Correlating Immune Status to Therapy Efficacy in Murine Tumor Models

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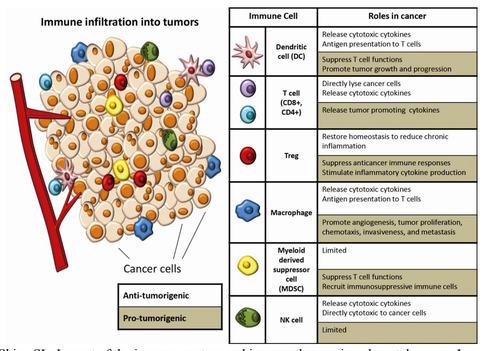
ABSTRACT

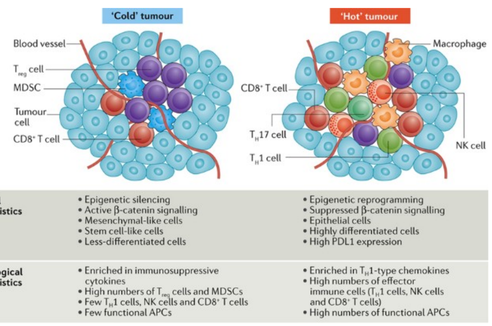
It is well established that there is a great deal of interplay between a patient's tumor and their immune system and this interplay can dictate overall outcomes and survival. As new therapeutics move from the pre-clinical to the clinical stage, data shows that some therapies, specifically immunotherapies, can be quite effective but only in small subsets of patients. Understanding why immunotherapies fail in a large majority of patient populations and how to make those populations into responders has become the aim of many. The immune phenotype of the tumor at the time of therapy initiation and during the treatment regimen can play a large role in responders. These data suggest that patients who have "*hot*" or "*inflamed*" tumor microenvironments, as marked by active immune infiltration, have a greater probability to respond to immunotherapy. Likewise, patients who develop a pro-immunogenic phenotype in their tumor microenvironment and/or periphery during the course of therapy also tend to have better overall outcomes. Based on these observations, many have geared attention towards immunophenotyping patients prior to and during therapeutic regimens to understand their tumor immune status to try and predict outcomes.

To this end, we have been phenotyping the immune status of the tumor microenvironment and peripheral organs such as the spleen in pre-clinical syngeneic tumor models. By understanding the immune status at the time of therapy initiation and the change in immune status with therapeutic intervention overtime, we hope to correlate these immune phenotypes to tumor burden and/or survival. The aim of this endeavor is to ultimately help predict responding patient populations and increase better overall outcomes.

BACKGROUND

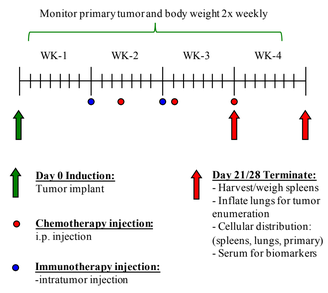


Shiao SL. Impact of the immune system and immunotherapy in colorectal cancer. J Gastrointest Oncol 2015;6(2):208-223.

