PDL-1 blockade and Sunitinib enhance the efficiency of oncolytic viral therapy

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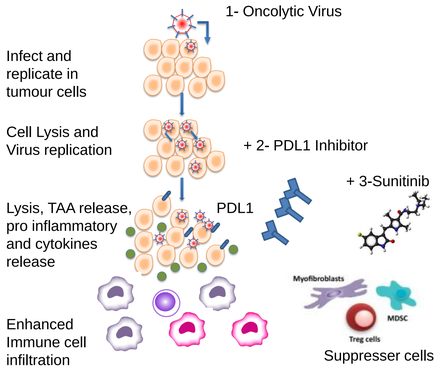
Introduction

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-1) receptor on the surface of activated cytotoxic T lymphocytes (CTL) and results in inhibition of the antitumor T-cell response.

Hypothesis and Rationale



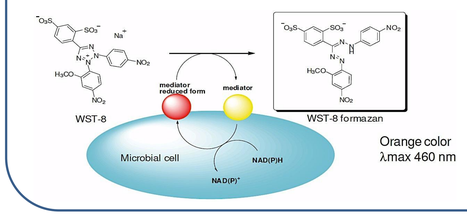
Objectives

1- In vitro assessment of effects on cytotoxcity and progeny release when virotherapy administration is combined with sunitinib against established breast and lung cancer cell lines.

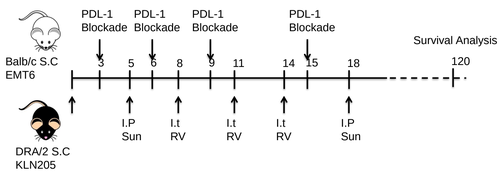
2 - In vivo assessment of Reovirus administration in combination with PDL1 blockade plus or minus Sunitinib in the established syngenic murine models for cancer.

Materials and Methods

WST- In Vitro Assay for detecting 50% of cell death
ED50 for Reovirus and Sunitinib

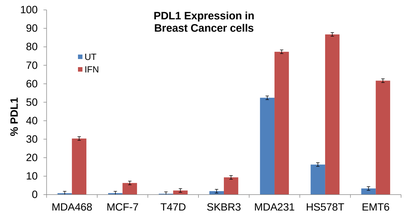


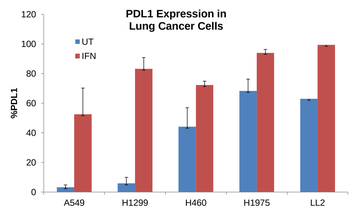
Orange color
λmax 460 nm



Results

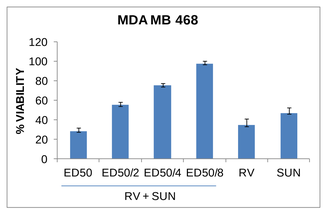
1- Surface Expression of PDL-1 in Established Breast and Lung Cancer Cell lines

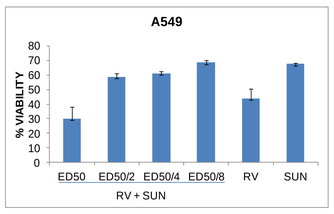


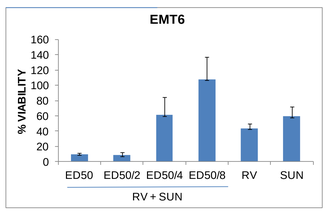


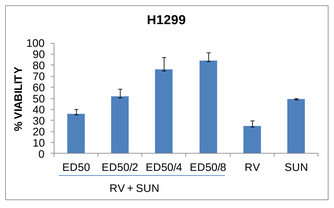
Cells were either stimulated or not with IFN-γ for 24 hours and PDL-1 expression was analyzed by surface flow cytometry.

2- Effect of Reovirus and Sunitinib Combination on Breast and Lung Cancer Cell lines Viability



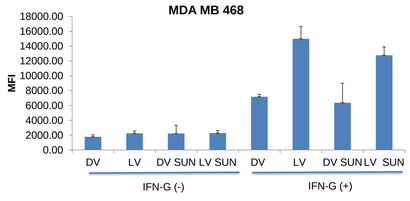


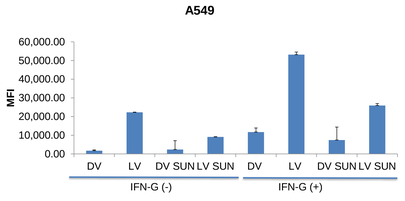


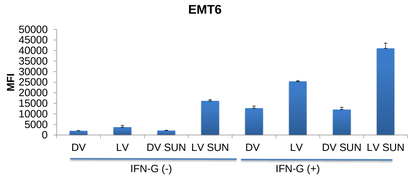


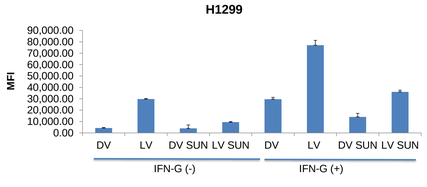
Cells were infected with constant value of ED50 for reovirus (MOI) and sunitinib (uM) and incubated for 48 hours.
Cytotoxcity was detected by measuring mitochondrial NADPH dehydrogenase using WST assay

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

Summary and Conclusion

1. PDL-1 is differentially expressed in lung and breast cancer cell lines.
2. Sunitinb 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

To the best of our knowledge, the idea of combination RV, PDL-1 and sunitinib have never been tried; however all the three mentioned therapies had been used solely before in clinical trials and in treatment of cancer. Taken together, these data will provide new treatment strategies with resultant improved efficacy and safety of our breast and lung cancer patients with the possibility of approaching phase I/II clinical trials.

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 CD8+ 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 Reolysin™ clinical trials (Breast and Non-Small Cell Lung Carcinoma) correlating MDSC enumeration with outcome.

Acknowledgments

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