NEW INVESTIGATOR AWARDS

Koichi Akashi, MD, PhD
Dana-Farber Cancer Institute, Boston, MA
$75,000 – *The Role of C/EBPα in Lineage Decision and Leukemogenesis*
Leukemia is a disease characterized by an accumulation of dysfunctional blood cells in the body. The accumulation occurs due to incapability of blood cells to mature, and to their unresponsiveness to death-inducing signals. We will introduce or disrupt several candidate genes related to cell maturation and death into normal mice to identify responsible genes for leukemia development. This approach will help establish a mouse model for acute leukemia that will be eventually useful to develop new treatment strategies for human leukemia.

David Fruman, PhD
University of California, Irvine CA
$73,230 – *Role of PI3K in Transformation by Abl Oncogenes*
Chronic myelogenous leukemia (abbreviated CML) accounts for about 20% of human leukemias. Bcr-Abl, a protein produced when two chromosomes break and rejoin at the wrong place, causes this disease. Bcr-Abl also causes many cases of acute lymphoblastic leukemia (ALL). The current treatments for these leukemias do not cure all patients and cause many side effects. A clear goal is to develop a less toxic treatment based on the molecular mechanism of leukemia development. A potential point of therapeutic intervention is an enzyme known as PI3K. PI3K helps coordinate several cellular processes that are important for growth and survival. Studies from several laboratories, including our own, have shown that PI3K activation plays a crucial role in the growth of cell types that are affected in CML and ALL. The research in this proposal will determine whether a specific from of PI3K is critical for development of CML and ALL. This could lead to new approaches for the treatment of these leukemias and other cancers in humans.

Jessica Tyler, PhD
University of Colorado Health Sciences Center, Denver, CO
$75,000 – *The Role of Chromatin Assembly in V(J)D Recombination*
The goal of this proposal is to better understand how changes in the arrangement of our genome, referred to as chromosomal translocation, occur and contribute to the development and progression of leukemia. Lymphoid cells are unique in undergoing a pre-programmed rearrangement of the genes that encode our immune system, and it appears that mis-regulation of this process leads to the chromosomal translocations that are characteristic of leukemia. It is becoming apparent that the manner in which the genome is packaged in the cell dictates the fidelity and accuracy of the rearrangements of the immune system genes. Accordingly, the proposed research will investigate how the packaging of the genome dictates the accurate pattern of rearrangements of the immune system genes. As such, these studies will help us better understand the aberrant gene rearrangements that lead to chromosomal translocation and leukemia.

Ricardo Aguiar, MD, PhD
Dana-Farber Cancer Institute, Boston, MA
$75,000 – *The Role of PDE 4B in the Biology and Clinical Heterogeneity of Diffuse Large B-Cell Lymphomas*
Additional advances in the treatment of aggressive Non-Hodgkin's Lymphoma will require a more precise understanding of the disease's cellular and molecular heterogeneity. We have recently used a very comprehensive approach to identify genes associated with the outcome in aggressive NHL. We identified a series of genes that are significantly more abundant in fatal diffuse large B-cell lymphoma (DLB-CL) than in curable DLB-CLs. One of the most prominently overexpressed genes in fatal DLB-CL is PDE4B. Based on the know functions of this gene in normal lymphocytes, we hypothesize that PDE4B plays a critical role in the poor response to treatment in fatal DLB-CL. We propose to characterize the functional relevance of this gene in the biology and clinical heterogeneity of NHL. These studies will provide the rationale for the development of new target-specific therapy for lymphomas.

Michael A. Barry, PhD
Baylor College of Medicine, Houston, TX
$75,000 – *Cell Targeting Ligands and Vectors for Chronic Lymphocytic Leukemia*
CLL cancer is the most common adult leukemia in the United States and currently has few effective therapies. The goal of this project is to create therapeutic agents with improved *in vitro* activity to improve current *ex vivo* clinical trials for CLL. At the same time, we hope to develop "smart" ligands and vectors that can seek out and specifically deliver therapy to CLL cells *in vivo* while sparing non-tumorigenic tissues. In this one-year project, we will perform work towards identifying proteins that can recognize and bind to CLL cells. We hope to find proteins that cannot only bind specifically to these tumor cells while avoiding normal cells. If identified in this project, these CLL-targeting proteins could be used in future work to target anti-cancer drugs or gene therapy delivery vehicles to CLL cancer cells directly in patients without the concern of delivering these agents to normal cells in the body to avoid the normal complications of cancer therapy. Proof of principle here against CLL will enable the application of this targeting technology in future clinical trials through the Center for Cell and Gene Therapy at Baylor College of Medicine. Development of this technology for CLL will also establish the protocols necessary to apply the same techniques against any other leukemia.