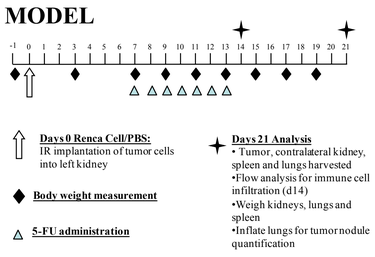


MODEL/METHODS

4T1 MODEL



RENCA MODEL



4T1 MODEL RESULTS

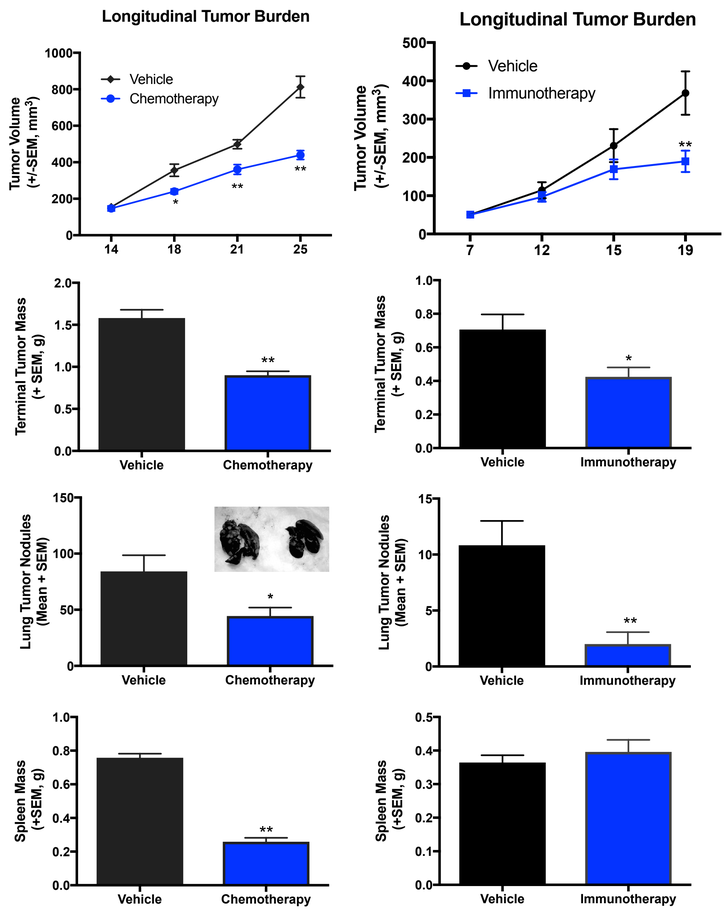


Figure 1: Standard Efficacy Data for Model. Animals were implanted with 4T1 tumor cells subcutaneously in the flank. Animals were then treated with either an immunotherapy or chemotherapy per model schematic. Tumors were monitored longitudinally with calipers. At termination, primary tumors and spleens were weighed and lungs inflated with India Ink and metastatic nodules manually counted. *p<0.05; **p<0.01.

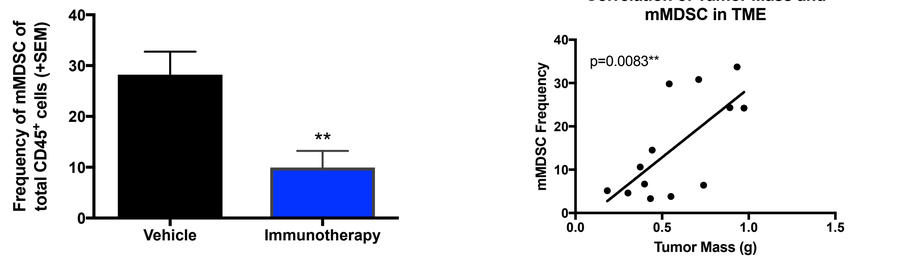


Figure 2: Example of Immunophenotyping at termination for immunotherapy treatment. Animals were implanted and treated similar to Figure 1. At termination, primary tumors were digested and prepared for flow cytometric analysis for assessment of immune cell populations including CD8 T cells and MDSC. The frequency of MDSC in the TME was correlated to primary tumor mass using Pearson Correlation Coefficient. **p<0.01.

