CDMRP

Department of Defense

Peer Reviewed Cancer Research Program

U.S. Army Medical Research and Materiel Command

History

The Office of the Congressionally Directed Medical Research Programs (CDMRP) was created in 1992 from a powerful grassroots effort that resulted in a Congressional appropriation of funds for breast cancer research. This initiated a unique partnership among the public, Congress, and the military. Since then, the CDMRP has grown to encompass multiple targeted programs and has received over $9.1 billion (B) in appropriations from its inception through fiscal year 2016 (FY16). Funds for the CDMRP are added to the Department of Defense budget to support individual programs, such as the Peer Reviewed Cancer Research Program (PRCRP), as directed by Congress.

The Peer Reviewed Cancer Research Program

Since its inception in FY09, the PRCRP has been charged by Congress to fund innovative basic, applied, and translational cancer research to support Service members, their families, Veterans, other military beneficiaries, and the American public.

To accomplish the vision of improving the quality of life of Service members and their families by decreasing the impact of cancer, the PRCRP promotes funding innovative and translational studies across the research landscape. The PRCRP offers funding opportunities that focus on addressing potential environmental cancer risks, and answering the gaps in the cancer care spectrum from prevention, through detection and diagnosis, prognosis, treatment, and survivorship. Through innovative award mechanisms, the PRCRP strives to enhance the health and well-being of military beneficiaries while supporting the research community to create a strong partnership between the Defense Health Agency, researchers, and patients.

The Program Cycle

The PRCRP holds an annual Vision Setting meeting to identify research gaps and define an investment strategy for the program year. Vision setting is conducted by the Programmatic Panel, a panel of experts including scientists, clinicians, and consumers (patients, or caregivers, affected by cancer). The Programmatic Panel recommends an investment strategy for soliciting the best applications that address the Congressionally directed areas of cancer research.

The PRCRP utilizes the CDMRP two-tier review of applications that is based on the recommendations set forth by the Institute of Medicine committee in 1993. The two tiers of review are peer review and programmatic review; although the two tiers have different goals, they are complimentary. Peer review, the first tier of application evaluation, is a scientific peer review of applications measured against established criteria for determining scientific merit. Programmatic review, the second tier of application evaluation, is conducted by the Programmatic Panel, which compares applications to each other and makes recommendations for funding based on scientific merit, portfolio balance, and relevance to program goals.

Malignancies Associated with Military Service

Members of the military are exposed to hazardous environments and dangerous deployments due to the nature of their service. Hazardous exposures can lead to the development of various cancers, many of which present a potential risk for Service members and their families. The table below lists many potential hazards identified in the environment—that Service members may be at higher risk of encountering—and the associated cancers that these hazards can cause.

Exposure-Related Cancer Risk

Ultraviolet Light
Melanoma; basal cell carcinoma; squamous cell carcinoma; other skin cancers

Agent Orange
Chronic B-cell leukemia; Hodgkin's disease; multiple myeloma; non-Hodgkin's lymphoma; respiratory cancers; soft tissue sarcomas; bladder cancer

Radiation
All cancers, but in particular, cancers of the bile ducts, bone, brain, colon, esophagus, gall bladder, liver, pancreas, pharynx, salivary gland, small intestine, stomach, thyroid, and urinary tract; leukemia (except chronic lymphocytic leukemia); lymphomas (except Hodgkin's); multiple myeloma

Asbestos
Mesothelioma; gastrointestinal, colorectal, throat, kidney, esophagus, and gall bladder cancers

Infectious Agents
Anogenital cancers; cervical cancer; Burkitt lymphoma; hepatocellular carcinoma; Kaposi sarcoma; leukemia; gastric cancers

Industrial Solvents
Leukemia; liver cancer; biliary tract cancer; kidney cancer; non-Hodgkin's lymphoma; brain cancer; blood cancer

Source: U.S. Department of Veterans Affairs, Public Health
http://www.publichealth.va.gov/exposures/index.asp
http://www.infectagentscancer.com
http://www.va.gov/vetapp07/files2/0717857.txt

Vision
To improve quality of life by decreasing the impact of cancer on Service members, their families, and the American public

Mission
To successfully promote high-impact research for cancer prevention, detection, treatment, and survivorship

Congressional Appropriations
FY09–FY15: $149.8M
FY16: $50M

PRCRP Invests in Different Types of Cancer

In each fiscal year, the PRCRP receives its funds and topic areas through the annual Congressional legislation known as the Defense Appropriations Act. The dollars to fund the PRCRP are added every year during the budget approval cycle by the members of the House or Senate in response to requests by the American public (consumers, disease survivors, caregivers); therefore, topic areas can vary from year to year. Since its inception in FY09 through FY16, the PRCRP has been tasked with funding research projects in 20 Congressionally directed topic areas. The topic areas span a wide range of cancer research areas. A complete list of topic areas and the associated fiscal year is provided below.

Topic Area	FY09 ($16M)	FY10 ($15M)	FY11 ($16M)	FY12 ($12.8M)	FY13 ($15M)	FY14 ($25M)	FY15 ($50M)	FY16 ($50M)
Bladder cancer								✔
Blood cancers		✔	✔	✔	✔	✔		
Cancers related to radiation exposure						✔		
Colorectal cancer		✔	✔	✔	✔	✔	✔	✔
Genetic cancer[1]	✔	✔	✔	✔	✔	✔	✔	
Immunotherapy								✔
Kidney cancer		✔	✔	✔	✔	✔	✔	✔
Listeria vaccine for cancer		✔	✔	✔	✔	✔	✔	✔
Liver cancer							✔	✔
Lymphoma								✔
Melanoma and other skin cancers[2]	✔	✔	✔	✔	✔	✔	✔	✔
Mesothelioma			✔	✔	✔	✔	✔	✔
Myeloproliferative disorders						✔	✔	
Non-Invasive cancer ablation[3]	✔							
Neuroblastoma					✔	✔	✔	✔
Pancreatic cancer			✔	✔	✔	✔	✔	✔
Pediatric brain tumors	✔	✔		✔	✔	✔		✔
Pediatric cancer			✔					
Radiation protection utilizing nanotechnology		✔	✔					
Stomach cancer							✔	✔

[1] Topic area includes FY09 congressional language: genetic cancer and its relation to exposures to the various environments that are unique to the military lifestyle, and the FY10 congressional language: genetic cancer research and genomic medicine.
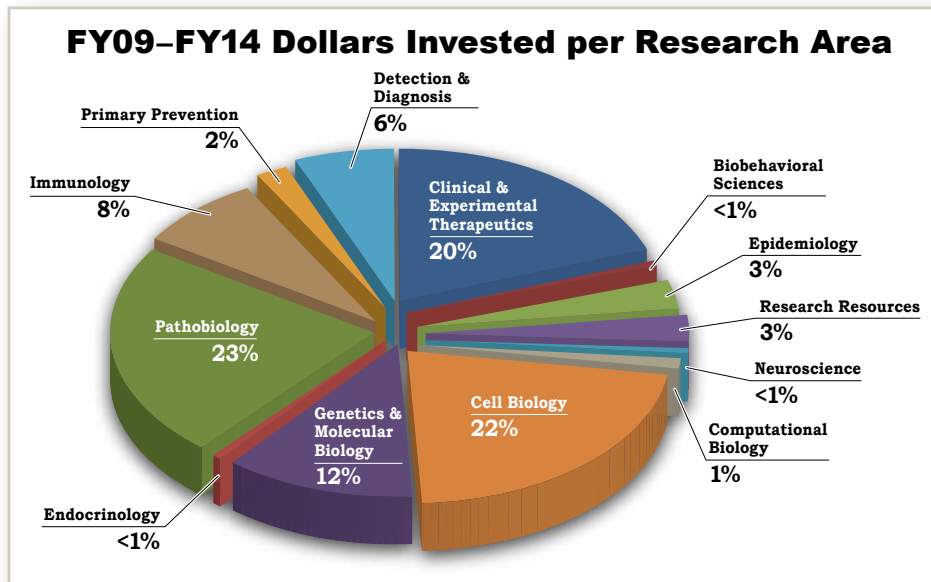
[2] Topic area includes FY09 congressional language: melanoma and other skin cancers as related to deployments of Service members to areas of high exposure, and the FY10-FY16 congressional language: melanoma and other skin cancers.

