



Continuing Umbrella of Research Experiences



**Scientific Presentations
Summer 2006**

Dana-Farber/Harvard Cancer Center



DANA-FARBER/HARVARD CANCER CENTER
A Comprehensive Cancer Center
Designated by the National Cancer Institute



Launched in 2002, the Continuing Umbrella of Research Experiences (CURE) at Dana-Farber/Harvard Cancer Center is an important building block in research training initiatives. This program is designed to provide high-school and college students with a highly stimulating and rewarding hands-on research experience with a view towards encouraging students to pursue advanced education and training in the biomedical sciences and future careers in basic, clinical and population science cancer research.





Dana-Farber/Harvard Cancer Center's Continuing Umbrella of Research Experiences (CURE) wishes to acknowledge and sincerely thank the CURE Advisory Committee members, mentors, scientific advisors, lecturers, and supporters for expanding and enhancing the academic and career horizons of our students.



Wednesday, August 23, 2006

Presentations

Mariaah Williams *

Healthy Solutions II

** This presentation was given at an earlier date.*

Chandreka Wright

The Role of Ffl-b in Sex Determination in Zebrafish (*Danio rerio*)

Sammy Kibet Sambu

Delineating the Gene Expression Patterns and Presumptive Tissue-Restricted Functions of Fox Family Genes in the Murine Gastro-Intestinal Tract

Christina Tabuteau

Perception of Suffering

Jazzmine Williams

Protein-Protein Interactions

Fittz-Patrick Noel

The Benefits of Array CGH and the Fight against Cancer

Olufunke Buraimoh

Developing a Genetically Engineered Breast Cancer Model for Drug Discovery

Afamefuna Nduaguba

Transforming Growth Factor-Beta (TGF β) Requirements for Tumorigenicity and Metastasis in Pancreatic Cancer





Healthy Solutions II

Mariaah Williams

Mentor: Gary Bennett, PhD

Scientific Advisor: Katrina Bond, MPH

Objective: To develop an electronic PDA device which monitors physical activity and specific eating behaviors, and to determine whether the device is more effective, in terms of promoting weight loss, when used independently or in combination with a coached weight loss intervention. **Methods:** Participants will include 60 overweight, adult women; approximately 80% of whom are expected to be lower income, ethnic minorities. Participants will all be given a PDA to record their daily step count, as measured by a pedometer, along with three other behaviors chosen from a list at baseline. Half of the participants will be randomized into a control group, and use only the device, while the other half (intervention group) will have a PDA plus interpersonal nutrition coaching. **Results:** PDA devices have been chosen, and Health Communications Core has developed a survey to install on them. Working groups have been formed to manage various aspects of program development, including the making of a guide to be utilized by coaches. User testing of intervention recommendations tracked on the PDA devices will be completed in August. Recruitment sites have been identified and will include churches, housing sites and other community organizations near the YMCA of greater Worcester. **Discussion:** The results of Healthy Solutions may prove to be useful in future weight loss intervention studies. The intervention is being designed with the aim of dissemination within large populations. The findings will identify successful methods of weight loss intervention, which can be used to reduce the incidence of obesity, along with heightened risks for many health conditions and diseases, including cancer.

John Howard is a rising sophomore at Bowdoin College. He returned this summer to his DFCI lab from last year to continue his research experience.

Tasmina Hyder graduated from high school this spring. She was able to enhance her research experience by working this summer in a clinical laboratory setting. This fall she will be attending the Albany College of Pharmacy.

Lois Luberice is entering her junior year at the University of Massachusetts, Boston. She is continuing her research experience by participating in the Research Experience for Undergraduates (REU). Lois is conducting research in the Huang lab.

Nicole Maddox graduated from Wellesley College in 2006 and is currently interning at the National Institute of Health's (NIH) National Cancer Institute (NCI). Nicole has been accepted into the Master's Medical Sciences program at Boston University.

Yanny Qin achieved a first place finish in the State's high school science fair. Consequently, she will attend a summer biomedical intern program at MIT. Additionally, she was accepted and will be enrolled in the Pharmacy program at Northeastern University.





Kayla Elliott graduated from the Winsor School in 2006. She will be attending Williams College this fall. This summer she was selected to participate in the Williams College Summer Science Program.

William Kaplan just completed his freshman year at Wesleyan University. He is currently serving as a TA this summer in a science class at the Carroll School in Lincoln.

Malti Sharma is a recent graduate of the CURE program. Malti will be entering her sophomore year at Boston University. In addition to taking summer courses, Malti will be volunteering at Brigham and Women's Hospital.

