Reactive Species From Air Plasma: Implications for Therapeutic Applications

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Plasma acts mainly to increase oxidative and nitrosative stress on tumors

This mechanism appears similar to radiation, PDT and chemotherapy anti-tumor action

*Plasma may offer advantages in treating radiation- and chemo-resistant tumors*

Air plasma-generated reactive nitrogen species example will be shown
What Might Lead to the Observed Anti-Tumor Effects of Plasma in Air?

Electric fields, charges and photons are known to be present, can certainly have biological effects, but a number of studies have suggested they are secondary. There may be exceptions, of course.

The evidence in favor of reactive oxygen and nitrogen species (RONS) as key anti-tumor agent is compelling.
What is our *Conceptual Model* for Plasma-Therapy?

Plasma device

- Gas Phase RONS
- Liquid Phase RONS products

Cell-cell communication: ‘bystander effect’ leading to tumor apoptosis?

Immune cell involvement?

Plasma-induced increased blood flow/O2?

Surface Cells

- RONS gas species: transfer into liquid
- (Liquid: water, salt, proteins, lipids; RONS react to form products)

Bulk Tissue/Tumor

Surface cells: RONS-macromolecule products induce stress that causes signaling to bulk tissue: *tumor apoptosis*
Air plasma interacting with water: initial model for plasma-cell interactions
Huge Literature on Pro-Oxidant Anti-Tumor Mechanism.....for example

The emerging role of reactive oxygen species in cancer therapy

Markus F. Renschler *

ROS stress in cancer cells and therapeutic implications

Helene Pelicano\textsuperscript{a}, Dennis Carney\textsuperscript{a,b}, Peng Huang\textsuperscript{a,*}


Oxidative stress and apoptosis: a new treatment paradigm in cancer

Ryan H. Engel, and Andrew M. Evens

[Frontiers in Bioscience 11, 300-312, January 1, 2006]
Oxidative Stress and Apoptosis: Impact on Cancer Therapy

TOMRIS OZBEN

JOURNAL OF PHARMACEUTICAL SCIENCES, VOL. 96, NO. 9, SEPTEMBER 2007

Tumor-targeted induction of oxystress for cancer therapy


J. FANG¹,†, H. NAKAMURA¹ & A. K. IYER¹,²

Cancer cell killing via ROS

To increase or decrease, that is the question

Jie Wang and Jing Yi*

[Cancer Biology & Therapy 7:12, 1875-1884; December 2008]
Redox-Directed Cancer Therapeutics: Molecular Mechanisms and Opportunities

ANTIOXIDANTS & REDOX SIGNALING
Volume 11, Number 12, 2009
Georg T. Wondrak

The causes of cancer revisited: “Mitochondrial malignancy” and ROS-induced oncogenic transformation – Why mitochondria are targets for cancer therapy

Molecular Aspects of Medicine 31 (2010) 145–170
Stephen J. Ralph a,*, Sara Rodríguez-Enríquez b, Jiri Neuzil c,d, Emma Saavedra b, Rafael Moreno-Sánchez b

Reactive Oxygen Species: The Achilles’ Heel of Cancer Cells?

ANTIOXIDANTS & REDOX SIGNALING
Volume 16, Number 11, 2012
Xiaojiang Cui
Upsides and Downsides of Reactive Oxygen Species for Cancer: The Roles of Reactive Oxygen Species in Tumorigenesis, Prevention, and Therapy

ANTIOXIDANTS & REDOX SIGNALING
Volume 16, Number 11, 2012
Subash C. Gupta, David Hevia, Sridevi Patchva, Byoungduck Park, Wonil Koh, and Bharat B. Aggarwal

Oxidative Stress and Lipid Peroxidation Products in Cancer Progression and Therapy

International Scholarly Research Network
ISRN Oncology
Volume 2012, Article ID 137289, 21 pages

Giuseppina Barrera

Mitochondria as a Source of Reactive Oxygen and Nitrogen Species: From Molecular Mechanisms to Human Health

ANTIOXIDANTS & REDOX SIGNALING
Volume 18, Number 16, 2013
Tiago R. Figueira, Mario H. Barros, Anamaria A. Camargo, Roger F. Castilho, Julio C.B. Ferreira, Alicia J. Kowaltowski, Francis E. Sluse, Nadja C. Souza-Pinto, and Anibal E. Vercesi
Oxidative stress and cancer: An overview

Venus Sosa, Teresa Moliné, Rosa Somoza, Rosanna Paciucci, Hiroshi Kondoh, Matilde E. LLeonart

Ageing Research Reviews 12 (2013) 376–390

Oxidative Stress in Cancer

U. Jakob and D. Reichmann (eds.), Oxidative Stress and Redox Regulation,
DOI 10.1007/978-94-007-5787-5_15,
© Springer Science+Business Media Dordrecht 2013

Peter Storz

Oxidants, antioxidants and the current incurability of metastatic cancers

Jim Watson

Open Biol. 2013 3, 120144, published 9 January 2013

Overcoming Drug Resistance Through Elevation of ROS in Cancer

Amit K. Maiti

B. Bonavida (ed.), Molecular Mechanisms of Tumor Cell Resistance to Chemotherapy,
Resistance to Targeted Anti-Cancer Therapeutics 1, DOI: 10.1007/978-1-4614-7070-0_7,
© Springer Science+Business Media New York 2013
“We propose to target the antioxidant mechanism of tumor adaptation by an anticancer therapy…by treating cancer cells either with ROS-inducing therapies or with antioxidant inhibiting therapies.”
The vast majority of all agents used to directly kill cancer cells (ionizing radiation, most chemotherapeutic agents and some targeted therapies) work either through directly or indirectly generating ROS that block key steps in the cell cycle....

