^2)

D

Selectivity

Tissue sterilization without visible or microscopic (histological) damage; selective development of apoptosis in cancer cells; difference in metabolism of peroxidation products; etc.





No treatment — 5 minutes @ 0.3W/cm² (90 J/cm²)

Skin is **sterilized in seconds** (<10 sec,<1 J/cm²);
Animals are fine up to **>10 minutes** of plasma (>240 J/cm²).

SKH1 hairless mice and regular swine: Differential toxicity escalation trials

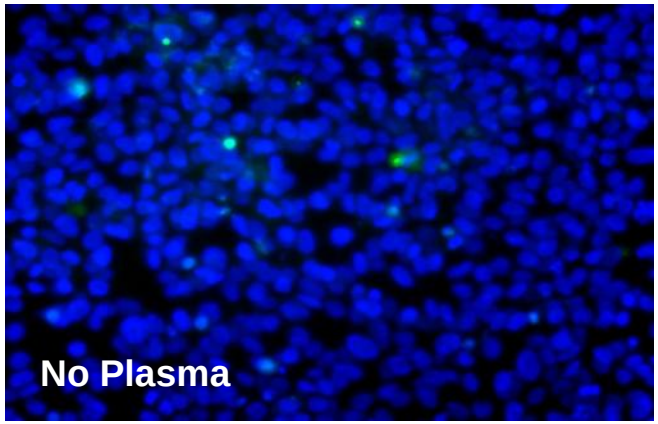
Safe and effective zones exist but toxicity is possible at higher dosage.

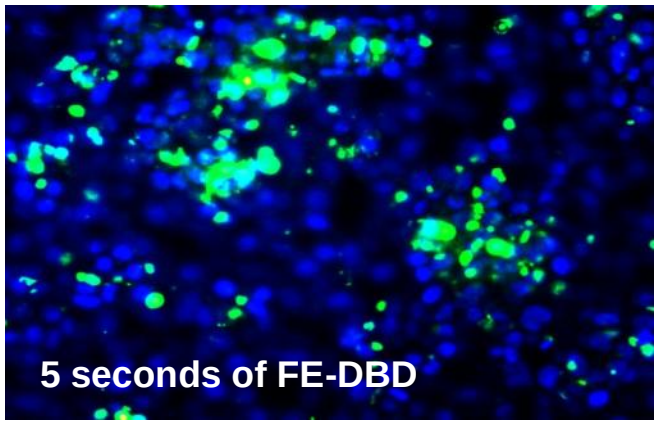
Animals survive the operation and stay healthy long after treatment (as observed for 2 weeks)

D

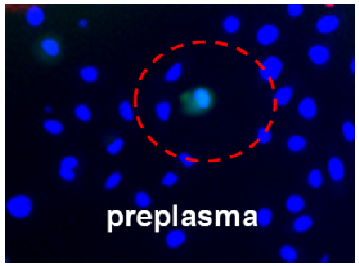
Selectivity

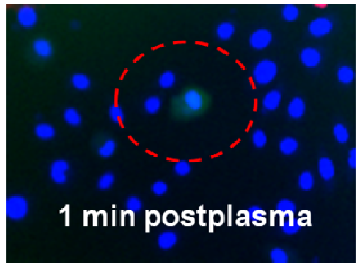
Tissue sterilization without visible or microscopic (histological) damage; selective development **of apoptosis in cancer cells**; difference in metabolism of peroxidation products; etc.

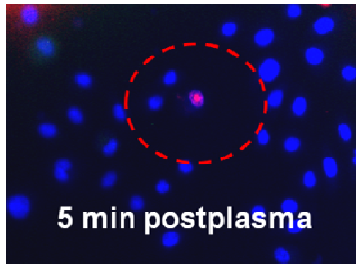




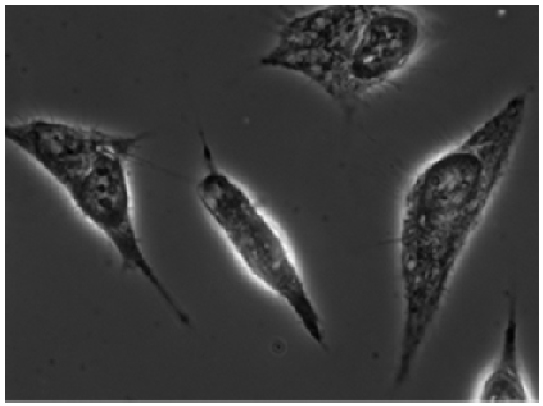
Apoptosis begins developing at the end of day 1, reaches it maximum at day 2 and returns to normal natural rate at day 3



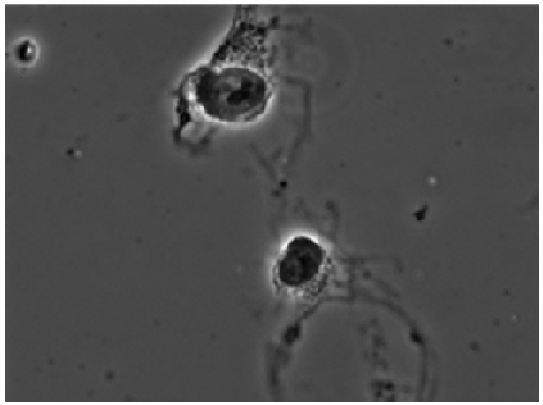




Plasma directly kills melanoma cells in co-cultures



Melanoma Cells untreated



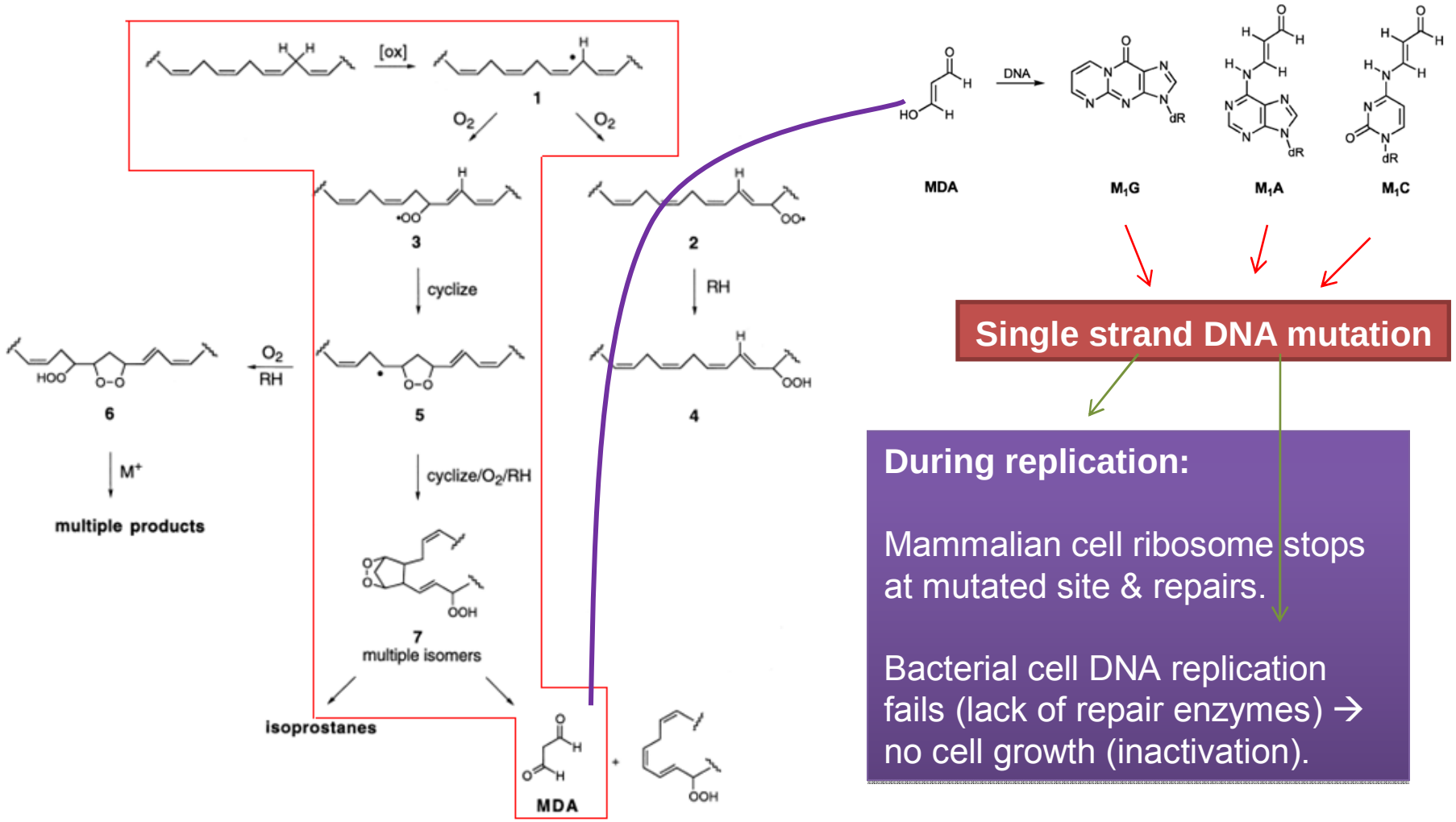
Melanoma Cells 10 s after Treatment

Shoshanna Zucker, Ben DesSoye, Archis Bagati, Thomas M. DiSanto, Jennifer Zirnheld and Kasra Etemadi

