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Summary of research accomplishments in 2013:

Thanks to the support of the Steven G. AYA Cancer Research Grant, I have been able to successfully transition from Dr. Kenneth Cooke's laboratory into Dr. Alex Huang's laboratory in the Division of Pediatric Hematology-Oncology at Case Western Reserve University School of Medicine since November 2012. Below is a summary of my research activities since joining Dr. Huang's laboratory during year 1 of the Steven G. AYA Cancer Research Grant funding period.

Project 1. *Examining the role of CCR5 expression in promoting the development of memory $CD8^+$ T cells.*

In previous studies from our laboratory, we identified an important role for the up-regulation of the chemokine receptor CCR5 in a subset of T cells that leads to the development of memory T cells. Continuing these studies, I am identifying the mechanisms that promote the up-regulation of CCR5 in $CD8^+$ T cells and determining how CCR5 expression leads to increased development of memory T cells. Success in this project will lead to the development of better cancer vaccines with long-lasting memory.

Project 2. *Examine the biology of intracellular Notch-1 expression in the development and function of T cell acute lymphoblastic leukemia (T-ALL)*

The major problem with treatment against T-ALL is the presence of a few cancer cells results in minimal residual disease (MRD), which can lead to relapse. Presence of cancer cells in the central nervous system (CNS) and in bone marrow after hematopoietic stem cell transplant (HSCT) can result in MRD. Using a model of T-ALL that was developed in our lab, I am currently characterizing two T cell populations from T-ALL that differ in the expression of CD4 ($CD4^-CD8^+$ = $CD8^+$ or $CD4^+CD8^+$ = DP). Because migration of T-ALL into CNS has been shown to be dependent on the expression of chemokine receptor CCR7 and its ligand CCL19, I examined expression of CCR7 and response to CCL19 by both T-ALL cell populations. While the DP population showed reduced expression of CCR7, DP T cells showed increased response to CCL19, suggesting that signaling pathway of CCR7 was different in these cells compared to $CD8^+$ T cells. This is important as DP population also showed increased migratory capacity into the CNS. This suggests that unique subpopulation of T-ALL cells may be responsible for migration into CNS and the establishment of MRD that may lead to relapse.

In collaboration with Dr. Goutham Narla, M.D., Ph.D. at the Harrington Institute, I am examining the effects of a novel class of drugs that promote protein phosphatase 2a (PP2A)

activation. This is a class of compounds derived from drugs that are already FDA approved, but are not currently being used to fight cancer. While many proteins that are involved in the development of cancer require modification through the attachment of the phosphate group, most current therapy is designed to inhibit these specific phosphorylation steps that are unique to a small number of proteins. Because more than one protein may be involved in cancer, targeting specific phosphorylation sites may help the cancer develop resistance to chemotherapy. By activating PP2A, which can target multiple sites for de-phosphorylation, we are able to target multiple oncogenic pathways making resistance less likely. I have shown that PP2A activation suppresses proliferation of T-ALL and promotes apoptosis of these cells. We will be using these drugs in animal models to look at their effects in the setting of minimal residual disease (MRD).

In collaboration with Dr. John Letterio, I am examining the effects of a class of anti-inflammatory drugs known as triterpenoids in preventing relapse and enhancing engraftment after hematopoietic stem cell transplantation (HSCT) as a treatment for leukemia and other malignancies. We will be looking at the ability of triterpenoids to limit the effect of MRD that can lead not only to relapse but may also reduce efficiency of hematopoietic stem cell engraftment.

Project 3. *Examining the role of Cdk5 in regulating graft-versus-host disease (GVHD).*

In collaboration with Dr. John Letterio, we are examining how the protein Cdk5 regulates donor T cells during the initial stages of GVHD. In previous work, I showed that Cdk5 dramatically influences T cell response to CCR7. I will be examining the ability of donor T cells to enter Lymph node and interact with host dendritic cells when Cdk5 is not functional. This is important as the interaction between donor T cells and host dendritic cells is needed to generate GVHD. These studies may show that Cdk5 is a potential target in HSCT for minimizing the risk of developing GVHD.

