Ambient Plasma Treatment of Cancer Cells

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Abstract

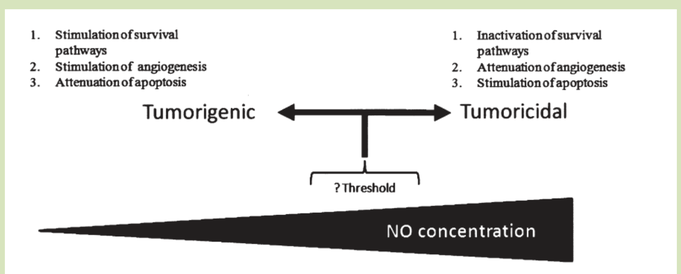
Ambient gas plasmas are cytotoxic to cancer cells and bacteria at least in part because they create reactive oxygen and reactive nitrogen species (RONS). It is known that these reactive species can also play important roles in conventional therapies, including cancer chemotherapy and antibiotics. For example, anti-tumor synergy between NO-donating compounds and some redox-active chemotherapeutics has been documented [1]. We therefore explored the possibility that plasma-generated RONS could act synergistically against cancer and bacteria with conventional chemotherapeutic agents.

Motivation

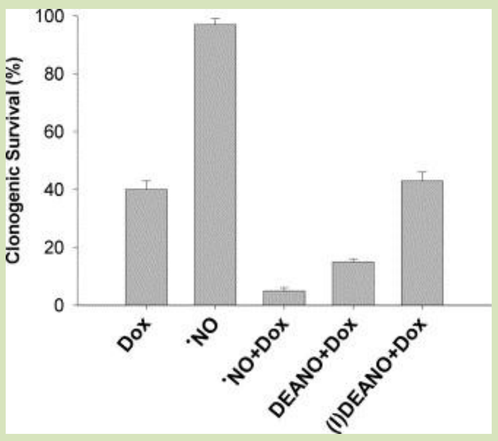
- Anti-cancer therapies are generally delivered simultaneously, usually as drug cocktails. Synergies increase cocktail effectiveness.
- Combining therapies and taking advantages of synergies can prevent the development of resistance to chemotherapeutics.
- Simultaneous plasma treatment and chemotherapy may attenuate existing resistance to chemotherapeutics.

Background

Reactions of NO and O_2 produce RONS, causing oxidative and nitrosative stress in the cell. The effects of this stress include DNA strand breaks, and inhibition of DNA repair, including DNA ligase.

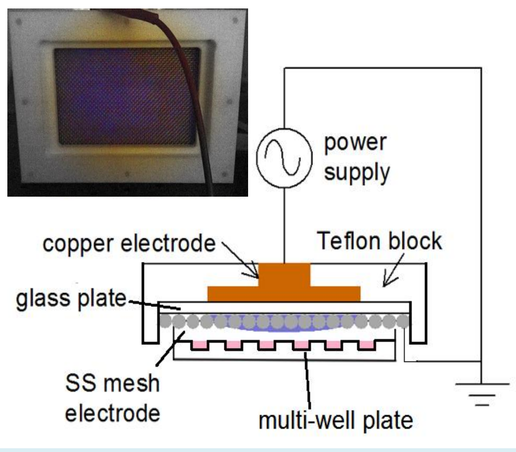


Sufficiently high concentrations of NO are known to be tumoricidal[2]. However, exposure to low doses of NO before treatment with chemotherapeutics such as doxorubicin and cisplatin increase the sensitivity of cancer cells to the chemotherapeutics. Notably , this effect has been observed for drugs known to have an RONS mechanism[1].



Doxorubicin and NO act synergistically. Treatment of MCF7 cells with different NO donors followed by doxorubicin drastically increased the toxicity of doxorubicin. [1]