D

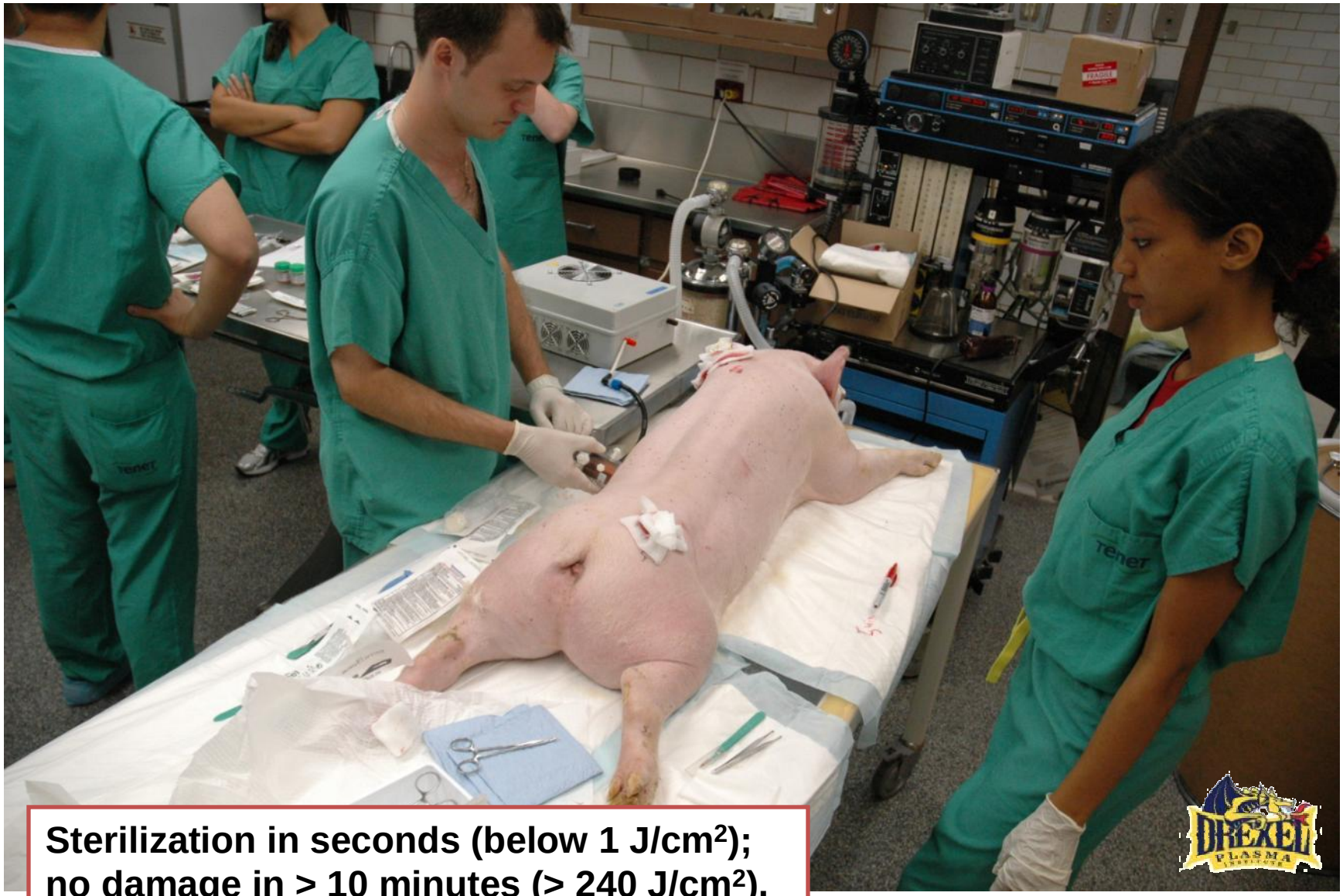
Selectivity

Tissue sterilization without visible or microscopic (histological) damage; selective development of apoptosis in cancer cells; **difference in metabolism of peroxidation products**; etc.



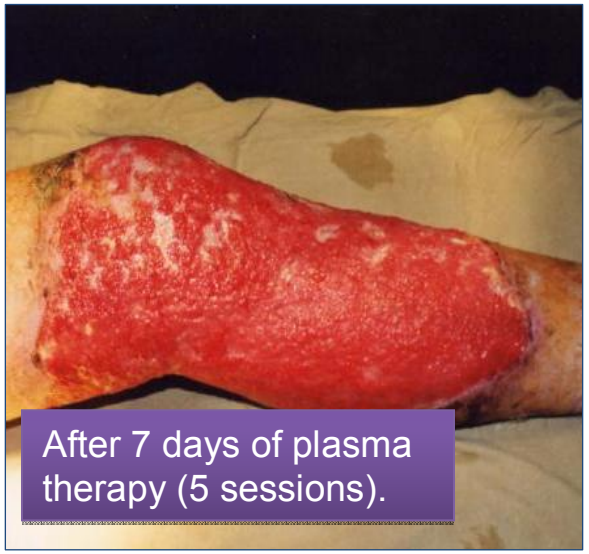
Marnett, Lawrence J. "Lipid Peroxidation - DNA Damage by Malondialdehyde." Mutation Research (1999): 83-95.

Wound Sterilization and Healing (FE-DBD)



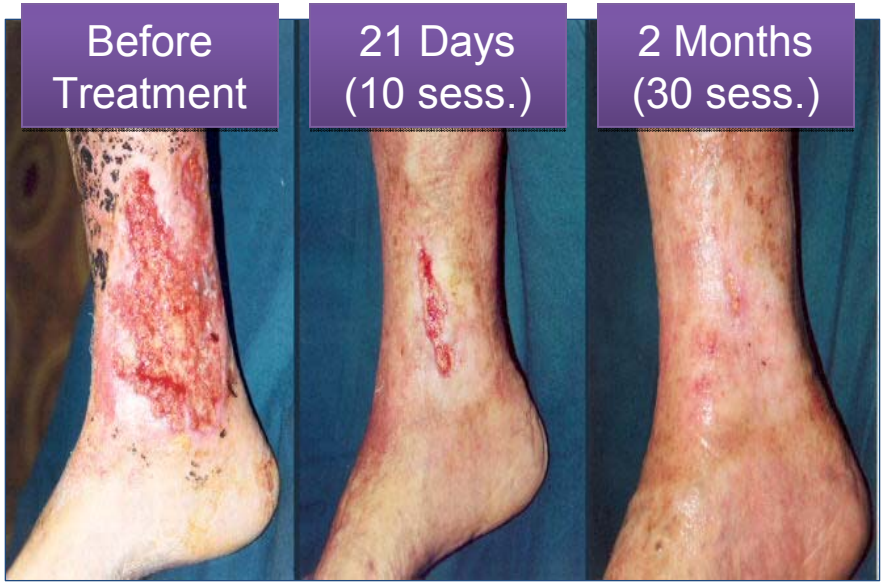
Sterilization in seconds (below 1 J/cm^2);
no damage in > 10 minutes (> 240 J/cm^2).