My ultimate goal as a researcher, with the critical support by the Steven G. AYA Cancer Research Award is to understand the interaction between tumor cells and the immune system to identify novel strategies that will result in better treatments. While tumor cells exhibit several mechanisms to subvert or hide from the immune system, we are increasing our ability to understand these mechanisms with the goal of enhancing the power of the patient's immune system to eliminate the tumor. Support from the Steven G. AYA Cancer Research Award has given me this tremendous opportunity to work with Dr. Huang and the lab environment that he has created, which is the BEST environment possible to achieve these goals.

Poster Presentations in 2013:

Askew D, Su CA, Barkauskas D, Myers J, Liou R, Chang N, and Huang AY. 2013. Dynamic regulation of CCR5 expression in naïve T cells within inflamed lymph nodes is essential for memory CD8 T cells. Case *ShowCase,* May 2013

Askew D, Othman Y, Durand D, Barkauskas D, Myers J, Wang G, Zhou L, and Huang AY. 2013. Identification and characterization of $CD8^+$ and $CD4^+CD8^+$ T cell acute lymphoblastic leukemia cells generated through constitutive expression of intracellular NOTCH1. Case Cancer Center Retreat, July 2013

Askew D, Othman Y, Durand D, Barkauskas D, Myers J, Wang G, Zhou L, and Huang AY. 2013. Identification and characterization of $CD8^+$ and $CD4^+CD8^+$ T cell acute lymphoblastic leukemia cells generated through constitutive expression of intracellular NOTCH1. Case Immunology Training Program, September 2013 (Selected as 1 of 3 Best posters)

Askew D, Othman Y, Durand D, Barkauskas D, Myers J, Wang G, Zhou L, and Huang AY. 2013. Identification and characterization of $CD8^+$ and $CD4^+CD8^+$ T cell acute lymphoblastic leukemia cells generated through constitutive expression of intracellular NOTCH1. Critical Mass: The Young Adult Cancer Alliance, November 2013

Accolades:

Askew D, One of three recipients for Best Poster Presenter at the 7th Annual CWRU Immunology Training Program Retreat held on September 27, 2013 at the Lerner Research Institute of the Cleveland Clinic.

Manuscript (In Press):

1. Scrimieri, F, **Askew D**, Corn DJ, Eid S, Bobanga ID, Bjelac JA, Tsao ML, Othman YS, Wang SG, Huang AY**.** Murine Leukemia Virus Envelope Gp70 is a Shared Biomarker for High-sensitivity Detection and Quantification of Murine Tumors. OncoImmunology, 2013 (in press).

Manuscripts in Preparation:

1. **Askew D**, Pareek T, Eid S, Myers J, Keller M, Guirdo-Wolff R, Huang AY, Letterio JJ and Cooke KR. A novel role for lymphocyte expression of cyclin-dependent kinase 5 (Cdk5) in the generation of allogeneic T cell responses after BMT.

2. **Askew D,** Su CA, Nthale J, Liou R, Barkauskas D, Myers J, and Huang AY. Transient surface CCR5 expression in naïve T cells within inflamed lymph nodes is dependent on LFA-1 and augments helper T-cell dependent memory response

3. **Askew D,** Eid S, Keller M, Guirdo-Wolff R, and Cooke KR. Conventional splenic dendritic cell subsets direct development of graft-versus-host disease.

Research Support:

Active:

1) NIH R01 HL111682: Novel Mechanism of immune activation following allogeneic hematopoietic stem cell transplant. P.I. John Letterio

2) Steven G. AYA Cancer Research Grant. **P.I. David Askew**

Pending:

1) NIH R21 NCI: Role of triterpenoids in regulating relapse and engraftment after stem cell transplant. **P.I. David Askew**