[3] Non-invasive cancer ablation treatment including selective targeting with nanoparticles.
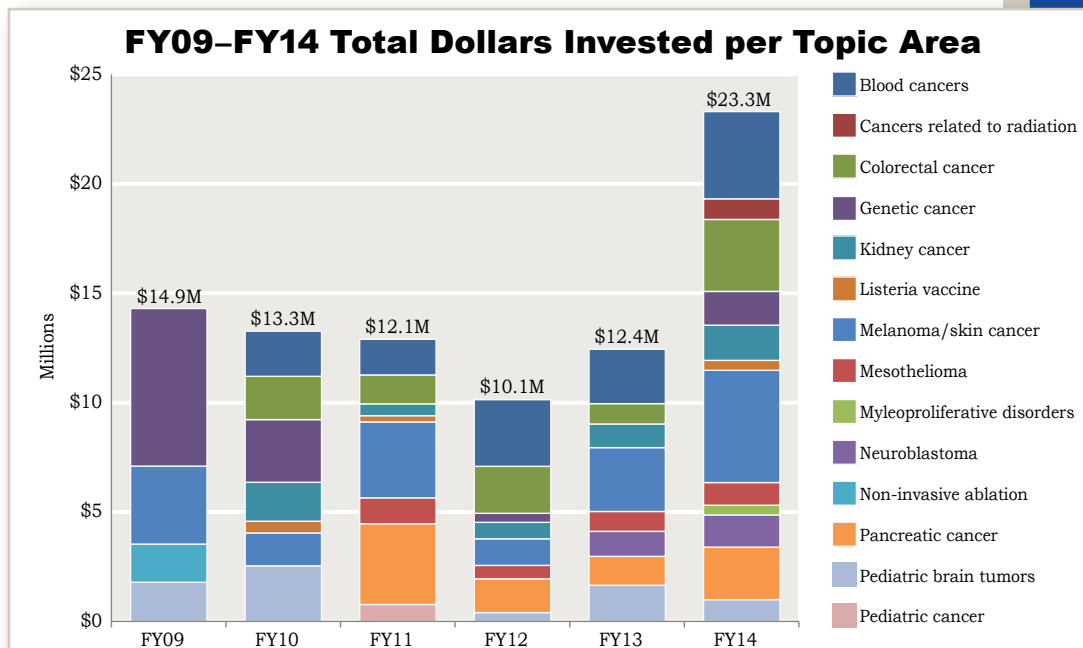
Investment Portfolio per Research Area

The research funded by the PRCRP targets the current unmet gaps in cancer research. The PRCRP awards have been categorized by the scientific classification system (as shown below). These areas include basic science studies in pathobiology, cell and molecular biology, and translational studies in clinical and experimental therapeutics, pathobiology, and detection and diagnosis.

FY09–FY14 Dollars Invested per Research Area

- Detection & Diagnosis 6%
- Primary Prevention 2%
- Immunology 8%
- Pathobiology 23%
- Endocrinology <1%
- Genetics & Molecular Biology 12%
- Cell Biology 22%
- Computational Biology 1%
- Neuroscience <1%
- Research Resources 3%
- Epidemiology 3%
- Biobehavioral Sciences <1%
- Clinical & Experimental Therapeutics 20%

Each year, the Congressional appropriation to PRCRP is distributed among applications with respect to scientific merit and portfolio balance within each of the topic areas (as indicated on page 4). Shown below is a depiction of the distribution of funds per topic area.

FY09–FY14 Total Dollars Invested per Topic Area

Millions

- FY09 $14.9M
- FY10 $13.3M
- FY11 $12.1M
- FY12 $10.1M
- FY13 $12.4M
- FY14 $23.3M

Legend:
- Blood cancers
- Cancers related to radiation
- Colorectal cancer
- Genetic cancer
- Kidney cancer
- Listeria vaccine
- Melanoma/skin cancer
- Mesothelioma
- Myleoproliferative disorders
- Neuroblastoma
- Non-invasive ablation
- Pancreatic cancer
- Pediatric brain tumors
- Pediatric cancer

Final or Current Invested Amounts (withdrawals, funds returned affect total amounts, etc.) Excludes USAMRMC, CDMRP, PRCRP management costs (~10% of the total appropriation).
No full applications were submitted for Radiation Protection Utilizing Nanotechnology (offered in FY10-FY11).

A unique aspect of the CDMRP is the active participation of consumers throughout the program's annual cycle. For the PRCRP, consumers are cancer survivors of the topic areas listed in the PRCRP program announcements, or family members of pediatric patients affected by the pediatric cancer topic areas.

Michael Holtz – Served 2013, 2015

Michael Holtz is many things—an author, a speaker, and a boot camp instructor. He is also a rectal cancer survivor. In December 2011, Michael began experiencing strange digestive symptoms. He underwent a colonoscopy in March 2012, during which he was diagnosed with stage 3B rectal cancer. During the following 11 months he underwent a barrage of chemotherapy, radiation therapy, and surgery. Because the tumor was much larger than the doctor had anticipated, and because of the scar tissue caused by the radiation treatments, Michael now lives with a permanent colostomy. Additionally, Michael continues to experience persistent neuropathy in his feet due to the chemotherapy. Although he lives with these continuing challenges, he has been cancer free since April 2013.

Michael's cancer experiences motivated his participation in activities that could raise awareness of rectal cancer. He works with organizations such as the American Cancer Society Cancer

Kay Kays – Served 2011–2015

As a 22-year pancreatic cancer survivor, Kay Kays brings a wealth of experience to the PRCRP. While undergoing the treatments for her own pancreatic cancer, Kay saw the need for more open communication and education between researchers, clinicians, and patients. To help bridge this gap, she co-founded the first Pancreatic Cancer Network in Arizona at The Wellness Community to disseminate information and build a support community for pancreatic cancer patients, families, and friends. She later co-founded the Tissue Donor Awareness Program to help educate patients about the importance of donating tissue for research. Kay has also participated in legislative visits and grant reviews for the Pancreatic Cancer Action Network (PanCAN).