Angelo A. Cardoso, MD, PhD
Dana-Farber Cancer Institute, Boston, MA
$75,000 – *Development of Anti-Angiogenic Strategies for the Treatment of Acute Lymphoblastic Leukemia*
Novel therapies are necessary for the treatment of acute lymphoblastic leukemia (ALL) as conventional strategies have reached their limits. These novel approaches ideally should target mechanisms that are critical for the development and progression of this cancer. It has been suggested de novo angiogenesis is associated with ALL. De novo angiogenesis is the formation of new vessels, which are primarily composed of endothelial cells. We have found that the leukemia environment contains factors that stimulate endothelial cells and promote the formation of new vessels, and that this leukemia-stimulated endothelium provides survival signals to the leukemia cells. Therefore, we reasoned that disruption of the leukemia-mediated formation of new vessels, and consequently, disruption of the interaction of leukemia cells with the endothelium, may function as an effective anti-leukemia strategy. In this project, we propose to determine whether the angiogenesis is critical for the establishment and progression of ALL, and to develop gene therapy strategies that attack leukemia by delivering anti-angiogenic molecules to cancer sites. If successful, this project should have a major impact in the management of this devastating disease.

Violetta Skalski, PhD
Ontario Cancer Institute, Toronto, Ontario Canada
$71,259 – *A Novel Exonuclease: Role in Resistance to Nucleoside Analogs in Leukemia Cells*
Nucleoside analogs (NAs) are chemotherapeutic agents designed to interfere with the replication of DNA in cancer cells resulting in the death of cancer cells. NAs are used in the treatment of leukemia, lymphoma and tumors such as pancreatic cancer. Exonucleases are proteins that normally function to preserve accuracy during DNA replication in cells. We identified and exonuclease (exoN) in human leukemia cells and determined that it removes NAs from DNA. The removal of NAs from DNA would enable leukemia cells to resume replicating their DNA and ultimately to survive. Therefore, the action of exoN would render NAs inactive in leukemia cells and may result in resistance to this class of drugs. We propose to characterize exoN in more detail and to study the mechanism(s) that regulate its activity. Since the onset of resistance during chemotherapy of leukemia is a major problem in the clinical management of this disease, our studies may offer important new information, which could result in improved efficacy of NAs and the development of more effective new drugs.

Jumin Zhou, PhD
The Wistar Institute, Philadelphia, PA
$75,000 – ***Molecular Genetic Analysis of a Novel cis-element, the Promoter Targeting Sequence***
To ensure normal biological functions, genes must be precisely regulated. Leukemia, in many cases, is the result of inappropriate regulation of genes, particularly a family of genes called the *Hox* genes. In this proposal, we are going to investigate the functions of individual regulatory DNA elements in the fruit fly homologue of the Human *Hox* genes called bithorax gene complex. The fruit fly is an excellent genetic model system to study such DNA elements. My previous work has isolated two types of these elements, insulators and anti-insulators, that play critical roles in regulating the bithorax gene complex. Understanding how these DNA elements work will help us to understand how *Hox* genes are regulated and therefore gain insight about the causes of leukemia.

Andrea Cerutti, MD
Weil Medical College, Cornell University, New York, NY
$75,000 – ***Role of CD30-CD30L Interaction in Leukemia-associated Immunodeficiency and Malignant B Cell Clonal Expansion***
In B cell leukemias and lymphomas, the progressive accumulation of malignant B cells is associated with recurrent infections that are secondary to the severe impairment of the immune response. This acquired immune deficiency stems from the ability of malignant B cells to alter the functionality of nonmalignant T and B lymphocytes. The observation that B cell tumors are often associated with increased expression of CD30 suggests that this molecule is involved in the pathogenesis of both immune deficiency and neoplastic B cell growth. The studies proposed in this project are aimed at elucidating the mechanisms by which CD30 interferes with the activation of nonmalignant lymphocytes and enhances the accumulation of malignant cells. A better understanding of these mechanisms might allow scientists to devise new strategies for the immunotherapy of leukemia patients.