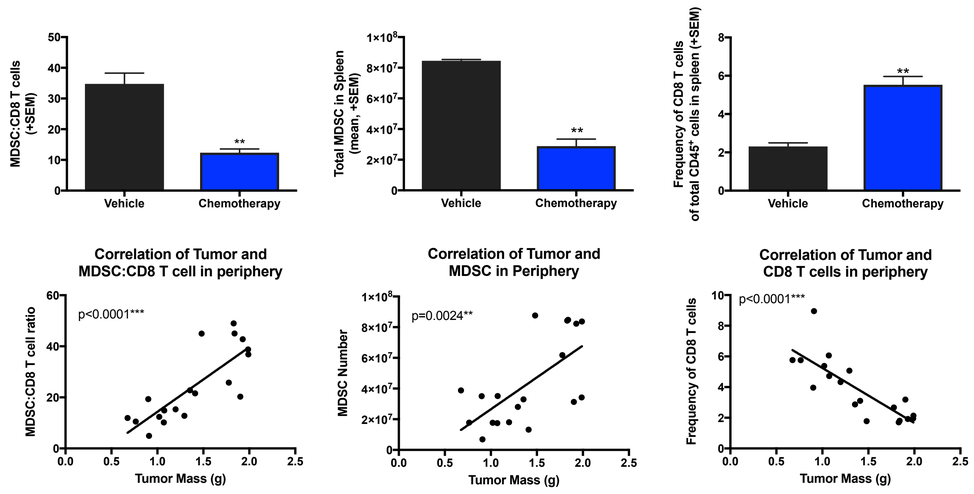


Figure 3: Example of Immunophenotyping at termination for chemotherapy treatment. Animals were implanted and treated similar to Figure 1. At termination, spleens were digested and prepared for flow cytometric analysis for assessment of immune cell populations including CD8 T cells and MDSC. The frequency of MDSC, CD8 T cells and their ratio in the periphery (spleens) were correlated to primary tumor mass using Pearson Correlation Coefficient. **p<0.01; ***p<0.0001

RENCA MODEL RESULTS

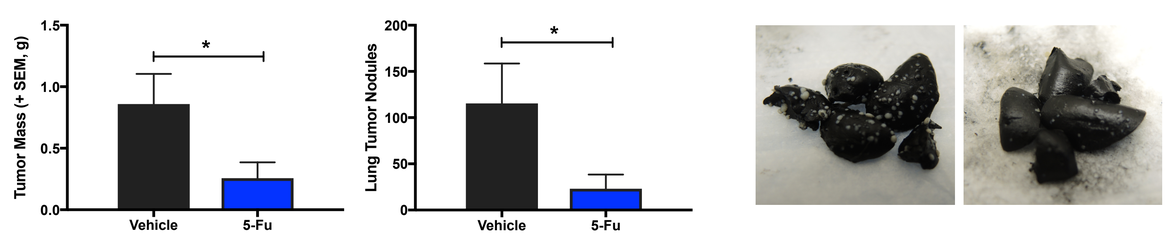


Figure 4: Standard Efficacy Data (d21 harvest). Animals were implanted with Renca tumor cells into the left kidney through the peritoneum. Animals were then treated with 5-FU chemotherapy per model schematic. At termination, primary tumors, and contralateral tumors were weighed and lungs inflated with India Ink and metastatic nodules manually counted. *p<0.05; **p<0.01

