2008 Rare Diseases Bench-to-Bedside Awards

1. Development of Immunotherapeutic Strategies to Overcome Tolerance in Leukemia

Principal Clinical Investigator/Institute	A John Barrett M.D., Hematology Branch, National Heart, Lung, and Blood Institute (NHLBI), National Institutes of Health (NIH)
Principal Basic Scientist/Institution	Katy Rezvani M.D., Ph.D., Imperial College School of Medicine, London, UK
Other Investigators/Institution/Institute	Jane Apperley M.D., Philip Ashton-Rickardt Ph.D., Imperial College School of Medicine, London, UK; Agnes Yong, M.D., Ph.D., Quan Le M.D., Ph.D., Hematology Branch, NHLBI, NIH

Abstract

Hematological malignancies are most common in individuals over 60 years who are least responsive to current treatments. Immunotherapy strategies could improve outcome in patients otherwise refractory to standard treatment. Most leukemia associated antigens (LAAs) that could be potential targets of immunotherapy regimens are overexpressed non-mutated self-antigens and as such are likely to induce immunological tolerance. We hypothesize that efficient leukemia vaccination may be achieved, provided that ideal LAA are used and the optimum immunological conditions to overcome tolerance are established. PRAME, Wilms tumor (WT1) and proteinase 3 (PR3) are tumor antigens of particular interest since they are widely expressed in hematological malignancies. We propose to investigate a multi-epitope vaccine approach, combining HLA-A*0201 restricted epitopes derived from PR3, WT1, and PRAME. We will use this multi-epitope vaccine approach as a platform to study and manipulate the immune response and overcome tolerance to achieve a successful vaccine-induced anti-leukemia effect.

2. Predicting the Response to Treatment Using Gene Mutation Profiling in Metastatic Melanoma Patients

Principal Investigator/Institute	Yardena Samuels, Ph.D., Cancer Genetics Branch, National Human Genome Research Institute (NHGRI), NIH
Associate Investigator/Institution	Phillip Buckhaults, Ph.D., University of

	South Carolina School of Medicine
Associate Investigator/Institution	Patrick Hwu, M.D., University of Texas, M. D. Anderson Cancer Center

Abstract

Despite years of research, the average survival time of patients with metastatic melanoma is less than 10 months, and new treatment strategies are needed. Pharmacological inhibition of activated kinases is a promising novel approach to cancer therapy. Fifty percent of metastatic melanomas harbor activating mutations in the Serine/Threonine kinase, BRAF, making it an especially promising target for inhibition. Additionally, sixty percent of melanomas contain activated AKT3, over half of which are caused by somatic inactivation of the PTEN tumor suppressor gene. Unknown molecular defects in one or more members of the PIK3CA pathway are presumed to be responsible for AKT3 activation in the remaining tumors. Finally, PTEN mutations, and AKT3 activation, result in activation of mTOR kinase, and its downstream targets. With evidence accumulating that both BRAF and the mTOR pathways are important to melanoma progression, these pathways may be appropriate therapeutic targets. Novel compounds have been developed that inhibit BRAF and mTOR. Sorafenib is a potent inhibitor of the BRAF kinase, and may be an effective treatment for melanomas harboring activating mutations of this enzyme. CCI-779 is an inhibitor of mTOR, and may be an effective treatment for melanomas harboring mutations in PTEN or other members of the PI3K pathway. We hypothesize that patients with activating mutations in both of these pathways are most likely to respond to treatment with a combination of these two compounds. An NIH-CTEP sponsored clinical trial utilizing a combination of sorafenib + CCI-779 in patients with metastatic melanoma is currently underway. Our interest is to look for the presence of somatic mutations in genes that lie in the BRAF and PI3K pathways (a total of 30 genes) and correlate positive response to treatment to the presence of somatic mutations in these pathways. If response to therapy is found to associate with somatic mutations, this information can be relayed back to the bedside to select patients based on gene mutations. The identification of novel genetic changes that underlie melanoma will allow us to better understand the phenotypes of individual tumors and better predict patients' responses to therapy.