Laporscha Wilson successfully completed her CNA training this summer. Currently enrolled in the nursing program at Simmons College, Laporscha is seeking to gain hands-on nursing experience.

2005

Yewande Adepoju is a senior at the University of Massachusetts, Boston. Yewande is spending her summer at Harvard Medical School conducting research on the interaction between Herpes Simplex Virus 1 and DNA replication proteins.

Carla Becerra graduated in 2006 from Matignon High School and will be entering the Class of 2010 at Wesleyan University. This summer she is continuing her exposure to research by participating in the HMS Project Success program.

Kya Grooms is a member of the class of 2006 at Buckingham Browne & Nichols School and is currently working in the Orthopedic Research Department at Children's Hospital. Kya will be attending Emory University in the fall with a plan on majoring in Anthropology and continuing on the pre-med track.



The Role of Ffl-b in Sex Determination in Zebrafish (*Danio rerio*)

Chandreka Wright

Mentor: Barry Paw, MD, PhD

Scientific Advisor: Chris Lawrence

Zebrafish (*Danio rerio*) are a very popular model organism for genetics and development. Despite this, mode of sex determination mechanism operating in this species is unknown. What is clear, however, is that the process of gonadal differentiation is directed by steroid hormones, the production of which are controlled by major sex determining genes that are sensitive to environment influences such as temperature and growth rate. There are a number of genes known to be involved in this process, any one of which may be a candidate gene for the master regulator of sex in this species. The gene *ffl-b* has been shown to be important for steroidogenic cell development and is required for the differentiation of the interrenal organs. Anecdotal evidence indicates that zebrafish heterozygous for the b333 mutation do not differentiate as females. The b333 mutant allele is the result of a gamma-induced deletion of chromosome 8. Bioinformatic analysis indicates that the deleted chromosomal segment may also include some or all of *ffl-b*. To test our assumptions, we screened DNA from b333 $-/-$ mutants for the presence or absence of *ffl-b* using PCR primers specific for the gene. Further, we formally tested for the presence of female b333 $+/-$ fish by scoring 100 non-genotyped progeny from a b333 $+/-$ individual mated to a wild type fish for the presence/absence of the b333 deletion and for phenotypic sex. The results of this experiment will help determine the extent to which *ffl-b* regulates sex differentiation in zebrafish.



Delineating the Gene Expression Patterns and Presumptive Tissue-Restricted Functions of Fox Family Genes in the Murine Gastro-Intestinal Tract

Sammy Kibet Sambu

Mentor: Ramesh Shivdasini, MD, PhD

Scientific Advisor: Mike Verzi, PhD

The role of Forkhead box (Fox) family transcription factors (TFs) in the development of the gastro-intestinal (GI) tract has gained increasing importance. However, researchers are yet to map completely, which Fox genes are expressed in the GI tract and the anatomic boundaries within which these transcription factors are transcribed. More importantly, it is yet to be determined whether the expression of these genes follows any patterns that may point to their functions in tissue differentiation. This investigation addresses these outstanding questions. Using reverse transcription polymerase chain reaction (RT-PCR) and *in situ* hybridization (ISH), we posit the mesenchymal and epithelial restriction of selected Fox-family TFs. Furthermore, the expression of these TFs is limited to certain regions of the adult murine gut as evidenced by the RT-PCR analysis of the following transcription factors: Foxa1, Onecut1, Foxp4, Foxp1, Foxq1, Foxk1 and Foxo3. Other patterns are observed. For instance, Mllt7, Foxm1, Foxf1a, Foxp3, Foxk2, Foxj2, Foxp2 and Ches1 are all expressed throughout the GI tract. Even so, this latter group shows some rostro-caudal variance in mRNA expression, thus necessitating a second means of categorization. Since the TFs responsible for gut epithelial differentiation are poorly understood, this study aims to delineate the gene expression patterns and presumptive tissue-restricted functions of Fox family genes expressed in the GI tract.

Alumni Update

2002

Ayodele Buraimoh is a rising junior majoring in pre-med at Harvard. Ayo is currently expanding his research experience at Brigham and Women's Hospital in the area of breast cancer.

Nsiong Ipka started his medical studies at the Chicago Medical School at Rosalind Franklin University of Medicine and Science. His plan will be to apply to the dual MD/PhD program.

Juan Molina graduated from Tufts this Spring and obtained employment as a Research Coordinator in the Division on Addictions at Cambridge Health Alliance. Additionally, Juan gained his first authorship on a recent article in *The Drinking Report for Addiction Medicine (DRAM)* entitled "Off the Sidelines: Women and Drinking Games."