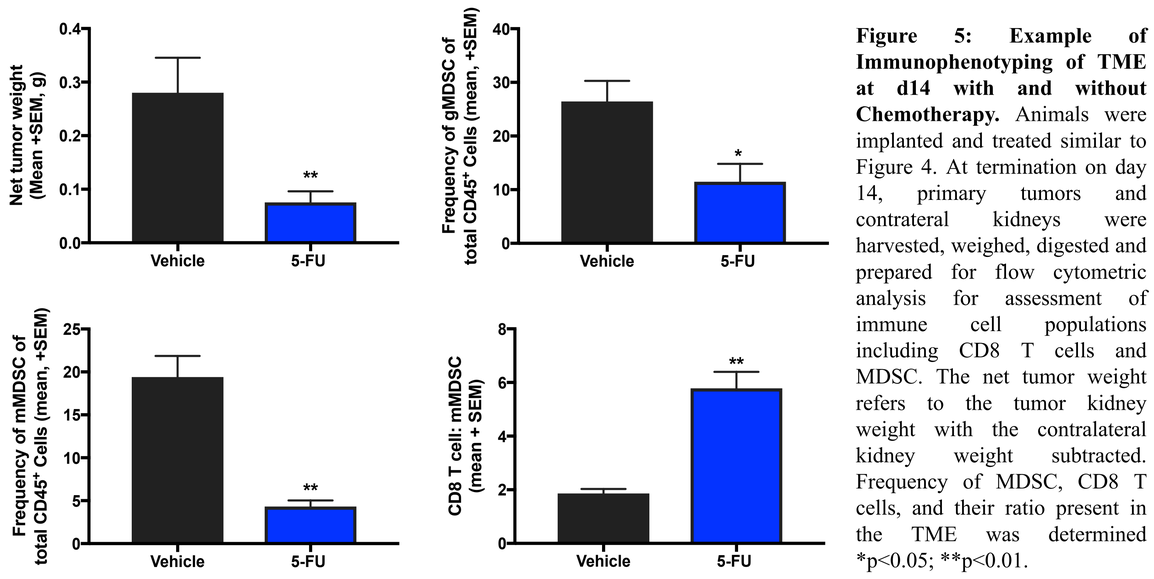


Figure 5: Example of Immunophenotyping of TME at d14 with and without Chemotherapy. Animals were implanted and treated similar to Figure 4. At termination on day 14, primary tumors and contrateral kidneys were harvested, weighed, digested and prepared for flow cytometric analysis for assessment of immune cell populations including CD8 T cells and MDSC. The net tumor weight refers to the tumor kidney weight with the contralateral kidney weight subtracted. Frequency of MDSC, CD8 T cells, and their ratio present in the TME was determined *p<0.05; **p<0.01.

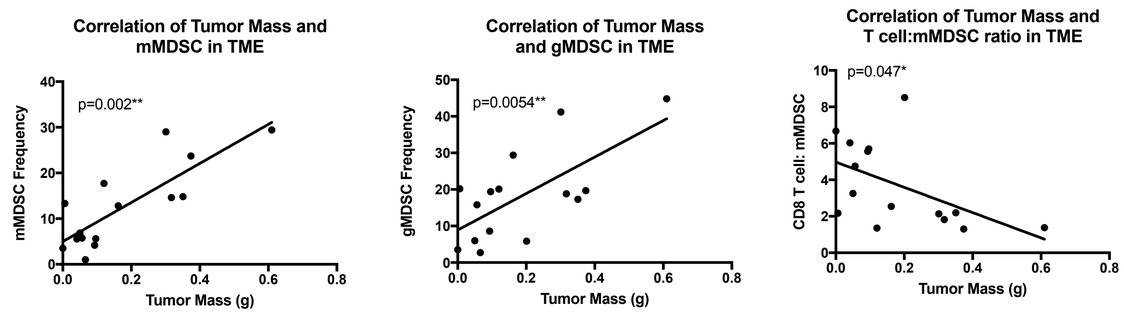


Figure 6: Example of Immune Status and Tumor Mass Correlation. Animals were implanted and treated similar to Figure 4. At termination primary tumors were digested and prepared for flow cytometric analysis for assessment of immune cell populations including CD8 T cells and MDSC. The frequency of MDSC, CD8 T cells and their ratio in the TME were correlated to primary tumor mass using Pearson Correlation Coefficient. *p<0.05; **p<0.01

CONCLUSIONS

4T1 Model

- 4T1 primary and metastatic tumor burden can be controlled with both chemotherapy and immunotherapy dosing regimens.
- The frequency of immunosuppressive cells, MDSC, positively correlates with tumor burden in the 4T1 model with immunotherapy treatment: a decreased MDSC population in the TME correlates with decreased tumor burden.
- The frequency of MDSC in the periphery positively correlates with tumor burden while the frequency of effector CD8 T cells in the periphery negatively correlates with tumor burden.

RENCA Model

- Renca primary and metastatic tumor burden can be controlled with a 5-FU chemotherapy dosing regimen.
- The frequency of immunosuppressive cells, MDSC, in the TME positively correlates with tumor burden with a chemotherapy dosing regimen.
- The frequency of effector CD8 T cells in the primary TME negatively correlates with tumor burden with a chemotherapy dosing regimen.
- 5-FU, or similar chemotherapeutic, may "prime" the immune system and TME to respond to subsequent or concomitant immunotherapy treatment.

- **In total these data suggest that syngeneic tumor models can be characterized to predict and/or correlate treatment efficacy to not only tumor burden but immune status in the TME and periphery.**

CONTACTS