Endogenous T Cell Redistribution in the Presence of Immuno-Oncology (I-O) Therapies

Farah Al Qaraghuli, WanYing Zhang, Xiaoying Yu, Dhaval K. Shah

Department of Pharmaceutical Science, School of Pharmacy and Pharmaceutical Sciences, University at Buffalo, The State University of New York, Roswell Park Cancer Institute, Buffalo, NY

Background

T lymphocyte is a subtype of white blood cell, which plays a central role in cell-mediated immunity. There are several subtypes of T lymphocyte within the body, and each has a distinct function in defending the body. Each subset of T lymphocyte also has a unique role in therapies. For example, CD4+ T lymphocyte plays a central role in the immune response, as they act by communicating and instructing other immune system cells. CD8+ T lymphocytes are cytotoxic and fight infected cells and cancer cells. These cells are further divided into memory and effector subsets.

Method

1. Prepare single cell suspensions of spleen, lymph node, blood, lung, bone marrow, and liver tissues using CountBright™. When comparing the three methods used to count total cells for staining, counts obtained from hemacytometer were very different than MACSQuant analyzer 10 and CountBright beads based methods. Especially in the peripheral tissues (liver and lung).

2. Different subsets of T lymphocytes were successfully identified using the staining method employed and analysis strategy performed on BD LSRRFortessa.

3. Cytotoxic T lymphocyte (CD8+) cells show higher central memory phenotype compared to the helper T lymphocyte (CD4+). Helper T lymphocyte has more effector/effector-memory cells compared to cytotoxic T lymphocytes.

Conclusion

Single cell suspensions of tissues were successfully obtained following mechanical and enzymatic dissociation. Absolute counts were obtained with CountBright®. When comparing the three methods used to count total cells for staining, counts obtained from hemacytometer were very different than MACSQuant analyzer 10 and CountBright beads based methods. Especially in the peripheral tissues (liver and lung).

Different subsets of T lymphocytes were successfully identified using the staining method employed and analysis strategy performed on BD LSRRFortessa. Absolute quantification of T cell subsets will be obtained at 1, 3, 5, and 14 days after each therapy. Mathematical PKPD model will be used to characterize the data.

Future direction

The C57Bl/6 mice will be inoculated with MC38 tumor cell line and will be treated with anti-CEA-Anti-CD3 bisppecific antibody, anti-PD-1 antibody, anti-CTLA-4 antibody, anti-OX40 antibody, and anti-GITR antibody. Absolute quantification of T cell subsets will be obtained at 1, 3, 5, and 14 days after each therapy. Mathematical PKPD model will be used to characterize the data.

References


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