

**NCI Extramural Kidney Cancer Research Grants FY 2004  
Grant Extracts for > 50% Kidney Cancer Relevance**

The following abstracts are for NCI-awarded grants for which 50% or greater share is for kidney cancer research. These grants comprise \$15 million of the \$22 million awarded in FY 2004 (September 1, 2004 – August 31, 2005) by the National Cancer Institute for kidney cancer related research. For a listing of all the grants refer to “NCI Kidney Cancer Budget” elsewhere on this website.

Project Number	Total Project Funding	Percent Relevant to RCC	Kidney Cancer Funding	Institution Being Funded
5P50CA101942-2	\$2,447,662	100.00	\$2,447,662	BETH ISRAEL DEACONESS MEDICAL CENTER
5R01CA79830-5	322,000	100.00	322,000	BOSTON MEDICAL CENTER
5R21CA103486-2	274,259	100.00	274,259	CHILDREN'S HOSPITAL MED CTR -CINCINNATI
5R01CA79495-4	267,750	100.00	267,750	CHILDREN'S HOSPITAL OF PHILADELPHIA
5R01CA56937-12	323,595	100.00	323,595	CLEVELAND CLINIC LERNER COL/MED-CWRU
7R21CA100078-2	137,700	100.00	137,700	CLEVELAND CLINIC LERNER COL/MED-CWRU
5R01CA102270-2	380,200	100.00	380,200	COLUMBIA UNIVERSITY HEALTH SCIENCES
1R21CA104940-1	153,900	100.00	153,900	DANA-FARBER CANCER INSTITUTE
5R01CA68490-9	283,286	100.00	283,286	DANA-FARBER CANCER INSTITUTE
5R01CA95539-3	281,240	100.00	281,240	DARTMOUTH COLLEGE
5R01CA95648-2	351,550	100.00	351,550	DARTMOUTH COLLEGE
5R01CA89102-4	237,174	100.00	237,174	DUKE UNIVERSITY
5R21CA98446-2	279,200	100.00	279,200	DUKE UNIVERSITY
1U01CA111242-1	304,448	100.00	304,448	FOX CHASE CANCER CENTER
1R01CA106512-1	313,199	100.00	313,199	FRED HUTCHINSON CANCER RESEARCH CTR
1R21CA101224-1	225,500	100.00	225,500	GREENVILLE GENERAL HOSPITAL
5R01CA79685-5	288,736	100.00	288,736	JOHNS HOPKINS UNIVERSITY
1R03CA110708-1	71,500	100.00	71,500	LOUISIANA STATE UNIV HSC NEW ORLEANS
5R37CA58596-12	432,500	100.00	432,500	MASSACHUSETTS GENERAL HOSPITAL
1R01CA104505-1	306,750	100.00	306,750	MAYO CLINIC COLL OF MED, JACKSONVILLE
5R03CA103494-3	75,000	100.00	75,000	MAYO CLINIC COLL OF MED, JACKSONVILLE
2R01CA78383-6	292,638	100.00	292,638	MAYO CLINIC COLL OF MED, ROCHESTER
5R01CA24652-24	317,250	100.00	317,250	MEDICAL COLLEGE OF WISCONSIN
5R01CA93871-3	282,108	100.00	282,108	OHIO STATE UNIVERSITY
5R21CA98842-2	189,245	100.00	189,245	ROSWELL PARK CANCER INSTITUTE CORP
5R21CA98969-2	156,741	100.00	156,741	ROSWELL PARK CANCER INSTITUTE CORP
5R37CA53370-14	408,563	100.00	408,563	RUTGERS - NEW BRUNSWICK, NJ
1R43CA106685-1	79,501	100.00	79,501	TWENTY-TWENTY GENE SYSTEMS
5R01CA102600-2	296,086	100.00	296,086	UNIVERSITY OF CALIFORNIA SAN FRANCISCO
1R01CA109446-1	272,138	100.00	272,138	UNIVERSITY OF IOWA
5R21CA97421-2	148,500	100.00	148,500	UNIVERSITY OF MARYLAND BALT PROF SCHL
5R01CA95572-3	290,363	100.00	290,363	UNIVERSITY OF NEVADA RENO
1R01CA100787-1	259,940	100.00	259,940	UNIVERSITY OF PENNSYLVANIA
5R01CA57840-10	265,245	100.00	265,245	UNIVERSITY OF PITTSBURGH
5R01CA63613-10	319,365	100.00	319,365	UNIV OF TEXAS MD ANDERSON CAN CTR
5R01CA90966-3	471,684	100.00	471,684	UNIV OF TEXAS MD ANDERSON CAN CTR
5R01CA98897-2	511,396	100.00	511,396	UNIV OF TEXAS MD ANDERSON CAN CTR
5R01CA61889-11	346,431	100.00	346,431	UNIVERSITY OF WASHINGTON

<b>Project Number</b>	<b>Total Project Funding</b>	<b>Percent Relevant to RCC</b>	<b>Kidney Cancer Funding</b>	<b>Institution Being Funded</b>
5R01CA92542-4	351,713	100.00	351,713	WEILL MEDICAL COLLEGE OF CORNELL UNIV
5R01CA85412-3	296,131	100.00	296,131	YESHIVA UNIVERSITY
1R21CA99069-1	186,788	75.00	140,091	DANA-FARBER CANCER INSTITUTE
1R01CA112533-1	410,762	50.00	205,381	BETH ISRAEL DEACONESS MEDICAL CENTER
5R21CA101629-2	175,000	50.00	87,500	CITY OF HOPE/BECKMAN RESEARCH INST
7R01CA89344-4	317,475	50.00	158,738	CLEVELAND CLINIC LERNER COL/MED-CWRU
1R01CA102309-1	315,495	50.00	157,748	DANA-FARBER CANCER INSTITUTE
5R37CA54358-14	405,419	50.00	202,710	JOHNS HOPKINS UNIVERSITY
5R01CA59998-11	322,262	50.00	161,131	MOUNT SINAI SCHOOL OF MEDICINE OF NYU
5R03CA106006-2	78,625	50.00	39,313	NORTHEASTERN UNIVERSITY
5R01CA95785-3	282,130	50.00	141,065	SLOAN-KETTERING INST FOR CANCER RES
5R01CA76035-6	325,358	50.00	162,679	UNIV OF COLORADO HLTH SCIENCES CTR
5R01CA89655-4	237,793	50.00	118,897	UNIVERSITY OF FLORIDA
1R21CA105293-1	128,329	50.00	64,165	WASHINGTON STATE UNIVERSITY
5R01CA91048-3	256,674	50.00	128,337	UNIV OF NORTH CAROLINA CHAPEL HILL
	<b>Total Relevant Funding</b>	<b>(for these projects)</b>	<b>\$15,079,942</b>	

<b>Project Number</b> 5P50CA101942-2	<b>Title</b> DF/HCC Renal Cancer SPORE	<b>City / State</b> BOSTON, MA		
<b>Institution</b> BETH ISRAEL DEACONESS MEDICAL CENTER		<b>Program Director</b> Andrew Hruszkewycz		
<b>Principal Investigator</b> ATKINS, MICHAEL B		<b>Cancer Activity</b> Organ Systems	<b>Total Project \$</b> DDES	\$2,447,662
<b>Start Date</b> 8/20/2004	<b>End Date</b> 05/31/2005			
<b>Science Area</b> KIDNEY		<b>Percent</b> 100.00	<b>Percent \$</b>	\$2,447,662

**Abstract**

DESCRIPTION (provided by applicant): This application represents the creation of a Specialized Program of Research Excellence in Renal Cancer originating from the Renal Cancer Program-in-Development of the newly configured Dana-Farber Harvard Cancer Center (DF/HCC) and the Beth Israel Deaconess Medical Center. The DF/HCC Renal Cancer SPORE includes investigators from all the Harvard affiliated hospitals - the Beth Israel Deaconess Medical Center, the Dana-Farber Cancer Institute, the Massachusetts General Hospital, the Brigham and Women's Hospital, and the Children's Hospital Medical Center, as well as the Harvard School of Public Health. Five major projects are supported through this application including: 1) Identification of markers for early detection and monitoring of high-risk RCC populations; 2) Inhibition of VHL-regulated growth factor pathways for treatment of RCC; 3) Combination radiofrequency ablation and antivascular/antiangiogenesis therapy for RCC; 4) Identification of molecular and immunologic correlates of RCC prognosis and responsiveness to therapy, and 5) Dendritic cell/tumor fusions in conjunction with IL-12 as novel immunotherapy for RCC. These projects are integrated by four cores. These are: 1) Administration, Evaluation and Planning; 2) Biostatistics; 3) Tissue Acquisition, Pathology and Clinical Data, and 4) Monitoring. This SPORE application outlines a Developmental Projects Program which includes a plan for selection and review of projects, and eight sample projects that could be considered for support. We also include a Career Development Award Program which outlines a mechanism for the identification, support, and review of talented young investigators in renal cancer. The overall goal of the DF/HCC Renal Cancer SPORE is the translation of biological and technological advances into clinically meaningful advances for patients with renal cancer.

<b>Project Number</b>	<b>Title</b>			
5R01CA79830-5	VHL, Jade-1 and protein stability in renal cancer			
<b>Institution</b>		<b>City / State</b>		
BOSTON MEDICAL CENTER		BOSTON, MA		
<b>Principal Investigator</b>		<b>Program Director</b>		
COHEN, HERBERT TOD		Judy Mietz		
<b>Start Date</b>	<b>End Date</b>	<b>Cancer Activity</b>		<b>Total Project \$</b>
7/2/2004	06/30/2005	Cancer Cell Biology	DCB	\$322,000
<b>Science Area</b>		<b>Percent</b>		<b>Percent \$</b>
KIDNEY		100.00		\$322,000

### Abstract

Revised Abstract: Renal cancer is a devastating malignancy that is highly resistant to medical therapy. The VHL tumor suppressor is mutated in most adult renal cancers. Yet, the mechanism of VHL tumor suppression is not known. Using a directed approach to find VHL-interacting proteins important in renal cancer, our laboratory has identified Jade-1 (gene for Apoptosis and Differentiation in Epithelia) as a particularly strong VHL interactor. Jade-1, a novel, kidney-enriched, PEST and plant homeodomain protein, is a member of a new protein family. VHL stabilizes Jade-1 protein, which is a new VHL function. Moreover, we have now shown that Jade-1 stabilization is VHL mutation-dependent, with non-renal cancer-causing VHL missense mutations able to stabilize Jade-1 like wild-type VHL. Jade-1 stabilization is therefore the first VHL activity to show substantial correlation with renal cancer risk, suggesting it has a disease relationship. Moreover, stabilization of Jade-1 by VHL is highly specific, as it has not been observed with known or potential VHL partners. We have established convincing evidence of the VHL-Jade-1 relationship's authenticity as well as Jade-1's compelling biological significance. We propose the following Aims: 1. The VHL-Jade-1 protein-protein interaction, and Jade-1 expression in renal cancer tissue 2. VHL-dependent Jade-1 stabilization and modification 3. Examination of the role of Jade-1 in apoptosis and modulation by VHL

<b>Project Number</b>	<b>Title</b>			
5R21CA103486-2	Utility of Rapamycin for the Treatment of Renal Angiomy*			
<b>Institution</b>	<b>City / State</b>			
CHILDREN'S HOSPITAL MED CTR (CINCINNATI)	CINCINNATI, OH			
<b>Principal Investigator</b>	<b>Program Director</b>			
BISSLER, JOHN J	Heng Xie			
<b>Start Date</b>	<b>End Date</b>	<b>Cancer Activity</b>	<b>Total Project \$</b>	
9/1/2004	08/31/2005	Clinical Oncology	DCTD	\$274,259
<b>Science Area</b>		<b>Percent</b>	<b>Percent \$</b>	
KIDNEY		100.00		\$274,259