Dean W. Felsher, MD, PhD
Stanford University School of Medicine, Stanford, CA
Targeting MYC Inactivation for the Treatment of Lymphoma
Lymphoma is caused by genetic events that activate oncogenes or inactivate tumor suppressor genes. Therapies that target the inactivation of these mutant gene products may be effective for the treatment of human lymphoma. I have shown that lymphoma caused by the activation of the *MYC* oncogene is reversed if *MYC* is inactivated. Now, I wish to determine to what extent must *MYC* be inactivated to cause tumor regression; how long must MYC be inactivated to cause tumor regression, what is required of *MYC* to maintain cancer; and finally, which genes regulated by MYC are responsible for its ability to cause cancer. The answers to these questions will be useful in developing new therapies that inactivate *MYC* to treat lymphoma.

Linzhao Cheng, PhD
John Hopkins University, Baltimore, MD
$75,000 – ***Towards Potent and Specific Immuno-therapies for Treating Leukemia Using Transduced Dendritic Cells and Their Hematopoietic Precursors***
Billions of blood-forming (hematopoietic) cells and immune cells are generated every day from their common mother cells called hematopoietic stem cells (HSC) residing in bone marrow. The mature blood and immune cells are short lived, and their numbers and ratio have to be exactly controlled. The abnormal growth of one type of these cells will tip the balance and generate blood disorders as well as immune deficiency, which is important for the body's self defense against viruses and cancer cells. Based on the latest advances in molecular and cell biology, we outline here a new strategy to develop a novel vaccination approach targeting cancer cells based on genetic enhancement of HSC and their daughter immune cells. Our approach combines the latest technologies of stem cell biology, immunology and gene therapy in order to mount a sustained, efficient and selective attack on leukemic cells but not on normal cells.

Ae-Kyung Yi, PhD
University of Tennessee Health Science Center, Memphis, TN
$75,000 – *Elucidation of Mechanisms by which MEK/ERK Negatively Regulates CpG DNA-mediated Th1 Response*
Our immune system rapidly responds to the DNA from bacteria, which contains high levels of CpG motif. Synthetic DNA that has CpG motif (CpG DNA) mimics bacterial DNA. CpG DNA helps our white blood cells to effectively fight with infections and cancers such as B lymphoma and melanoma. CpG DNA strongly induces white blood cells to make antibiotics and various hormone-like proteins called cytokines. These cytokines dictate function of white blood cells. For example, a cytokine called IL-12 promotes natural killer cells, a type of white blood cell, to secrete cytokines called interferon and to kill infected cells and cancerous cells. Other cytokines like IL-10, on the other hand, inhibits IL-12 and interferon production by white blood cells. CpG DNA can induce production of both sets of cytokines with a slightly different time course. If we know details about how CpG DNA activates and deactivates white blood cells, we could come out with a better way to use CpG DNA as an effective and safe lymphoma therapeutic agent. Therefore, we will investigate how proteins activated by CpG DNA in white blood cells regulate function and production of other proteins and how inhibition of one protein's activity promotes the secretion of several cytokines, while suppressing the production of other cytokines. Our study will enhance our understanding of the body's immune system and will lead to more effective and safe strategies to fight the cancerous white cells.

Huaizhong Hu, PhD
University of Wisconsin, Madison, WI
$73,000 – *Graft-Versus-Leukemia Effect of Donor T Cells Modulated in the Homing Process*
Leukemia is a malignant disease involving the white blood cells, and occurs in people of all ages. If not treated properly, the disease is fatal to patients. Bone marrow transplantation is a therapeutic method that can cure leukemia. However, the donor cells transplanted into a patient cause a side effect called graft-versus-host disease (GVHD), which causes damage in the patient's liver, intestines, and skin. GVHD is mediated by donor T cells, and importantly these T cells are also responsible for depleting leukemia cells. Therefore, it is important to keep the T cell function from depleting leukemia cells, and meanwhile suppress their role in mediating GVHD. We recently found a novel method that could prevent or ameliorate GVHD without changing donor T cells. In this project, we will explore whether this novel method will be able to maintain the T cell function for depleting leukemia cells. If it does, this method will be clinically useful for bone marrow transplant patients.