In 2011, with the inclusion of pancreatic cancer under PRCRP, a call for consumer reviewers was released. PanCAN nominated Kay to serve on the pancreatic cancer peer review panel for PRCRP. As a long-term survivor of pancreatic cancer, Kay serves the panel not only as a reviewer

Richard Mosca – Served 2011, 2013, 2015

Mesothelioma is an orphan disease with about 3,000 cases being diagnosed each year. Therefore, funding for research is difficult to obtain. Patients, doctors, and others who make up the mesothelioma community were thrilled when the disease was added as a topic area to the Peer Reviewed Medical Research Program in FY08. The Meso Foundation nominated Richard Mosca to be a consumer reviewer that first year. Richard is a 9-year survivor of mesothelioma, and looking back, it is a milestone he did not think he would reach because the average survival period of a mesothelioma patient is only 12 months. Richard was a little scared leading up to serving on the panel, thinking of the prospect of being in a conference room with 15 scientists listening to his every word. However, each person on the panel went out of their way to make him feel comfortable. He has served as a consumer reviewer for the last 7 years, continuing his service on the panel after mesothelioma became a topic area under the PRCRP in FY11. For Richard, two of his most important activities are his participation with the Mesothelioma

Perspectives

Consumers bring their unique perspectives and experiences to panels across all levels of review, and instill a sense of urgency and focus to the panels on which they serve. They are active in outreach, and support organizations, and are the representative voice of the consumer community. As integral members of the review panels, consumers are full voting members and have great potential to make a significant impact on the various cancer communities.

Action Network, the Subway Race Against Cancer to support his treatment center's community outreach, and the Colon Cancer Alliance. His book, *It's Not Harder Than Cancer,* offers encouragement and includes life lessons he learned during his own cancer journey. In 2013, Michael was nominated by the Colon Cancer Alliance to serve as a consumer reviewer for PRCRP during peer review. He was honored and thrilled to have a role in the peer review process, assessing the impact of applications that may lead to improved detection and treatment of colorectal cancer.

Consumer reviewers *"are the voice of hope to help scientists understand what is at stake."* Michael feels that the scientific community genuinely appreciates the consumer reviewers, who *"bring real-life perspective of cancer survivors to the academic process of [peer reviewing] research proposals."*

but also a mentor to the other consumer reviewers included on the PRCRP pancreatic cancer review panels. When asked to describe her experiences with the PRCRP, she feels that her views are respected during peer review discussions. Kay is pleased that the voices of individuals whose lives have been impacted by cancer are included in the process.

"The honor of representing our Service men and women, and their families [during peer review] is very emotional and priceless for me."

From her years working in different advocacy areas, Kay has had the opportunity to personally speak with military pancreatic cancer patients. As she participates on the pancreatic cancer peer review panel, she feels *"their words ring strong [as she] reviews research grants for military relevance."*

Applied Research Foundation (Meso Foundation) and serving as a peer reviewer for the CDMRP. In retirement now, Richard spends his time traveling and playing golf with his wife, going to the theater, and visiting with his grandchildren. He also urges others who may have the opportunity to serve as a consumer reviewer to do so.

"There is nothing more rewarding than sitting in a room with some of the most intelligent people you will ever meet, and changing their evaluation of a grant proposal because they didn't see it from a patient's point of view."

For more information about Richard, please visit the CDMRP website: *http://cdmrp.army.mil/cwg/stories/2010/mosca_profile.shtml.*

Below are representative highlights for each of the topic areas for which the PRCRP has funded projects.

BLOOD CANCERS

Left: Dr. Michaela Reagan, now an Associate Professor at the Maine Medical Center Research Institute. Right: Dr. Irene M. Ghobrial, Dr. Reagan's mentor at the Dana-Farber Cancer Institute.

Michaela Reagan, PhD
Dana-Farber Cancer Institute
Improved models of how multiple myeloma (MM) progresses in the bone marrow are urgently needed to facilitate the development and clinical translation of MM inhibitors. Dr. Reagan developed a novel cell culture model based on a silk-protein scaffold that better mimics tumor growth in the bone than traditional culture models. This new model was used to study how MM grows in a bone marrow-like environment and how MM impairs bone growth. She identified the first microRNA (miR), a short regulatory nucleic acid, abnormally expressed in bone cancer patients. Targeting this miR could lead to therapeutics that enhance bone healing to fight cancer-induced bone disease and potentially reduce tumor burden. Dr. Reagan is now more fully elucidating the effects of bone cells on cancer cells in her new independent laboratory at the Maine Medical Center.

Dr. Reagan's work is important for MM, a disease particularly relevant to our Veterans. Male Veterans using VA hospitals are at 51% increased risk of MM compared to the general public.

Swami A, Reagan MR, Basto P, et al. 2014. *Proc Natl Acad Sci U S A* 111(28):10287-10292.

Reagan MR, Mishima Y, Glavey SV, et al. 2014. *Blood* 124(22):3250-3259.

CANCERS RELATED TO RADIATION

Nelson Chao, MD, Duke University
Using an FY14 Idea Award with Special Focus, Dr. Chao will study a novel therapeutic target for radiation-induced hematological malignancies. Exposure to radiation can transform hematopoietic stem cells, immature cells that develop into different types of specialized blood cells. These transformed cells evade the immune system and may become malignant. Using mouse models, Dr. Chao is working on verifying the effects of protein deletion due to radiation exposure on lymphoma and myeloma progression. Additionally, Dr. Chao's team will test whether an identified inhibitor represents a new class of therapeutics that could cure radiation-induced blood cancer.

Research Achievements

John Jessup, MD, National Cancer Institute
Dr. Jessup demonstrated that the NANOGP8 gene directly modulates and maintains stem cell characteristics of colorectal carcinoma (CRC) cells. Furthermore, he used a lentiviral vector delivered short hairpin RNA (shRNA), an artificial RNA, in human CRC cells to show that the transduction of shRNA targeting NANOGP8 induces apoptosis and inhibits further cell proliferation. Unfortunately, when Dr. Jessup tested the shRNA in human CRC growing in mice, the shRNA had only a transient effect on slowing tumor growth. He and his colleagues then developed an adenovirus that contains the shRNA and grows preferentially in human CRC cells, and he demonstrated that this continuously inhibits the growth of large human CRC tumors in mice. These findings could help improve drug activity and provide a new route in therapy regimens for CRC. Dr. Jessup's research focuses on the growth of established human colorectal xenografts in preclinical models, which will benefit military personnel and dependents once it is matured into a clinical treatment.

Mattoo AR, Zhang J, Espinoza LA, and Jessup JM. 2014. *Clin Cancer Res* 20(21):5446-5455.

Zhang J, Espinoza LA, Kinders RJ, et al. 2013. *Oncogene* 32(37):4397-4405.

Sivanesan Dakshanamurthy, PhD, Georgetown University
In this FY14 award, Dr. Dakshanamurthy will develop novel in-silico methods to identify cellular pathways adversely affected by environmental chemicals to which members of our military may be exposed during their service. In collaboration with Dr. Stephen Byers, he plans to use this information to explain how these interactions are important in the formation of colorectal cancer.

Dr. Robert L. Moritz   Dr. Leroy Hood   Dr. Gregory Folz   Dr. Charles Cobbs

Robert L. Moritz, PhD, Leroy Hood, PhD, MD, Gregory Folz, PhD, MD, and Charles Cobbs, MD, Institute for Systems Biology and the Swedish Neuroscience Institute
Drs. Moritz, Hood, Folz, and Cobbs have developed techniques in proteogenomic analysis for whole-genome sequencing of cancer patients' tumors, individual quantized tumor cells, non-cancerous cells, and non-cancerous cells from the patients' family members to perform inheritance analysis and identify candidate genes involved in neoplastic glioblastoma formation. These techniques, combined with proteomic analysis tools, have led to the identification of perturbed networks of genes, providing promising insights for the discovery of novel targeted therapeutics for glioblastoma and identification of novel blood biomarkers for early detection. Identification of biomarkers for early detection of cancer answers an unmet gap in the cancer care spectrum and, therefore, supports the long-term health and well-being of military beneficiaries.

Roach JC, Glusman G, Smit AF, et al. 2010. *Science* 328(5978):636-639.