Wound Healing: Suppurated Burns



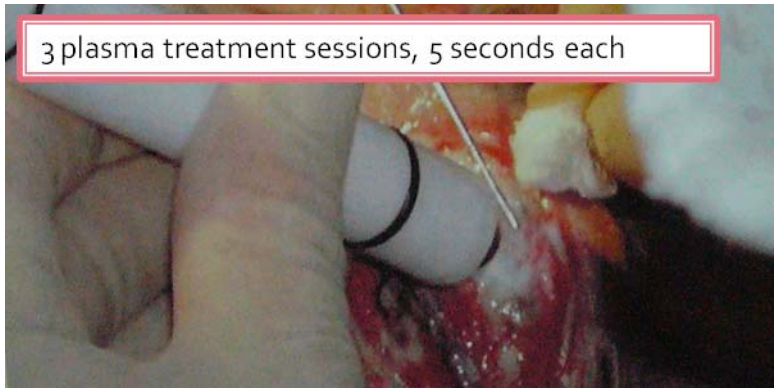
Plasma Technologies, Inc.

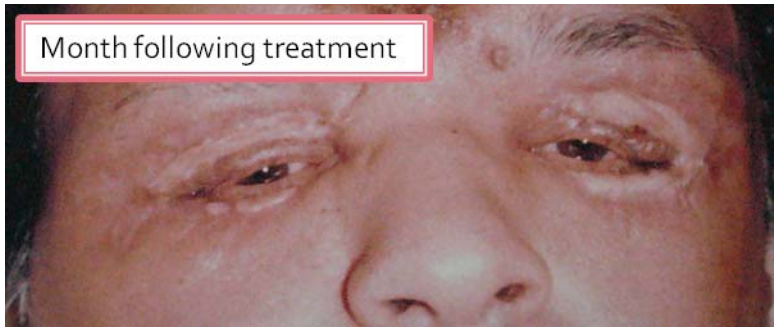
Wound Healing: Trophic Venous Ulcers



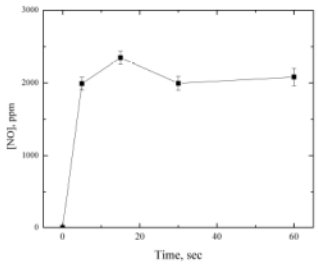
Broad Necrotic Suppurated Ulcer (Diabetic Peripheral Neuropathy)

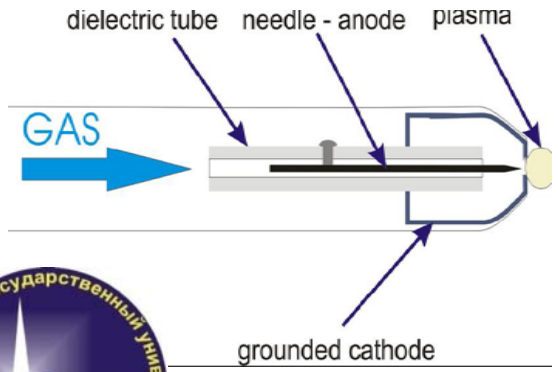
Phlegmonous Eyelid Defeat treatment by Pin-to-Hole spark Discharge (PHD)





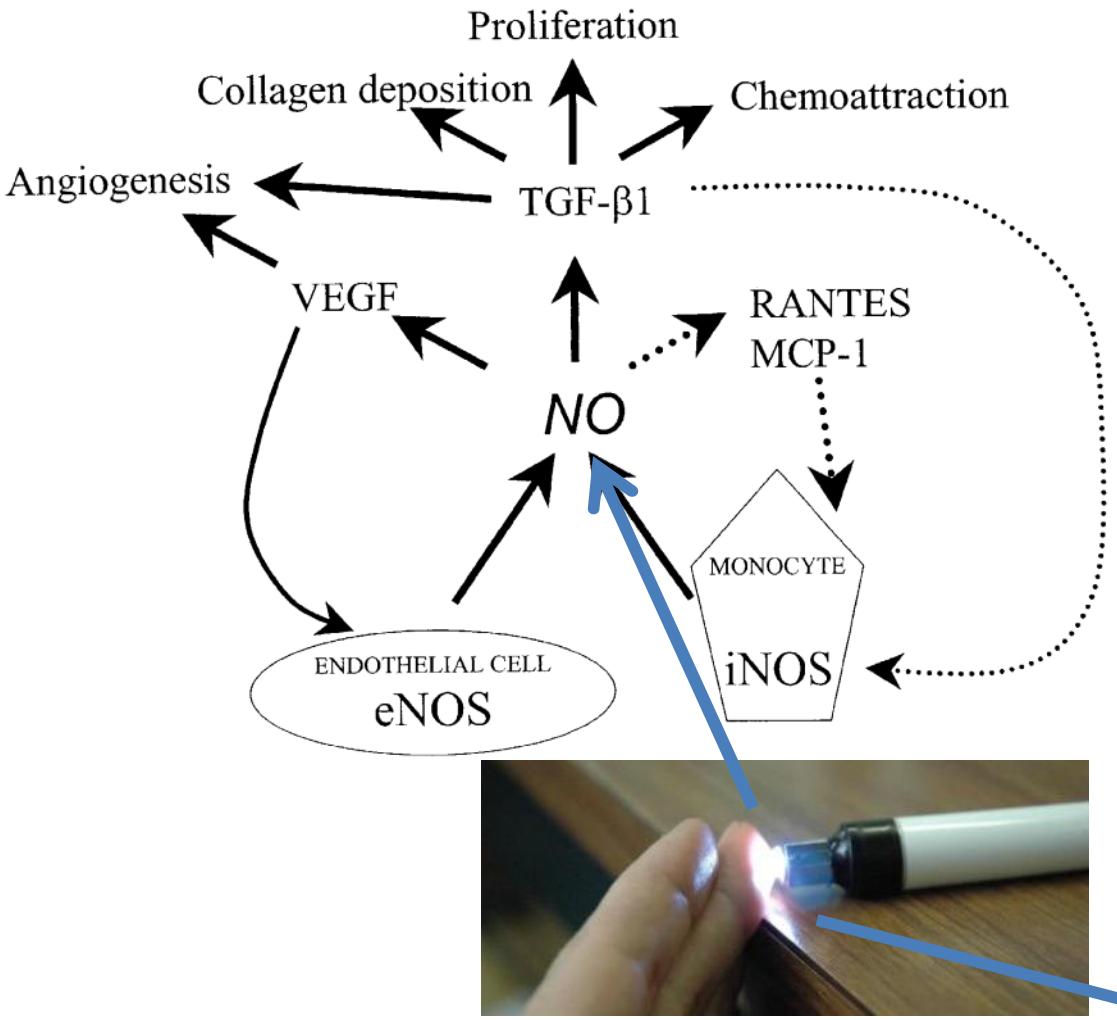
- Discharge voltage: 1 – 5 kV
- Plasma diameter: 3 – 5 mm
- Frequency: 1 – 7 Hz
- Pulse duration: ~30 ms
- Energy per pulse: 0.5 - 1 J



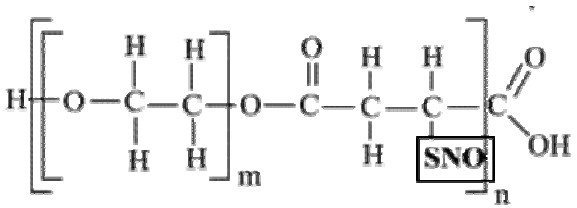


Danil Dobrynin, Valery Gostev, Sameer Kalghatgi, Gregory Fridman, Kenneth Barbee, Gary Friedman, Alexander Fridman, Andrew Wu, Erica Podolsky, Ari Brooks

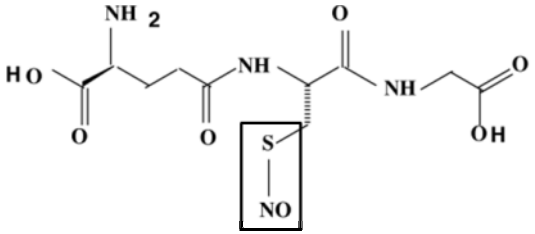
Nitric oxide (NO) plays a major role in wound healing



