NEW INVESTIGATOR AWARDS

Majed M. Hamawy, PhD
University of Wisconsin, Madison, WI
$75,000 – *Targeting the Signaling Molecule LAT for Preventing Graft-Versus-Host Disease*
Graft-Versus-Host Disease (GVHD) remains one of the major complications of bone marrow transplantation and is a significant cause of morbidity. GVHD occurs when donor T cells containing the donor bone marrow become activated upon interacting with the organs of the recipient. Thus, blocking the activation of T cells is important for preventing GVHD. Current immunosuppressive drugs have reduced the incidence of GVHD; however, these drugs cause side effects and toxicities because they target and inhibit the function of many different types of cells. LAT is one of the proteins involved in T cell activation and is found primarily in T cells. The aim of our project is to examine if blocking LAT expression in T cells inhibits T cell activation, and in turn, prevents the development of GVHD. Because LAT is found primarily in T cells, blocking its expression should affect mainly T cells. This, in turn, could sharply reduce the side effects and toxicity associated with the use of current immunosuppressive drugs. Preventing GVHD without the side effects associated with current immunosuppressive drugs would have an enormous clinical impact by enhancing patient longevity and quality of life.

Scott T. Handy, PhD
State University of New York at Binghamton, Binghampton, NY
$64,171 – *Stegane: A Scaffold for New Anti-Cancer Agents*
In light of the importance and challenge in treating all forms of cancer (including leukemia), this proposal seeks to develop a new class of anti-cancer compounds. These compounds will address the difficulties associated with the variability and drug resistance in cancer cells by acting at multiple cellular targets. The structure for these compounds is based on the general framework of the stegane family of natural products. The steganes have been known as potential anti-cancer drugs for over 20 years and are reported to interact with the same cellular target – tubulin – that the highly useful anti-cancer drugs taxol and vinblastine do, although at a different site. At the same time, the steganes have similarities to drugs that interact with other cellular targets and also compounds that are effective against resistant cancer cell lines. By preparing hybrid molecules that contain features present in the steganes and these other anti-cancer agents, compounds can be created that have the potential to target cancer by multiple mechanisms. A major challenge in realizing this therapeutic potential is that previous synthetic efforts have only prepared the steganes with great difficulty. This proposal utilizes a much shorter and more flexible method to synthesize the steganes, which will enable rapid exploration of the stegane structure itself as well as hybrid molecules. In combination with an existing means of testing these compounds, this new route will enable the full exploration of the chemotherapeutic potential of this family of compounds and ideally develop a new class of versatile anti-cancer agents.

Barbara L. Kee, PhD
The University of Chicago, Chicago, IL
$75,000 – *Analysis of the Mechanism of E2A-Mediated Suppression of T Cell Lymphoma*
The E2A proteins, E12 and E47, are transcription factors that regulate the expression of genes required for the proper growth, survival and differentiation of developing lymphocytes. Mice that lack E2A proteins do not make B lymphocytes and produce a reduced number of T lymphocytes. Despite the decreased number of lymphocytes, these mice invariably develop T cell lymphoma. In humans, greater than 60% of T lymphoblastic leukemias have alterations that lead to the loss of E2A activity demonstrating the importance of these transcription factors in suppressing lymphocyte cancers. The goal of the research proposed in this application is to determine how E2A proteins prevent the development of leukemia and lymphoma and to identify the targets of E2A's tumor suppressing abilities. Our studies will provide significant insight into the mechanisms underlying tumor formation and may provide targets for therapeutic intervention in T cell leukemia and lymphoma.

Michelle A. Kelliher, PhD
University of Massachusetts Medical School, Worchester, MA
$75,000 – *The Mechanism(s) of TAL-1/SCL Mediated Leukemogenesis*
TAL-1 activation is the most common gain of function mutation associated with pediatric T cell acute lymphoblastic leukemia (T-ALL). In contrast to T-ALL with activation of *HOX-11* gene, T-ALL patients that express TAL-1 respond poorly to conventional therapy, with greater than 50% of patients relapsing after standard therapy. Although much has been learned about the normal function of TAL-1 function in regulating blood cells, less is known about how TAL-1 causes leukemia. The goal of this research project is to apply genetic methods in the mouse to define how TAL-1 contributes to leukemia. This research project will lead to an improved understanding of TAL-1 mediated transformation and ultimately to the development of more specific therapies for T-ALL.