### Abstract

DESCRIPTION (provided by applicant): Targeted molecular therapy is the ultimate objective for the management of neoplasia, but only a few examples exist in practice, due in large part to the complexity of genetic events that result in unregulated cell growth. Tuberous sclerosis is an inherited cancer syndrome associated with the formation of hamartomas in multiple organs, including angiomyolipomas in the kidney, caused by well-characterized inactivating mutations at genetic loci that encode the interacting proteins, tuberin or hamartin. Elegant studies have recently elucidated the pivotal role of the tuberin/hamartin complex in the checkpoint control of the Akt signaling pathway that regulates cell growth and division. Rapamycin, an FDA immunosuppressive approved drug used to prevent renal transplant rejection, mimics the function of the tuberin/hamartin complex by binding to a protein downstream of Akt called mammalian target of rapamycin (mTOR) and inhibiting the phosphorylation of more distal elements that control cell cycle and protein translation. Rapamycin has been shown to specifically inhibit the growth of tuberin and hamartin deficient cells from humans, rodents and flies, and to produce tumor regression in rats and mice. The consensus opinion of the recent Tuberous Sclerosis Complex Research conference in Chantilly, Virginia was that the preclinical evidence for the use of rapamycin in TSC was sufficiently compelling to warrant a human trial. The objective of the current study is to determine if rapamycin reduces the volume of angiomyolipomas. This goal will be accomplished by treatment of thirty patients with angiomyolipomas, either in the setting of tuberous sclerosis, or a related disease associated with mutations in tuberous sclerosis genes called sporadic lymphangiomyomatosis, with dose-adjusted rapamycin for a period of one year. The size, number, volume and tissue composition of renal angiomyolipomas will be monitored by MRI scans of the kidney, performed prior to treatment, at two months, four months, and every six months. Other manifestations of TSC, including brain, skin and lung lesions, will also be monitored with appropriate clinical, functional and imaging techniques. The minimal rapamycin dose that produces an effect, defined as a greater than 10% decrease in angiomyolipoma volume, will be titrated beginning with doses that result in subimmunosuppressive serum levels to those that produce levels in the low to modestly immunosuppressive range. Toxicities, as defined by the NCI common toxicities criteria, will be carefully monitored, reported, and expeditiously addressed. Successful completion of the aim of this study will help to establish tuberous sclerosis as a valuable model for targeted molecular therapy for neoplasia.

<b>Project Number</b> 5R01CA79495-4	<b>Title</b> PREVENTION OF IFOSFAMIDE INDUCED NEPHROTOXICITY	<b>City / State</b> PHILADELPHIA, PA	
<b>Institution</b> CHILDREN'S HOSPITAL OF PHILADELPHIA		<b>Program Director</b> Mary Wolpert	
<b>Principal Investigator</b> NISSIM, ITZHAK		<b>Cancer Activity</b> Biochemistry and Pharmacology	
<b>Start Date</b> 8/1/2004	<b>End Date</b> 07/31/2005		<b>Total Project \$</b> DCTD \$267,750
<b>Science Area</b> KIDNEY		<b>Percent</b> 100.00	<b>Percent \$</b> \$267,750

### Abstract

Ifosfamide (IFO), an alkylating oxazaphosphorine, has been found to be very effective for the treatment of relapsed solid tumors and in patients who respond poorly following treatment with other chemotherapeutic agents. However, the efficacy of IFO is severely limited by a high incidence of nephrotoxicity. This proposal entails a comprehensive investigation of the as yet unknown mechanism(s) involved in IFO- induced renal injury and prevention of such injury by administration of glycine (Gly), which we found to be an effective cytoprotective agent both in vitro and in vivo. Our ultimate goal is to develop a clinically applicable protocol involving administration of glycine with IFO to prevent nephrotoxicity in cancer patients treated with this antineoplastic drug. The main hypothesis to be explored is that the induction of renal injury during IFO treatment is mediated via accumulation in the kidney cortex of one or more of the active metabolites of IFO, i.e., 4-hydroxy-IFO (4- OH-IFO) and/or isophosphoramidate mustard (IPM), secondary to depletion of [GSH] by chloroacetaldehyde (CAA) and/or acrolein (ACR). These metabolites may react with SH-groups of the plasma membrane or mitochondrial membrane proteins, thereby damaging cellular integrity. An alternative, but not mutually exclusive hypothesis is that the primary mechanism in evoking renal injury during IFO treatment is mediated via inhibition of mitochondrial oxidative metabolism by CAA and/or ACR, resulting in defective energy production, multiple metabolic abnormalities, and thereby, cellular damage. However, concomitant oral supplementation of Gly with IFO will attenuate IFO-induced nephrotoxicity by maintaining the renal proximal tubule integrity without diminishing the antitumor action of IFO. Unique features of the current proposal are: (a) the successful development of a rat model system for investigation of IFO-induced renal toxicity; (b) the use of Nuclear Magnetic Resonance (NMR), Gas Chromatography-Mass Spectrometry (GC-MS), LC-MS-MS, Laser- Scanning Confocal Microscopy and techniques of molecular biology to explore the biochemical/molecular lesions responsible for IFO-induced renal injury; and (c) a prevention of such injury by oral supplementation of Gly. The proposed studies are of clinical as well as scientific significance. The data to be generated will potentially have considerable importance for prevention of renal dysfunction associated with cancer chemotherapy, and thus allow for a greater therapeutic efficacy and enhanced survival of cancer patients.

<b>Project Number</b>	<b>Title</b>			
5R01CA56937-12	T Cell Unresponsiveness in Human Tumors			
<b>Institution</b>		<b>City / State</b>		
CLEVELAND CLINIC LERNER COL/MED-CWRU		CLEVELAND, OH		
<b>Principal Investigator</b>		<b>Program Director</b>		
FINKE, JAMES H		Susan McCarthy		
<b>Start Date</b>	<b>End Date</b>	<b>Cancer Activity</b>	<b>Total Project \$</b>	
9/1/2004	08/31/2005	Immunology	DCB	\$323,595
<b>Science Area</b>		<b>Percent</b>	<b>Percent \$</b>	
KIDNEY		100.00		\$323,595

**Abstract**

DESCRIPTION (provided by applicant): There is growing evidence to suggest that the immune dysfunction noted in T cells from cancer patients may be linked to the ability of tumors to alter the sensitivity of T cells to apoptosis. This idea is supported by our observation that a subset of tumor infiltrating T cells (TIL) is apoptotic, and that an even higher percentage of the viable TIL demonstrates a heightened sensitivity to activation induced cell death (AICD). Similar findings have been observed by others in different tumor types, and can also be seen in circulating T lymphocytes. We have also found that supernatants derived from renal cell carcinoma (RCC) explants can either directly induce apoptosis of peripheral blood T cells from normal individuals or sensitize them to AICD. Our recent findings implicate gangliosides within the tumor supernatants as one class of products that can induce both phenotypes in T cells. Gangliosides isolated from some RCC supernatants induce apoptosis, while gangliosides isolated from other RCC supernatants only sensitize T cells to AICD. Additional evidence suggests that gangliosides expressed on RCC lines induce T cell apoptosis. We have also found that aldehyde products of fatty acid oxidation, likely resulting from oxidative stress within the tumor microenvironment, represent another class of products within RCC supernatants that can induce apoptosis of T cells. Among the most prominent aldehyde products is 4-hydroxynonenal (HNE), which is present in our tumor supernatants. Moreover, commercial HNE can induce apoptosis of T cells. We propose that the heterogeneous capacity of RCC tumors to directly induce apoptosis, or sensitize T cells to AICD, is related to the variations in the specific products expressed by individual tumors. We further hypothesize that specific structural features of these molecules dictate whether individual products induce apoptosis or only sensitize. Thus, in Aim 1, we will determine the relationship between the structural characteristics and functional activities of these molecules. Other studies will determine the impact that blocking the formation of oxidative products or ganglioside synthesis in the RCC explants has on structure and function of apoptogenic and sensitizing products present in the tumor supernatants. Aim 2 will determine whether apoptogenic gangliosides and HNE induce cell death via similar or distinct mechanisms. This will include testing whether these products differ in the reactive oxygen species (ROS) they induce, the order of signaling events they activate, and their ability to down regulate Bcl-2. Other experiments will determine the mechanism by which gangliosides sensitize T cells AICD. This will involve defining the effect that sensitizing gangliosides have on mitochondrial stability. A better understanding of immune dysfunction may lead to ways in which to improve anti-tumor immunity.

<b>Project Number</b>	<b>Title</b>			
7R21CA100078-2	Urinary MMP activity to detect renal cell carcinoma			
<b>Institution</b>		<b>City / State</b>		
CLEVELAND CLINIC LERNER COL/MED-CWRU		CLEVELAND, OH		
<b>Principal Investigator</b>		<b>Program Director</b>		
WEIMBS, THOMAS		James Tricoli		
<b>Start Date</b>	<b>End Date</b>	<b>Cancer Activity</b>	<b>Total Project \$</b>	
6/3/2004	05/31/2005	Diagnostics Research	DCTD	\$137,700
<b>Science Area</b>		<b>Percent</b>	<b>Percent \$</b>	
KIDNEY		100.00		\$137,700

### Abstract

DESCRIPTION (provided by applicant): The survival rates for patients diagnosed with renal cell carcinoma (RCC) -the most common type of kidney cancer- are extremely low. A major reason is that early-stage RCC is usually asymptomatic leading to a high frequency of patients that present already with metastatic disease. No clinically relevant marker is available that would allow the detection of early-stage RCC. Ideally, a screening assay for RCC should be non-invasive, and should be possible to be developed into a cost-effective, high-throughput assay. An RCC-specific urinary marker may fulfill these criteria. Our preliminary results indicate that urine from RCC patients contains increased levels of matrix metalloproteinase (MMP) activity which causes the degradation of normally excreted extracellular matrix (ECM) proteins. In a preliminary analysis, the detection of the absence of urinary ECM proteins has allowed the detection of RCC with a sensitivity of 95% (21/22) and specificity of 95% (21/22). Importantly, all early-stage cases were correctly identified. The over-all goal of this project is to develop a rapid screening assay based on the detection of urinary MMPs or MMP activity, and to test its clinical usefulness for the detection of RCC. First, the excreted MMP(s) will be identified using specific antibodies or by affinitychromatography and sequencing. Second, a micro-titer plate screening assay will be developed based on the degradation of fluorogenic substrates and/or on the immunological detection of MMPs. Third, using larger RCC patient and control populations, the sensitivity and specificity of the developed screening assay(s) will be determined.

<b>Project Number</b>	<b>Title</b>			
5R01CA102270-2	WT1 and beta-catenin targets in Wilms tumor			
<b>Institution</b>		<b>City / State</b>		
COLUMBIA UNIVERSITY HEALTH SCIENCES		NEW YORK, NY		
<b>Principal Investigator</b>		<b>Program Director</b>		
TYCKO, BENJAMIN		Judy Mietz		
<b>Start Date</b>	<b>End Date</b>	<b>Cancer Activity</b>	<b>Total Project \$</b>	
7/1/2004	06/30/2005	Cancer Cell Biology	DCB	\$380,200
<b>Science Area</b>		<b>Percent</b>		<b>Percent \$</b>
KIDNEY		100.00		\$380,200

### Abstract

DESCRIPTION (provided by applicant): Major efforts have gone into identifying the targets of oncogenic transcription factors, but data from tissue culture experiments have seldom been vetted in primary cancers. Here we address this problem, using Wilms tumor as an experimental system. Wilms tumors occurring in Denys-Drash and WAGR syndromes carry inactivating mutations of the WT1 tumor suppressor gene. In contrast, WT1 mutations are rare in sporadic Wilms tumors. We have confirmed previous data indicating that mutations in beta-catenin (CTNNB1), a component of the Wnt signaling pathway that acts as an oncogene in many of the most common human malignancies, are restricted to the WT1-null tumors. We also find that these mutations are strikingly clustered at codon Ser45. By expression profiling we have identified a panel of genes that distinguish the WT1-null from WT1-positive tumors. This dataset should be a powerful tool to identify downstream targets of WT1 and beta-catenin/TCF, which we hypothesize are enriched among these differentially expressed genes. Aim 1. We will determine whether the Wnt/beta-catenin signaling axis is universally activated in the WT1- null class of Wilms tumors. Aim 2. To narrow the list of WT1 and beta-catenin target genes, we will manipulate WT1 levels, and components of the beta-catenin pathway in tissue culture, and compare the results with our "gold standard" data from the primary Wilms tumors. Aim 3. To validate the candidate beta-catenin target genes in vivo, we will express mutant beta-catenin to the developing kidney, using a gene knock-in approach. By creating an isogenic series, these mice will also allow us to assay for the relative potency of mutation at Ser45 vs. other phosphorylation sites in affecting the proliferative/oncogenic potency of beta-catenin. We will subsequently cross these mice with Wt1-mutant heterozygotes, possibly generating a mouse model for Wilms tumor.