Applying NO to wound:



Polynitrosated polyester [2]



S-nitrosoglutathione [3]

Continuous administration:

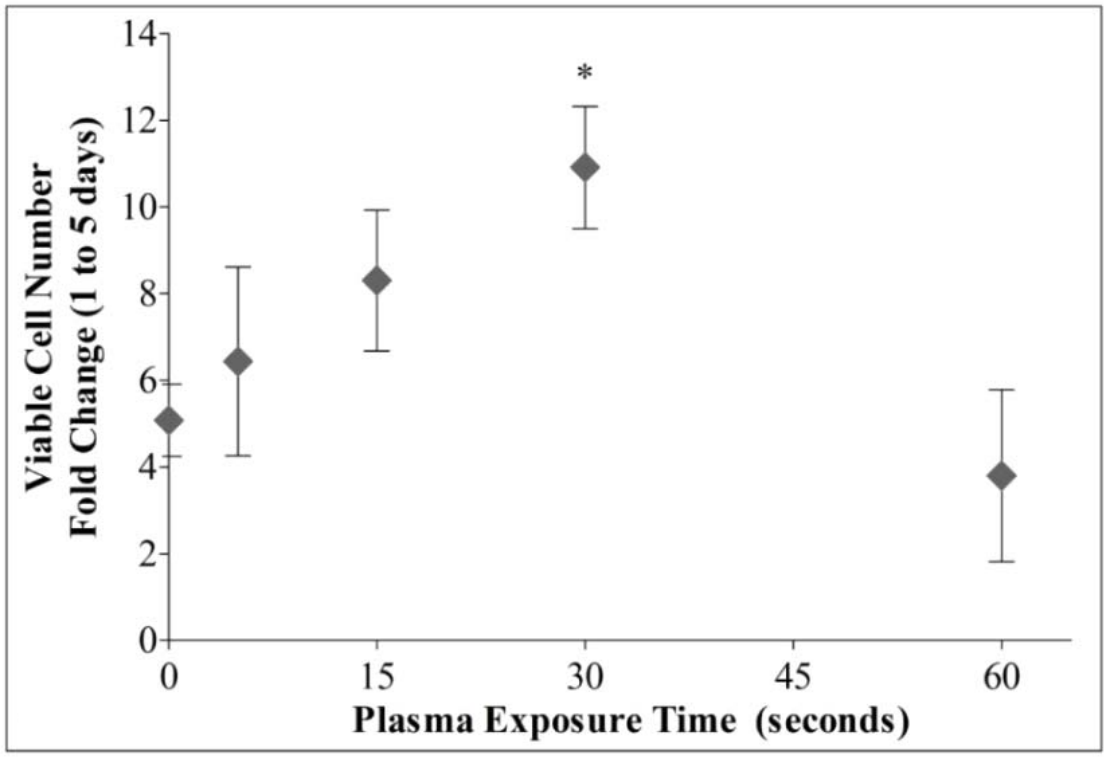
- NO bound to polymer "hydrogel", applied to wound
- NO released from hydrogel as NO• or as S-nitrosothiol

Intermittent administration:

- Apply NO• to wound
- gNO
- **Plasma afterglow**

1. Schwentker, A., et al., Nitric Oxide, 2002. 7(1): p. 1-10.
2. AB Seabra, R da Silva, MG de Oliveira, Biomacromolecules, 2005.
3. AB Seabra, et al., British Journal of Dermatology, 2004.

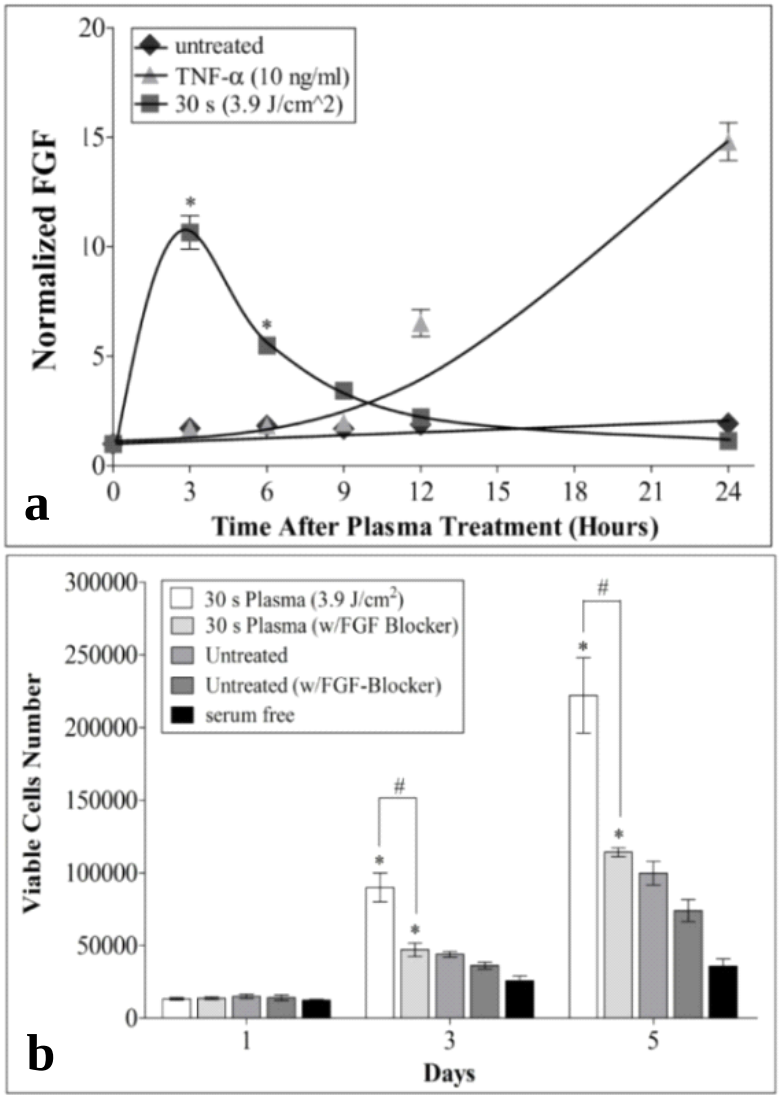
Plasma treatment promotes endothelial cell growth



Viable endothelial cell (Porcine Aortic Endothelial Cell, PAEC) number was enhanced by low dose FE-DBD plasma treatment. FE-DBD plasma-treated cells showed greater viable cell number than control up to 30 seconds of plasma treatment. However, plasma treatment greater than 30 seconds resulted in similar cell number to controls.

S. Kalghatgi, et al., Proceedings of the ASME 2009 Summer Bioengineering Conference (SBC2009), June 17-21, Lake Tahoe, CA, USA
S. Kalghatgi, et al., 30th International Conference of the IEEE EMBS, August 20-24, 2008, Vancouver, British Columbia, Canada.
S. Kalghatgi, et al., Proceedings of the IEEE 35th International Conference on Plasma Science, Jun 15-19, 2008, Karlsruhe, Germany

Treated endothelial cells release FGF-2 growth factor

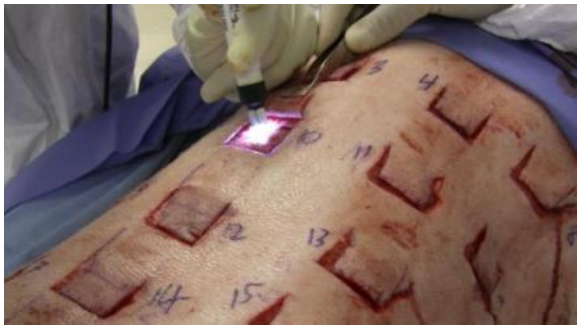


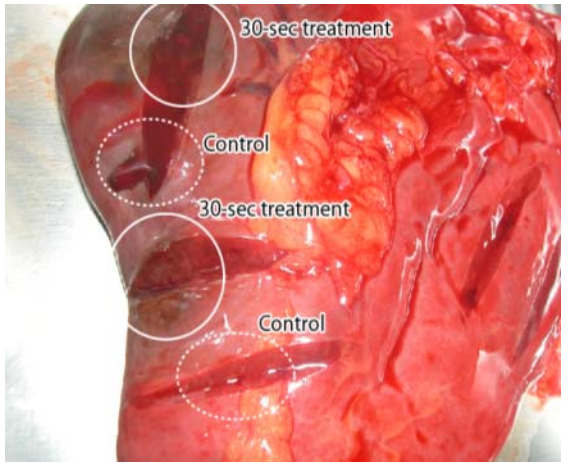
1. Cell-released FGF2 increased up to 3 h after plasma treatment, and then rapidly decreased up to 24 h after plasma treatment for 30 s.