Tannishtha Reya, PhD
Duke University Medical Center, Durham, NC
$75,000 – *Regulation of Hematopoietic Stem Cell Self-Renewal by Wnt Signaling*
Every day, billions of new blood cells are produced in the body, each one derived from a hematopoietic stem cell (HSC). Because most mature blood stem cells have a limited lifespan, the ability of HSC to perpetuate themselves through self-renewal and generate new blood cells for the lifetime of an organism is critical to sustaining life. A key problem in hematopoietic stem cell biology is how HSC self-renewal is regulated. We propose to determine whether HSC self-renewal is regulated by the Wnt signaling pathway. Our studies will not only lend insight into normal self-renewal but also into oncogenesis, since emerging evidence suggests that similar signaling pathways regulate the self-renewal of normal stem cells and the uncontrolled self-renewal of cancer cells. Progress in the basic understanding of regulation of HSC self-renewal has significant practical ramifications since identification of factors that influence HSC growth will be a critical step towards defining ways to expand stem cells *in vitro*, and thereby improve transplantation based therapies for leukemia and other cancers.

Charles P. Scott, PhD
Kimmel Cancer Center, Philadelphia, PA
$74,935 – *Activating Apoptosis in Leukemia and Lymphoma*
Every day, millions of white blood cells are generated by the immune system, but most of them are eliminated by programmed cell death processes that are essential to prevent the development of autoimmune diseases. One important cell death process involves two groups of similar proteins (called Bcl proteins) with opposite effects on cell death. Bcl-mediated cell death is regulated by balancing the levels of Bcl proteins that promote cell death with those that prevent it. Leukemia is often the result of upsetting this balance by producing too much of a Bcl protein imbalance found in cancerous cells. Others have shown that peptides from Bcl proteins that promote cell death (Bcl homology domain 3 or BH3 peptides) can be used in place of the whole protein. Although these smaller molecules are leads toward the development of drugs to treat leukemia, they are not ideal molecules to use as drugs. Peptides don't enter cells easily, so we are testing a number of different modifications that should improve the ability of BH3 peptides to enter cells and turn off Bcl proteins that prevent cell death. Peptides are also degraded rapidly in the body, so we are modifying the structure of BH3 peptides to slow down this degradation. Finally, peptides cannot be taken by mouth so we are developing efficient screening methods to identify other small molecules with the same effects on cancerous cells, but with better properties for use as drugs.

Trygve Tollefsbol, PhD, DO
University of Alabama at Birmingham, Birmingham, AL
$75,000 – *Telomerase Regulation in Terminallyl Differentiating Human Leukemia Cells*
Telomerase is an enzyme that stabilizes chromosomes and has been found to be elevated in leukemia cells but not in normal cells. This enzyme allows leukemia cells to continue proliferating indefinitely leading to many of the features of cancer. Considerable interest has been focused on the role of telomerase in leukemia cells and how the gene that encodes this enzyme is regulated in these cells. Our studies as well as those of others have shown that the telomerase gene becomes inactivated when cancer cells are treated with compounds such as retinoic acid (a form of vitamin A). Telomerase inhibition through these methods causes leukemia cells to slow their growth and differentiate into benign cells that lose their cancer properties. Our aim is to elucidate the early therapeutic anticancer modalities utilizing gene therapy will be facilitated in the treatment of leukemia. Control of the telomerase gene has tremendous potential in the treatment of most if not all leukemias and it is the goal of this study to understand the basic biology involved in the regulation of this important gene.