<b>Project Number</b>	<b>Title</b>		
1R21CA104940-1	Using Synthetic Lethality to Select Cancer Drug Targets		
<b>Institution</b>		<b>City / State</b>	
DANA-FARBER CANCER INSTITUTE		BOSTON, MA	
<b>Principal Investigator</b>		<b>Program Director</b>	
KAELIN, WILLIAM G		Ronald Dubois	
<b>Start Date</b>	<b>End Date</b>	<b>Cancer Activity</b>	<b>Total Project \$</b>
3/12/2004	02/28/2005	Biochemistry and Pharmacology	DCTD \$153,900
<b>Science Area</b>		<b>Percent</b>	<b>Percent \$</b>
KIDNEY		100.00	\$153,900

**Abstract**

DESCRIPTION (provided by applicant): Loss of function tumor suppressor gene (TSG) mutations are common in cancer. Two genes are 'synthetically lethal' if inactivation of either gene alone is compatible with viability but inactivation of both genes leads to death. In theory, the product of a gene that was synthetically lethal to a mutated TSG would be an attractive drug target because its inhibition should kill TSG(-/-) cancer cells but not normal [TSG(+/+)] cells. We have chosen the VHL tumor suppressor gene, which is mutated in the majority of clear cell renal carcinomas, to explore this paradigm further. Importantly, inactivation of the VHL gene product, pVHL, does not grossly affect cell growth in vitro, which might otherwise confound our studies. Moreover, we have multiple matched (isogenic) renal carcinoma cell (RCC) lines that do or do not contain wild-type pVHL as well as VHL(+/+) and VHL (-/-) C. elegans. Cells lacking pVHL overproduce a transcription factor called HIF (hypoxia-inducible factor). Consequently, the transcriptomes of VHL (-/-) RCCs resemble that of hypoxic cells, which are common in solid tumors and known to be relatively chemoresistant. In specific aim 1 we will use chemical biological approaches to look for compounds that selectively kill VHL(-/-) RCCs. Such compounds might then be used as probes to identify their protein targets. In specific aim 2, we will use a genome-wide RNAi approach to identify genes that selectively kill VHL (-/-) C. elegans. These genes would then be interrogated in human RCCs. Secondary screens will address whether dysregulation of HIF is responsible for differential sensitivity of VHL (-/-) and VHL (+/+) cells in specific aims 1 and 2. These experiments may identify new drug targets for renal carcinoma, in particular, and hypoxia tumor cells, in general. Moreover, they may establish a new paradigm for the selection of anticancer drug targets based on inactivating mutations of TSG.

<b>Project Number</b>	<b>Title</b>			
5R01CA68490-9	Functional Analysis of the von Hippel Lindau Protein			
<b>Institution</b>		<b>City / State</b>		
DANA-FARBER CANCER INSTITUTE		BOSTON, MA		
<b>Principal Investigator</b>		<b>Program Director</b>		
KAELIN, WILLIAM G		Judy Mietz		
<b>Start Date</b>	<b>End Date</b>	<b>Cancer Activity</b>	<b>Total Project \$</b>	
6/1/2004	05/31/2005	DNA Chromosome Aberrations	DCB \$283,286	
<b>Science Area</b>		<b>Percent</b>	<b>Percent \$</b>	
KIDNEY		100.00	\$283,286	

**Abstract**

DESCRIPTION: (Adapted from the investigator's abstract) Germ line mutation of the von Hippel-Lindau (VHL) tumor suppressor gene causes a hereditary cancer syndrome characterized by blood vessel tumors (hemangioblastomas) of the central nervous system and retina, renal cell carcinomas, and pheochromocytomas. Somatic VHL inactivation is common in sporadic renal cell carcinomas and hemangioblastomas. The VHL gene product, pVHL, forms a multimeric complex that contains elongin B, elongin C, Cullin 1, and Rbx1. This complex is an E3 ubiquitin ligase in which pVHL targets specific proteins for degradation by the proteasome. The best understood targets are HIF1a and HIF2a, two related transcription factors that regulate hypoxia-inducible genes such as VEGF. In normal cells HIF1a and HIF2a are rapidly degraded in the presence of oxygen and only become stable when cells are made hypoxic. In cells lacking pVHL these proteins can not be degraded and accumulate to high levels leading to activation of HIF target genes. How pVHL is prevented from degrading HIF under hypoxic conditions is not known and is the subject of specific aim 1. Specific aim 2 asks to what degree the set of pVHL-regulated genes and hypoxia-regulated genes overlap and specific aim 3 asks whether inhibition of HIF function is necessary and/or sufficient for tumor suppression by pVHL. Identification of additional proteins that bind to, and are regulated by, pVHL is the goal of specific aim 4.

<b>Project Number</b> 5R01CA95539-3	<b>Title</b> Novel Targets of the Von Hippel Lindau Gene	<b>City / State</b> HANOVER, NH		
<b>Institution</b> DARTMOUTH COLLEGE		<b>Program Director</b> Judy Mietz		
<b>Principal Investigator</b> RIGBY, WILLIAM F		<b>Cancer Activity</b> Cancer Cell Biology		
<b>Start Date</b> 4/1/2004	<b>End Date</b> 03/31/2005		<b>Total Project \$</b> DCB	\$281,240
<b>Science Area</b> KIDNEY		<b>Percent</b> 100.00	<b>Percent \$</b>	\$281,240

#### Abstract

DESCRIPTION: (provided by applicant) The regulation of neoplastic transformation, mRNA turnover, and the adaptive response to hypoxia are each of profound biologic and clinical importance. These apparently distinct cellular events are linked through the function of a single protein, the Von Hippel-Lindau (VHL) gene product, pVHL. Renal cell carcinomas (RCC) associated with VHL mutations exhibit increased expression of Glucose Transporter 1 (GLUT 1) and Vascular Endothelial Growth Factor (VEGF) mRNA, which results from increased mRNA stability and transcription. The interaction of the VHL gene product, pVHL, with elongin B, C, Cu12, and Rbx-1 has conclusively demonstrated a role of this complex (VBC) in regulating protein turnover. However, the absence of pVHL in RCC cells has also been associated with activation of the phosphatidylinositol 3 (PI 3)-kinase pathway, and disordered fibronectin (FN) assembly. Neither observation accounts for the increased stability of hypoxia-inducible (VEGF and possibly GLUT1) mRNA observed with pVHL-deficient RCC lines. We have made four observations that potentially identify mechanism(s) of increased GLUT1 mRNA stability in pVHL deficient RCC cell lines: i) The GLUT1 3'UTR alters gene expression in a pVHL-dependent manner; ii) pVHL specifically regulates the expression of hnRNP A2, which binds the GLUT1 3'UTR; iii) Regulation of hnRNP A2 levels by pVHL requires functioning proteasomes; iv) pVHL regulates p38 Stress-Activated Protein Kinase (SAPK) activation, which modulates VEGF and other AURE-dependent mRNA turnover. We hypothesize that the absence of pVHL results in the activation of the p38 SAPK pathway and hnRNP A2 overexpression and propose to address this and its relevance to GLUT1 mRNA turnover.

<b>Project Number</b> 5R01CA95648-2	<b>Title</b> Immunotherapy for Renal Cell Carcinoma	<b>City / State</b> HANOVER, NH		
<b>Institution</b> DARTMOUTH COLLEGE		<b>Program Director</b> Heng Xie		
<b>Principal Investigator</b> ERNSTOFF, MARC S		<b>Cancer Activity</b> Clinical Oncology		
<b>Start Date</b> 9/1/2004	<b>End Date</b> 08/31/2005		<b>Total Project \$</b> DCTD	\$351,550
<b>Science Area</b> KIDNEY		<b>Percent</b> 100.00	<b>Percent \$</b>	\$351,550

### Abstract

DESCRIPTION (provided by applicant): Medical treatment for RCC has primarily focused on biological therapies designed to mobilize immune effector cells that recognize and destroy cancer. Treatment with interleukin-2 (IL-2) and interferon alpha (IFNalpha), either singly or in combination, have shown dramatic clinical efficacy in a minority of metastatic RCC patients. The absence of benefit in the majority of patients may be due to tumor resistance and/or inadequate effector cell response. In vitro and animal studies show that dendritic cells (DC) are powerful initiators of cellular and humeral immune response and have therapeutic benefit in cancer models. Preliminary human studies suggest DC therapies have clinical and biological effectiveness in RCC patients. However, evidence suggests that immune inhibitory pathways may limit effectiveness of immunotherapy, including DC vaccines. The primary goal of combination immunotherapies is to successfully exploit complementary pathways of immune activity. DC vaccine, IL-2, and IFNalpha influence different pathways of immune activation and inhibition. DC vaccines can initiate, and at times sustain, an effective anti-tumor immune response. IL-2 induces T-cell activation and proliferation, and can overcome the negative inhibitory action of CTLA-4 on activated T-cells. IFNalpha enhances DC and T-cells function as well as tumor immunogenicity by inducing expression of MHC molecules and tumor associated antigens. We hypothesize that combined sequential DC vaccine and IL-2/IFNalpha therapy will decrease tumor-specific immune inhibition and increase tumor-specific immune activation in RCC patients. We propose to test this hypothesis in a phase H clinical trial of 33 RCC patients. Primary tumor removed as standard care will be processed for autologous vaccine. Eligible and consented patients with metastatic RCC will undergo leukapheresis to obtain peripheral blood monocyte derived DCs. DCs loaded with autologous tumor lysate administered by ultrasound guided injection into inguinal lymph nodes will be combined with IL-2 and IFNalpha therapy. We propose to determine 1) the objective clinical response rate to treatment, 2) the toxicity profile of this combined therapy, 3) the treatment related tumor-specific immune response and 4) the relationship of tumor-specific immune response and objective clinical response.

<b>Project Number</b> 5R01CA89102-4	<b>Title</b> IMMUNOTHERAPY WITH RENAL TUMOR RNA TRANSFECTED CELLS			
<b>Institution</b> DUKE UNIVERSITY		<b>City / State</b> DURHAM, NC		
<b>Principal Investigator</b> VIEWEG, JOHANNES W.G.		<b>Program Director</b> Heng Xie		
<b>Start Date</b> 2/1/2004	<b>End Date</b> 01/31/2005	<b>Cancer Activity</b> Clinical Oncology	<b>Total Project \$</b> DCTD	\$237,174
<b>Science Area</b> KIDNEY		<b>Percent</b> 100.00	<b>Percent \$</b>	\$237,174

### Abstract

The overall hypothesis of this proposal is that the activation of tumor antigen specific T cells will prevent tumor recurrence and metastasis. The frequency of cytotoxic T cells circulating in the peripheral blood of patients with metastatic renal cell carcinoma (RCC) and directed against RCC associated antigens is low or undetectable. In this proposal, we hypothesize that administration of escalating doses of tumor RNA transfected DC to patients with metastatic RCC will lead to detectable levels of circulating, functionally active tumor specific T cells. We have developed a novel, broadly applicable, and clinically feasible immunization strategy, using dendritic cells (DC) transfected with RNA encoding tumor antigens to elicit significant levels of tumor antigen specific T cells in patients with RCC. This program proposes to extend the basic research and clinical observations from this group of investigators to explore the clinical use of immunotherapy with tumor RNA transfected DC and address the overall hypothesis, specifically to generate and characterize tumor antigen specific T cells in patients with cancer. An ongoing phase I clinical trial of active immunotherapy with DC transfected with RNA encoding a widely expressed tumor antigen, prostate specific antigen (PSA), has demonstrated the safety and feasibility of this approach in patients with advanced prostate carcinoma. Preliminary analysis of patients following immunization demonstrates the induction of PSA specific T cells, suggesting the bioactivity of this vaccine strategy. To broaden the potential of this strategy and to avoid the potential pitfalls of targeting any single defined tumor antigen, we propose immunizing patients using total: tumor derived RNA transfected DC which we have demonstrated to elicit immune responses against a broad repertoire of tumor antigens and will not require prior determination or knowledge of the antigenic profile of each patient. The proposed program will set the stage for definitive trials designed to demonstrate a clinical benefit of inducing tumor antigen specific T cells in cancer patients using RNA transfected DC vaccines to reduce cancer recurrence and metastasis.