2. Conditioned media samples collected 3 h after plasma treatment (30 s) were applied to non-treated cells, and cell number was assessed 1, 3, and 5 days after conditioned media addition. While media from plasma treated cells significantly increased cell number, an **FGF2 neutralizing antibody cancelled this effect**.

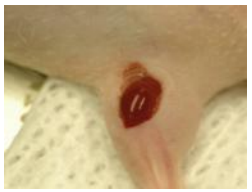
S. Kalghatgi, et al., Proceedings of the ASME 2009 Summer Bioengineering Conference (SBC2009), June 17-21, Lake Tahoe, CA, USA

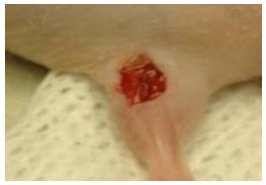
FE-DBD stimulated blood coagulation



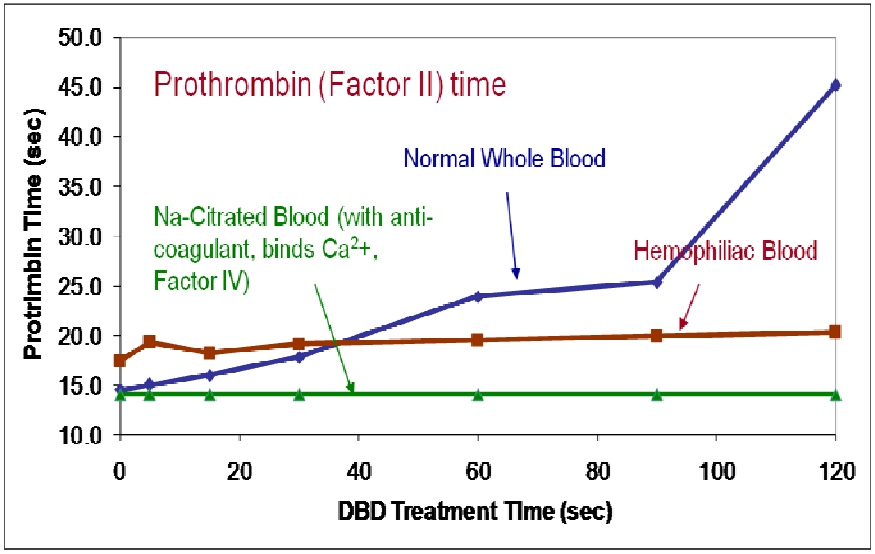


10 seconds to stop bleeding (10+ minutes w/o plasma).

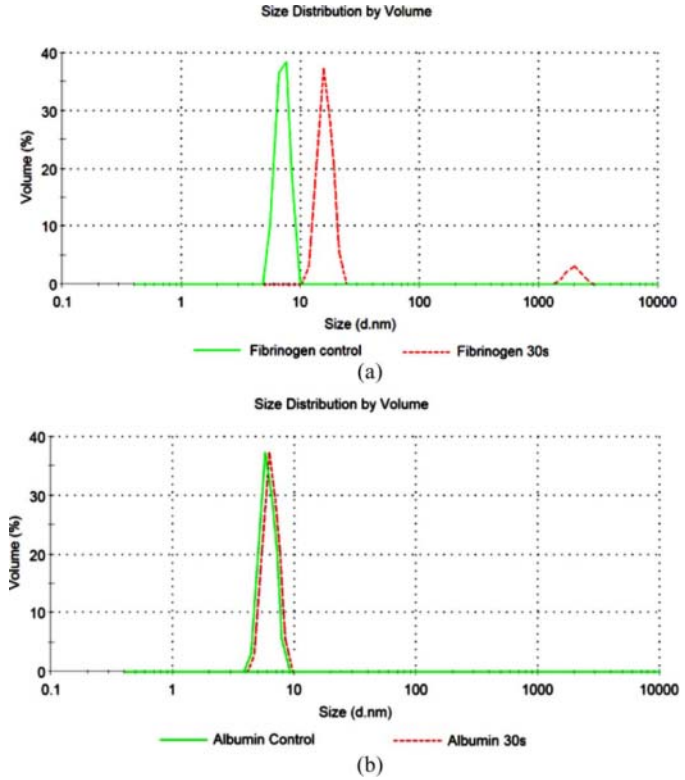




Plasma Influences the Natural Blood Coagulation Mechanism



Prothrombin Time (PT) analysis: Prothrombin (Factor II) time of residual blood increases 3 times after 120 seconds (120 J/cm^2) of DBD treatment.



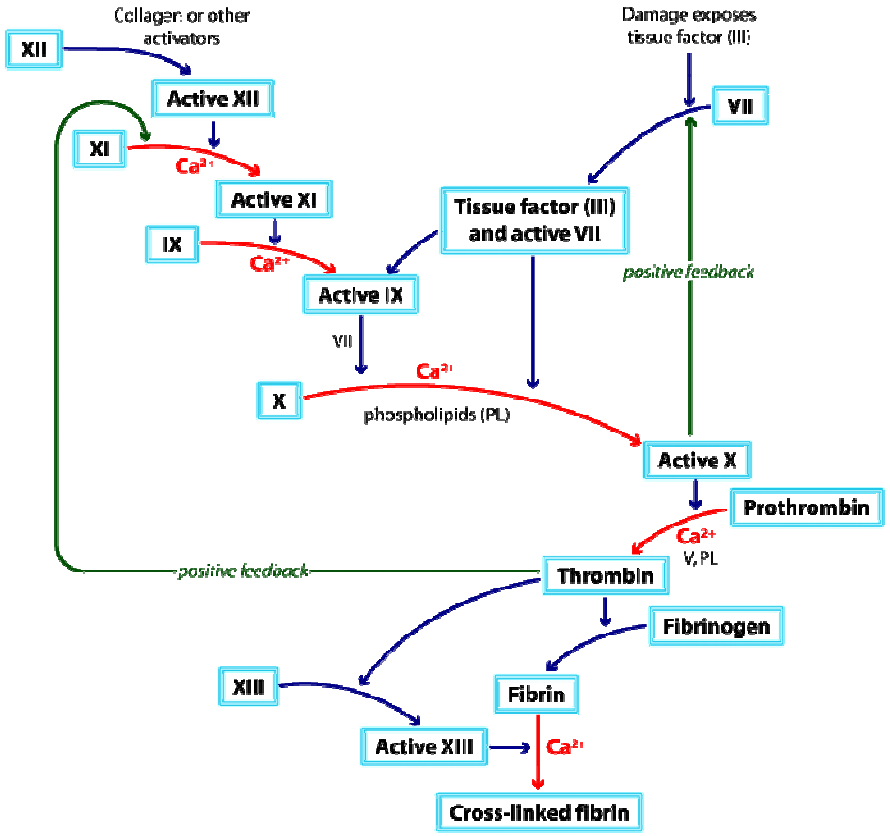
Dynamic Light Scattering: (a) Fibrinogen and (b) Albumin

Albumin treated by plasma does not coagulate; Fibrinogen, treated for the same time period, coagulates, forming a cloudy solution

G. Fridman, et al., *Plasma Chemistry and Plasma Processing, 26, 425-442 (2006)*.

S.U. Kalghatgi, et al., *IEEE Transactions on Plasma Science, Volume 35, Issue 5, Part 2, Oct. 2007, pp. 1559-1566*.