Chau Tran will be entering into her senior at Harvard. During this past summer, Chau participated in a research fellowship through a program at Harvard College.

2003

Erick Kamau continues to gain experience in the biotech industry as a research technician. Erick will be applying to medical school for the Fall of 2007.

2004

True Brewster graduated in 2006 from Brook Farm on the West Roxbury High School complex. True will be attending Bridgewater State College this fall.



The Origin of Carcinoma Associated Fibroblasts (CAFs)

Louka Abed

Mentor: Ragu Kalluri, PhD

Scientific Advisor: Elisabeth Zeisberg, MD

Carcinoma Associated Fibroblasts (CAFs) have a significant role in cancer progression and metastasis, but their origin is unclear. Are they localized in normal tissue, or are they bone-marrow-derived, possibly attracted by the cancer cells? Moreover it is controversial whether fibroblasts facilitate or inhibit metastasis. To address these questions, we transplanted nine female mice with bone marrow from FSP-GFP transgenic mice. In these mice GFP is expressed under the FSP1 (FSP= Fibroblast Specific Protein) promoter, and thus, in the transplanted mice green fluorescence identifies fibroblasts derived from the bone marrow. Three weeks after transplantation, we injected 4T1 cells, a breast cancer cell line which metastasizes to the lung, into the breast pads of six of these mice. The tumor growth will be monitored, and one group of mice will be sacrificed at a tumor size which is typically not yet associated with metastasis. The other group will be sacrificed at a tumor size which is typically associated with metastasis. The tumor and all organs will be collected and screened for GFP expression. We anticipate that cancer cells recruit bone marrow fibroblasts in order to help their spread, and in that case, we may find a significant number of fibroblasts of bone marrow origin. If we find insignificant GFP-positive cells, this would suggest that localized fibroblasts are transformed. In addition, if we find bone-marrow-derived fibroblasts in the lung of tumor cell-injected mice, even before the tumor has metastasized to the lung, this might stress the role of fibroblasts in mediating metastasis.

Perception of Suffering

Christina Tabuteau

Mentor: Richard T. Penson, MD, MRCP

There is very little data examining perceptions of suffering in patients undergoing treatment. The purpose of this study is to examine differences between the manner in which staff perceive suffering in head and neck, ovarian, breast and cervical cancer patients and patients suffering from rheumatoid arthritis. Additionally we want to know how they evaluate suffering. It is important to proactively respond so that caregivers deliver optimal care. The procedure for this study is as follows: an investigator will approach doctors and nurses from two infusion units. Female patients with ovarian, head and neck, breast and cervical cancer patients and patients suffering with panic disorder and rheumatoid arthritis, and who has had a mastectomy will be contacted by a psychologist or Clinical Research Coordinator (CRC) after identification through their treating clinics. Each participant will be asked to complete a survey and Body Image Scale (BIS), before reading a series of 4 scenarios describing typical experiences of patients with each of the aforementioned forms of cancer, panic disorder and rheumatoid arthritis. They will rate their perceptions of the patient's suffering on the Body Image Scale (BIS). They will then participate in a focus group containing a nurse, a doctor, and a patient from each disease-type cohort, before re-rating their perceptions of suffering, using the same scales. The results and information gained from this study will advance the understanding of nursing care and identification of key issues experienced by various patient groups. This may help to identify a focus for further caregiver training regarding perceptions of patient suffering. It may also prove therapeutic for patients to discuss their own experiences as a patient and understand the issues faced.

Protein-Protein Interactions

Jazzmine Williams

Mentor: David Hill, PhD

Scientific Advisor: Xiping Yang, PhD

The mutation in the RB (Retinoblastoma protein) gene causes a common eye cancer, mostly found in younger children, called Retinoblastoma. We introduced the RB protein into a pool of genes that connect to AD, the transcription activation domain, into the yeast. The yeast already contains RB fused to DB, the DNA binding domain. This process is known as the Yeast Two Hybrid technique, Y2H. When using this technique it will show the interaction partners of the RB protein. We know that RB will interact with certain proteins, but the question is what proteins will RB interact with? It is predicted that RB will interact with many of the proteins, but not all of them. We are conducting this experiment to assess which proteins interact with the Retinoblastoma protein.