3. Recombinant IL-7 (CYT107) as Immunomodulatory Therapy for Idiopathic CD4 Lymphopenia: A Phase I/IIa Open-label Pilot Study

Bench/Translational: **Basic Science Principal Investigator/Institute**	Jason Brenchley, Ph.D., Laboratory of Molecular Microbiology (LMM), National Institute of Allergy & Infectious Diseases (NIAID), NIH
Associate Investigator/Institute	Jake Estes, Ph.D., AIDS Vaccine Program, National Cancer Institute (NCI), NIH
Associate Investigator/Institute	Jessica Hodge, B.S., Laboratory of Immunoregulation (LIR), NIAID, NIH
Associate Investigator/Institute	Brian Porter, M.D., Ph.D., M.P.H., LIR, NIAID, NIH

Associate Investigator/Institute	Peter Mannon, M.D., Laboratory of Host Defenses (LHD), NIAID, NIH

Abstract

Idiopathic CD4 lymphopenia (ICL) is a rare (affecting ~0.05% of adults), heterogeneous disorder characterized by persistently low CD4 T cell counts in the absence of an underlying primary infectious cause, such as HIV-1 or HIV-2. ICL patients are at risk for potentially life-threatening opportunistic or otherwise severe infections, autoimmune diseases, and malignancies. In addition to aggressive treatment for concurrent infections, novel immunomodulatory therapies are needed to address underlying deficits in T cell-mediated immunity, as concurrent CD8 lymphopenia appears to confer an even worse prognosis. Exogenous human recombinant interleukin-7 (rhIL-7), a master regulatory cytokine of both central and peripheral T cell development, is one such candidate therapy. Our hypothesis is that IL-7 treatment will improve T cell function and T cell numbers and decrease morbidity in patients with ICL. Initially, we will characterize extensively the immunophenotype of specific lymphocyte subsets from preexisting samples of peripheral blood lymphocytes (PBL) from ICL patients. This analysis will include proliferative responses to IL-7 and other common γ chain (γc)-utilizing cytokines as well as the ability of these cytokines to enhance T cell responses to specific pathogens that commonly infect ICL patients. These studies will be followed by a prospective, Phase I/IIa clinical trial of exogenous IL-7 (CYT107) to treat ICL patients with a history of serious infection but not autoimmunity, evaluating clinical measures to determine the safety (primary outcome: adverse events) and biologic activity (secondary outcomes: numerical and functional immune reconstitution) of this novel immunotherapy.

4. Characterization of Jak/Stat Activation in Patients with Monosomy 7 and the Development of Targeted Therapy for Patients Using a Jak2 Inhibitor

Principal Investigators/Institute	Elaine M Sloand, M.D., Neal Young, M.D., Hematology Branch, NHLBI, NIH
Principal Scientific Investigator/Institution	Jerome Groopman, M.D., Chief of Experimental Medicine, Beth Israel Deaconess Medical Center
Co-investigators/Institute/ Organization	Ramesh Kumar, Ph.D., President and CEO of Onconova Therapeutics Inc., Susan Leitman, M.D., Deputy Director, Department of Transfusion Medicine, Clinical Center (CC), NIH Loretta Pfannes, B.S., post-baccalaureate fellow, Hematology Branch, NHLBI Phillip Scheinberg, M.D., Staff Clinician, Hematology Branch, NHLBI Mathew Olnes, M.D., Clinical Associate, Hematology Branch, NHLBI

Abstract

Patients with bone marrow failure frequently develop cytogenetic abnormalities following immunosuppressive treatment. Monosomy 7 is most frequent and is associated with severe cytopenias and a high propensity to develop leukemia. Patients who do develop acute myelogenous leukemia (AML) are more difficult to treat and relapse more quickly or die of infection. Previous work from this laboratory showed that an abnormal granulocyte colony stimulating factor receptor (GCSFR) was present on hematopoietic cells from patients with Monosomy 7. This receptor though defective at low GCSF concentrations facilitated the expansion of monosomy 7 cells at high concentrations of granulocyte colony stimulating factor (GCSF). At ambient concentrations of GCSF, present in healthy individuals, monosomy 7 cells showed decreased proliferation and increased cell death. Recently, our laboratory demonstrated increased numbers of monosomy 7 cells (measured by fluorescence in situ hybridization) in bone marrows of severely neutropenic patients with aplastic anemia, who had high endogenous GCSF concentrations. The behavior of monosomy 7 cells at high concentrations of GCSF, which contributes to their expansion, is likely a result of constitutive expression of Jak2 and STAT1 in these cells. The purpose of this study is to: 1) determine if monosomy 7 is present in low frequency in the bone marrows of normal individuals; 2) to explore the possible clinical utility of using a Jak2 inhibitor (ON44580 provided by Onconova) as targeted therapy to suppress monosomy 7 hematopoietic colony formation. Should pre-clinical studies be successful we aim to establish a clinical pilot study in patients with monosomy 7 and MDS.