PHYSICIAN-SCIENTIST AWARDS

Corey Cutler, MD, FRCPC
Dana-Farber Cancer Institute, Boston, MA
$90,000 – ***A Decision Analysis of Allogeneic Stem Cell Transplantation for Myelodysplastic Syndrome***
The myelodysplastic syndromes (MDS) are a collection of pre-leukemic conditions that affect between 3.5 and 12.6/100,000 people each year. Allogeneic stem cell transplantation (SCT) is the only curative therapy for patients with MDS, however, the procedure is associated with significant risks. The optimal timing of SCT for MDS is not known; patients and physicians often delay the procedure as long as possible, but the net effects of this strategy on survival and transplantation success are unknown. We hypothesize that transplantation early after the diagnosis of MDS and prior to leukemic progression leads to improved survival overall. Decision analysis is a statistical technique that is used to aid medical decision-making under conditions of uncertainty and is particularly useful when randomized clinical trials are difficult or impossible to perform. We propose to perform a decision analysis to identify the optimal timing of SCT for patients with MDS. The result of this analysis will potentially influence the treatment of all MDS patients who are candidates for transplantation.

Jean-Pierre Bourquin, MD, PhD
Dana-Farber Cancer Institute, Boston, MA
$90,000 – ***Role of the Tel/ETV6 Gene in Hematopoiesis and Leukemia***
A gene called TEL was discovered by the study of chromosomal rearrangements characteristic for several forms of human leukemia. Such gene rearrangements produce abnormal proteins, in which the products of two genes are brought together and then initiate cancer formation. Relatively little is known about the normal functions of TEL. TEL is a transcription factor, a protein that regulates the expression of specific genes, and plays an important role in modulating genetic programs for the normal development of blood cells (hematopoiesis) both in the embryo and the adult organism. Loss of TEL gene function is lethal for the developing mouse embryo. To further understand the role of this gene in hematopoiesis, TEL will be inactivated specifically in blood cell precursors of adult mice, both in the intact mouse and in cell cultures. This aim is to discover the molecular pathways regulated by the TEL in hematopoietic cells. This knowledge is fundamental to an improved understanding of the consequences of TEL gene rearrangement for leukemia causation.

Leonard J. Appleman, MD, PhD
Dana-Farber Cancer Institute, Boston, MA
$90,000 – ***Targeted Degradation of p27kip1 in T Lymphocytes: A Model for Leukemia and Other Cancers***
Leukemia cells undergo uncontrolled multiplication until their massive numbers eventually overwhelm the patient. In contrast, normal cells divide only when they receive appropriate signals from the environment. A nuclear protein called $p27^{kip1}$ prevents these resting cells from multiplying. When outside stimuli signal the cells to divide, $p27^{kip1}$ is degraded in cancer cells. In order to address this question, normal human T cells (obtained from volunteers as a byproduct of platelet donation) will be compared to a T cell leukemia cell line with respect to mechanism of $p27^{kip1}$ degradation. The normal T cells and the leukemia cells share receptors and associated intracellular proteins that sense environmental signals to undergo cell division. However, these mechanisms are somehow uncoupled from cell division in the leukemia cells. By using the normal and leukemia cells as a “matched pair”, we hope to identify the molecular control mechanisms that have failed in the leukemia cells. This information may help to develop drugs to target these abnormal processes. In addition, understanding how T cell proliferation is controlled will help in the design of effective T cell immunotherapies directed against leukemia and other cancers.