Li XJ, Hayward C, Fong PY, et al. 2013. *Sci Transl Med* 5(207):207ra142.

KIDNEY CANCER

Dr. Tewari

Dr. Pantuck

Muneesh Tewari, MD, PhD, Fred Hutchinson Cancer Research Center
Allan Pantuck, MD, University of California, Los Angeles

Dr. Tewari and Dr. Pantuck are using a novel technology of droplet digital PCR (ddPCR) with high precision and sensitivity to identify and evaluate miR-210, a circulating regulatory fragment of RNA, called miRNA, as a blood-based biomarker for clear cell renal carcinoma. They studied and have reported in detail the sensitivity and reproducibility of the ddPCR method for measuring miR-210 and other plasma biomarkers. They hope that miR-210 can be used as a diagnostic tool in blood-based early detection of clear cell kidney cancer. The third most common urological diagnosis at the VHA system are renal masses; thus, research that helps to find these masses earlier will benefit military beneficiaries.

Hindson CM, Chevillet JR, Briggs HA, et al. 2013. *Nat Methods* 10(10):1003-1005.

LISTERIA VACCINE FOR CANCER
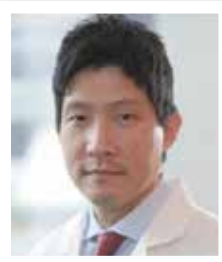
David Chung, MD, PhD, Memorial Sloan Kettering Cancer Center
A promising approach to cancer treatment is to make vaccines using specialized cells of the immune system called dendritic cells. Dr. Chung found that a noninfectious strain of Listeria monocytogenes bacteria could activate dendritic cells while avoiding excessive activation of immune-dampening factors that could impede vaccine responses. These results support the use of Listeria to boost dendritic cell vaccine efficacy. The insight gained from this project could be used to develop cancer vaccines for multiple cancers that are prevalent among our Service members, Veterans, and their families.

Chung DJ, Romano E, Pronschinske KB, et al. 2013. *J Transl Med* 11:166-175.

MELANOMA and OTHER SKIN CANCERS

Douglas Faller, MD, PhD, Boston University Medical Campus
Dr. Faller used a small molecule inhibitor of the signaling protein Protein Kinase Cδ (PKCδ) for specific targeting of melanoma with mutations in the NRAS gene, and melanomas that have developed resistance to therapeutic inhibitors of BRAF. The PKCδ inhibitor lead compound has been refined for potency and specificity. Studies are ongoing to optimize a "final" compound that will be tested for clinical relevancy using a mouse melanoma model. Military Service members are at greater risk for the development of melanoma due to increased sun exposure. As the incidence of melanoma has increased in recent years, Dr. Faller's research aims to develop a targeted and effective therapeutic approach to treat melanoma.

Chen Z, Forman LW, Williams RM, and Faller DV. 2014. *BMC Cancer* 14: 90-98.

Takashima A, English B, Chen Z, et al. 2014. *ACS Chem Biol* 9(4):1003-1014.

Daniel Bikle, MD, PhD, Northern California Institute for Research and Education
Many of our Service members are currently stationed in areas of the world where sunlight exposure is intense, and their duties often require long periods of exposure to the sun. Dr. Bikle's work aims to determine if Vitamin D can play a role in preventing the development of skin cancers. He has demonstrated that, following acute UVB exposure, there is delayed clearance of specific indicators of DNA damage in Vitamin D Receptor (VDR) knockout mice. These novel data indicate that VDR plays an important role in facilitating DNA damage repair, and any damage not repaired could potentially lead to the formation of melanoma or non-melanoma skin cancers.

Bikle DD and Jiang Y. 2013. *Cancers (Basel)* 5(4):1426-1438. doi: 10.3390/cancers5041426.

Ravi Salgia, MD, PhD, University of Chicago

Dr. Salgia targeted malignant pleural mesothelioma (MPM) with a trio of small molecule inhibitors that block three cellular signaling molecules: MET (also known as haptocyte growth factor receptor) receptor tyrosine kinase, phosphatidylinositol 3-kinase (PI3K), and mammalian target of rapamycin (mTOR). The combined use of these inhibitors was more effective than using any single drug in suppressing MPM cell motility and growth in vitro, and tumor growth in an in vivo mesothelioma mouse model. Many of our military personnel are exposed to asbestos while serving in developing countries or in Navy shipyards. Dr. Salgia's studies show promise in the development of a novel and effective treatment for mesothelioma.

Kanteti R, Dhanasingh I, Kawada I, et al. 2014. MET and PI3K/mTOR as a potential combinatorial therapeutic target in malignant pleural mesothelioma. *PLoS One* 9(9):e105919. doi: 10.1371/journal.pone.0105919.

Bridget Wilson, PhD
University of New Mexico Health Sciences Center

Drs. Wilson and Cleyrat plan to apply state-of-the-art techniques to define the interactions between three proteins (Mpl, Jak2, and calreticulin), which are key to the development of myeloproliferative neoplasm (MPN). This research could lead to preclinical studies that translate this mechanistic information into new therapeutic strategies for MPN patients, which include Service members, who are at increased risk for developing MPN.

Dr. Bridget S. Wilson (left) and Dr. Cédric Cleyrat of the University of New Mexico Comprehensive Cancer Center.

Clinton Stewart, PharmD
St. Jude Children's Research Hospital

The health and welfare of our Service members is partially determined by the health and welfare of their families. Missions benefit when the Service members' families are healthy and well. Dr. Stewart is building a novel pharmacokinetic model with an individualized tumor compartment and 3D transport model to better understand and quantify chemotherapeutic uptake in neuroblastoma tumors. This model will more accurately predict drug effect within the 3D geometry of individual neuroblastoma tumors. Completion of this model will allow clinicians to make more informed decisions on treatment dosing and scheduling.

Dr. Clinton Stewart (left) and Co-Principal Investigator Dr. Andras Sablauer.

Anthony Berdis, PhD
Case Western Reserve University

Technological advances supported by the military increase the ability of caretakers to answer the needs of Service members and their families. Over the course of this award, Dr. Berdis synthesized several novel gold-containing anti-cancer agents and demonstrated that these compounds could reduce cancer growth, with minimal signs of toxicity, when combined with clinically relevant doses of ionizing radiation. This combination could provide a new strategy to noninvasively ablate tumors.

Craig S, Gao L, Lee I, et al. 2012. Gold-containing indoles as anticancer agents that potentiate the cytotoxic effects of ionizing radiation. *J Med Chem* 55:2437-2451.

PANCREATIC CANCER

Pinku Mukherjee, PhD, University of North Carolina at Charlotte

Dr. Mukherjee observed that MUC1-overexpressing pancreatic cancer cells (>80% of pancreatic ductal adenocarcinomas) also express high levels of proteins that induce angiogenesis and metastasis, namely, Neuropilin-1 (NRP1) and vascular endothelial growth factor (VEGF), which interact with each other within the tumor microenvironment. She demonstrated that by blocking the interaction of NRP1 and VEGF, tumor burden was reduced in MUC1-overexpressing pancreatic tumors. Certain behavioral risk factors, such as smoking and the consumption of alcohol, are known to increase the chances of developing pancreatic cancer. Dr. Mukherjee's research provides insight into the mechanisms that contribute to the aggressive and deadly nature of pancreatic cancer.

Zhou R, Curry JM, Das Roy L, et al. 2016. *Oncogene* doi: 10.1038/onc.2015.516 [E-pub ahead of print].