Materials and Methods



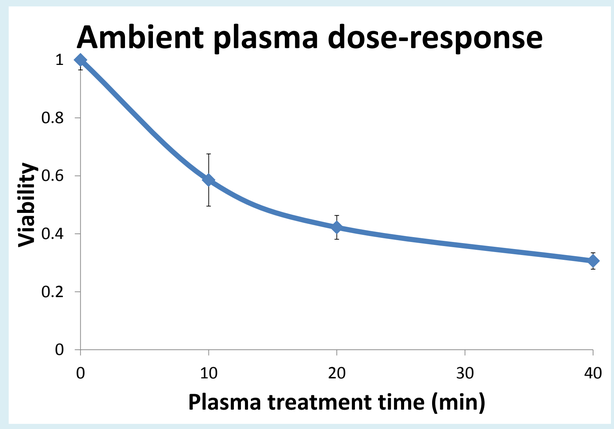
- Dielectric barrier discharge device
- Operated in indirect mode
- Operating conditions: 5kV, 5 kHz
- Power: 0.15-0.2 W/cm^2

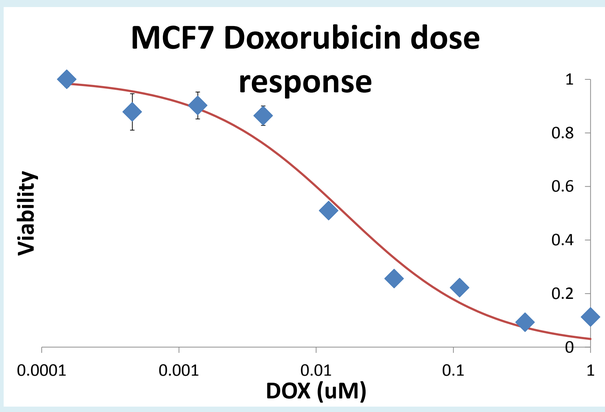
Procedure:

- Plasma treat cells (MCF7 breast cancer line) covered with media (DMEM high glucose), incubate for two days
- Change media, incubate for three days more, and measure viability with Alamar blue fluorescence

Results

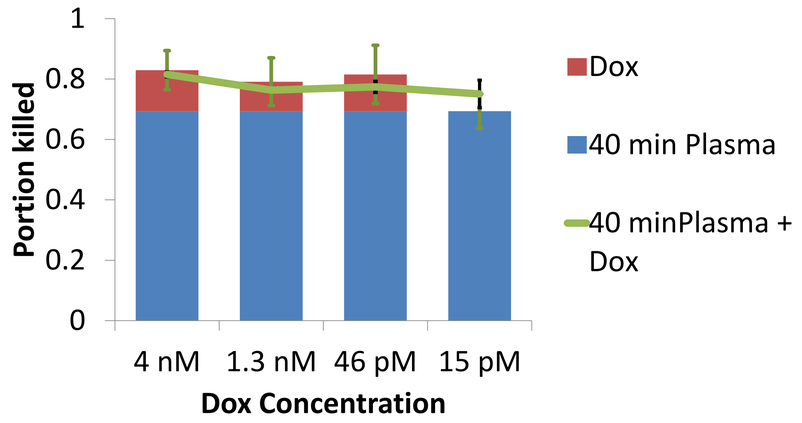
- Ambient plasma treatment of MCF7 breast cancer cells kills 50% of cells after 15 minutes.
- Exposure times are much longer than comparable studies in the literature [3] because cells are completely immersed in growth medium.
- Cells covered only by a thin layer of medium are completely killed after 4 minutes of exposure.
- Doxorubicin dose -response is fitted to a Hill equation, giving an IC50 of 16 nM.





- Plasma treatment of cells following addition of doxorubicin shows that the two effects are at least additive.
- For doxorubicin concentrations ranging from 15 pM-4nM, plasma treatment under these conditions does not impact doxorubicin cytotoxicity.

Additive effects of doxorubicin and plasma treatment



Conclusions

- For low concentrations of doxorubicin, plasma treatment and doxorubicin treatment are additive.
- More data are required to determine if synergy exists between ambient plasma treatment and doxorubicin, and if so, in what range of treatments.
- While studies on NO-donating compounds support the existence of synergy between doxorubicin and NO, our methodology may need to be changed to reveal it.
- In particular, changing the order of plasma treatment and chemotherapy, and the plasma treatment conditions (power and growth medium coverage) may be essential.
- Similarly, glutathione depletion may allow for better resolution of any synergy, facilitating determination of statistical significance.

References

1. CB Evig et al. *Nitric Oxide*. **2004,** *10*, 119–129.
2. S Huerta et al. *Int J Oncol.* **2008,** *33,* 909-927.
3. Sensenig et al. *Ann Biomed Eng*. **2011**, *39*, 674-687.