Carthene R. Bazemore Walker, PhD
University of Virginia, Charlottesville, VA
$52,367 – *2-Methoxyestradiol-Induced Phosphorylation in Human Jurkat Leukemia Cells*
Tumor cells are characterized by a lack of normal control over cell growth, and leukemic cells exhibit this characteristic in that they proliferate in an uncontrolled and invasive manner. The normal life cycle of the cell is extended because it has mutated in a way that allows it to forgo the natural process of cell death (apoptosis). The treatments that are currently used to manage leukemia damage the cells, causing them to die. Disappointingly, these drugs do not have the ability to distinguish between normal cells and those of leukemia. However, a naturally occurring estrogen derivative, 2-methoxyestradiol shows significant promise as a chemotherapeutic agent. It kills a wide variety of tumors, appears to be nearly nontoxic to normal cells, and does not have the unpleasant side effects of standard chemotherapy treatments. This is a special combination of features that could be further exploited if we had a better understanding of exactly how the compound works. The goal of Dr. Bazemore-Walker's research is to more clearly define the mechanism by which 2-methoxyestradiol causes apoptosis in leukemia cells. She will use a combination of biochemical and state-of-the-art analytical techniques to facilitate this process.

PHYSICIAN-SCIENTIST AWARDS

Karl Hsu, MD
Dana-Farber Cancer Institute, Boston, MA
$90,000 – *Zebrafish Models of FLT3 Leukemias*
This proposal uses the zebrafish to determine the significance in blood development and leukemia of mutations in the FLT3 gene, a class III receptor tyrosine kinases that is abnormal or mutated in 20-30% of acute myeloid leukemia (AML) patients. It has been proposed that AMLs are caused by more than one mutation in the same cell. These mutations include a mutated tyrosine kinases gene causing abnormal cell growth and survival and a second mutation, which prevents the cell from maturing. Dr. Hsu will examine the effect of a mutated FLT3 tyrosine kinase in zebrafish white blood cells. He will also establish zebrafish families that have a mutated FLT3 gene in their white blood cells and monitor these fish for the development of AML. These families are vital for the investigator's future goal of discovering additional important genes which interact with the FLT3 gene. Discovery of these genes may lead to discovery of mutated genes in the 60-70% of AMLs that do not have an FLT3 mutation. These genes may be future drug targets in the treatment of AML.

Aleksandra Kirovski, MD
Memorial Sloan Kettering Cancer Center, New York, NY
$90,000 - *T cell Homing in Graft-versus-Host Disease*
Hematopoietic stem cell transplantation is an effective treatment for leukemia and other hematopoietic malignancies. A major complication of stem cell transplantation is graft-versus-host disease (GVHD), in which cells, namely T cells from the donor attack the host organs; in particular liver, intestine, and skin. The donor T cells have to home to these target organs, a process believed to be regulated by special proteins in the vasculature (blood vessels), on the target organs and on the surface of the donor T cells. We will use mouse bone marrow transplantation models to further define the role that these proteins play in the development of GVHD. We will employ T cell selection, knockout mice which lack certain homing proteins and neutralizing antibodies to specifically inhibit homing proteins and their receptors. We will analyze the effect of this selective inhibition of T cell homing on the development of GVHD and graft-versus-tumor activity (GVT), which is also dependent on donor T cell activity. With these studies, we hope to pave the way for more organ specific treatment for graft-versus-host disease without interfering with graft-versus-tumor effect.

Matthew J. Walter, MD
Washington University, St. Louis, MO
$90,000 – *The Role of Interstitial Deletion of Chromosome 2 for Murine Acute Promyelocytic Leukemia Progression*
In order to study the biology of acute promyelocytic leukemia, a transgenic mouse has been created that co-expresses two fusion genes found in human acute promyelocytic leukemia (APL). This model produces a fatal myeloid leukemia that is very similar to human APL in 60% of transgenic mice. APL cells that arise in these doubly transgenic mice almost always contain a specific deletion of one copy of chromosome 2 (del 2), which also occurs in mice that develop radiation or benzene-induced AML. These independent models strongly implicate del 2 as an important event in murine AML progression. It is our goal to recreate del 2 in mice using chromosomal engineering techniques in order to study the contribution of del 2 for AML progression in the mouse. Our long-term goal is to determine whether similar genetic lesions contribute to the progression of human AML, and to define new molecular targets for the therapy of patients with this disease.