Forkhead Family of Transcription Factors and Pancreatic Cancer

Meera Sreedhara

Mentor: Roya Khosravi-Far, PhD

Scientific Advisor: Ken Ndebele, PhD, MPH

FOXO3a, a member of the forkhead transcription factor family, acts as a tumor suppressor by inducing apoptosis in cells. FOXO3a has been shown to be a critical regulator of the expression of several apoptotic factors such as TNF-Related Apoptosis Inducing Ligand (TRAIL). TRAIL is a tumor selective death inducing ligand that is a key regulator of tumor surveillance by the immune system. The expression of members of the forkhead family is altered in many cancer cells. In our studies, we hypothesize that the proteasome degradation pathway is involved in the alteration of the expression of Forkhead proteins. For these studies we will analyze the expression of FOXO3a and response to TRAIL in several pancreatic cancer cell lines. We will then determine whether velcade, a proteasome inhibitor, can increase the expression of FOXO3a and alter the response of these tumor cells to TRAIL. These studies will provide novel insight into the mechanism by which proteasome inhibitors such as velcade induce apoptosis and likely generate effective anti-cancer therapies.

***Renal Cell Carcinoma: The Study
of MDA-7/IL-24 and NSAIDS***

Alexandre Swayne

Mentor: Towia Libermann, PhD

Scientific Advisor: Luiz Zerbini, PhD

Structurally different non-steroidal anti-inflammatory drugs (NSAIDS) have been found to induce apoptosis in cancer cells. Furthermore, it has also been shown that these particular NSAIDS also induce mda-7/IL-24. Mda-7/IL-24 is of great interest because, it has been found to mediate apoptosis in human cancer cells. The strong correlation between NSAIDS and IL-24 leads the project, which we are working on: to test if IL-24 is a critical gene for the induction of apoptosis in Renal Cell Carcinoma (RCC) by NSAIDS. In order to test this we will infect two RCC cell lines (ACHN, UOK) with a lentivirus called siRNA (Silencing interference RNA) against IL-24, which will block IL-24 gene expression. The GFP gene in the same vector will allow us to see which cells have been successfully infected with the siRNA. When completed we will treat the two cell lines with NSAIDS and check the apoptosis induction in the RCC cell lines that have IL-24 gene blocked. The apoptosis of the cell lines will be tested in comparison with the control in order to see if in fact, IL-24 has a essential role in the induction of apoptosis by NSAIDS in RCC.

***The Benefits of Array CGH
and the Fight against Cancer***

Fittz-Patrick Noel

Mentor: John Quackenbush, PhD

Scientific Advisor: Renee Rubio

Cancer is a disease that involves both genetic and environmental factors. If scientists had a better understanding of what genes were causing this disease or what problems occur in the DNA, of an individual, then it could make all the difference when it comes to finding a "cure." Researchers have developed a method called array Comparative Genomic Hybridization (aCGH). This method involves taking genomic DNA from tumor samples and from normal tissue, then attaching florescent probes to the DNA and hybridizing them onto human DNA microarray chips. Through analysis of this data, scientists hope to discover actual mutations in the genetic code of individuals with cancer; for example, a deletion or an insertion. Results are still unknown but in progress. However, the projected outcome could be beneficial to detect what genes are involved in cancer and the corresponding mutations that occur in the DNA.

Developing a Genetically Engineered Breast Cancer Model for Drug Discovery

Olufunke Buraimoh

Mentor: J. Dirk Iglehart, MD

Scientific Advisors: Jean Zhao, PhD, and Erin Allgood

The purpose of this study is to develop an advanced breast tumor model for preclinical test of drugs that target most prevalent cancer causing genes, such as PIK3CA, in breast tumors. Recently, mutations have been identified in the PIK3CA gene in more than 25% of patient breast tumors. The PIK3CA gene encodes a kinase termed phosphatidylinositol 3-kinase (PI3K) that regulates many cellular activities. The mutations in the PIK3CA gene result in the constitutive activation of the enzyme PI3K, thus making PI3K a potential drug target. A previously established human mammary epithelial cell (HMEC) transformation system with defined genetic alterations, including inactivation of p53 pathway and activation of the PI3K pathway will be used in this study. It has been subsequently shown that the tumor mutant alleles of PIK3CA are able to induce these HMEC cells to grow as tumors in host animals. In order to facilitate drug discovery using this model system, a retroviral construct expressing Green Florescent Protein (GFP) will be utilized. This GFP marker will then be introduced to HMEC cells transformed with oncogenic PIK3CA. These GFP-expressing cells will be injected into mammary tissue of experimental mice to raise a breast tumor carrying tumorigenic PIK3CA and GFP. This model can be used to monitor in vivo tumor growth and to observe the effects of drug treatments, such as PI3K inhibitors. By using a murine model for breast cancer research, potential drugs that target PI3K may be discovered for human breast cancer patients.