PEDIATRIC BRAIN TUMOR/ PEDIATRIC CANCER
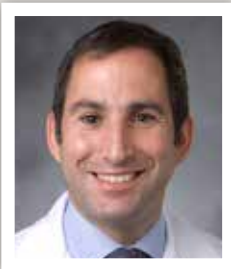
Oren Becher, MD, Duke University

Dr. Becher identified the transcription factor Pax3 as differentially expressed between cortical gliomas and brainstem gliomas, including Diffuse Intrinsic Pontine Glioma (DIPG). DIPG afflicts the pediatric population and is the leading cause of death for children with brain tumors. Expression of the Pax3 transcription factor enhances the growth and survival of murine brainstem glioma cells and is increased in 40% of human DIPGs, results that provide a novel marker with which to characterize and subtype the disease. These findings increase our understanding of DIPG classification and are important for the development of targeted therapeutics.

Misuraca KL, Barton KL, Chung A, et al. 2014. *Acta Neuropathologica Communications* 2(1):134.

Patrick Paddison, PhD, Fred Hutchinson Cancer Research Center

Dr. Paddison performed multiple genome-wide RNAi screens in patient-derived glioblastoma multiforme stem cells (GSCs) and discovered that the plant homeodomain finger domain protein PHF5A is required for GSC expansion. Additional studies revealed another protein, PKMYT1, as necessary for GSC viability. Both PHF5A and PKMYT1 are novel candidate drug targets for adult and pediatric glioblastoma tumors.

Toledo CM, Ding Y, Hoellerbauer P. et al. 2015. *Cell Reports* 13:2425-2439.

Hubert CG, Bradley RK, Ding Y. et al. 2013. *Genes Dev* 27(9):1032-1045.

Xuanming Shi, PhD, University of Texas Southwestern Medical Center

Overactive Sonic hedgehog (Shh) signaling in cerebellum granular neural precursor (CGNP) is the leading cause of childhood medulloblastoma (MB). Dr. Shi identified Brg1, a chromatin remodeler required to express the target of oncogene Shh. He observed Brg1 was integral in CGNP and tumor cell proliferation. Through deletion of Brg1, he demonstrated inhibition of MB progression via a reduction of mitogenic target genes and tumor cell proliferation.

Shi X, Wang Q, Gu J. et al. 2016. *Oncogene* (in press).

Shi X, Zhang Z, Zhan X. et al. 2014. *Nature Commun* 5:5425.

The work of Drs. Becher, Paddison, and Shi represents an important area of study for the health and well-being of military families. In order to support active-duty Service members and ensure a fit force, the military members' families must be healthy as well.

Peer Reviewed Cancer Research Recent Advancements

Mentor/Mentee Team Makes Advances in Developing a Noninvasive Tool for Assessing Renal Cancer Aggressiveness

Zhen Jane Wang, MD, and Renuka Sriram, PhD
Concept Award and Visionary Postdoctoral Fellowship Award

Drs. Zhen Jane Wang and Renuka Sriram from the University of California, San Francisco, epitomize the PRCRP's goal of growing and developing the future of research by supporting early career researchers. They are conducting impactful research by developing new tools to noninvasively inform on renal tumor aggressiveness.
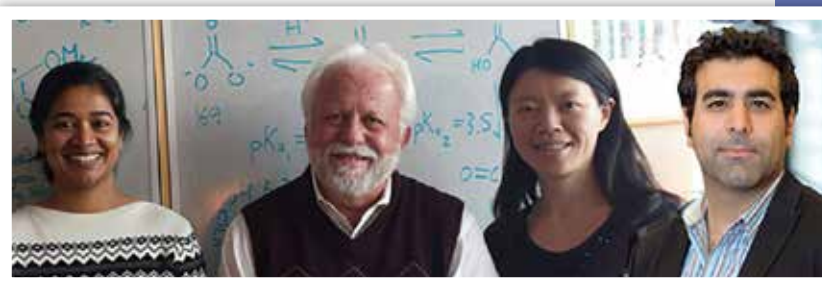
From left to right: Dr. Renuka Sriram, Dr. John Kurhanewicz, Dr. Zhen Jane Wang, and Dr. Kayvan Keshari

In FY10, Dr. Wang received a one-year Concept Award to investigate whether a newly developed imaging technique, hyperpolarized 13C magnetic resonance, could differentiate indolent renal cell carcinomas from potentially aggressive ones based on tumor glycolysis ("Warburg effect"). Glycolysis produces the metabolite pyruvate, which is then often converted to lactate in tumor cells to support growth and invasion. Dr. Wang applied the novel hyperpolarized 13C pyruvate magnetic resonance to differentiate renal cancer cells with low metastatic potential from those with high metastatic potential in an ex vivo system. She showed that high metastatic potential was linked to increased rate of lactate production and export, potentially making this a new class of renal cancer biomarkers. The results of these studies were published in a 2013 *Cancer Research* article, and a 2015 *NMR in Biomedicine* article.

This preliminary work paved the way for Dr. Wang's postdoctoral fellow, Dr. Sriram, to continue these studies after Dr. Sriram obtained an FY11 PRCRP Visionary Postdoctoral Fellowship Award. During the three years of this award, Dr. Sriram investigated renal tumor metabolism using a novel combination of patient-derived tumor samples maintained in an ex vivo three-dimensional culture system and hyperpolarized 13C MR techniques. Dr. Sriram observed that renal cancer tissues exhibited higher production and export of lactate than benign renal tumors. These studies suggest that such metabolic phenotypes can be exploited to differentiate benign renal tumors from renal cancers, and indolent from aggressive renal cancers.

To allow translation of the biomarkers from ex vivo studies to the clinic, Drs. Sriram and Wang also began collaboration with their colleagues to apply a novel diffusion-weighted hyperpolarized 13C MR technique to determine tumor lactate compartmentalization in vivo. These studies will provide the necessary foundation to move this emerging technology, hyperpolarized 13C MR, closer to clinical evaluation of renal tumors, a disease of increasing frequency.

Keshari KR, Sriram R, Koelsch BL, et al. 2013. Hyperpolarized 13C-pyruvate magnetic resonance reveals rapid lactate export in metastatic renal cell carcinomas. *Cancer Res* 72:529-538.

Sriram R, Van Criekinge M, Hansen A, et al. 2015. Real-time measurement of hyperpolarized lactate production and efflux as a biomarker of tumor aggressiveness in an MR compatible 3D cell culture bioreactor. *NMR Biomed* 28:1141-1149.

The XactMouse: A Xenochimaeric Mouse with Tumor and Hematopoietic System Obtained from the Same Patient

Antonio Jimeno, MD, PhD, Professor of Medicine/Oncology and Otolaryngology, University of Colorado, Denver

The development of relevant and effective therapeutic options for the treatment of cancer requires research platforms that accurately model the human tumor microenvironment. This need has led to the use of patient-derived xenograft models. Xenograft models use cancer cells isolated from patients that are implanted into mice for the most accurate recapitulation of a specific patient's disease. However, the interplay between the implanted human cancer cells and recruited mouse stroma and mouse immune cells can alter the tumor microenvironment. This infiltration of mouse cells can result in genetic drift toward mouse-specific gene expression within the tumor microenvironment and reduce the validity of results.