Andrew P. Weng, MD, PhD
Brigham & Women's Hospital, Boston, MA
$90,000 – *Deltex Antagonism of Notch Signaling in T Cell Development and Neoplasia*
A common theme in cancer is the misregulation of a normal developmental process that goes awry and leads to unchecked cell proliferation. One example of this situation occurs in a subset of acute T cell lymphoblastic leukemia in which a normal cellular signal is delivered in excess by a molecule called Notch. Notch plays an important role in the normal development of the immune system, and a thorough understanding of this role greatly facilitates the design of therapeutic approaches to combating the Notch signal when it is delivered inappropriately. For example, there is another molecule called Deltex that is known to alter Notch signaling in other organisms. By studying the function of Deltex as it relates to Notch, it will allow a deeper understanding of how Notch works both in normal development and in cancer. Moreover, as some previous work has shown that Deltex can block the signal delivered by Notch, Deltex becomes a potentially useful means of combating Notch-driven cancers.

Makoto Yawata, MD, PhD
Stanford University School of Medicine, Stanford, CA
$90,000 – *Study on the Diversity of HLA Class I-Recognizing Natural Killer Cell Receptors*
Immunologicallly, we, as human beings, are all "different" in the sense that our reactions against invading microbes and ever-occurring cancer cells differ between individuals. Natural killer (NK) cells are cells that provide critical defense against compromising malignant transformation or viral infection. A group of receptors called Killer cell Immunoglobulin-like Receptors (KIRs) are important in enabling these cells to distinguish between normal cells of our body and cancerous cells including leukemia cells. Research performed in this laboratory and other institutes is indicating that there are large differences in the KIR genes that has been previously known. This diversity in KIR genes could account for person to person differences in the strength and specificity in the ability of NK cells to recognize and eliminate cancer cells. My research project proposes to conduct an investigation in the differences of these KIR genes between individuals and to determine the functional relevance of these differences. Genetic analysis will be conducted on KIR genes in various subjects to assess differences in these genes in the population. From this analysis, unique KIR types will be found and tested at the cell level to determine the differences in their function. Results from this research hold important clinical implications especially in the treatment of leukemia. NK cells preferentially interact with other blood cells hence the study on the regulation of these cells has particular potential for preventing the progress of leukemia. Concerning bone marrow transplantation, although it has become one of the mainstays in treatment of leukemia, it is still not a robust therapeutical option because there still unknown factors complicating the outcome of this technique. Existing studies indicate that matching KIR genes in addition to the routine HLA allotype matching leads to a high success rate. The drawback is that there is presently no comprehensive data on the many types and combinations of KIR genes contributing to the success of transplantation. Results from this project will provide essential information in this aspect. The importance of this systematic study of the population diversity of KIR genes is not only to study clinical correlation, but also to ensure that the clinical benefits will be available to all patients.

POSTDOCTORAL FELLOWSHIP AWARDS

Jean Marie Bruey, PhD
The Burnham Institute, La Jolla, CA
$60,000 – *NAC Expression and Function in Leukemia*
Leukemic cells arise as a result of uncontrolled cell growth in the body. While cell division certainly plays an important role in this uncontrolled cell growth, it has become clear that the cell accumulation typical of leukemic disorders is also facilitated by a failure of these malignant cells to die. In this regard, the average adult produces and in parallel eradicates 50-70 billion cells each day in his or her body, offsetting the similar amount of daily cell production and thus achieving homeostasis. This cell eradication is achieved through a natural process that triggers cells in the blood, bone marrow, and other tissues to commit suicide, a phenomenon also known as "programmed cell death" and which has also been called "apoptosis."

The development of effective anti-cancer drugs continues to be a challenge for the successful treatment of leukemia and other malignancies, primarily because resistance mechanisms often develop which make it difficult to kill malignant cells. Most chemotherapy drugs eradicate tumor cells by activating the physiological pathway for apoptosis, but treatment failures can result if tumor cells become, or are already, insensitive to apoptosis.