<b>Project Number</b> 5R21CA98446-2	<b>Title</b> Elimination of Regulatory T Cells	<b>City / State</b> DURHAM, NC		
<b>Institution</b> DUKE UNIVERSITY		<b>Program Director</b> Heng Xie		
<b>Principal Investigator</b> VIEWEG, JOHANNES W.G.		<b>Cancer Activity</b> Clinical Oncology	<b>Total Project \$</b> DCTD	\$279,200
<b>Start Date</b> 8/1/2004	<b>End Date</b> 07/31/2005			
<b>Science Area</b> KIDNEY		<b>Percent</b> 100.00	<b>Percent \$</b>	\$279,200

**Abstract**

DESCRIPTION (provided by applicant): The long-term goal of this application is to develop a clinically effective vaccination strategy for patients with metastatic renal cell carcinoma (RCC) by using mature dendritic cells (DC) transfected with autologous tumor RNA. In two pilot trials, we have shown that vaccination with RNA transfected DC represents a safe and effective strategy to elicit potentially therapeutic T cell responses in patients with metastatic renal and prostate cancers. More recent studies conducted in murine and human systems have demonstrated that the effects of vaccine protocols can be dramatically enhanced by the elimination of CD25+ regulatory T cell subsets preceding active immunotherapy. Here we propose to perform a clinical trial administering mature, renal tumor RNA transfected DC to patients with metastatic RCC with or without prior CD25+ regulatory T cell depletion (Aim 1). In Aim 2 of this application, we propose to analyze the vaccine-mediated and renal tumor-specific T cell responses among patients enrolled in each treatment arm (efficacy end point of this study). Immune monitoring will be performed by determining the presence and magnitude of tumor-specific T cells prior to and after vaccination by analyzing changes in the post-treatment cytokine profiles of activated T cells as assessed by an automated ELISPOT assay. We further propose to complement the ELISPOT data by measuring the functional capability of the in vivo generated CTL to specifically recognize and lyse autologous tumor targets. In order to optimize protocol efficacy in future trials, we will longitudinally monitor the kinetics of CD4+/CD25+ regulatory subsets prior and after vaccination with RCC RNA transfected DC. The proposed trial will allow us to proceed with sufficiently powered phase II clinical trials to directly determine the clinical efficacy of RNA transfected DC vaccines in patients with metastatic renal cell carcinoma.

<b>Project Number</b>	<b>Title</b>			
1U01CA111242-1	Methylation & Proteomics-Based Detection of Renal Cancer			
<b>Institution</b>		<b>City / State</b>		
FOX CHASE CANCER CENTER		Philadelphia, PA		
<b>Principal Investigator</b>		<b>Program Director</b>		
CAIRNS, PAUL		Jacob Kagan		
<b>Start Date</b>	<b>End Date</b>	<b>Cancer Activity</b>		<b>Total Project \$</b>
9/23/2004	08/31/2005	Early Detection	DCP	\$304,448
<b>Science Area</b>		<b>Percent</b>		<b>Percent \$</b>
KIDNEY		100.00		\$304,448

**Abstract**

DESCRIPTION (provided by applicant): Novel approaches for the early detection of renal cancer are urgently needed. Adult sporadic cancers are known to arise through the accumulation of multiple clonal genetic or epigenetic alterations which can be used as targets for the detection of neoplastic cells in bodily fluids that surround or drain from an organ. Since most renal tumors arise from the tubular epithelium and the renal parenchyma itself is in close proximity to the urinary collecting system, we hypothesized that urine from patients with renal cancer may contain shed neoplastic cells or DNA amenable to analysis. Using the sensitive methylation specific PCR (MSP) we have established that cancer specific hypermethylation of a panel of normally unmethylated tumor suppressor and cancer genes is frequent and can be detected in a simple voided urine from patients with early renal cancer. We have demonstrated for the first time that highly sensitive and specific MSP-based detection of renal cancer in urine is feasible. Our specific goals are 1) to construct a hypermethylation progression model and profile for early renal cancer by determining the frequency and timing of hypermethylation of the panel of cancer genes in defined pathological stages, 2) to validate the MSP assay for detection of renal cancer cell DNA in urine from 100 organ-confined renal cancers and to examine specificity in urine from 100 normal and benign controls, 3) to elucidate the methylome profile of early renal cancer and 4) to examine the diagnostic utility of proteomic based detection of early renal cancer. A progression model of early renal cancer will identify the most useful markers for early diagnosis and verify that hypermethylation is a valid detection target. Validation of a simple, non-invasive test for renal cancer would be of immense benefit towards the earlier diagnosis, and thereby cure, of this disease. The methylome profile will provide signatures for early renal cancer and an optimal detection panel of methylated genes. Serum proteomics based detection can be compared with methylation-based technology. Our long range goal is to make this molecular detection test a clinical reality. We have preliminary evidence which indicates that a sensitive and specific molecular test providing diagnosis of early renal cancer is feasible from a simple urine specimen. We will test this further.

<b>Project Number</b>	<b>Title</b>			
1R01CA106512-1	Allogeneic T Cell Responses Against Renal Cell Carcinoma			
<b>Institution</b>		<b>City / State</b>		
FRED HUTCHINSON CANCER RESEARCH CENTER		SEATTLE, WA		
<b>Principal Investigator</b>		<b>Program Director</b>		
WARREN, EDUS H		Susan McCarthy		
<b>Start Date</b>	<b>End Date</b>	<b>Cancer Activity</b>		<b>Total Project \$</b>
4/1/2004	03/31/2005	Immunology	DCB	\$313,199
<b>Science Area</b>		<b>Percent</b>		<b>Percent \$</b>
KIDNEY		100.00		\$313,199

### Abstract

DESCRIPTION (provided by applicant): Renal cell carcinoma (RCC) is distinguished amongst solid tumors for its resistance to conventional chemo- and radiotherapy but its susceptibility, in a minority of patients, to immunologic manipulation. Regression of metastatic disease is seen in 10-20% of patients who are treated with the immune-modulating cytokines Interleukin-2 and/or Interferon-alpha, and pilot studies have demonstrated that adoptive cellular therapy with ex vivo-expanded autologous lymphocytes can increase the response rate. More recently, regression of metastatic RCC has also been seen in up to 40% of patients who undergo nonmyeloablative allogeneic hematopoietic cell transplantation (HCT) from donors who are matched at the major histocompatibility complex (MHC). Responses in this setting are typically seen several months or more after HCT, after the establishment of complete donor T cell engraftment, and are closely associated with development of graft-versus-host disease (GVHD). This has suggested that tumor regression may in part be attributable to a graft-versus-tumor (GVT) effect mediated by donor cells reacting with minor histocompatibility (H) antigens expressed on recipient tumor cells. Preliminary data presented in this application demonstrate that T cell clones specific for recipient minor H antigens that are expressed on RCC tumor cells can be isolated from RCC patients experiencing tumor regression after nonmyeloablative allogeneic HCT. Identification of the genes encoding minor H antigens expressed on RCC cells and evaluation of their expression in normal and malignant tissues will facilitate the development of therapeutic strategies for augmenting GVT activity after allogeneic HCT. In this proposal, we will isolate CD8+ and CD4+ RCC-reactive minor H antigen-specific T cell clones from RCC patients after allogeneic HCT and use cellular and molecular methods to identify the genes encoding class I MHC-restricted minor H antigens expressed by RCC cells. The specific aims are: (1) Isolate and characterize CD8+ and CD4+ minor H antigen-specific and tumor-specific T cell clones from RCC patients after nonmyeloablative MHC-identical HCT. (2) Identify clonally expanded T cells that are associated with tumor regression in patients with metastatic RCC following nonmyeloablative MHC-identical HCT. (3) Identify the genes encoding antigens recognized by RCC-reactive CD8+ T cell clones isolated from patients with metastatic RCC who exhibit tumor regression after nonmyeloablative HCT.

<b>Project Number</b>	<b>Title</b>			
1R21CA101224-1	Dendritoma Vaccine on Renal Cell Carcinoma			
<b>Institution</b>		<b>City / State</b>		
GREENVILLE GENERAL HOSPITAL		GREENVILLE, SC		
<b>Principal Investigator</b>		<b>Program Director</b>		
WEI, YANZHANG		Heng Xie		
<b>Start Date</b>	<b>End Date</b>	<b>Cancer Activity</b>	<b>Total Project \$</b>	
7/19/2004	06/30/2005	Clinical Oncology	DCTD	\$225,500
<b>Science Area</b>		<b>Percent</b>	<b>Percent \$</b>	
KIDNEY		100.00		\$225,500

**Abstract**

DESCRIPTION (provided by applicant): Dendritic cell based tumor immunotherapy is a very promising approach for cancer treatment. Recent studies have shown that vaccines using the fused cells made from tumor cells and dendritic cells can stimulate tumor cell specific antitumor immune responses both in clinical studies and in pre-clinical animal studies. However, because of the difficulty of selecting and purifying the fused cells, fusion mixtures containing unfused and self-self fused cells were used in these studies. We reasoned that vaccines using purified fused cells would make the immune responses against tumors more effective. A method that purifies the fused cells (Instant Dendritomas) from the fusion mixture has been developed in our laboratory. Animal studies have shown that instant dendritomas are better activators of anti-tumor immunity than fusion mixtures. In vitro human studies using this technology have shown that instant dendritomas made from patients' own blood cells and primary tumor cells are effective in activating patients' peripheral lymphocytes to differentiate into functional cytotoxic T lymphocytes (CTLs) that efficiently lyse autologous tumor cells. Furthermore, our completed Phase I clinical trial in melanoma patients using the dendritoma vaccine demonstrated that the vaccine is safe and able to stimulate tumor cell specific immune responses. More importantly, some patients in the trial had benefited by having complete remission or stabled diseases. Based on these information, we, therefore, propose to conduct a phase II trial in patients with advanced renal cell carcinoma in order to determine if Instant Dendritoma vaccines have anti-tumor activity in a different type of cancer. The specific aims of this study are 1) to determine if the Instant Dendritoma vaccine has anti-tumor activity (defined as complete response, partial response) in patients with stage IV renal cell carcinoma and 2) to assess if the Instant Dendritoma vaccine produces an immune response as measured by an increase in interferon production by T-cell subsets, CTL activity, and immune response in tumor used for vaccination production. Patients with advanced renal cell carcinoma who have exhausted all available therapies will be considered for this trial. The vaccine will initially be given in combination with low dose IL-2; subsequent doses of the vaccine will be given at three month intervals for up to six doses. The study will employ a two-stage approach for evaluating efficacy. Initially, 10 patients will be evaluated; if one or more patients show evidence of a clinical response, then an additional 19 patients will be enrolled in the second stage such that 29 patients are ultimately included. We anticipate it will require at least two years to complete this study. This trial has already been approved by FDA and started to enroll patients. The preliminary data from the first 5 patients enrolled in this trial are encouraging.