POSTDOCTORAL FELLOWSHIP AWARDS

Roger T. Luo, PhD
University of Chicago, Chicago, IL
$60,000 – *Structure Function Studies of MLL, ell, and EAF Family Members in Myeloid Leukemogenesis*
Leukemias can be classified by their particular chromosomal abnormalities, and 11q23 translocation are one of the most frequent of these chromosomal aberrations. Patients with acute leukemia and 11q23 translocation represent a distinct pathological entity with similar clinical manifestations and unfortunately, constitute a subtype of leukemia that is most refractory to conventional chemotherapy strategies. Thus, new insights into successful treatments will require a greater understanding of the pathogenesis of these leukemias. My overall goal is to determine the role of the unique fusion gene involving *MLL* and *ELL* in the development of human acute leukemia. These leukemias arise as a result of a chromosomal rearrangement followed by a selective advantage to the cell gained from the expression of the *MLL-ELL* fusion gene. In addition to its fusion to *ELL*, the *MLL* gene fuses to over 20 different partner genes that share few obvious similarities. To understand the mechanisms that underlie the formation of these translocations, it is essential to characterize this process on several levels. Our working hypothesis is that the formation of *MLL* fusion proteins disrupt the normal protein-protein interactions of *MLL* and its partner genes. To test this hypothesis, we have sought to identify proteins that interact with *ELL*. Recently, we have cloned a novel gene that we named *EAF1* for *ELL Associated Factor 1*. Using retroviral gene transfer into bone marrow cells to generate a mouse model of leukemia, I have recently found that the interaction between *ELL* and *EAF1* plays a critical role in the development of this subtype of leukemia. In this grant, I propose to use retroviral gene transfer into normal bone marrow cells from mice to determine the critical features of *ELL* and *EAF1* that result in the development of acute leukemia. I also plan to develop a novel mouse model of leukemia by introducing the ELL gene into the mouse chromosomal location of the *MLL* gene. This will provide me the opportunity to characterize the development of these leukemias in greater detail. The generation of these models will be critical for unraveling the molecular pathogenesis of 11q23 leukemia and this knowledge will provide a basis for the development of future treatment strategies for this common subtype of leukemia.

Paolo Vigneri, MD, PhD
University of California San Diego, La Jolla, CA
$60,000 – *Unconventional Therapeutic Approach to CML: Converting Bcr-Abl into an Anti-oncoprotein*
Chronic Myelogenous Leukemia (CML) is a deadly form of cancer caused by the abnormal Bcr-Abl protein. Bcr-Abl is confined to the cytoplasm of CML cells. I have discovered that Bcr-Abl can be forced to enter the nucleus and, while in the nucleus, can kill CML cells. Hence, I have found a way to transform the culprit of CML into the terminator of the disease. I will further investigate the mechanisms determining Bcr-Abl cellular localization with the aim of trapping increasing amounts of this oncogenic protein inside the nucleus. I will also investigate how nuclear Bcr-Abl forces leukemic cells to commit suicide. Bcr-Abl sequestration inside the nucleus represents an unconventional therapeutic approach to CML.

Andreas Villunger, PhD
The Walter and Eliza Hall Institute, Melbourne, Australia
$60,000 – *The Role of BH3-only Proteins in Hemopoietic Development and Tumorigenesis*
Apoptosis, the Greek term for the cell suicide program intrinsic to all cells of the body, is a physiological process for the deletion of unwanted and potentially harmful cells. During development, the "death" program" is activated in certain cells of the organism to allow modeling of tissues but also for the maintenance of the equilibrium within the formed tissue. Abnormalities in cell death control, allowing malfunctioning cells to survive, can be one of the initial steps in the development of cancer or auto-immunity. Under physiological conditions, apoptosis is mediated either by so called "death receptors," present on the surface of a cell, after activation by corresponding ligands or by members of the Bcl-2 protein family. In the latter case, pro-apoptotic members of the Bcl-2 family neutralize the function of the pro-survival members of the family and promote the assembly of a signaling complex within the cell which causes activation of a cascade of "cell death proteases," termed caspases. These enzymes cleave vital cellular proteins and thereby lead to cell collapse. For

my fellowship, I propose to analyze the biologic function of a new, so far unknown pro-apoptotic Bcl-2 family member, termed Bmf (Bcl-2 modifying factor). Bmf is a protein which promotes cell death. When its function is lost, for example due to mutations or deletions of the gene, I anticipate that this may contribute to tumor formation and/or auto-immunity. I want to provide evidence for this by generating mice that lack *bmf* by targeted deletion of the gene. This will allow investigation of the function of this novel cell death regulator in normal physiology and in neoplastic disease in a model organism. During these studies, I will focus particularly on the hemopoietic system. I anticipate that these studies will provide interesting insights into the normal control of hemopoiesis and into tumor formation. In addition, I hope that this information can be used to develop new or to improve current treatment strategies to allow better treatment of patients suffering from hemopoietic cancers.