One new avenue to controlling the growth of tumors, including leukemia, is to understand the regulatory components of the apoptosis machinery within cells and to design new drugs to target them specifically, thus restoring apoptosis-sensitivity to cancer cells, and reactivating the natural pathway for cell turnover. Indeed, in many ways, this approach to cancer treatment envisions utilizing cancer-specific aberrations in cell division control mechanisms to selectively trick cancer cells into committing suicide, leaving normal cells intact.

We have discovered a new gene, NAC, that we believe to be a critical regulator of programmed cell death. We have determined that the levels of the NAC protein are increased in normal blood cells as they move closer to their time to die. In this proposal, we hope to understand more about the NAC protein so that we might devise strategies for encouraging leukemia cells to commit suicide, using the same natural pathway responsible for eradication of billions of cells each day in the human body.

Michelle Miranda, PhD
University of Pittsburgh, Pittsburgh, PA
$60,000 – *Role of the MEK/ERK Signaling Pathway in Cytokine-induced Monocytic and Granulocytic Differentiation*
Myeloid leukemias result from the inability of primitive myeloid blood cells to complete maturation or differentiation. As a result, there is a tremendous increase in the number of immature myeloid cells, leading to the development of leukemia. Treatments that promote differentiation of immature myeloid cells hold considerable promise because they promote maturation of the leukemic cells to more mature cells that have limited lifespans. However, the implementation of differentiation therapies has been slow due to incomplete understanding of the signaling mechanisms that drive myeloid differentiation. This proposal will examine the role of a central signaling pathway, the MEK/ERK pathway, in the differentiation of immature myeloid cells into mature cells following treatment with proteins that drive the differentiation process in normal cells. The proposed studies will employ pharmacologic and protein inhibitors of the MEK/ERK pathway to determine whether this pathway is required for myeloid differentiation. In addition, the MEK/ERK pathway will be artificially activated to determine whether this is sufficient to drive the differentiation of immature myeloid cells. Together, these studies will enhance our understanding of the signals that promote normal differentiation and provide targets for developing new therapies that can overcome the differentiation blockades that are key features of myeloid leukemias.

Lisa A. Porter, PhD
University of California San Diego, La Jolla, CA
$60,000 – *Human Speedy – Characterization of a Novel Cell Cycle Regulator*
A central requirement in understanding the causes and mechanisms regulating cancer is determining how cells grow and divide, both in regard to normal proliferation and in response to cellular damage. In mutlicellular organisms, genotoxic stress triggers surveillance mechanisms, referred to as checkpoints, which allow the cell to temporarily exit from the cell cycle and repair DNA. The capacity to delay cell cycle progression prevents the accumulation of genetic alterations leading to neoplastic evolution. Therefore, proteins normally involved in cell cycle progression must function differently following DNA damage to allow a cell to trigger checkpoint responses. This proposal focuses on characterizing a novel human cell cycle protein, designated Speedy (Spy1). Preliminary data indicate that Spy1 is a pivotal protein in controlling both cell proliferation as well as cell survival following DNA damage. The objectives in this proposal are directed at further elucidating the molecular mechanism by which Spy1 functions to enhance normal cell proliferation, as well as determining the role that Spy1 plays in making cellular decisions following DNA damage. Furthermore, we will examine the involvement of Spy1 in oncogenesis through classical oncogenic potential assays as well as determining the extent of protein expression in a variety of tumor tissues. In its entirety, elucidating the function of this protein will contribute significantly to our understanding of both the etiology and the treatment of oncogenesis.