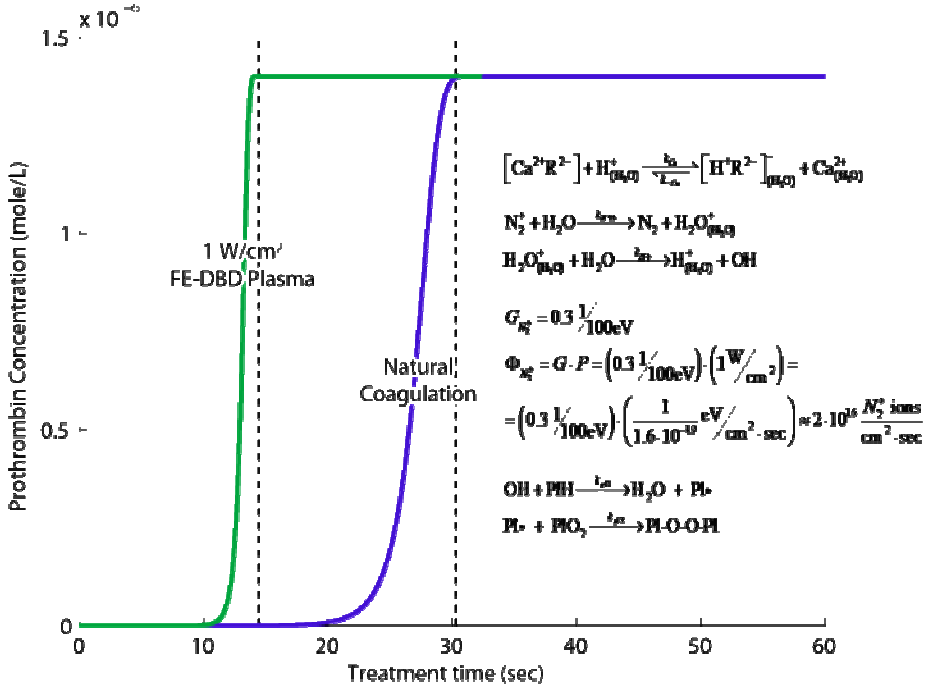
Natural Blood Coagulation: Plasma Influenced Increase in Effective Ion Concentrations



Blood coagulation cascade

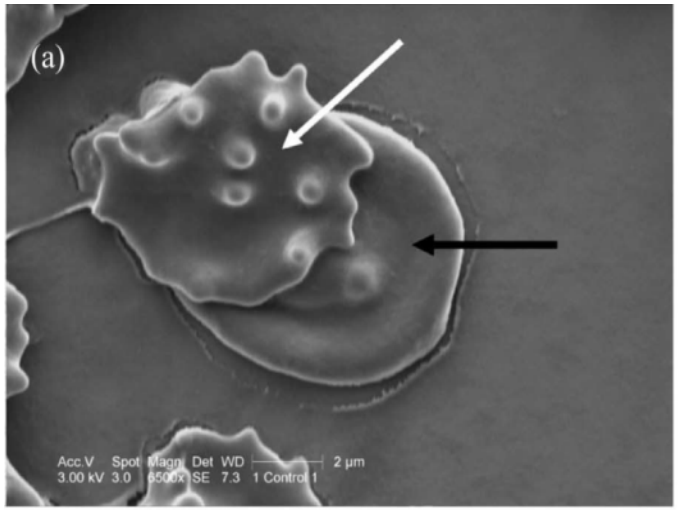
- Ions catalyze many coagulation processes
- Increase in effective ionic concentration leads to rapid coagulation

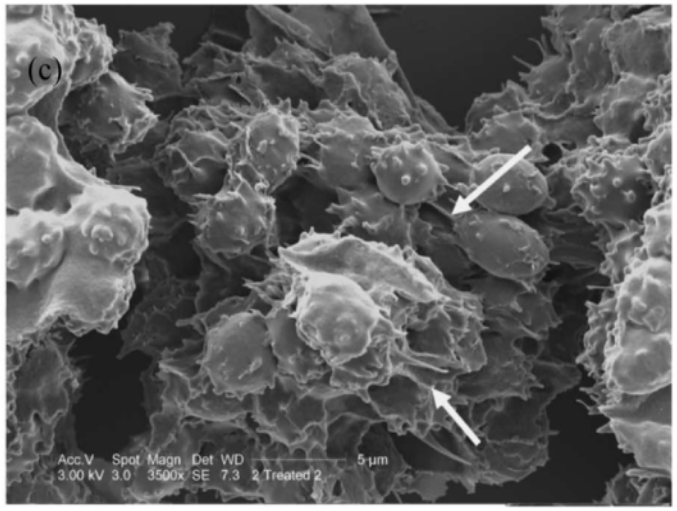
- Plasma influences effective ionic concentrations in blood and thus may be able to catalyze the coagulation process

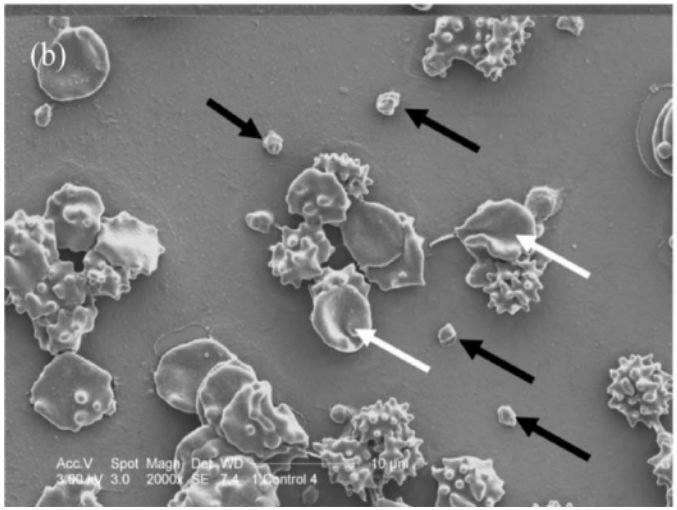


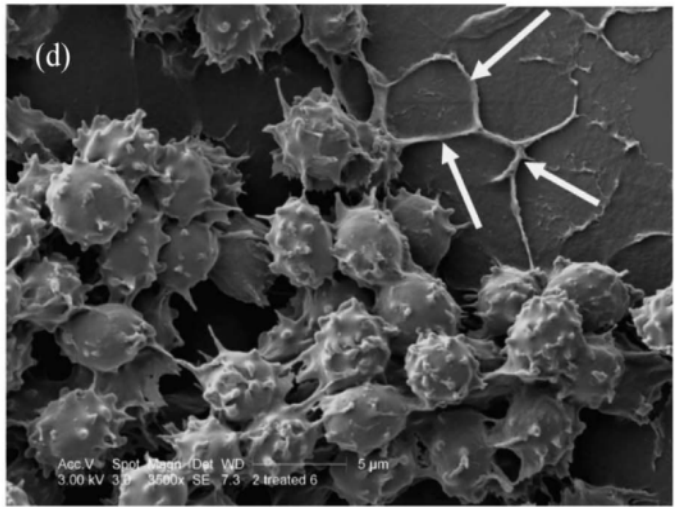
G. Fridman, et al., *Plasma Chemistry and Plasma Processing, 26, 425-442 (2006)*.

Natural Blood Coagulation: Platelet Activation by Plasma









Untreated:

(a) Citrated whole blood (control) showing a single activated platelet (white arrow) on a red blood cell (black arrow).

(b) Citrated whole blood (control) showing many non-activated platelets (black arrows) and intact red blood cells (white arrows).

Treated:

(c) Citrated whole blood (treated) showing extensive **platelet activation** (pseudopodia formation) and **platelet aggregation** (white arrows).