Promoter Sequence Labeling Using GFP Markers

Ezra Luna Star

Mentor: Robert Handin, MD

Scientific Advisor: Jing Zhang, PhD

Janus Kinase (JAK) is a tyrosine kinase that has been found in numerous biological systems to be a pre-signaling device in cytokine signaling pathways. There are numerous forms of JAK, such as JAK1, JAK2, JAK3, TYK2. These JAK proteins are important because cytokines regulate proliferation and cell differentiation as well as controlling cell functions during hematopoiesis. The JAK2 protein has two variants within fish: JAK2a and JAK2b. Fish eggs (specifically Zebrafish [*Danio rerio*]) allow us to examine the development cycle (in vivo) of an organism because the Zebrafish eggs are translucent therefore allowing for observation of cell lines and systems. This technique is also useful as a way to witness directly (as opposed to the numerous in vivo techniques available) whether or not a specific region of the gene has been isolated. Using fish embryos is very effective in experiments that are seeking to find the location and size of specific promoter sequences. To do this, a GFP marker is placed within the gene sequence (by way of a plasmid vector), cut with enzymes, and then placed within the selected area of the DNA. In trying to find the Promoter sequence for JAK2, which is primarily expressed within the hematopoietic system the marker and Plasmid are placed within the egg, as the egg develops and if the correct region of the promoter sequence has been labeled, than the GFP marker will be expressed within the blood of the developing fish. In this way it is immediately possible to determine whether or not the selected region of the gene contains the promoter sequence.

Factors Contributing to the Regression of Infantile Hemangioma

Jonathan Pham

Mentor: M. Judah Folkman, MD

Scientific Advisors: Carmen Barnes, PhD and
Emily Christison-Lagay, MD

Transforming Growth Factor-Beta (TGFb) Requirements for Tumorigenicity and Metastasis in Pancreatic Cancer

Afamefuna Nduaguba

Mentor: Nebeel El-Bardeesy, PhD

Scientific Advisor: Kuang Cheng, PhD

Introduction: Infantile hemangioma (IH) is the most common pediatric tumor, affecting 4-5% of Caucasian infants. It is a benign endothelial cell neoplasm, which exhibits rapid growth during infancy followed by slow involution during childhood. We are interested in investigating the unusual timing of IH growth and involution in infants. It is our hypothesis that the cellular basis for this change may be attributed to differentiation of resident mesenchymal stem cells in proliferating IH to adipocytes. As the stromal compartment in IHs plays a role in tumor growth by stimulating proliferation of IH ECs in a paracrine fashion, stromal differentiation into fatty tissue may result in deprivation of local growth factors, thereby contributing to involution. Conversely, IH EC apoptosis during the involuting phase may trigger MSC differentiation. Thus, by targeting the IH stromal compartment with adipogenic drugs, differentiation may accelerate involution by reducing local fibroblast (FB) and MSC production of stimulatory factors. **Methods:** The ability of various ages of IH to differentiate into fat was assessed through incubation of stromal cells for 14 days with 7 types of adipocyte differentiation media. Following differentiation, the cells were stained with oil-red-o and quantitation using computer software (IP Lab). We subsequently used Western blot to investigate the angiogenic profiles of tissue lysates and cultured stromal cells. Human Microvascular Endothelial Cells (HMVEC) were co-cultured with undifferentiated and differentiated stroma derived from IH. Proliferation assays were performed after 3 days of co-culture. **Results:** IH demonstrated a comparable ability to differentiate into adipocytes as our positive control: MSC. Fibroblasts and a related vascular tumor, the non-involuting congenital hemangioma (NICH), showed significantly less differentiation. The greatest degree of differentiation was observed using human serum (10%) in the presence of isobutylmethylxanthine, dexamethasone, and insulin. The stroma of later stage lesions appeared to express a slightly greater amount of thrombospondin, a known antiangiogenic agent. The degree of stromal differentiation did not affect HMVEC proliferation in our 3 day assay.