“A common ROS-mediated way through which almost all anti-cancer agents induce apoptosis explains why cancers that become resistant to chemotherapeutic control become equally resistant to ionizing radiotherapy.”

Plasma RONS-based therapy must offer advantages over existing therapies!

Can plasma therapy overcome resistance??
NO\textsubscript{x} Cancer Therapy: A Possible Mechanism for Plasma to Overcome Resistance?

Need Reliable Targeting in Tumors at High Concentration

*Targeting nitric oxide for cancer therapy*
David Hirst and Tracy Robson
JPP 2007, 59: 3–13
NOx leads to radio- & chemo-sensitization or apoptosis
Surface Microdischarge (SMD) Air Plasma

Diagram showing the components of the SMD air plasma system, including a powered copper electrode, Teflon block, glass dielectric barrier, grounded steel mesh, plasma region, treated surface, and acrylic reactor. A graph below shows absorbance peaks for various compounds such as N₂O, NO, NO₂, HNO₃, and O₃, with wavenumbers on the x-axis ranging from 800 to 2400 cm⁻¹.
$\text{N}_2\text{O}$ and NO Concentrations

$\text{N}_2\text{O}$

$\text{NO}$
NO$_2$ and O$_3$ Concentrations

NO$_2$

O$_3$
Efficient NOx-Generating Plasma

in 150 μL water
Plasma Increases Local Blood Flow/O$_2$ Concentration *In Vivo*

Plasma jet-induced tissue oxygenation: potentialities for new therapeutic strategies

Possible role of NO generated via plasma/NO$_2^-$ route?
NO$_2$(aq) + NO$_2$(aq) + H$_2$O(l) $\rightarrow$ NO$_2^-$ + NO$_3^-$ + 2H$^+$

NO(aq) + NO$_2$(aq) + H$_2$O(l) $\rightarrow$ 2NO$_2^-$ + 2H$^+$.

NO$_2^-$ + H$^+$ $\rightarrow$ HNO$_2$

2 HNO$_2$ $\rightarrow$ 2 N$_2$O$_3$ + H$_2$O

N$_2$O$_3$ $\rightarrow$ NO + •NO + •NO$_2$

NO, NO$_2$ and OH in gas phase will form H$_2$O$_2$; NO$_2^-$; H$^+$; and NO$_3^-$ in water.

NO$_2^-$; H$^+$ will result in **NO and NO$_2$**.

H$_2$O$_2$; NO$_2^-$ and H$^+$ will form **ONOO$^-$**.

(UVA photolysis of NO$_2^-$ can replace acid; Pavlovich et al. 2013)

(Lundberg et al., 2008; Lukes et al., 2014)
NOx leads to radio- & chemo-sensitization or apoptosis: can plasma do the same?
Inorganic nitrite therapy: historical perspective and future directions

Christopher G. Kevil, Gopi K. Kolluru, Christopher B. Patillo, Tony Giordano

Free Radical Biology & Medicine 51 (2011) 576–593

**NO$_2^-$ (nitrite) as systemic therapy:**

What are implications for plasma therapy?

- Cerebral Vasospasm: ↑ vasodilation
- Pulmonary Hypertension: ↑ vasodilation
- Ischemia / Reperfusion Injury (heart, brain, liver, kidney):
  - ↑ cytoprotection
  - ↑ heme oxygenase-1
  - ↑ mitochondrial function
  - ↑ tissue preconditioning
- Cystic Fibrosis / BioFilm Infection:
  - ↑ bacterial killing
- Gastric Ulcer:
  - ↑ mucus formation
  - ↑ ulcer healing
- Inflammatory Bowel Disease:
  - ↑ colitis
  - ↓ TNF-α
  - ↓ colon shortening
- Sickle Cell Disease:
  - ↑ blood flow
- Hypertension:
  - ↓ mean arterial pressure
- Peripheral Artery Disease:
  - ↑ angiogenesis
  - ↑ arteriogenesis
- Hypercholesterolemia:
  - ↓ triglycerides
  - ↓ low density lipoproteins
  - ↓ leukocyte recruitment

**Endogenous:**

- 200-600 nM blood
- low µM in tissue
- low mM in saliva
Plasma-Generated NO\textsubscript{x}: Does it Match Known NO/NO\textsubscript{2}/NO\textsubscript{2}⁻/ONO\textsubscript{2}⁻ Biochemical Effects?

Biochemistry literature shows NO/NO\textsubscript{2}/NO\textsubscript{2}⁻/ONO\textsubscript{2}⁻ have important effects.

Early results suggest plasma-generated NO\textsubscript{x} may give similar results.

One vision of plasma biomedicine: *plasma acts as a RONS delivery device, suitable for localized therapy in cancer and other disease treatments like infected tissue, wound healing, etc.*
Key Question

Even though plasma kills tumors with pro-oxidant mechanism (like existing therapies), if resistance develops in plasma therapy as it does for current RONS-based therapies, there will be no great advantage to plasma cancer treatment.

NOx-based therapy shows promise for treating radiation- and chemo-resistant tumors and we know air plasma can create similar chemistry.

But will this promise eventually translate to plasma therapy in clinical trials?
Summary of Ideas

- Atmospheric pressure, low temperature air plasmas create large quantities of reactive oxygen and nitrogen species, as well as photons, electric fields and charges; RONS effects probably dominate biochemically.

- Plasma can deliver locally high concentrations of key RONS to cells at tissue surface; RONS-based cell-cell communication probably transmits effects to adjacent tissue.

- $\text{H}_2\text{O}_2/\text{OH}$ are key ROS; $\text{NO}/\text{NO}_2/\text{NO}_2^-/\text{ONOO}^-$ are key RNS.