<b>Project Number</b> 5R01CA79685-5	<b>Title</b> IDENTIFICATION OF CD8+ T CELL TARGETS ON RENAL CANCER	<b>City / State</b> BALTIMORE, MD	<b>Total Project \$</b> DCB \$288,736
<b>Institution</b> JOHNS HOPKINS UNIVERSITY	<b>Program Director</b> Susan McCarthy		
<b>Principal Investigator</b> JAFEE, ELIZABETH M	<b>Cancer Activity</b> Immunology		
<b>Start Date</b> 4/1/2004	<b>End Date</b> 12/31/2004		
<b>Science Area</b> KIDNEY	<b>Percent</b> 100.00	<b>Percent \$</b> \$288,736	

### Abstract

Murine studies employing tumor cells genetically-modified to express cytokines have demonstrated the induction of potent immunity that can cure mice of pre-established tumors. Experiments aimed at dissecting the mechanism of this response have revealed that both CD8+ and CD4+ T cells are critical effectors of the systemic immunity generated. These studies have led to on-going clinical trials that are testing this approach for the treatment of patients with cancer. Although the specificity of the immune response seen in these models suggests that renal-specific antigens are being expressed by the tumor and recognized by the ensuing immune response, in most cases the identity of these antigens is unknown. Recently, the identification of several murine and human melanoma antigens has validated these earlier hypotheses, and has provided a strong impetus for the identification of other human tumor antigens that can be exploited therapeutically. Antigen identification requires methods for generating T cell lines and clones, and for isolating either the gene encoding the tumor antigen or the antigen itself. We have developed methods for routinely generating tumor-specific CD8+ T cells from lymphocytes isolated from vaccinated patients. In this proposal, we will test our hypothesis that T cells isolated from vaccinated individuals will identify relevant tumor rejection antigens that can be used for immunization. Drawing on our previous experience with antigen identification, and utilizing our unique source of banked pairs of autologous lymphocytes and tumor cells obtained from patients receiving a renal cell carcinoma vaccine (RCC), we will identify tumor antigens expressed by human RCC. CD8+ renal tumor-specific T cell lines and clones will be generated from lymphocytes isolated from patients with renal cancer following vaccination with a GM-CSF secreting renal tumor vaccine. A genetic approach will be used to identify genes encoding for MHC class I-restricted antigens. Defined antigens will be analyzed to determine their MHC binding kinetics, and employed to analyze the frequency of antigen-specific T cells in banked pre- and post-vaccination lymphocytes. This was originally submitted as an interactive RO1. In the previously accompanying funded proposal, my colleague Dr. Pardoll is attempting to isolate genes encoding for MHC class II-restricted RCC tumor antigens. These studies should shed light on the immunobiology of tumors. Ultimately, the identification of common renal antigens that are recognized by both CD4 and CD8 T cells will allow the development of generalized gene therapy vaccine approaches that can generate immune responses potent enough to treat RCC.

<b>Project Number</b>	<b>Title</b>		
1R03CA110708-1	Analysis of Somatic Mutations in Cancer of the Kidney		
<b>Institution</b>		<b>City / State</b>	
LOUISIANA STATE UNIV HSC NEW ORLEANS		NEW ORLEANS, LA	
<b>Principal Investigator</b>		<b>Program Director</b>	
HUNT, JAY D		Virginia Hartmuller	
<b>Start Date</b>	<b>End Date</b>	<b>Cancer Activity</b>	<b>Total Project \$</b>
9/30/2004	08/31/2005	Epidemiology	DCCPS \$71,500
<b>Science Area</b>		<b>Percent</b>	<b>Percent \$</b>
KIDNEY		100.00	\$71,500

**Abstract**

DESCRIPTION (provided by applicant): Obesity, hypertension, and heavy cigarette smoking are suspected risk factors for kidney cancer, although the actual mechanisms by which these factors exert their action remain to be elucidated. Because the type of p53 gene mutation has provided clues as to etiology for several cancers (e.g., in bladder cancer and renal pelvic carcinoma it has been established that G:C->A:T transitions in p53 result from tobacco smoke carcinogens), and because we have access to DNA from 1,141 newly diagnosed renal cell carcinoma cases in the INCO Central Europe Health Study (CEHS), this NCI R03 project will evaluate whether there is a relationship between p53 gene mutation characteristics and certain selected risk factor characteristics in renal cell carcinoma. The results will help elucidate potential etiologic pathways for kidney cancer that will be pursued in subsequent studies. The CEHS is a hospital-based case-control study of environmental and genetic risk factors for kidney cancers at six centers across Central and Eastern Europe. Funded intramurally by NCI Division of Cancer Epidemiology and Genetics and IARC, the CEHS used an extensive in-person questionnaire to assess general lifestyle, medical and drug use history, family history, use of tobacco products, exposure to environmental tobacco smoke, residential history, alcohol and dietary information, and a detailed occupational history in the 1,141 cases and in 1,157 age-matched controls. Blood samples were collected on all 2,300 participants for genotyping of specific single nucleotide polymorphisms in genes encoding enzymes that are in well characterized metabolic pathways for a variety of environmental and occupational carcinogens, as well as for genes involved in DNA repair, immune function, and cell cycle control. Tumor tissue was collected and DNA was extracted at the NCI. Aim 1 is to determine the frequency, spectrum and specificity of p53 gene mutations from the DNA of each CEHS kidney cancer case. Exons 4-9 will be evaluated for mutations using denaturing high-performance liquid chromatography, which will be followed by sequencing to determine the exact nature of the mutation. Aim 2, in collaboration with Environmental Cancer Epidemiology Unit at IARC, is to use the p53 gene mutation data together with the lifestyle and occupational data on the cases to test specific etiologic hypotheses: (A) Is there a correlation between tobacco smoke exposure and p53 gene mutation spectrum? Do the kidney cancer patients with the highest levels of tobacco smoke exposure have a higher prevalence of p53 gene mutation and gene mutation spectrums that resemble the patterns associated with exposure to tobacco in bladder cancer and renal pelvic carcinoma patients? These analyses will be stratified by (i) never smokers versus ever smokers, (ii) duration and (iii) intensity of exposure. (B) Is there a correlation between other suspected risk factors, hypertension and obesity, and p53 gene mutation prevalence and spectrum? (C) Is there a correlation between survival and p53 gene mutation prevalence and spectrum?

<b>Project Number</b>	<b>Title</b>			
5R37CA58596-12	Functional Properties of the Wilm's Tumor Gene WT1			
<b>Institution</b>		<b>City / State</b>		
MASSACHUSETTS GENERAL HOSPITAL		BOSTON, MA		
<b>Principal Investigator</b>		<b>Program Director</b>		
HABER, DANIEL A.		Judy Mietz		
<b>Start Date</b>	<b>End Date</b>	<b>Cancer Activity</b>	<b>Total Project \$</b>	
1/20/2004	12/31/2004	DNA Chromosome Aberrations	DCB \$432,500	
<b>Science Area</b>		<b>Percent</b>	<b>Percent \$</b>	
KIDNEY		100.00	\$432,500	

**Abstract**

DESCRIPTION (provided by applicant): The genetic pathways that underlie the development of the pediatric kidney cancer Wilms tumor provide unique insight into the link between normal development of the kidney and its deregulation during malignant transformation. Inactivating mutations in the WT1 tumor suppressor, a zinc finger transcription factor encoded by multiple alternative splicing variants, underlie a subset of Wilms tumors, pointing to critical transcriptional targets that contribute to kidney development and tumorigenesis. We have used cells with inducible expression of the transcriptionally active isoform, WT1 (-KTS), combined with expression profile analysis to identify physiologically regulated target genes, and we have established a model for WT1-directed cellular differentiation using hematopoietic precursors. Here, we propose to focus on the functional properties of the most abundant isoform, WT1 (+KTS), whose function is unknown. In Aim I, we will search for endogenous genes whose expression is regulated by WT1 (+KTS), using both hybridization to microarrays and subtractive PCR approaches. The mechanisms by which this WT1 isoform regulates expression of its target genes (transcriptional as well as postulated post-transcriptional mechanisms) will be studied. The functional properties of these downstream effectors and their potential contribution to renal differentiation and Wilms tumorigenesis will be addressed. In Aim II, we will pursue protein-protein interactions that have been implicated in WT1 (+KTS) function, as well as in its characteristic subnuclear localization within "speckles". A combination of yeast-two hybrid assays and mass spectrometry sequencing of coprecipitated proteins will be employed, and the functional significance of confirmed interactors will be addressed. In Aim III, we will study another recently isolated transcription factor, BF2, which is also essential for kidney development, but through a distinct mechanism. Remarkably, BF2 is expressed only in stromal cells of the fetal kidney, yet its inactivation in the mouse suppresses epithelial differentiation without affecting stromal cells themselves. Using inducible expression of BF2 and expression profile analysis, we have identified downstream targets, including secreted growth factors, whose contribution to renal differentiation and to stromal-epithelial interactions will be explored. Taken together, this proposal aims at identifying downstream targets of two transcription factors that are essential to kidney development, defining the mechanism by which these targets are regulated, and studying their potential contribution to Wilms tumorigenesis.

<b>Project Number</b> 1R01CA104505-1	<b>Title</b> TGF Beta receptor biology in human renal cell carcinoma	<b>City / State</b> JACKSONVILLE, FL		
<b>Institution</b> MAYO CLINIC COLL OF MED, JACKSONVILLE		<b>Program Director</b> Donald Blair		
<b>Principal Investigator</b> COPLAND, JOHN A		<b>Cancer Activity</b> Cancer Cell Biology		
<b>Start Date</b> 8/10/2004	<b>End Date</b> 07/31/2005		<b>Total Project \$</b> DCB	\$306,750
<b>Science Area</b> KIDNEY		<b>Percent</b> 100.00	<b>Percent \$</b>	\$306,750

### Abstract

DESCRIPTION (provided by applicant): Renal cell carcinoma (RCC) is a major health issue. While localized disease can be cured surgically, there is no effective treatment for metastatic disease. The development of therapy awaits understanding of the molecular pathways that underlie RCC carcinogenesis. Using genomic profiling of conventional RCC patient matched specimens, we identified aberrations in the transforming growth factor beta (TGFbeta) pathway. We observed loss of type III TGFbeta receptor (TbetaR3) in all samples. This suggests that loss of TbetaR3 is an early, sentinel event in the genesis of RCC. This is the first clear demonstration linking loss of TbetaR3 to a disease state. We also observed loss of type II TGFbeta receptor (TbetaR2) in metastatic RCC's. These data suggest that aberrations in TGFbeta signaling are important in RCC carcinogenesis and progression, and are mediated through down regulation of TbetaR. We hypothesize that loss of TbetaR3 promotes RCC tumorigenesis through dysregulation of TGFbeta signaling, mediated through Smad dependent and/or independent mechanisms. Our preliminary data also support the hypothesis that TbetaR3 has growth inhibitory activity independent of TGFbeta signaling and TbetaR2. These hypotheses will be tested in models of RCC, in vitro and in vivo, through the following specific aims: 1) We will test the hypothesis that TbetaR3 inhibits cell proliferation in RCC, in vitro, through both Smad dependent and independent mechanisms. We will further test whether TbetaR3 growth inhibition is mediated through TGFbeta/TbetaR2 independent pathways through interaction with, as yet, unknown intracellular proteins. 2) We will test the hypothesis that TbetaR3 inhibits tumorigenicity in vivo, using relevant animal models of RCC. We will test the efficacy of adenoviral gene therapy targeting TbetaR. 3) We will test the hypothesis that TbetaR3 is silenced in RCC through transcriptional regulation of the TbetaR3 promoter. Completion of these studies will define the role of TbetaR3 loss in RCC carcinogenesis, the function of TbetaR3 in normal renal biology and carcinogenesis, and the mechanism of regulation of TbetaR3 in RCC biology.