Pancreatic ductal adenocarcinoma (PDAC) is the fourth leading cause of cancer death in the United States. Due to a dearth of early detection methods, and lack of effective therapies, the prognosis is very poor at the time of diagnosis with a 5-year survival of only 3%. An understanding of the molecular basis of PDAC could lead to earlier detection, targeted therapies and improved prognosis. Mutational activation of the KRAS oncogene and inactivation of the INK4A tumor suppressor gene are observed in virtually all cases of PDAC whereas mutations in the SMAD4 tumor suppressor occur in 50% of cases. Smad4 is a central regulator of the TGFb signaling pathway that regulates many cell functions such as differentiation, proliferation and apoptosis. A genetically engineered mouse model demonstrated that Kras activation initiates pre-malignant pancreatic lesions, and that the inactivation of the INK4A tumor suppressor gene facilitates malignant progression of PDAC. Furthermore, Smad4 inactivation in this model leads to more rapid PDAC development. Interestingly, tumors lacking Smad4 exhibit a less aggressive phenotype than those with Smad4 intact, suggesting that Smad4 may contribute to suppression of early tumors but promote invasion and metastasis in advanced cancer. In this study, we address the hypothesis that that TGFb-SMAD4 signaling may serve to enhance the malignant growth of the subset of established PDAC with SMAD4 intact. We engineered retroviral vectors, encoding dominant negative mutants of Smad4 and of the TGFb type II receptor that genetically inactivate TGFb-SMAD4 signaling at different levels. Introducing these vectors into a series of Smad4 deficient and proficient PDAC cell lines, we expect to see different changes of morphology and aggressive behavior between genotypes. We expect the loss of tumorigenicity in cell lines with functional SMAD4, while SMAD4 null cell lines will be resistant to this treatment. Our findings suggest that the subset of patients with PDAC expressing Smad4 could benefit from the introduction of therapies that target TGFb-SMAD4 signaling.



Thursday, August 24, 2006
Presentations

Julia Kimotho

The Characterization of TCRb-FCp33 Receptor Signaling within the Facilitating Cell

Marilyn M. Gomez

Her2/Neu Minigene and Full Gene Nucleofection into K562/A2 Cells

Sheila Jeudy

The Role of the TSP-1 – CD36 Complex in Breast Cancer Progression

Lisa Barros

Prevalence and Correlates of Smoking Behavior among Adults with Cancer

Jean T. Loreus, Jr.

Preoperative Breast MRI: Who Should Get It?

Emily Caulfield


The Mechanism of APF Function in Interstitial Cystitis

Blessing Agunwamba

Does Mullerian Inhibiting Substance Cause Cellular Regression Through Interference with Tyrosine Phosphorylation?

Nancy J. Roy

Understanding Tumor Vasculature



Friday, August 25, 2006
Presentations

Jonathan Pham

Factors Contributing to the Regression of Infantile Hemangioma

Ezra Luna Star

Promoter Sequence Labeling Using GFP Markers

Alexandre Swayne

Renal Cell Carcinoma: The study of MDA-7/IL-24 and NSAIDS

Meera Sreedhara

Forkhead Family of Transcription Factors and Pancreatic Cancer

Louka Abed

The Origin of Carcinoma Associated Fibroblasts (CAFs)



Understanding Tumor Vasculature

Nancy J. Roy

Mentor: David Louis, MD

Scientific Advisor: Jennifer Roy

In treating tumors, the recent therapy of choice is the use of anti-angiogenic therapies to destroy the source of blood and nutrients the tumor has created for itself by forming its own blood vessels. In implementing this method of therapy the hope is that the tumor will diminish when it no longer has the nutrients it needs. In our study, we examine the expression of PDGFRB (platelet-derived growth factor receptors, specifically beta) in anaplastic oligodendrogliomas. The goal of this study is to better understand when and how tumor vasculature is created in this particular subset of malignant gliomas. In studying PDGFRB expression, IHC (Immunohisto Chemistry) will be the primary method of study. By optimizing a protocol for this particular antibody, we can then study PDGFRB expression on various tissues, particularly oligodendrogliomas. The projected outcome is that fully understanding the expression of this gene we can better understand more effective ways of therapy for these and possibly other gliomas.

The Characterization of TCRb-FCp33 Receptor Signaling within the Facilitating Cell

Julia Kimotho

Mentor: Yolanda Colson, MD, PhD

Scientific Advisor: Kendra Taylor, PhD

Bone marrow transplantation is currently the major therapy for treatment of some hematologic diseases. However, the clinical application of this therapy is limited by the occurrence of graft vs. host disease (GVHD). Recent studies have shown that the transplantation of Facilitating Cells (FC), a bone marrow-derived cell population, promotes stem cell reconstitution without the occurrence of GVHD. However, the mechanism required for FC function is not known. Recent developments have demonstrated that FC-mediated SC reconstitution and transplantation tolerance are dependent on the expression of a unique TCRb-FCp33-FcRg receptor complex. Moreover, Fc receptor g-chain, (FcRg), is capable of transmitting intracellular TCRb signal through the activation of protein tyrosine kinases Zap70 or Syk. We have analyzed the gene expression profiles of Syk and Zap70, using real time PCR. The current studies demonstrate that Syk gene expression is greater than Zap70 expression within the FC, suggesting that Syk may be utilized in the TCRb-FCp33-FcRg signaling pathway. Therefore, the goal of my project is to determine if indeed Syk is utilized at a molecular level and biochemical level by determining its phosphorylation state within the FC.