Aparna Raval, PhD
Ohio State University, Columbus, OH
$60,000 – *DNA Methylation in Chronic Lymphocytic Leukemia*
Cancer is a genetic disease. This is true for both solid tumors as well as leukemias. Genetic research in the past focused mainly on genetic alterations in cancer genes. It could be shown that either activation of growth promoting genes (oncogenes) or silencing of growth suppressing genes (tumor suppressing genes) is resulting in a malignant phenotype. This is also true for Chronic Lymphocytic Leukemia (CLL), the most common leukemia in the Western world. Research has identified defects in genes that normally trigger cell death, or apoptosis, of a cell. Altering the genetic code or inactivation in these genes result if uncontrolled growth. More recently, a modification of the DNA, DNA methylation, has been described. DNA methylation does not change the sequence code of genes, however, it results in silencing of tumor suppressor genes. It also has been shown that removal of the methylation reactivates the gene. The contribution of DNA methylation to human malignancies has been completely underestimated and it is now very clear the genetic and epigenetic (DNA methylation) changes contribute equally to malignancies. In this proposal it is planned to study, for the first time, methylation changes in CLL on a genome wide level. Novel methylated genes will be identified and studied in more detail. This research will benefit CLL patients in two ways. First, a better understanding of DNA methylation in CLL will foster the development of therapeutic strategies that use demethylating drugs. These drugs are available and currently under investigation in clinical trials. Second, this project will identify novel CLL related genes and thus improve our understanding of the biology of the disease. This in turn will allow the development of novel therapeutic drugs targeting specific genes, a strategy that usually reduces the toxicity of a treatment.

Wen Tang, PhD
Dana-Farber Cancer Institute, Boston, MA
$60,000 – ***Dissection of the Regulatory Roles of Proto-oncoprotein Gfi-1B Essential for Erythrocyte and Megakaryocyte Development***
Transcription factors are nuclear regulatory proteins that control the expression of other genes. The growth of blood cells is under tight control of transcription factors. In particular, many transcription factors that are essential for normal blood cell development often malfunction in human leukemias, which leads to the abnormal, uncontrolled growth of blood cells. Gfi-1B is a transcription factor that plays an essential role in regulating red blood cell and megakaryocyte development. It is also implicated in the tumor genesis of mouse lymphoid cells. The proposed studies are aimed to understand how Gfi-1B regulates genes in blood cell development and to identify those genes that are regulated by Gfi-1B. These goals can be accomplished by first purifying and analyzing the proteins that interact with Gfi-1B and coordinate Gfi-1B's regulatory function. The target genes of Gfi-1B can be defined by isolating the DNA fragments that Gfi-1B binds. Studies of Gfi-1B-mediated regulation in blood cell development will lead to better understanding of the process of blood cell development and may have two important implications in treatment of leukemia. First, understanding how Gfi-1B functions may suggest the pathways interrupted or perturbed in genesis of leukemia. Second, identification of target genes of Gfi-1B in blood cell development may provide eventual targets for developing new therapeutic strategies to treat leukemia.

Clifford Wang, PhD
University of California, San Francisco, San Francisco, CA
(2nd year funding)
$30,000 – ***Investigating Tumorigenesis and Oncogenes by Directing B-cell Hypermutation***
Researchers have searched long and hard for the magic bullet to treat cancer. For patients with chronic myelogenous leukemia, such a bullet now exits and those patients now have a very good chance of survival. Patients with chronic myelogenous leukemia have a mutation that creates an oncogene, Bcr-Abl. The treatment is a drug that specifically targets and inactivates the Bcr-Abl protein. This example illustrates the importance of identifying oncogenes and the mutations that activate them. When the underlying cause of cancer is known, a drug to target and eliminate that cause can be developed. Point mutations are mutations in which individual base changes are made in DNA. They can be caused by external mutagens such as nitrites, cigarette smoke, and oxygen radicals. They also can result due to imperfect DNA replication and repair, mutations that invariably occur as we age. Currently, there is no systematic method with which to discover cancer causing point mutations and their corresponding oncogenes. By engineering a virus to carry an immuglobulin gene segment that confers high mutability to cellular DNA, we will introduce point mutations and induce leukemia in mice. Using the inserted virus as a genetic tag, we will locate and identify the tumor inducing genes and point mutations. We will attempt to obtain a complete listing of mutations leading to leukemia. In addition, because a gene can be responsible for several types of tumors, this method will likely identify oncogenes not only from leukemia, but also from other types of cancers.