<b>Project Number</b> 5R03CA103494-3	<b>Title</b> Differential Gene Expression in Renal Cancers	<b>City / State</b> JACKSONVILLE, FL		
<b>Institution</b> MAYO CLINIC COLL OF MED, JACKSONVILLE		<b>Program Director</b> Virginia Hartmuller		
<b>Principal Investigator</b> PARKER, ALEXANDER S		<b>Cancer Activity</b> Epidemiology	DCCPS	<b>Total Project \$</b> \$75,000
<b>Start Date</b> 9/1/2004	<b>End Date</b> 08/31/2005			
<b>Science Area</b> KIDNEY		<b>Percent</b> 100.00		<b>Percent \$</b> \$75,000

#### Abstract

DESCRIPTION (provided by applicant): Incidence and mortality rates for renal cell carcinoma (RCC) have increased steadily over the past three decades, and these trends are largely unexplained. While it is widely acknowledged that a history of obesity increases the risk of developing RCC, little is known regarding the actual biologic mechanisms that underlie this association. Indeed, the Kidney Cancer Progress Review Group recently recommended that investigators focus efforts on illuminating the "biological mechanisms underlying the known risk factors for kidney cancers". A logical first approach to identifying specific molecular alterations that link obesity and RCC would be to compare the somatic gene expression profiles in tumors from RCC patients with and without a history of obesity. In this application, we propose to employ the high throughput capabilities of commercially available DNA microarray technology in order to scan the genome for genes that are differentially expressed in RCC tumors that develop in obese and non-obese individuals. To do this, we will use the Mayo Nephrectomy Registry to identify all patients treated surgically for stage I, clear cell RCC at the Mayo Clinic Rochester during a one year period who report no history of smoking. From this list, we will sample 10 individuals with and 10 individuals without a history of obesity. We will then request fresh-frozen tissue samples (both tumor and normal) on each individual and use laser capture microdissection to obtain samples for RNA extraction. The Mayo Clinic DNA Microarray Core Facility will conduct measurements of gene expression in the sampled tissues using the Affymetrix U133 GeneChip platform. Members of our investigative team with experience in microarray analysis and bioinformatics will then conduct analysis to determine differential gene expression patterns between the two study groups. Explicitly, we wish to test the hypothesis that specific gene expression profiles can be identified that will distinguish between RCC tumors that develop in obese and nonobese individuals. Results from this study will provide direction and support for larger, more candidate-specific investigations of the biologic mechanisms behind the obesity/RCC association. In summary, this application represents an effort to harness an innovative, high throughput laboratory technology in order to improve our understanding of RCC etiology by broadening the search for risk-factor-specific molecular heterogeneity in human RCC. The long-term goal of the proposed study is to potentially inform novel prevention and treatment strategies for RCC at both the individual and population level.

<b>Project Number</b>	<b>Title</b>			
2R01CA78383-6	Regulatory pathways and role of VPF/VEGF in renal cancer			
<b>Institution</b>		<b>City / State</b>		
MAYO CLINIC COLL OF MEDICINE, ROCHESTER		ROCHESTER, MN		
<b>Principal Investigator</b>		<b>Program Director</b>		
MUKHOPADHYAY, DEBABRATA		Neeraja Sathyamoorthy		
<b>Start Date</b>	<b>End Date</b>	<b>Cancer Activity</b>	<b>Total Project \$</b>	
4/1/2004	03/31/2005	Tumor Biology	DCB	\$292,638
<b>Science Area</b>		<b>Percent</b>		<b>Percent \$</b>
KIDNEY		100.00		\$292,638

## Abstract

DESCRIPTION (provided by applicant): Angiogenesis plays a pivotal role in several important pathological disease processes as well as in normal physiology. It is widely anticipated that modulation of angiogenesis (inhibition in tumors, stimulation in vascular insufficiency) will provide important therapeutic benefits. Like many human carcinomas, renal cell carcinomas (RCC), which are characteristically highly vascular, are thought to induce angiogenesis by secreting an angiogenic cytokine: vascular permeability factor, also known as vascular endothelial growth factor (VPF/VEGF). The long-term goal of this proposal is to investigate in depth the regulatory role of VPF/VEGF in RCC. RCC, arising sporadically or in the von Hippel Lindau (VHL) syndrome, exhibits loss of function of a tumor suppressor gene, VHL. Previously, our laboratory as well as others have shown that wild type VHL (pVHL) selectively interacts with and thereby down-regulates the activities of PKCzeta and Sp-1 and also degrades alpha subunits of hypoxia inducible factors (HIFs); when pVHL is afunctional, as in RCC, VPF/VEGF is over-expressed. But the mechanism of VPF/VEGF over-expression in the absence of pVHL is not yet clear. In Aim 1, we will study how PKC zeta interplays with HIF alphas, p300 and other transcription factors as well as trans-inhibitors like FIH. The endogenous promoter activity of VPF/VEGF will also be examined in RCCs in regard to PKC zeta and its regulators. In Aim 2, we will investigate the mechanism of VPF/VEGF mRNA stability in RCCs. We will examine the involvement of PKC zeta and its associated molecules in the modulation of RNA-binding protein, HuR, that usually binds at 3'-translated regions of VPF/VEGF mRNA. This aim will also elucidate the physical interactions between HuR and pVHL and its biological relevance. Finally, Aim 3 will extrapolate tissue culture data and reagents to animal models of RCC, seeking to understand the importance of VPF/VEGF as well as the mechanisms by which VPF/VEGF is activated in renal tumors; it is likely that we will propose new, specific targets for therapies that block angiogenesis and thereby tumor growth and metastasis. These experiments will provide new and important information on the mechanisms of carcinogenesis and suggest new targets for intervention in RCC, a common, highly vascular, angiogenesis-dependent carcinoma that is resistant to currently available therapies.

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<b>Project Number</b>	<b>Title</b>			
5R01CA24652-24	Kidney Response to Radiation and Chemotherapy			
<b>Institution</b>		<b>City / State</b>		
MEDICAL COLLEGE OF WISCONSIN		MILWAUKEE, WI		
<b>Principal Investigator</b>		<b>Program Director</b>		
MOULDER, JOHN E		Helen Stone		
<b>Start Date</b>	<b>End Date</b>	<b>Cancer Activity</b>		<b>Total Project \$</b>
2/1/2004	01/31/2005	Radiotherapy	DCTD	\$317,250
<b>Science Area</b>		<b>Percent</b>		<b>Percent \$</b>
KIDNEY		100.00		\$317,250

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**Abstract**

DESCRIPTION (provided by applicant): This project is based on the hypothesis that late radiation-induced normal tissue injuries can be prevented and/or treated with post-irradiation pharmacologic intervention. The rat radiation nephropathy model is used to study the pathophysiological mechanism(s) of late radiation-induced normal tissue injuries, and these mechanistic understandings are used in the development of methods for prophylaxis and treatment of these injuries. The discovery that radiation nephropathy is a major complication of bone marrow transplantation (BMT) conditioning, and that the rat was an excellent model for this nephropathy, added a preclinical component to the project. The discovery that angiotensin converting enzyme (ACE) inhibitors and an angiotensin II (AII) blockers could be used for the prophylaxis and treatment of BMT nephropathy led directly to clinical studies. The studies are strongly influenced by the finding that blocking the renin-angiotensin system can permanently interfere with the development of radiation nephropathy even when treatment is started weeks after irradiation and/or is not continued indefinitely. These latter findings cast considerable doubt on the standard mechanistic explanations for late radiation injuries, and suggest that injuries caused by radiation therapy, radiation accidents or nuclear terrorism could be treated or prevented with post-irradiation pharmacological interventions. The specific aims of this proposal fall into four related groups:

- 1) Refute the hypothesis that radiation-induced activation of the renin-angiotensin system is the proximal mechanistic cause of radiation nephropathy.
- 2) Confirm the hypothesis that prophylaxis and treatment of radiation nephropathy with ACE inhibitors and AII blockers operate by different mechanisms.
- 3) Determine the mechanistic basis for the efficacy of ACE inhibitors and AII receptor blockers in the prophylaxis of radiation nephropathy.
- 4) Complete the randomized, prospective, trial of the use of ACE inhibitors to prevent the development of radiation nephropathy in BMT patients.

<b>Project Number</b> 5R01CA93871-3	<b>Title</b> Chemoresistance in Renal Cell Cancer	<b>City / State</b> COLUMBUS, OH	
<b>Institution</b> OHIO STATE UNIVERSITY		<b>Program Director</b> Heng Xie	
<b>Principal Investigator</b> AU, JESSIE L.-S.		<b>Cancer Activity</b> Clinical Oncology	<b>Total Project \$</b> DCTD \$282,108
<b>Start Date</b> 7/1/2004	<b>End Date</b> 06/30/2005		
<b>Science Area</b> KIDNEY		<b>Percent</b> 100.00	<b>Percent \$</b> \$282,108

## Abstract

DESCRIPTION (provided by applicant): Patients with metastatic renal cell cancer (RCC) have a bleak prognosis with an average survival time of less than 18 months. Hence, there is an urgent need of more effective treatments. RCC show an overall response rate of <10% to chemotherapy and is resistance to different classes of drugs that do not share the same action or resistance mechanisms. We recently discovered a new epigenetic mechanism of anticancer drug resistance, that is caused by two fibroblast growth factors expressed in solid tumors, i.e., acidic and basic fibroblast growth factors (aFGF and bFGF). These two proteins at clinically relevant concentrations induce an up to 10-fold resistance to drugs with diverse structures and action mechanisms. Inhibitors of these FGF, including the monoclonal antibodies and suramin (at low concentrations with no cytotoxicity), completely reverse the FGF-induced resistance and enhance the activity of chemotherapy. In animals, low and nontoxic doses of suramin significantly enhanced the efficacy of chemotherapy, resulting in eradication of well-established tumors in immunodeficient mice. In addition to these earlier studies conducted with prostate tumor cells, we have obtained data showing the following. (a) RCC cell lines and RCC tissues obtained from patients contained high levels of aFGF/bFGF. (b) The FGF-induced resistance was observed for drugs that have been used to treat RCC, and was reversed by FGF inhibitors. (c) aFGF/bFGF induced chemoresistance in other human solid tumor cells (i.e., lung ovarian, colon, pharynx). (d) FGF inhibitors (suramin and/or pentosan polysulfate) enhanced the antitumor activity of 5-fluorouracil and gemcitabine in two RCC cell lines and histocultures of RCC patient tumors. (e) bFGF expression was a better predictor of paclitaxel resistance in multiple types of human tumors, compared to other known prognostic indicators (i.e., mutated p53, overexpression of Bcl2 and the mdr1 p-glycoprotein, and tumor pathology). (e) Results of cDNA microarray analysis show that bFGF enhanced the expression of genes involved in several known chemoresistance mechanisms (GST, Bcl-2 family proteins, topoisomerase, and drug efflux proteins), whereas suramin reduced the expression of these genes and FGF receptors. To evaluate the clinical application of our findings, we initiated a phase I/II trial using low dose suramin to enhance the efficacy of paclitaxel and carboplatin, in advanced non-small cell lung cancer patients. The preliminary results of the completed phase I trial support the hypothesis that low and nontoxic doses of suramin enhanced the response rate, prolonged the progression-free survival and prolonged the median survival time, as compared to the historical data in this patient group treated with only paclitaxel/carboplatin. Based on these findings, we hypothesize that (a) aFGF/bFGF is an important resistance mechanism of RCC to chemotherapy, and (b) aFGF/bFGF inhibitors can enhance the efficacy of chemotherapy in RCC. The overall goal of this application is to test these hypotheses in preclinical (Aims 1 to 3) and clinical (Aims 4 and 5) studies. The proposed research has the potential of identifying a new treatment paradigm for RCC.

<b>Project Number</b>	<b>Title</b>			
5R21CA98842-2	Molecular Cloning of the Wilms tumor Gene from 7p15-21			
<b>Institution</b>		<b>City / State</b>		
ROSWELL PARK CANCER INSTITUTE CORP		BUFFALO, NY		
<b>Principal Investigator</b>		<b>Program Director</b>		
SOSSEY-ALAOUI, KHALID		Kelly Kim		
<b>Start Date</b>	<b>End Date</b>	<b>Cancer Activity</b>		<b>Total Project \$</b>
8/1/2004	07/31/2005	DNA Chromosome Aberrations	DCB	\$189,245
<b>Science Area</b>		<b>Percent</b>		<b>Percent \$</b>
KIDNEY		100.00		\$189,245

**Abstract**

DESCRIPTION (provided by applicant): Wilms tumor is pediatric lesion of the kidney and is one of the most common solid malignancies of the childhood. Disease can occur in one or both kidneys, approximately 8% of cases being bilateral. The suggestion of a genetic component in the etiology of the tumor has come from several observations. Firstly, bilateral disease is associated with an early age of onset. Secondly, there is a high incidence of bilateral tumors in cases with a family history of Wilms tumor and in patients with associated congenital anomalies. Histological features indicate that the tumor occurs as a result of aberrant embryological development of the kidney. The kidney is therefore a model for studying the association between processes involved in tissue development and predisposition to malignancy. Although a gene for Wilms tumor (WT1) has been cloned, less than 10% of cases could be explained by mutations and/or alterations of this gene. Several other loci have been implicated in the etiology of Wilms tumors, including the 7p15-21 locus which was shown to be involved in 15-25% of Wilms tumors cases, strongly suggesting that a tumor suppressor gene for this disease must lie within this region. Since homozygous deletions are hallmarks of tumor suppressor genes, a homozygous deletion has been described in a Wilms tumor within the 7p15-21 locus and we have now characterized the extent of this deletion as a first step towards the identification of the Wilms tumor suppressor gene using a very powerful mutation analysis technology (DHPLC) and a large cohort of Wilms tumors, including those tumors that we have identified to show loss of heterozygosity at the 7p15-21 locus. By studying the expression pattern of this gene we will be able to identify the population of stem cells which give rise to these tumors. Understanding the nature of the genetic events which allow these cells to escape their normal growth regulation may also provide an opportunity for therapeutic intervention.