Her2/Neu Minigene and Full Gene Nucleofection into K562/A2 Cells

Marilyn M. Gomez

Mentor: Karen Sue Anderson, MD, PhD

Scientific Advisor: Donna Drakoulakos

Does Mullerian Inhibiting Substance Cause Cellular Regression Through Interference with Tyrosine Phosphorylation?

Blessing Agunwamba

Mentor: David MacLaughlin, PhD

Her2/neu (also called ErbB-2) is a gene member of the epidermal growth factor receptor which when amplified can lead to over expression and uncontrolled cell division. Several mutations associated with ErbB-2 are a predisposition for cancer. The purpose of this study is to determine if a full gene and a mini gene construct for Her2/neu can be effectively transfected into K562/A2 cells and recognized by T-cells. To examine these possibilities the minigene construct was made and TOPO cloned into a mammalian expression vector. PCR analysis was performed to test the construct. The DNA sequencing of the Her2/minigene indicate that the construct was effective. Also, recent flow cytometry analysis demonstrate that a full Her2/neu gene version can be successfully transfected into K562/A2 cells. Further analysis needs to be performed to confirm the mini gene's ability to transfect into K562/A2 cells as well as testing for T-cell recognition.

Mullerian Inhibiting Substance (MIS) is a glycoprotein that causes the regression of the Mullerian duct in male embryos. The Mullerian duct gives rise to the internal female reproductive organs: the fallopian tubes, ovaries, uterus, cervix and upper third of the vagina. These organs arising from the Mullerian duct retain receptors for MIS even in adulthood. Just as the ability of MIS to cause the regression of the Mullerian duct in male embryos depends on attachment to these receptors, the inhibiting function of MIS in adult female reproductive organs still depends on the presence of receptors on the organs. Cancers arising from Mullerian tissues often retain functioning MIS receptors on their cell surfaces. MIS has been tested and proven to cause regression of cancers of this type both in vitro and in vivo in mice. This study aims to clarify how MIS interferes with cell growth to cause regression. On the cell surface of Mullerian tissues, epidermal growth factor receptor (EGFR) plays an important role in regulating cell growth through its ability to be a tyrosine kinase in the presence of epidermal growth factors (EGF). Tyrosine is an amino acid that prompts a signal transduction pathway leading to cell growth when phosphorylated. Therefore, this study tries to answer whether or not MIS inhibits cell growth by stopping the phosphorylation of tyrosine at EGFR. Two reagents will be used to measure the interaction between MIS and EGF. Vanadate will mimic EGF by causing tyrosine to remain phosphorylated. The cell growth prompted by Vanadate should diminish with the introduction of MIS if MIS blocks the production of phosphotyrosine. The second reagent, Okadaic acid blocks EGFR's ability to phosphorylate tyrosine while preventing the dephosphorylation of serine and threonine on MIS receptors. The amino acids, serine and threonine, both prevent cell growth when phosphorylated. If Okadaic acid mimics MIS, its ability to keep serine and threonine phosphorylated should cause regression in Mullerian tissues.

The Mechanism of APF Function in Interstitial Cystitis

Emily Caulfield

Mentor: Michael Freeman, PhD

Scientific Advisor: Jayoung Kim, PhD

Previous studies have shown that a peptide biomarker found in the urine of IC patients can inhibit cell growth in human urothelial cells. IC, or Interstitial Cystitis, is a chronic bladder syndrome characterized by inflammation of the bladder wall with pelvic and/or perineal pain and urinary frequency and urgency. Although the peptide has been detected, the underlying molecular cause of IC is unknown. The main objective of this study is to have a better understanding of the mechanism stimulating the peptide biomarker, antiproliferative factor (APF), into action. Observations were made from increasing APF levels and regulating p53 protein levels in human urothelial cells and T24 bladder carcinoma cells. The increasing of APF levels resulted in increased expression of p53. When p53 levels were altered, biological responses were detected. From these findings, it was identified that APF regulates p53 expression levels. The results also demonstrate that APF's functions may possibly be mediated by p53.