Min Huang, PhD
University of North Carolina, Chapel Hill, NC
$60,000 – *The Role of CAD Degradation in the Progression of Cell Death of Leukemia Cells*
We are investigating the basic mechanisms by which cell growth is regulated and our research has important applications for leukemia where unregulated growth is a causative agent in this disease. In particular, we are interested in the processes by which the cell regulates the production of "building blocks" needed for cell growth. This involves a series of "switches" that are turned on and off as the building blocks are or are not needed. The specific questions that we are asking revolve around an enzyme that makes nucleotide for the synthesis of RNA and DNA, the nucleic acid building blocks of the cell. We are characterizing the regulation of a key enzyme in this process by a molecular "switch" known as phosphorylation. Phosphorylation controls many aspects of cell function including cell growth and cell death. We are particularly interested in determining how hormones and other signals switch these processes on or off. The long-term significance of our research is that once we understand the important "switches" in the cell, we can design new and better drugs to control unregulated cell growth as needed.

Go Totsukawa, PhD
Rutgers University, Piscataway, NJ
(2nd Year Funding)
$30,000 – *Role of Myosin Phosphatase in Cell Division*
One of the definitive characteristics of cancer cells including leukemia is uncontrolled cell division. This proposal is aimed at understanding the functions of myosin phosphatase in cell division control. Myosin phosphatase regulates phosphorylation of myosin. Because myosin is an essential motor to drive cytoplasmic cell division, and because phosphorylation of myosin controls the activity of the myosin motor, myosin phosphotase is a key molecule controlling cell division. I have found that myosin phosphatase activity is controlled during cell division. The major aim of this proposal is to elucidate what is the mechanism that controls myosin phosphatase activity during cell division. I will also manipulate the activity of myosin phosphatase during cell division to see whether such manipulation blocks cell division. My studies will help us understand why cancer cells lose cell division control and may help to develop new therapies to treat leukemia.

Sharon A. Matthews, PhD
University of Washington, Seattle, WA
$60,000 – *Cloning and Characterization of Two Novel Calcium Channels Expressed in the Immune System*
A detailed understanding of the precise mechanisms that control the activation and growth of cells of the immune system is essential in order to understand how beneficial immune responses are generated. Study of these regulatory signals provides insight into the mechanisms underlying pathological diseases such as leukemia and lymphoma and thus the opportunity to identify novel targets and strategies for therapeutic intervention. Calcium is a key intracellular signal that controls immune cell activation and growth. However, the mechanisms by which calcium levels are maintained in immune cells and the identity of the membrane channels which control calcium entry are mostly unknown. Two novel calcium channels, TPC1 and TPC2, have been cloned which are expressed in the immune system elsewhere. These channels are distinguished by the presence of the two homologous transmembrane spanning regions, a unique feature that

has not previously been described for any other channel type. This proposal aims to investigate the regulation and function of these novel channels. TPC1/2 function will be explored through two specific aims: TPC1 and TPC2 will be expressed in lymphocyte and endothelial cell lines and the effect on cellular growth and death will be determined. Secondly, the role of TPC1/2 in regulating calcium levels and signaling in these cell lines will be investigated. In addition, electrophysiological studies will examine the role of the TPC1/2 channels in regulating calcium influx into these cells. Overall, these studies will provide significant insight into the regulation of TPC1 and TPC2 and also into the role of these new channels in calcium signaling and homeostasis of the immune system.

Andrea A. Duina, PhD
Harvard Medical School, Boston, MA
$60,000 – ***Characterization of Novel Functions for Histone H3 in Transcriptional Regulation***
DNA is the genetic material that contains the information necessary for the growth and development of living beings. Specialized proteins, called histones, interact with DNA and other factors to create a compacted protein/DNA complex that can be accommodated inside the small confines of a cell's nucleus. These interactions, however, have a strong impact on the way genetic information is utilized by the cell and, by extension, play a crucial role in preventing uncontrolled cell growth and cancer. Mutations that alter the interactions between histones and DNA have been correlated with a number of human cancers, including Burkitt's lymphoma and leukemia. My proposed studies will explore the roles of histones in the expression of genetic information.

Oleg V. Kovalenko, PhD
Dana-Farber Cancer Institute, Boston, MA
$60,000 – ***Association of Novel Human Hematopoietic Stem Cell Marker AC133/Prominin with Tetraspanins***
The proposed project is aimed at the understanding of new protein interactions that occur on the surface of tumor cells during cancer growth and metastasis. I have discovered an interaction between a novel hematopoietic stem cell protein, AC133, and several members of the tetraspanin family of membrane proteins. The AC133 protein has previously been implicated in the growth and expansion of blood cell precursors. Tetraspanin proteins have been implicated in various aspects of tumor cell invasion and metastasis. Since both AC133 and tetraspanin proteins are highly expressed in leukemic cells, the observed associations between these two types of molecules are likely to be important in formation and progression of hematopoietic malignancies. I will carry out detailed structural and functional analyses of AC133-tetraspanin associations to establish their mechanisms and relevance for the tumor cells. These studies should lead to better understanding of the ways by which leukemic cells appear, interact with their environment and metastasize.

