PDL-1 blockade and Sunitinib enhance the efficiency of oncolytic viral therapy
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Introduction
Oncolytic viruses such as reovirus (RV) are non-pathogenic viruses, which specifically target and lyse cancer cells due to genetic abnormalities, with no effect on normal cells. Recently, RV has been used in hundreds of human clinical trials in the form of monotherapy or in combination with chemotherapy against different histological malignancies. The challenge of these trials is the elicitation of anti-viral immune response, which results in viral clearance. Moreover, the uses of immunosuppressive agents have only resulted in modest improvement. Immune checkpoint receptors such as programmed cell death 1 ligand (PDL-1) that is upregulated on the surface of cancer cells, binds to the programmed cell death 1 (PD-L1) receptor on the surface of activated cytotoxic T lymphocytes (CTL) and results in inhibition of the antitumor T-cell response.

Hypothesis and Rationale
1. Oncolytic Virus
   - Infect and replicate in tumour cells
   - Cell Lysis and Virus replication
   - Lysis, T&G release, pro-inflammatory and cytokines release
   - Enhanced immune cell infiltration

2. Reovirus and Sunitinib Combination on Breast and Lung Cancer Cell lines Viability
   - Cells were infected with constant value of ED50 for reovirus (MDI) and sunitinib (uM) and incubated for 48 hours.
   - Cytotoxicity was detected by measuring mitochondrial NADPH dehydrogenase using WST assay

3. Effect of Reovirus and Sunitinib Combination on Breast and Lung Cancer Cell lines PDL-1 expression
   - Cells were either infected with live reovirus, UV inactivated reovirus and/or Sunitinib and left stimulated or not with IFN-γ for 24 hours
   - PDL-1 expression was analyzed by surface flow cytometry

Materials and Methods
WST-1 In Vitro assay for detecting 50% of cell death
ED50 for Reovirus and Sunitinib

Results
1. Surface Expression of PDL-1 in Established Breast and Lung Cancer Cell lines
   - Cells were either stimulated or not with IFN-γ for 24 hours and PDL-1 expression was analyzed by surface flow cytometry

Summary and Conclusion
1. PDL-1 is differentially expressed in lung and breast cancer cell lines.
2. Sunitinib enhances the sensitivity of Reovirus killing in some breast and lung cancer cell lines.
3. Both Sunitinib and Reovirus differentially modulate PDL-1 expression on some breast and lung cancer cell lines

Future Directions
1. In vivo assessment of OV administration in combination with PDL1 blockade plus or minus sunitinib in the established murine cancer models.
2. FACS assessment of circulating MDSC and memory CD8+ T cells levels in spleen after combinational treatment.
3. Assessment of CD69+ mononuclear splenocytes from established murine cancer model after treatment and adoptive transfer of mononuclear splenocytes from donor mice into untreated murine cancer model.
4. Randomized Phase II Reovysin™ clinical trials (Breast and Non-Small Cell Lung Carcinoma) correlating MDSC enumeration with outcome.

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