The Role of the TSP-1 – CD36 Complex in Breast Cancer Progression

Sheila Jeudy

Mentor: John Lawler, PhD

Scientific Advisor: Shideh Kazerounian, PhD

Thrombospondin-1 (TSP-1) and CD36 are naturally synthesized by most cell types including microvascular endothelial cells, platelets, and some tumor cells such as breast cancer cells. CD 36 is a transmembrane glycoprotein and a critical receptor for TSP-1 in endothelial cells. TSP-1 is an extracellular glycoprotein that binds to 12 different receptors depending on the cell type. In endothelial cells, TSP-1 binds to CD36 to mediate apoptosis and antiangiogenic events. TSP-1 also inhibits endothelial cell proliferation by inhibiting VEGF-activated survival pathways. Physiological levels of TSP-1 support breast cancer metastasis, but the role of CD36 and its effect on TSP-1 signaling pathways in these cells is not known. Our hypothesis is that TSP-1 binds to CD36, which may bind to other specific proteins, initiating signaling pathways that regulate tumor progression. One known signaling protein is Syk tyrosine kinase. Syk is expressed in tumor cells at growth, but as cells become more metastatic, its expression level decreases. Therefore, one possibility is that TSP-1 binds to CD36 and changes either the expression or phosphorylation of Syk, stimulating tumor progression. To investigate our hypothesis, various antibodies are being used in western blots of mice tumor tissues at various stages of tumor progression to determine the expression level of TSP-1, CD36 and Syk. We will also perform adhesion and migration assays to determine the importance of these proteins in tumor cell invasion and migration. Immunofluorescence assays of human and mice breast cancer cell lines will also be performed to determine the subcellular localization of TSP-1, CD36 and Syk in breast cancer cell lines. All together, our results will enable us to establish a mechanism by which binding TSP-1 to CD36 regulates breast cancer progression.

Prevalence and Correlates of Smoking Behavior among Adults with Cancer

Lisa Barros

Mentor: Mary E. Cooley, PhD, CRNP

Scientific Advisor: Elizabeth Xiarhos

Problem: The time surrounding the diagnosis of an illness presents an opportunity to prompt smokers to change their smoking behaviors. Smoking cessation after cancer is important because it is associated with increased quality of life and increased survival. Because lung and head and neck cancers are the most common smoking-related malignancies, these cancers are the focus for the study. The information gathered will provide direction to develop a smoking cessation intervention.

Purpose: 1) Estimate point-prevalence abstinence and continued abstinence rates among adults with newly diagnosed lung and head and neck cancers at 3 and 6 months after study entry; 2) Describe smoking cessation interventions received by smokers cancer; 3) Describe participant flow related to screening and enrollment, 4) Identify the most common reasons for participant exclusion. **Methodology:** Data will be collected at entry into the study and at 3 and 6 months later. Interviews, standardized questionnaires, biochemical verification of smoking status, and medical records will be used to gather information. A database will be maintained to monitor recruitment. **Results:** One-thousand-fifty-seven participants have been screened for eligibility. One-hundred-fifty-seven participants were eligible and of these patients, 102 were enrolled. The most common reasons for exclusion into the study were being a former or never- smoker, having an ineligible diagnosis and not receiving follow up at the study sites. **Conclusion:** Recruitment for studies examining smoking behavior in adults with cancer is challenging. Many patients are former or never-smokers. Future studies should expand inclusion criteria to encompass multiple cancer types and study sites.

Preoperative Breast MRI: Who Should Get It?

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Background: Prospective randomized studies of surgical therapy for breast cancer have shown that surgical decision-making, based on clinical data and conventional imaging, results in low rates of IBTR and contralateral cancers. Breast MRI has emerged as an imaging adjunct suggested to better define the true extent of subclinical ipsilateral and contralateral disease. Due to high sensitivity but low specificity of MRI, its accuracy and cost-effectiveness require further evaluation. The purpose of this study was to identify the subset of breast cancer patients with the greatest potential to benefit from preoperative MRI. **Methods:** The records of approximately 300 primary breast cancer patients treated between 12/97 and 12/03 were reviewed. Mammography and ultrasound (bilateral or focused) were supplemented by bilateral breast MRI before definitive surgery. Patients in whom MRI changed surgical plans (group 1) were compared to those in whom it did not (group 2). **Results/Conclusion:** Data extraction and statistical analysis are ongoing. Based on prior literature we anticipate that breast MRI will be beneficial in a small group of primary breast cancer patients in planning their definitive treatment.