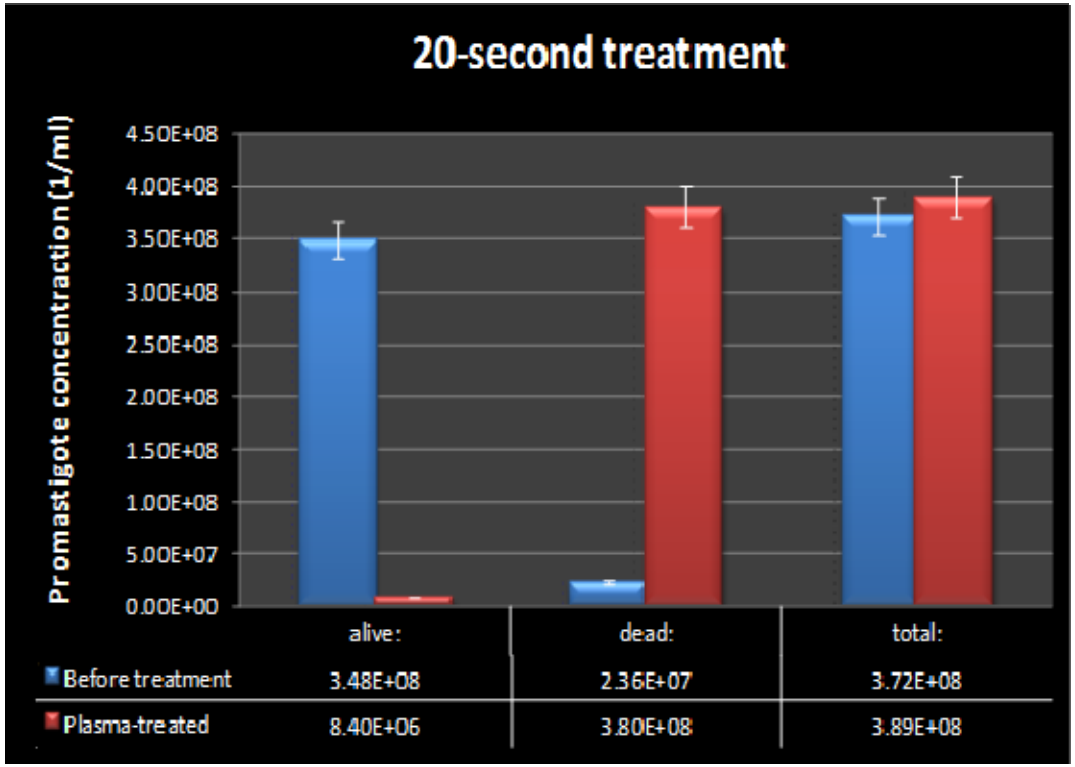
(d) Citrated whole blood (treated) showing platelet aggregation and **fibrin formation** (white arrows).

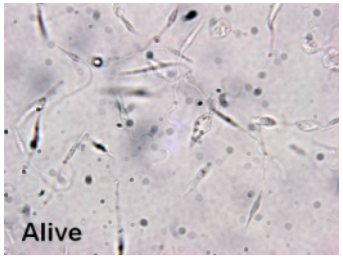
Plasma →
*** Platelet activation**
*** Platelet aggregation**
*** Fibrin formation**

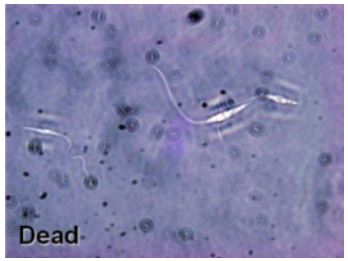
S.U. Kalghatgi, et al., *IEEE Transactions on Plasma Science, Volume 35, Issue 5, Part 2, Oct. 2007, pp. 1559-1566*.

Skin Diseases

Plasma treatment of cutaneous Leishmaniasis

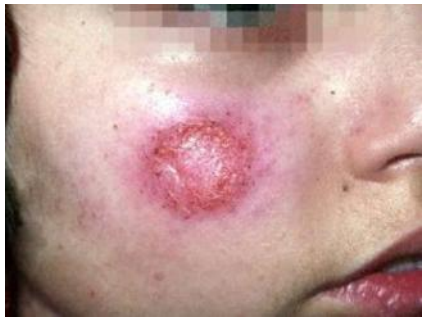


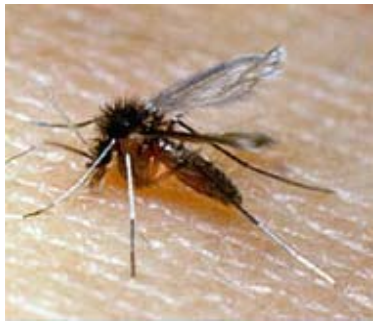


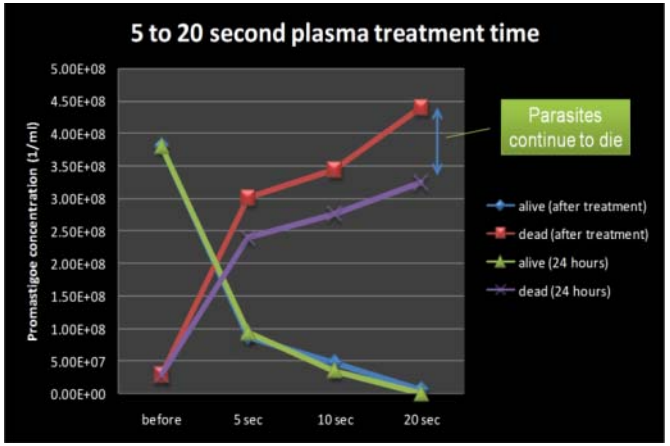


20 seconds of FE-DBD: 100% inactivation of *C. Leishmania* promastigotes

2 minutes of FE-DBD: ~20-50% inactivation of Macrophages







G. Fridman, et al., *Plasma Process. Polym., Volume 5 Issue 6 (2008), Pages 503 – 533.*

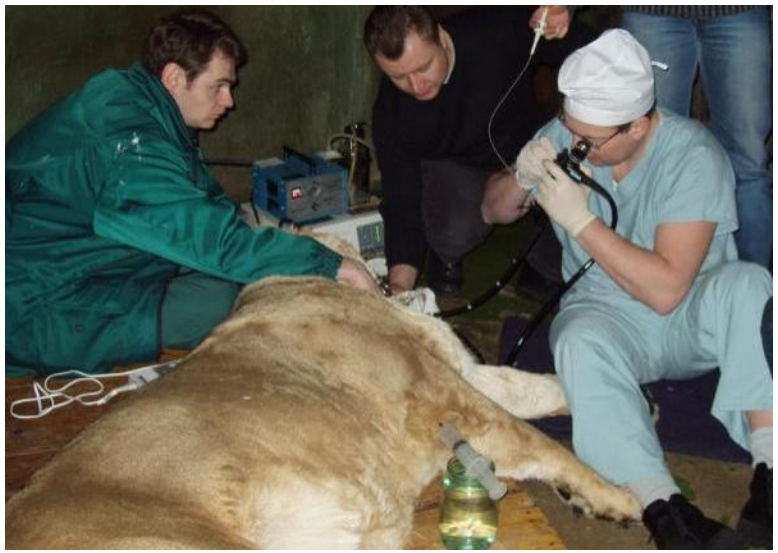
Plasma treatment inactivates *Helicobacter pylori* in Gastro-Duodenal Ulcers

Gastroenterology in-Vitro

Treatment of bleeding peptic ulcers (BPU) for coagulation and eradication of **Helicobacter pylori** (Hp) was compared for argon plasma coagulation (APC) and cold plasma elimination (CPE).

FE-DBD cold plasma elimination was found to be an effective treatment against Hp without the extensive deep tissue damage observed in APC (0.5 – 3 mm).