Using an FY09 PRCRP New Investigator Award, Dr. Antonio Jimeno sought to develop the XactMouse, a more relevant xenograft mouse model that could better mimic the human tumor microenvironment. Dr. Jimeno hypothesized he could create a mouse model with both tumor and hematopoietic stem cells (HSC) from the same patient. This fully humanized and homologous cancer model would faithfully replicate the patient's tumor. Specifically, Dr. Yosef Refaeli (a co-investigator in this application), also at the University of Colorado in Denver, developed an ex vivo technique for the expansion of patient-derived human HSC and progenitor cells, which are precursor cells for the human immune system. The researchers used these cells to reconstitute radiation-depleted bone marrow of immune-compromised mice for production of patient-specific immune cells within the mouse. This recapitulation of the human immune system within the XactMouse was stable over 6 months as demonstrated by consistent expression of human blood marker levels as measured by flow cytometry. Bone marrow-derived human immune cells homed into the growing tumor and interacted with human cancer cells as they would in a patient (see figure). Using RNA transcriptome analysis, Dr. Jimeno demonstrated that the gene transcription within the implanted tumor of the XactMouse containing both patient-specific immune and cancer cells reversed the initial genetic drift observed after tumor implantation in immune-compromised hosts. These data indicate the XactMouse allows for more human-specific in vivo tumor modeling than the current standard xenograft models. Further, these studies demonstrated the patient-specific engrafted immune system was capable of interacting with the corresponding patient-specific grafted tumor with observed
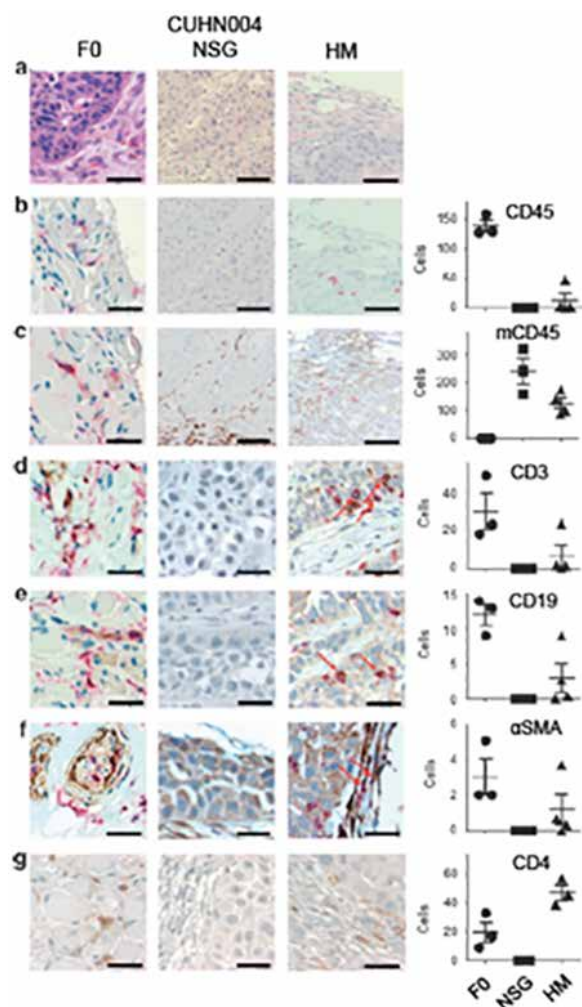
Stroma and immune marker staining in F0 (patient), NSG and aHM tumors, with CD45 (red) as co-marker. IHC markers are: a) H/E; b) human cells (CD45); c) mouse cells (mCD45); d) T cells (CD3); e) B cells (CD19); f) fibroblasts (aSMA); and g) T-helper and T-reg cells (CD4).

changes in immune cell infiltration, cytokine expression, and lymphangiogenesis for generation of a tumor microenvironment similar to that observed originally in the patient.

These studies have demonstrated that the XactMouse model recreates the native tumor environment that is absent in current animal models and can more accurately address studies targeting invasion and metastasis. The XactMouse model provides a novel research platform and improved tool for the development of next-generation therapeutic options to treat a variety of cancers, and it will be integral in the rapidly expanding field of immune-directed drug development.

Morton JJ, Bird G, Keysar SB, et al. 2016. XactMice: Humanizing mouse bone marrow enables microenvironment reconstitution in a patient-derived xenograft model of head and neck cancer. *Oncogene* 35(3):290-300.

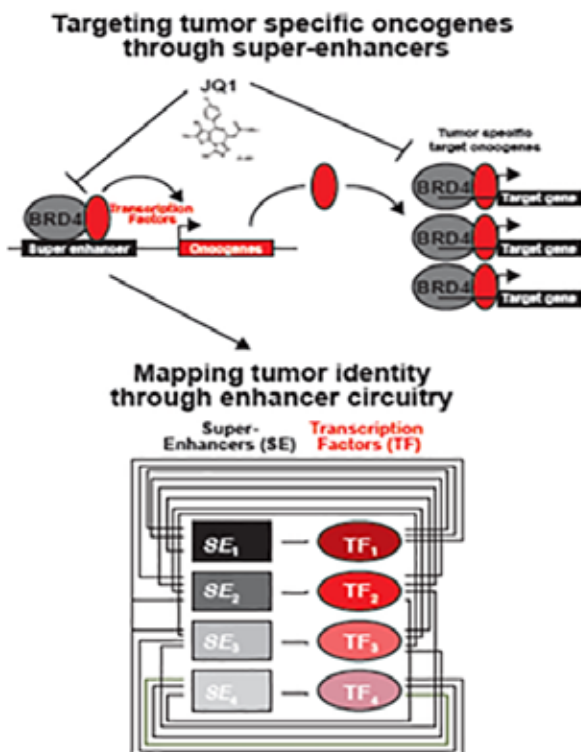## Discovery of "Super-Enhancers" Reveals New Avenue for Pursuing Therapeutic Strategies to Treat Cancer

Charles Lin, PhD, Dana-Farber Cancer Institute
Visionary Postdoctoral Fellowship Award

Multiple myeloma (MM) is an aggressive and incurable cancer that affects plasma cells, a type of white blood cell. In a subset of MM, mutations that alter how cells package and read the genome can result in the overexpression of oncogenes and their resulting oncoproteins. Recent studies demonstrate that inhibitors of a general class of proteins that help organize the genome, called BET bromodomains, can selectively turn off the overexpression of oncoproteins. This results in the death of the cancerous cells, while leaving the normal cells intact. The mechanisms that explain how inhibition of a class of proteins with broad functionality, rather than targeting a particular pathway, can appear to specifically target cancer cells was not understood. In FY12, Dr. Charles Lin received a three-year Visionary Postdoctoral Fellowship to investigate these mechanisms under the guidance of his mentor, James Bradner, MD, at the Dana-Farber Cancer Institute.

Previously, Dr. Bradner's laboratory developed a new small molecule, JQ1, that binds to the BET bromodomain protein, BRD4, and inhibits BRD4's role in regulating protein production. As part of a multidisciplinary team, Dr. Lin began his award by identifying all of the locations where BRD4 can localize to regions of both the MM genome and normal blood cell genome. The results of these studies revealed the existence of larger than normal regulatory regions in the MM genome called super-enhancers, which drive the production of key oncoproteins. Furthermore, the studies showed that BRD4 localized to super-enhancers at a much higher frequency than

Targeting tumor specific oncogenes through super-enhancers

Mapping tumor identity through enhancer circuitry

normal enhancers that control normal cellular functions. Thus, when cells were treated with JQ1, the abnormally high level of oncoprotein production driven by super-enhancers was most affected by the inhibitor, while the protein production driven by normal enhancers was relatively unaffected. These results provided key insight into how JQ1, and similar therapeutics, are able to selectively target cancer cells.