Pranay D. Khare, PhD
Mayo Clinic, Rochester, MN
$60,000 – ***Selection of Multiple Myeloma and Angiogenesis Specific-targeted Peptides from Glycosylated Peptide Libraries Displayed on Avian Leukosis Virus***
The growth and metastasis of malignant tumors depends on angiogenesis; the development of new blood vessels. Recently, angiogenesis and the expression of angiogenic growth factors have also been recognized in multiple myeloma (MM). The proliferating MM cells and blood vessels in tumors may express markers that could be used as anti-tumor targets. The majority of known antigens and cell surface markers are either non-selectively expressed, shed or secreted, or not fully characterized, resulting in limited therapeutic and clinical role. This project will develop a novel approach for the identification of peptide ligands that recognize specifically MM cells and/or activated vascular endothelial cells that are expressed at site(s) of angiogenesis using a retrovirus, avian leucosis virus (ALV). Eukaryotic viruses have not previously been used for the presentation or selection of libraries of cell-binding peptides. Retroviruses offer several advantages over phage (prokaryotic virus) as vehicles both for the generation of unique peptide display libraries (e.g., carrying post-translational modifications such as N-linked glycosylation), and for selection of novel eukaryotic cellular targets. Many of the proteins manufactured in mammalian cells (particularly cell surface and secreted proteins) are subjected to post-translational modifications such as glycosylation and proteolytic cleavage. ALV provides significant advantages as a platform to generate stable retrovirus peptide display libraries that can be selected against mammalian cells *in vitro* and

tumors *in vivo*. In our preliminary studies, we have demonstrated that two different peptides can be stably displayed on ALV envelope glycoprotein and incorporated into infectious virions: the eight amino-acid FLAG peptide and the 53-amino-acid epidermal growth factor peptide. Over the fellowship period, we plan to generate an ALV peptide display library comprising a diverse array of glycosylated peptides to use to select peptides that bind MM cells and/or activated endothelial cells. Peptides ligands identified by this screen will have great potential as therapeutic targets to combat cancer.

Takashi Nagata, PhD
The Rockerfeller University, New York, NY
(2nd year funding)
$30,000 – *Structure and Function of the t(12;21) Leukemogenic Fusion*
Mutations in proteins which control gene expression are commonly associated with the onset of acute human leukemia. The most frequently mutated protein is AML1, a factor which is essential for normal blood cell development. AML1 forms intimate relationships with several proteins to initiate and regulate development of blood. These proteins include ETS-family proteins which associate with AML1 and enhance its activity. Mutated AML1 fused to the ETS-family protein TEL (TEL-AML1) results in acute leukemia. We propose to investigate the structure and function of TEL-AML1. This study will provide molecular insights into the role of AML-1 in leukemogenesis and blood cell development.

Benjamin T. Kile, BSc(Hons) LLBR
The Walter and Eliza Hall Institute, Melbourne, Australia
$60,000 – *A Mutagenesis Screen for Genes Controlling Murine Hemopoiesis*
Normal blood cell growth and development is a complex process regulated by a large number of different genes. Leukemia is the result of uncontrolled blood cell growth, caused by mutations of those genes within individual cells. Identifying the genes that regulate normal blood cell growth and determining their function is critical to understanding the genetic mutations which result in leukemia. The mouse is the most useful experimental animal model for examining human diseases, and this project therefore aims to identify genes in mice which are involved in normal blood cell growth, and isolate the mutations that affect them. The project involves treating mice with a chemical that causes mutations in all types of genes, and examining their offspring to see if they suffer from blood cell growth abnormalities or leukemias. Once mice that do have been identified, the genes that are involved can be located, and the mutations affecting them can be determined. This will enhance our understanding of the processes by which blood cell growth is regulated, how leukemias arise, and potentially provide novel targets for drug discovery and the development of new therapeutic agents for the treatment of leukemia.