Electrochemotherapy Augments Pancreatic Ductal Adenocarcinoma Tumor Cell Killing by Inducing Apoptosis and Disrupting Cell Adhesion

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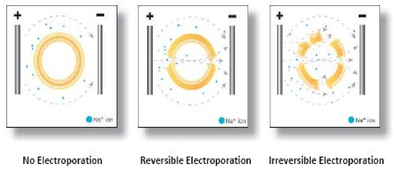
Background

- Stage III PDAC is an extremely lethal disease with a 5 year survival of only 3%
- Irreversible electroporation (IRE) is a surgical technique that consists of delivering controlled pulses of electricity to the tumor, and has been shown to improve survival in stage III PDAC patients
- Standard of care chemotherapy regimens include Gemcitabine and FOLFIRINOX
- No previous work has been done to study the mechanistic actions underpinning the benefits of IRE in combination with standard of care chemotherapy



Schematic of Irreversible electroporation demonstrating disruption of the plasma membrane after exposure to the high voltage, short duration electric field. This porous membrane will result in higher intracellular chemotherapy concentrations.[1]

Objectives

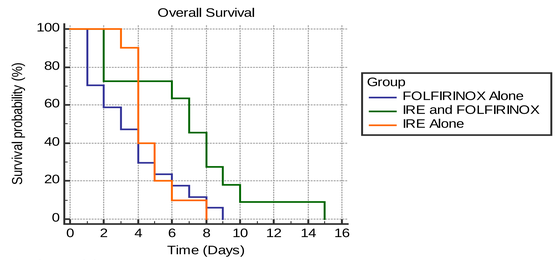
- The objective of this study was to investigate the tumor cell killing potential and mechanisms of electrochemotherapy (ECT) in comparison to IRE or chemotherapy alone

Methods

- Clinically relevant doses of IRE, chemo, or ECT were administered to S20-13 cells and PanC-1 PDAC cell lines *in vitro*
- Cells were assessed for post-treatment viability via colony assay
- Apoptosis proteins (caspases and BCL-2) were analyzed via western blot
- The tight junction protein Claudin-1 was assessed by Immunofluorescence
- Identical treatments were used *in vivo* in an athymic nude mouse model inoculated with S20-13 cells (conducted by Bhutiani, et al.)

Results

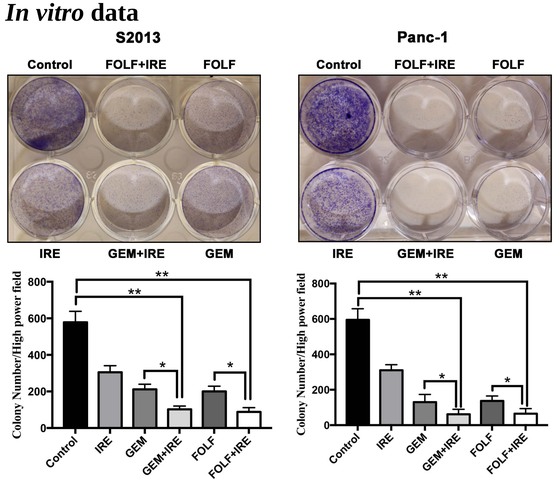
In vivo data



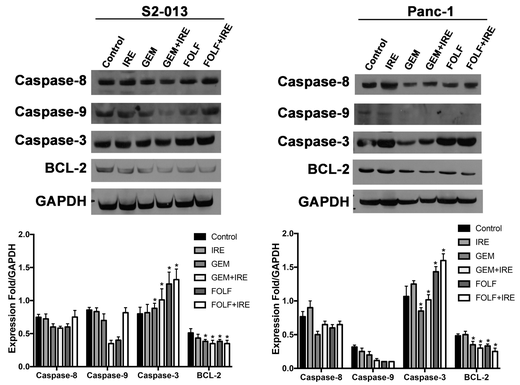
Mouse experiments conducted by Bhutiani, et al. demonstrated a clear survival benefit in those treated with ECT compared to IRE or chemo alone[2].

Results

In vitro data



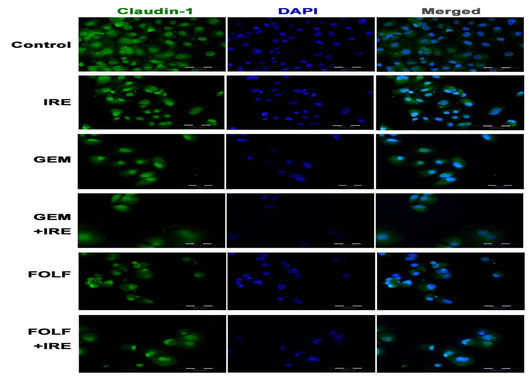
Colony assay for both cell lines reveals significantly reduced cell viability after ECT treatment compared to control, IRE alone or chemo alone ($p<0.01$).



Western blot data for both cell lines shows significantly increased caspase-3 expression in response to ECT ($P<0.01$). Additionally, the anti-apoptotic BCL-2 protein was downregulated in ECT groups ($p<0.01$).

Results

In vitro data



Immunofluorescence images of S20-13 cells demonstrating reduced expression of claudin-1 in response to ECT, implying disruption of the cell-cell adhesions.

Conclusions

- ECT demonstrated a survival benefit in a mouse model of stage III PDAC
- *In vitro* cell viability was synergistically diminished by ECT treatment in comparison to IRE or chemo alone
- Mechanistically, the benefit of ECT involves upregulation of apoptotic machinery (namely caspase-3) and disruption of the intercellular tight junctions (claudin-1)

Resources

1) Meijerink, M. *'the NanoKnife' Irreversible Electroporation-from bench to bedside*. Angiodynamics Lunch Symposium, CIRSE. 2015.

2)Bhutiani, et al. *Electrochemotherapy with irreversible electroporation and FOLFIRINOX improves survival in murine pancreatic adenocarcinoma by increasing apoptosis and decreasing tumor cell proliferation.* AACR. 2018

Acknowledgements

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