Dr. Lin and his colleagues expanded on this work and characterized super-enhancers to provide information on tumor classification in another type of tumor—MB tumors. Clinically, MB is classified in four subgroups, where the subgroup classifications are based on the types of proteins that are predominantly expressed in a particular tumor, with Group 4 MB being the least understood. Dr. Lin helped to develop experimental and computational techniques to map what types of proteins were bound in MB super-enhancer regions, thus predicting the genes that were being regulated. Analysis of this data identified "signatures" for each MB subgroup, including Group 4. With further development, Dr. Lin hopes that these signatures may be used as biomarkers to aid in discovering new therapeutic targets and informing clinicians on treatment strategies.

In summary, within two years of working as part of a multidisciplinary team, Dr. Lin was the first to identify and describe super-enhancer regions in a cancer genome. This information was then used to develop methods to better characterize MB tumors. These exciting achievements have been published in six scientific articles. Additionally, Dr. Lin has helped to develop publicly available analysis software and databases, and his research has guided pharmaceutical companies' preclinical research on therapeutic strategies for MM and other cancers. Now, Dr. Lin is preparing to start up his own laboratory as an Assistant Professor at the Baylor College of Medicine.

Anand P, Brown JD, Lin CY, et al. 2013. BET bromodomains mediate transcriptional pause release in heart failure. *Cell* 154: 569-582.

Development of Novel Cancer Therapies that Target Polyamine Metabolism

Otto Phanstiel, PhD, University of Central Florida

Current therapies for cancer are frequently toxic to non-cancerous cells, and often have limited efficacy due to tumor cells rapidly building resistance and evading treatment. To sustain an increased growth rate, cancer cells rely on the native polyamines (putrescine, spermidine, and spermine), which are essential growth factors either synthesized by the cell and/or actively transported from the environment. These low molecular weight aliphatic amines play critical roles in chromatin remodeling, translation, and transcription. Ornithine decarboxylase (ODC) is a key enzyme involved in the first step of polyamine biosynthesis, and polyamine production is inhibited by difluoromethylornithine (DFMO). However, the success of DFMO in the clinic has been limited because cells compensate for reduced
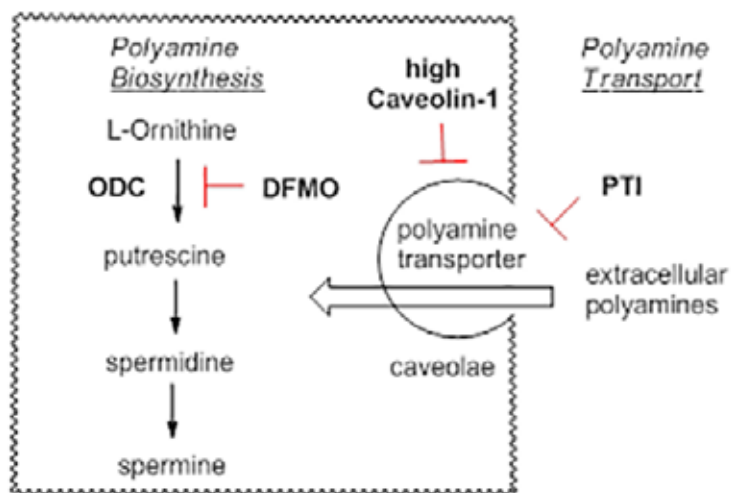

Targeting Polyamine metabolism. DFMO is an inhibitor of ODC, (biosynthesis) and Cav-1 is an inhibitor of polyamine import via caveolae. The small molecule PTI blocks import of the native polyamines from exogenous sources to effectively deplete DFMO-treated cancer cells of the polyamine metabolites needed for growth.

cellular polyamine production by increasing transport of exogenous polyamines. Therefore, polyamine transport inhibitors (PTIs) have become a target of research for the development of novel cancer therapeutics (see figure).

Using an FY11 PRCRP Discovery Award, Dr. Otto Phanstiel and collaborator Dr. Deborah Altomare tested novel polyamine derived PTIs alone and in combination with DFMO to target pancreatic cancer cells in vitro and in vivo. Dr. Phansteil demonstrated that these PTIs can inhibit the uptake of spermidine, an exogenous polyamine source for cell growth. This combination therapy showed efficacy in two mouse models of pancreatic cancer. Dr. Phanstiel's team also investigated potential biomarkers for the development of resistance to DFMO-induced inhibition of the polyamine biosynthesis pathway. These studies initially focused on caveolin-1 (Cav-1), a scaffolding protein found in the plasma membrane of most cell types. Cav-1 is a tumor suppressor gene and has been shown to impair polyamine transport activity and reduce proliferation in cancer cell lines. However, there are conflicting reports in the literature regarding Cav-1 expression in pancreatic cancers. The research team demonstrated that high levels of Cav-1 resulted in reduced polyamine uptake in the pancreatic cancer cell line BxPC-3, even in the presence of DFMO. In addition, the investigators studied the transcription factor c-Myc, which upregulates ODC transcription and affects polyamine biosynthesis. Cells that overexpress c-Myc have elevated levels of polyamines sufficient for tumor promotion. Dr. Phanstiel demonstrated a positive correlation between the expression of the transcription factor c-Myc and uptake of polyamines in untreated pancreatic cancer cells. Additionally, the protein ATP13A3 was targeted as a potential biomarker for DFMO-induced resistance due to its analogue in the *C. elegans* model, CATP-5, having a known role in polyamine transport. Similarly, high basal expression of the novel P5 type ATPase (ATP13A3) was observed in untreated pancreatic cancer cells that were resistant to DFMO therapy. The team demonstrated that pancreatic cancer cell lines with high ATP13A3 expression could be readily rescued from DFMO with exogenous polyamine exposure, whereas cell lines with very low expression of ATP13A3 could not. Specific knockdown of ATP13A3 expression using siRNA methods showed a significant reduction in the ability of DFMO-treated pancreatic cancer cells to be rescued by exogenous polyamines. Collectively, these data suggest that the protein ATP13A3 is involved in polyamine transport.