5. Graft-Versus-Host Disease: Novel Cellular Therapy Using Selective Thawing Of Umbilical Cord Blood to Obtain an Aliquot for Ex-Vivo Natural Killer Cell Expansion and Infusion Following Allogeneic Hematopoietic Stem Cell Transplantation

Principal Clinical/Translational Investigator/Institute	Richard Childs, M.D., Hematology Branch, NHLBI, NIH
Principal Basic Scientist/Center	Sumithira Vasu, MD, Department of Transfusion Medicine, CC, NIH

Abstract

Umbilical cord blood (UCB) transplants have been increasingly used as a source of hematopoietic stem cells for patients with hematological disorders requiring an allogeneic stem cell transplant (SCT) who lack an HLA matched sibling or unrelated donor. Graft-versus-Host disease (GVHD), an orphan disease, is a major obstacle to successful allogeneic stem cell transplantation. Severe GVHD resulting in morbidity and death occurs in up to 15% of recipients of an UCB transplant. Recently, natural killer (NK) cells have been shown to play an important role in preventing GVHD. Our group has observed that an adoptive infusion of allogeneic in vitro expanded NK cells reduced GVHD and improved survival in recipients of a T-cell replete SCT. Further, in collaboration with the NIH Clinical Center's Department of Transfusion Medicine,

we have developed a method to expand NK cells by greater than 500 fold in vitro under GMP (Good manufacturing practices) conditions. Although umbilical cord blood units are easily available for transplantation, adoptive NK cell and/or T-cell infusions at the time of transplantation are currently not possible, since the entire cord blood unit is infused during the transplant, eliminating the donor cell source from which these cells would be expanded. The inability to thaw a small portion of the UCB unit to expand NK cells while maintaining the viability of the non-thawed portion of the cord unit has precluded investigators from evaluating the impact of adoptive NK cell infusion following UCB transplantation. Here we propose to design a device that selectively thaws a very small portion of the UCB unit while maintaining cold temperatures in the remainder of the non-thawed unit. The ability to successfully acquire a small sample from the cord unit without compromising the integrity/sterility or viability of the unthawed portion of the cord unit would provide the opportunity to expand ex vivo cord blood NK cells. Expanded UCB NK cells will then be adoptively infused at the time of UCB transplantation in a clinical trial that evaluates whether an NK cell infusion can be used to reduce GVHD and improve survival in patients with hematological disorders undergoing UCB transplantation.

6. Evaluation of the Platelet Transcriptome Expression Profile in Pulmonary Arterial Hypertension

Translational Research Principal Investigator/Institute	Roberto Machado, M.D., Pulmonary and Vascular Medicine Branch, NHLBI, Critical Care Medicine Department, CC, NIH
Basic Science Principal Investigator/Center	Nalini Raghavachari, Ph.D., Critical Care Medicine Department, CC, NIH
Co-investigators/Institute/ Center/Institution	Mark T. Gladwin, M.D., Pulmonary and Vascular Medicine Branch, NHLBI, NIH; Michael J Cuttica, M.D., Critical Care Medicine Department, CC, NIH; Steven D. Nathan, M.D., Advanced Lung Disease and Transplant Program, Inova Fairfax Hospital

Abstract

Pulmonary Arterial Hypertension (PAH) is a rare, debilitating and fatal disease for which there is currently not a cure. PAH can be classified as either idiopathic associated with an underlying illness which worsens the morbidity and mortality of those suffering from the underlying illness. These classifications are linked by common histologic changes and cellular dysfunction of the vasculature of the lung. Proliferation of smooth muscle and endothelial cells, which normally exist in a quiescent state leads to remodeling of the vessels with obliteration of the lumen of the pulmonary vasculature. Within the lesions characteristic of PAH fibrin, thrombin and platelets are present. These pathologic changes cause a progressive rise in pulmonary vascular resistance as blood is pumped through an ever-decreasing vascular area, producing an abnormal workload on the right ventricle. Overwhelmed by this

workload the right ventricle eventually fails and leads to premature death in these patients. In situ thrombotic lesions and platelet dysfunction are important in PAH as it further impairs pulmonary vascular flow. Recent evidence also suggests that interactions between platelets and the pulmonary artery wall can contribute to the functional and pathologic vascular changes associated with PAH. We have validated methodologies to characterize the global transcriptome of amplified platelet RNA derived from human peripheral blood. To this end we will characterize the transcriptome of amplified platelet RNA in patients with PAH in comparison to those of patients with other forms of pulmonary hypertension and healthy individuals. We will also evaluate the effects of specific treatment of PAH with pulmonary vasodilators and anti-remodeling agents on the global transcriptome of amplified platelet RNA derived from these patients. These studies will provide an improved understanding of the intrinsic signaling pathways that affect the transcriptome of platelets in general and in patients with pulmonary arterial hypertension. These investigations have the promise of increasing the understanding of the interactions between platelets and the pulmonary artery wall and their potential effects on the pathogenesis of PAH. Finally we hope that this approach will help identify novel prognostic markers and therapeutic targets for this devastating disease.