<b>Project Number</b>	<b>Title</b>			
5R21CA98969-2	Array Based CGH for Genome-Wide Analysis of Wilms' Tumor			
<b>Institution</b>		<b>City / State</b>		
ROSWELL PARK CANCER INSTITUTE CORP		BUFFALO, NY		
<b>Principal Investigator</b>		<b>Program Director</b>		
NOWAK, NORMA J.		Kelly Kim		
<b>Start Date</b>	<b>End Date</b>	<b>Cancer Activity</b>	<b>Total Project \$</b>	
5/1/2004	04/30/2005	DNA Chromosome Aberrations	DCB \$156,741	
<b>Science Area</b>		<b>Percent</b>	<b>Percent \$</b>	
KIDNEY		100.00	\$156,741	

**Abstract**

DESCRIPTION (provided by applicant): Wilms' tumor (WT) is the most common primary malignant solid tumor of childhood, accounts for 6-7% of all childhood malignancies in the United States, and is an important model for the study of the fundamental mechanisms of tumorigenesis. The WT1 gene was identified and positionally cloned in 1991, but we have shown that this gene is only involved in a very small percentage (-5-7%) of WT patients. Since the cloning of WT1, no additional genes have been shown to be directly involved in the development of WT. Several groups, including our own, have attempted to identify new regions associated with WT through genome wide LOH (Loss of Heterozygosity) analysis and CGH (Comparative Genomic Hybridization) analysis on metaphase chromosomes, resulting in the association of chromosomes 5q (22%) 7p (15%) and 16q (12%) with WT. While these studies have provided some evidence for genomic regions harboring genes involved in Wilms' tumorigenesis, no gene has yet been cloned to coincide with these regions. These efforts have been hampered by the lack of robust, high throughput, high resolution techniques for detecting submicroscopic deletions and amplifications. Thus, the genetic defect(s) in the majority of these patients still remains unclear. It is clear that WT results from abrogation of the normal developmental pathways within the kidney. Identification of additional genes involved in WT pathogenesis will not only advance our understanding of Wilms' tumorigenesis, but also uncover crucial events required for proper cellular differentiation and normal kidney development. We have been developing a high throughput array based technology that will allow us to scan the genome for deletions and amplifications, or copy number aberrations (CNAs), at 0.5-1mb level of resolution. Hybridization of tumor DNA to these arrays readily identifies losses and gains in the tumor samples using a normal DNA reference. We will identify the recurrent copy number aberrations present in our WT bank, tile the implicated chromosomal regions on WT subarrays, and identify candidate genes involved in WT and kidney development.

<b>Project Number</b>	<b>Title</b>			
5R37CA53370-14	Function of the Adenovirus E1B Oncogene			
<b>Institution</b>		<b>City / State</b>		
RUTGERS THE ST UNIV OF NJ NEW BRUNSWICK		NEW BRUNSWICK, NJ		
<b>Principal Investigator</b>		<b>Program Director</b>		
WHITE, EILEEN		May Wong		
<b>Start Date</b>	<b>End Date</b>	<b>Cancer Activity</b>	<b>Total Project \$</b>	
7/1/2004	06/30/2005	Comp Min Biomed Prog	DDES	\$408,563
<b>Science Area</b>		<b>Percent</b>	<b>Percent \$</b>	
KIDNEY		100.00		\$408,563

#### Abstract

DESCRIPTION (provided by applicant): The human DNA tumor virus adenovirus replicates productively in human cells and transforms primary rodent epithelial cells. Integral to these activities is the deregulation of cell cycle control and the inhibition of programmed cell death (apoptosis) by the viral oncogenes E1A and E1B. E1A induces S-phase, which is required for replication of viral DNA and stimulation of cell proliferation in transformation. E1A also induces apoptosis that it requires E1B to inhibit to sustain productive infection and permit oncogenic transformation. E1A deregulates the cell cycle by binding to and inhibiting negative regulators of cell growth, one of which is the retinoblastoma tumor suppressor gene product (Rb). E1B encodes two proteins that block apoptosis by binding to and inhibiting pro-apoptotic proteins: 55K binds and inhibits the p53 tumor suppressor protein; 19K binds and inhibits Bax and other related pro-apoptotic components of the apoptotic machinery. I propose to extend these studies in two specific areas that center around the mechanism of modulation of death receptor signaling by E1A and E1B during infection of human cells, and the mechanism of inhibition of p53-dependent apoptosis downstream of mitochondria by the E1B 19K protein in rodent cells. Finally, mice mutant for known components on the apoptotic machinery will be utilized to establish the requirement and ordering of these gene products in cell death signaling pathways in viral infection and transformation. By executing these aims we hope to illuminate novel mechanisms of cell death regulation and how they relate to virus replication and the development of cancer. Knowledge gained from this approach will be useful in the development of new anti-viral and anti-cancer regimens.

<b>Project Number</b>	<b>Title</b>		
1R43CA106685-1	LAYERED MEMBRANES IN PROTEOMIC PROFILING OF RENAL CANC*		
<b>Institution</b>		<b>City / State</b>	
TWENTY-TWENTY GENE SYSTEMS		ROCKVILLE, MD	
<b>Principal Investigator</b>		<b>Program Director</b>	
KNEZEVIC, VLADIMIR		James Tricoli	
<b>Start Date</b>	<b>End Date</b>	<b>Cancer Activity</b>	<b>Total Project \$</b>
4/6/2004	03/31/2005	Diagnostics Research	DCTD \$79,501
<b>Science Area</b>		<b>Percent</b>	<b>Percent \$</b>
KIDNEY		100.00	\$79,501

**Abstract**

DESCRIPTION (provided by applicant) "Personalized medicine" is a new clinical paradigm where a specific cancer therapeutic ("smart drug") is prescribed based upon the individual biology of the patient. A goal of personalized medicine is to select individual therapies based upon the correlation of proteomic profiles from diseased tissues with patient response to drug therapy. Therefore, there is a critical need for a practical tool that can generate protein expression profiles from small amounts of tissue samples collected from patients. We hypothesize that cancer patients will be treated more successfully if a correlation between proteomic profiles and drug efficacy is established and that a diagnostic test using our innovative and novel technology, Layered Protein Scanning (LPS), will be able to establish that correlation. The long-term goal of this proposal is to, by the end of Phase II, develop a clinical diagnostic tool that would be used to select and monitor patients most likely to respond to "smart drugs". We will initially focus on clear cell renal carcinoma patients being treated with inhibitors of Epidermal Growth Factor Receptor (EGFr) signaling. To establish proof of principle, in Phase I, we will test the ability of LPS to profile EGFr activity in clear cell renal carcinoma cell lines and tissues. Specifically, we will use LPS to quantitate differences in the levels of total EGFr, activated EGFr, total Erk1/2, activated Erk1/2, total TGF-alpha, and total VHL in: 1) Cell line samples (by comparing LPS-tissue blot with standard western blot) 2) Tissue sections of RCC (by comparing LPS-tissue blot to immunohistochemistry).

<b>Project Number</b>	<b>Title</b>			
5R01CA102600-2	Renal cancer genomic alterations and environmental risk			
<b>Institution</b>		<b>City / State</b>		
UNIVERSITY OF CALIFORNIA SAN FRANCISCO		SAN FRANCISCO, CA		
<b>Principal Investigator</b>		<b>Program Director</b>		
WALDMAN, FREDERIC M.		Kumi Iwamoto		
<b>Start Date</b>	<b>End Date</b>	<b>Cancer Activity</b>	<b>Total Project \$</b>	
9/24/2004	05/31/2005	Epidemiology	DCCPS	\$296,086
<b>Science Area</b>		<b>Percent</b>	<b>Percent \$</b>	
KIDNEY		100.00		\$296,086

### Abstract

DESCRIPTION (provided by applicant): A number of environmental, occupational and life style factors have been associated with renal cell cancer (RCC) incidence. However, the mechanism by which these risk factors cause RCC, presumably acting through alteration of specific genes, is unknown. We hypothesize that specific genomic alterations detected in renal cancers (DNA gain or loss) are associated with renal cancer risk factors. We propose to analyze associations between risk factors and genomic alterations in renal tumors collected as part of the Eastern European Renal Cell Cancer Study (EERCC), being carried out by the National Cancer Institute and the International Association for Research on Cancer (IARC). We hypothesize that specific genomic alterations in renal cancers (DNA gain or loss) are associated with renal cancer risk factors and with tumor progression. Our Aims are: The overall design of this study is to characterize over 700 renal tumors by array CGH to define genomic alterations and their associations with clinical variables, and with exposure, and genetic risk factors. 1. Identify genomic alterations associated with tumor stage in conventional (clear cell) renal cancer. Array based CGH will be used to define copy number gains and losses, including DNA amplifications and homozygous deletions, in DNA from 200 conventional RCC grouped according to stage (I-IV). 2. Identify associations of genomic alterations with statistically significant exposure, occupational, and genetic risk factors defined using this IARC case-control cohort of renal cancer patients. 500 additional tumors will be characterized by array CGH beyond those studied in Aim 1. Patients will be selected based on their exposure history, to generate the most power for analysis (enriching for highest exposure groups). Risk factors to be considered include smoking, occupational, and environmental exposures (trichloroethylene, polycyclic aromatic hydrocarbons, petroleum products, asbestos, and heavy metals) and factors that alter renal function (obesity and hypertension). 3. Validate genes identified in Aims 1 and 2 using immunohistochemical analysis of renal tumor tissue microarrays containing the entire cohort of tumors.

<b>Project Number</b> 1R01CA109446-1	<b>Title</b> Recombinant Immunotherapy for Renal Cell Carcinoma	<b>City / State</b> IOWA CITY, IA		
<b>Institution</b> UNIVERSITY OF IOWA		<b>Program Director</b> Toby Hecht		
<b>Principal Investigator</b> GRIFFITH, THOMAS S		<b>Cancer Activity</b> Biological Resources Branch	<b>Total Project \$</b> DCTD	\$272,138
<b>Start Date</b> 6/15/2004	<b>End Date</b> 05/31/2005			
<b>Science Area</b> KIDNEY		<b>Percent</b> 100.00	<b>Percent \$</b>	\$272,138

### Abstract

DESCRIPTION (provided by applicant): Although many agents induce apoptosis, they are commonly associated with side effects that compromise health. TRAIL (TNF-related apoptosis-inducing ligand)/Apo-2L is generating excitement because it induces apoptosis in a wide range of tumor cells but not in normal cells and tissues. Preclinical studies using systemic TRAIL/Apo-2L doses are safe and can suppress tumor growth in vivo. Large amounts of TRAIL/Apo-2L are, however, needed to inhibit tumor formation, primarily because of the short in vivo half-life of the protein. Therefore, an alternative means of delivery may increase the relative activity of TRAIL/Apo-2L such that larger, more established tumors can be eradicated as efficiently as smaller tumors. This year in the U.S. approximately 30,000 new cases of renal cell carcinoma (RCC) will be diagnosed and nearly 12,000 deaths are expected from RCC. Metastatic RCC carries a median survival of 8 months and almost 30% of RCC patients are diagnosed with advanced metastatic disease. Furthermore, RCC is highly resistant to chemotherapy, a possible consequence of its association with the multidrug-resistance P-glycoprotein. RCC is regarded as an immunogenic tumor, with many reports of spontaneous regression and evidence of tumor-specific immune responses being a strong indicator of the immunogenicity of RCC. Thus, immunotherapy is being intensely studied as a treatment for RCC. Unfortunately, the response rates have been poor and significant toxicity reported, limiting the use of immunotherapy in the treatment of RCC. Gene transfer therapy offers new alternatives in the treatment of RCC.