Taken together, these studies demonstrated that PTIs in combination with DFMO can inhibit the uptake of polyamines for reduced pancreatic cancer cell growth, and that Cav-1, c-Myc, and ATP13A3 are potential biomarkers for the development of DFMO-induced resistance. Dr. Phanstiel's team demonstrated that Cav-1, c-Myc, and ATP13A3 proteins play a role in pancreatic cancer cells' ability to escape DFMO therapy through enhanced import of exogenous polyamines. These studies represent promising movements toward developing effective treatments for pancreatic cancer.
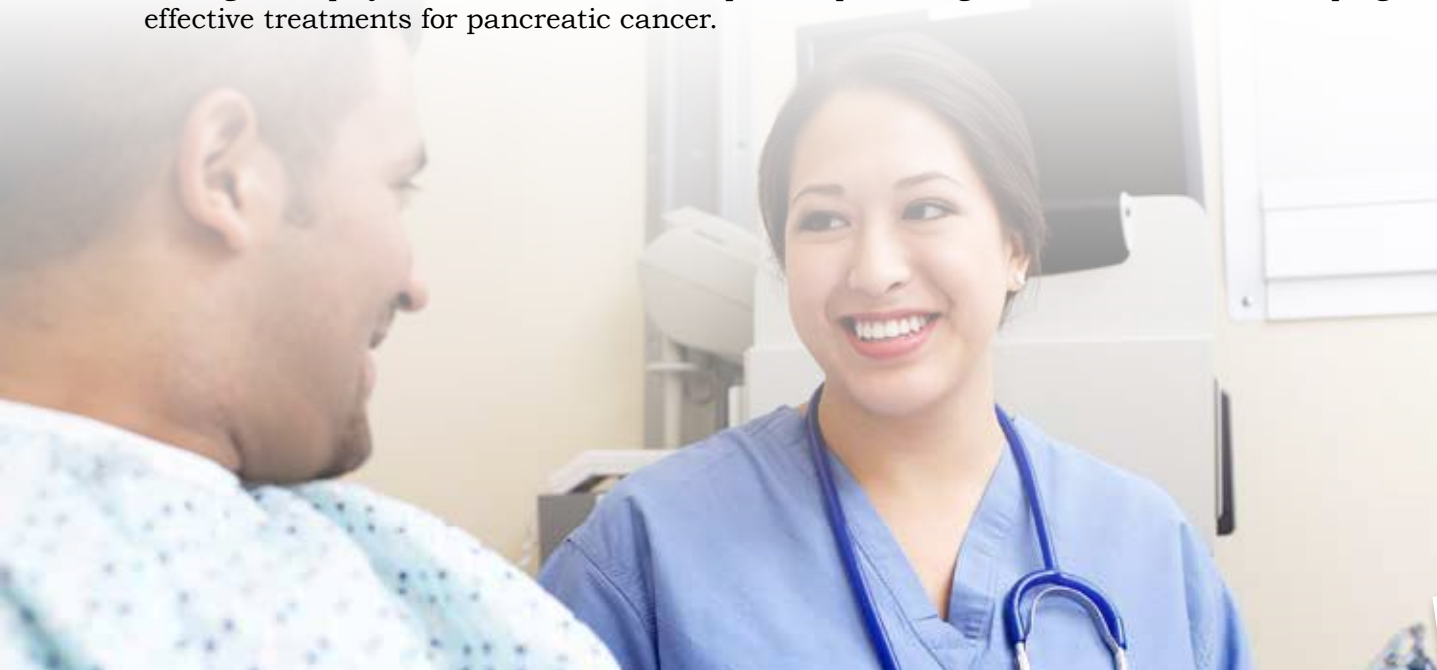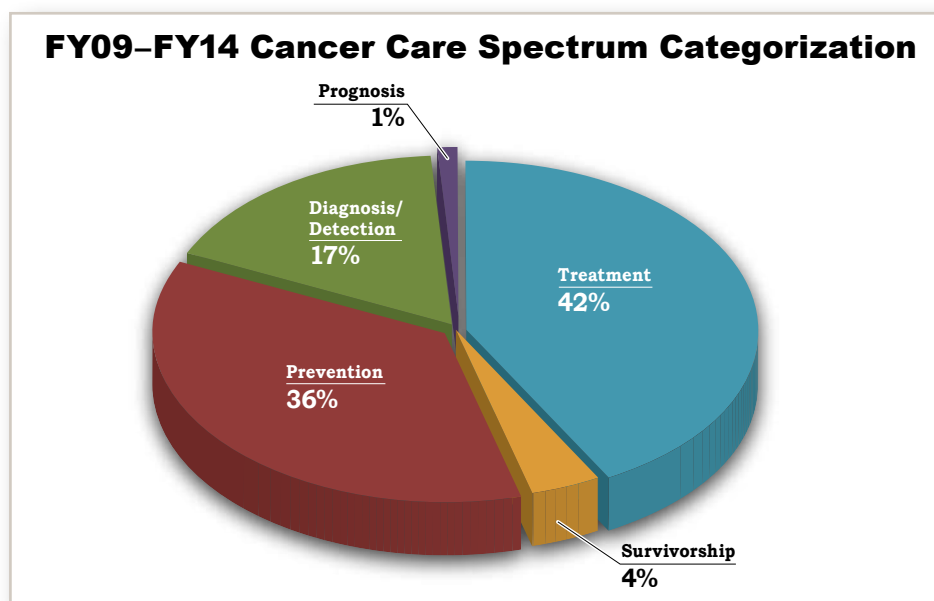
Investing in the Cancer Care Spectrum

The ultimate goal of medical research is to impact the clinic in ways that provide patients and their families with advancements in clinical care including prevention, diagnosis/detection, prognosis, treatment, and survivability of cancer. PRCRP awards have aimed to impact the clinic in various ways. Categorization of the PRCRP awards based on a cancer care spectrum (as shown below) identifies the targeted areas of impact on outcomes for patients and military families affected by cancer.

	Prevention	Diagnosis/Detection	Prognosis	Treatment	Survivorship
Definition*	Risk and prevention factors identification, implementation	Identifying a cancer by signs and symptoms	Likely outcomes of a cancer diagnosis	Therapeutics and surgical interventions	Impact of treatments, short- and long-term quality of life
Research Spectrum	• Identification of risk factors • Environmental exposures including biological, chemical, ionizing radiation • Genetic risk factors	• Imaging techniques • Early detection methodologies • Biomarker repositories	• Personalized medicine • Tailored treatments for long-term outcomes	• Chemotherapy • Immunotherapy • Radiation therapy • Personalized medicines • Drug-delivery systems	• Quality-of-life studies • Behavioral factors • Compliance factors • Relapse • Remission

*Definitions from the NCI Dictionary of Cancer Terms

PRCRP awards focus on both basic and translational aspects of cancer research that aim for long-term development of highly impactful clinical outcomes.

FY09–FY14 Cancer Care Spectrum Categorization

Prognosis 1%

Diagnosis/Detection 17%

Treatment 42%

Prevention 36%

Survivorship 4%

Military Relevance and Cancer Research

Our military Service members are often physically challenged in ways that increase their risk for the development of a variety of cancers as described on page 3.

The PRCRP seeks to support cancer research that addresses the cancer health needs of military personnel, their dependents, retirees, and Veterans by soliciting applications responsive to military-relevant focus areas. The military-relevant focus areas from FY16 are listed below:

- Military-relevant risk factors associated with cancer (e.g., ionizing radiation, chemicals, infectious agents, and environmental carcinogens).

- Gaps in cancer prevention, diagnosis, early detection, diagnosis, treatment and/or survivorship that may affect the general population but have a particularly profound impact on the health and well-being of military members, Veterans, and their beneficiaries.

There are over 300,000 military beneficiaries with a cancer diagnosis, for a prevalence of 4.1%, comprised of over 60 different cancer types. The cost of cancer care within the Military Health System is over $1B. Funding studies on the detection, diagnosis, treatment, and prevention of these diseases benefits both the Service member and the American public, ultimately leading to increased survival rates and decreased costs of medical care.

In summary, the PRCRP directly impacts military welfare by providing research into cancers that may develop due to exposure in various environments that are unique to the military, and answering unmet knowledge gaps in the cancer care spectrum that may profoundly affect Service members, their families, Veterans, and the American Public.

Crawford RS, Wu J, Park D, and Barbour GL. 2007. A study of cancer in the military beneficiary population. *Military Medicine* 172:1084-1088.

> "It gives me great pride to be an active duty military member on the PRCRP panel. This opportunity allows me to bring a voice, as a clinician and researcher, to the panel from someone who is actively treating our military beneficiaries. My goal is to ensure only the best research is being developed for the people who have served our country."

LCDR Corey Carter, MD
Walter Reed National Military Medical Center
Three years of service on the PRCRP Programmatic Panel (FY13-FY15)

For more information, visit
http://cdmrp.army.mil
or contact us at:

usarmy.detrick.medcom-cdmrp.mbx.cdmrp-public-affairs@mail.mil
(301) 619-7071

05-2016