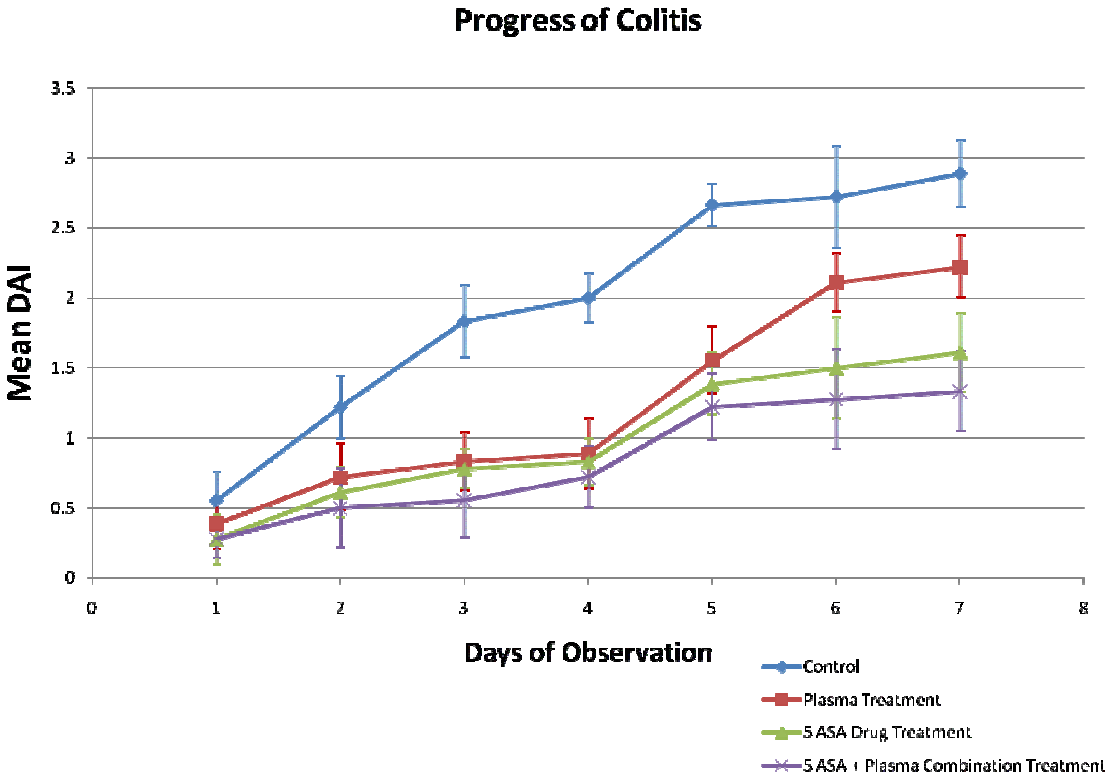
Treatment was performed on Hp-infected mucosal membrane of the swine stomach.



G. Fridman, et al., *Plasma Process. Polym., Volume 5 Issue 6 (2008), Pages 503 – 5*

Gastroenterology in-Vivo

Treatment of Crohn's Disease



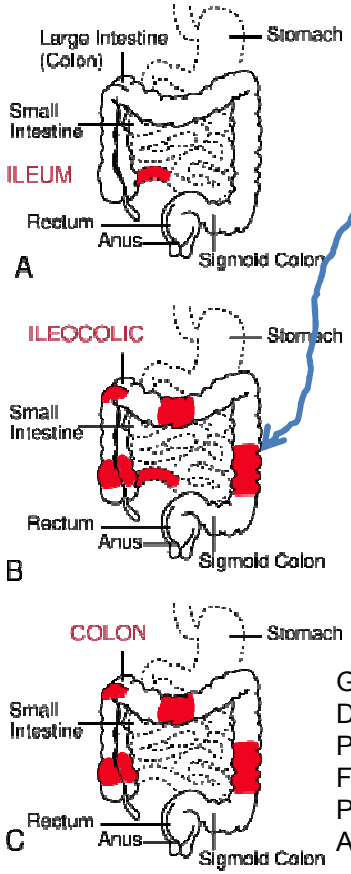
- Qualitative experiment: oxidation of Cu in H_2O_2(aq) with and w/o Ar plasma.
- Result: oxidation rate of **H_2O_2** < **H_2O_2 +plasma**

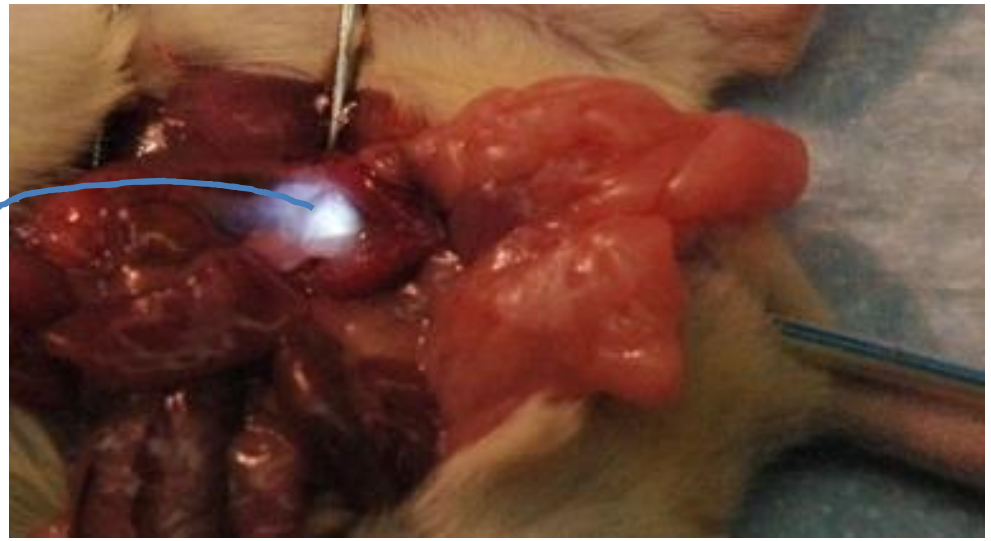
$H_2O_2 \cdot R\ (aq) + H^+\ (aq) \rightarrow RH + H_2O + O_2$

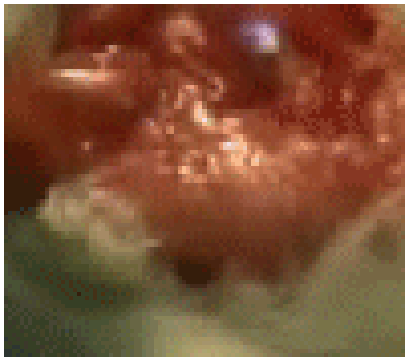
- Animals are fed 2.5% DSS all through the study.
- Plasma probe is inserted 4 cm into the colon through the anal verge and plasma is applied for stipulated time.
- Plasma treatment is given on every alternate day (3 treatments / experiment).
- 5 – ASA drug is administered everyday through the 7 days.
- Results of "mouse" experiments: **plasma treatment stopped progression of colitis**. Through "antioxidant" properties of plasma (due to ↑acidity (positive ions $\Rightarrow H^+(H_2O)$)

K. Chakravarthy, et al., ICPM-2, March 16-20, 2009, San Antonio, Texas.
K. Chakravarthy, et al., DDW 2009, Digestive Disease Week, Chicago, IL, May 30 - June 4, 2009.

Treatment of **Inflammatory Bowel Diseases**: Crohn's Disease & Ulcerative Colitis

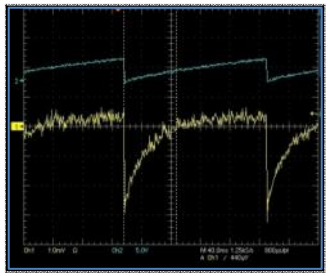


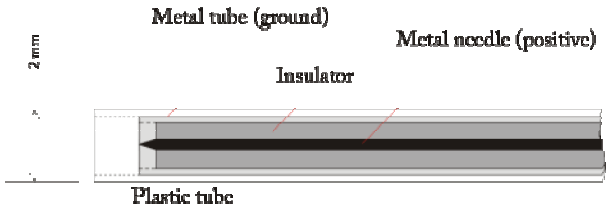


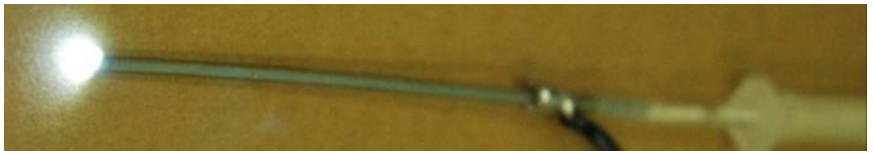


Plasma can be safely applied to treatment of gastrointestinal diseases.

Gas: atmospheric air
Discharge voltage: 0.5 - 1 kV
Plasma afterglow diameter: 2 - 3 mm
Frequency: 1 – 7 Hz
Pulse duration: 30 ms
Average plasma gas temperature: 300 K







K. Chakravarthy, et al., ICPM-2, March 16-20, 2009, San Antonio, Texas.

K. Chakravarthy, et al., DDW 2009, Digestive Disease Week, Chicago, IL, May 30 - June 4, 2009.

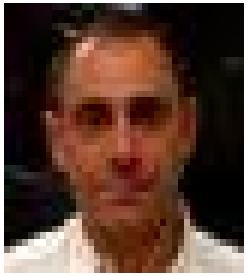
Multi-Disciplinary Effort Requires a Multi-Disciplinary Team




College of Engineering




College of Medicine

College of Arts and Sciences



School of Biomedical Engineering

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