Employment of non-replicative viral gene delivery systems is making it possible to administer genes directly into tumors in situ. Previously, we described the cytotoxic activity of recombinant TRAIL/Apo-2L protein against human RCC cell lines, and the development and testing of a recombinant, replication-deficient adenoviral vector encoding the human TRAIL gene (Ad5-TRAIL). Transfer of the TRAIL gene into human tumor cells in vitro and in vivo, using immunodeficient mice, led to the rapid production and expression of TRAIL/Apo-2L protein, and apoptotic death of the tumor cells. However, it remains unknown whether Ad5-TRAIL will inhibit tumor growth in immunocompetent animals, and if the Ad5-TRAIL-induced tumor cell death will activate systemic antitumor immunity. With this in mind, the proposed project will employ a novel adenoviral vector encoding the mouse TRAIL gene (Ad5-mTRAIL) combined with agents to boost systemic immune responses through augmenting antigen presentation and stimulating T cell expansion to develop unique approaches for the treatment of RCC. Specific Aims: (1) Investigate the ability of DC to present antigens derived from apoptotic Renca cells to stimulate antitumor immunity and analyze the effector cells and mechanism of tumor rejection; and (2) Examine the ability of Gelfoam R and depsiptide (FR901228) to augment Ad5-mTRAIL infectivity and transgene expression, making Ad5-mTRAIL gene transfer therapy more potent.

<b>Project Number</b> 5R21CA97421-2	<b>Title</b> Proteotoxicity Biomarkers for Cisplatin Nephrotoxicity	<b>City / State</b> BALTIMORE, MD	<b>Total Project \$</b> \$148,500
<b>Institution</b> UNIVERSITY OF MARYLAND BALT PROF SCHOOL	<b>Program Director</b> Mary Wolpert		
<b>Principal Investigator</b> FLAWS, JODI A	<b>Cancer Activity</b> Biochemistry and Pharmacology		
<b>Start Date</b> 9/1/2004	<b>End Date</b> 08/31/2005	DCTD	
<b>Science Area</b> KIDNEY	<b>Percent</b> 100.00	<b>Percent \$</b> \$148,500	

**Abstract**

DESCRIPTION (provided by applicant): There is a pressing need to better understand the mechanisms of platinum-based anti-cancer drug toxicity to the kidney and to utilize this basic scientific information for the development of biomarkers to detect early manifestations of renal damage in patients treated with these and other nephrotoxic anti-cancer agents. It is our hypothesis that treatment of renal proximal tubule cells to platinum-based drugs will produce specific oxidative cellular effects that will be intrinsically linked to increased excretion of specific oxidized proteins of renal derivation into the urine via chaperoning by specific stress protein families. In vitro studies which will compare the responses of primary cultures of both human and rat kidney cells to cisplatin using a short term dose-response design. These studies will provide a mechanistic basis for testing the hypothesis that stress proteins play a central role in the excretion of specific oxidized proteins from this target cell population. Data from these studies should will also determine whether this proposed mechanism of platinum-induced toxicity is similar in rat and human proximal tubule cells. This hypothesis will be further tested via studies involving acute in vivo exposure of rats to platinum- based drugs using a dose-response, time course design. Examination of urine samples from these animals will be focused on those proteins found to be preferentially excreted by the proximal tubule cells in culture. Data from these studies will be essential for understanding the extent to which urinary biomarkers derived in rats may be used for predicting platinum-induced human nephrotoxicity. The utility of these new biomarkers for earlier detection of platinum-induced nephrotoxicity in humans will hopefully be evaluated under a future R33 grant application that will apply knowledge derived from these studies to other nephrotoxic anti-cancer agents.

<b>Project Number</b> 5R01CA95572-3	<b>Title</b> IT by CD40 stimulation and IL2 against renal cell carcin	<b>City / State</b> RENO, NV		
<b>Institution</b> UNIVERSITY OF NEVADA RENO		<b>Program Director</b> Susan McCarthy		
<b>Principal Investigator</b> MURPHY, WILLIAM JOSEPH		<b>Cancer Activity</b> Immunology	<b>Total Project \$</b> DCB	\$290,363
<b>Start Date</b> 9/1/2004	<b>End Date</b> 08/31/2005			
<b>Science Area</b> KIDNEY		<b>Percent</b> 100.00	<b>Percent \$</b>	\$290,363

### Abstract

DESCRIPTION (provided by applicant): CD40/CD40 ligand interactions have been demonstrated to be critical co-stimulatory molecules in the generation of a successful immune response. We hypothesized that it may be possible to augment the immunostimulatory effects of CD40 stimulation by combining it with cytokine therapy. IL2 has been demonstrated to augment both adaptive and innate immune responses. Thus far, cytokine therapy has concentrated on using combinations of cytokines. We reasoned that the use of CD40 stimulation to promote dendritic cell development and function coupled with the properties of IL2 to promote T cell function would result in synergistic anti-tumor effects in vivo. Using a highly metastatic murine renal cancer model we found that strong synergistic anti-tumor effects resulted if CD40 stimulation was applied with IL2 therapy whereas no effect was observed using either agent alone. We now propose to delineate the mechanism underlying these anti-tumor effects and optimize the immunotherapeutic potential of this regimen. Toward this goal we have developed 5 specific aims: Specific Aim 1 will assess the effects of CD40 stimulation using agonist antibodies to CD40 in combination with IL2 on various metastatic renal tumor models (using both CD40+ and CD40- tumor lines) to determine the extent at which this approach is efficacious and to determine if specific immunity results upon rechallenge. Specific Aim 2 will explore the immunomodulatory effects of this regimen through the assessment of various components of both the innate (dendritic, NK) and adaptive (T and B) immune system as well as the role of cytokines. Specific Aim 3 will determine the role of CD40 on various tissues in response to this regimen through the use of CD40 +/- mice and various chimeras constructed by them. Specific Aim 4 will ascertain the efficacy of a recombinant soluble ligand for CD40 (srCD40L) to determine if similar immunomodulatory and anti-tumor effects can be obtained with this combination with IL2, particularly in light of recent data demonstrating extreme toxicities associated with antibodies to CD40 given after cytoreductive conditioning. Specific Aim 5 will then examine the effects of CD40 stimulation and IL2 in models in which cytoreductive conditioning is applied to advanced tumor-bearing mice. Both potential toxicities (which will be sought to be overcome), effects on immune reconstitution, and anti-tumor effects will be evaluated. This last model will most closely resemble advanced tumor bearing patients undergoing a debulking procedure followed with immunotherapy to remove the minimal residual disease. This proposal examining induction of costimulation with CD40 followed with IL2, should yield significant insights, not only to the efficacy of this combination approach in cancer, but also as an immunostimulatory regimen for infectious disease.

<b>Project Number</b>	<b>Title</b>			
1R01CA100787-1	Hypoxia VHL nad HIF in Renal Tumor Development			
<b>Institution</b>		<b>City / State</b>		
UNIVERSITY OF PENNSYLVANIA		PHILADELPHIA, PA		
<b>Principal Investigator</b>		<b>Program Director</b>		
HAASE, VOLKER H		Judy Mietz		
<b>Start Date</b>	<b>End Date</b>	<b>Cancer Activity</b>	<b>Total Project \$</b>	
4/1/2004	03/31/2005	Cancer Cell Biology	DCB	\$259,940
<b>Science Area</b>		<b>Percent</b>		<b>Percent \$</b>
KIDNEY		100.00		\$259,940

### Abstract

DESCRIPTION (provided by applicant): Clear cell carcinoma of the kidney (RCC), the most common form of kidney cancer, is associated with the inactivation of the von Hippel-Lindau (VHL) tumor suppressor. Mutations in the VHL gene can be found in approximately 70% of sporadic RCCs. One of the major functions of the VHL gene product, pVHL, is the targeting of the oxygen sensitive alpha-subunit of hypoxia-inducible factor (HIF) for ubiquitination and subsequent proteasomal degradation. Inactivation of VHL is felt to be an early event during RCC tumorigenesis and results in constitutive expression of two major HIF isoforms, HIF-1 and HIF-2. This results in increased transcription of genes that regulate glycolysis, angiogenesis and erythropoiesis. HIF-1 has furthermore been shown to up-regulate factors that promote growth arrest and apoptosis. The role of increased HIF expression in VHL associated tumorigenesis remains controversial. The hypothesis that the ratio of HIF-1 to HIF-2 levels is important for VHL associated renal tumor development will be investigated. Conditional gene targeting technology based on Cre-loxP mediated recombination as well as targeted transgenesis will be used to manipulate the expression levels of both pVHL and HIF in vivo and in vitro. This system enables the study of the functional relationship between VHL deficiency and HIF activation in regard to renal cell growth and viability in primary renal epithelial cells of different histogenetic origin. Specifically, the proposed investigations will examine the effects of increased or absent HIF-1 and increased HIF-2 expression in VHL deficient and wild type backgrounds. Studies will be performed in primary cell culture and in vivo with kidney specific conditional knock out mice. Gene expression studies will provide information regarding differential HIF-1 and HIF-2 target gene expression in different nephron segments with a special emphasis on genes involved in the regulation of cell survival. Taken all together, the proposed in vivo and in vitro studies will not only provide novel insights into the early events of renal oncogenesis and the histogenetic origin of RCC, but also examine fundamental aspects of HIF-1 and HIF-2 function in different nephron segments. Ultimately they have the potential to create a murine model of VHL associated renal tumors.

<b>Project Number</b>	<b>Title</b>			
5R01CA57840-10	Dendritic Cell Strategies to Tumor Reactive T Cells			
<b>Institution</b>		<b>City / State</b>		
UNIVERSITY OF PITTSBURGH AT PITTSBURGH		PITTSBURGH, PA		
<b>Principal Investigator</b>		<b>Program Director</b>		
STORKUS, WALTER J		Kevin Howcroft		
<b>Start Date</b>	<b>End Date</b>	<b>Cancer Activity</b>		<b>Total Project \$</b>
4/1/2004	03/31/2005	Immunology	DCB	\$265,245
<b>Science Area</b>		<b>Percent</b>		<b>Percent \$</b>
KIDNEY		100.00		\$265,245

### Abstract

DESCRIPTION (provided by applicant): Melanoma and renal cell carcinoma represent two of the most receptive cancer histologies to immunotherapy. While a myriad of melanoma-associated antigens have been identified and vaccines based on these antigens have been evaluated in the clinic, comparatively few tumor antigens have been characterized and specific therapies have been developed for renal cell carcinoma (RCC). We have recently identified a series of peptides that derive from RCC-associated gene products that serve as target epitopes for anti-tumor CD4+ or CD8+ T cells. Of major note, our preliminary data suggest that patients with active disease, particularly those with advanced malignancy, tend to display Th2-type CD4+ T cell responses to target epitopes, while normal donors and patients that have been treated and are disease-free at the time of analysis display either mixed "Th0"-type or predominant Th1-type CD4+ T cell responses to these same epitopes. In all of these patients, Th1-type dominated responses are observed against epitopes derived from EBV or influenza viruses. In the current proposal we will extend these preliminary studies of HLA-DR4+ patients to discern the correlation of tumor antigen-specific Th1/Th2-biased CD4+ T cell responses with disease stage, the durability of these responses in the disease-free state and the correlation of deviation in the Th1/Th2-type balance of tumor-specific CD4+ T cells with disease recurrence. Since the polarization of specific CD4+ T cell responses may also modulate the quality of anti-tumor CTL responses in situ, we will also evaluate whether patients with active disease exhibit Tc2-type dominated CD8+ T cell responses to RCC-associated epitopes and if Tc1-type reactivity is associated with better prognosis and disease free status. Lastly, we will analyze whether and to what extent, dendritic cell (DC)-based vaccines can "repolarize" Th2 (and Tc2)-type anti-tumor T cell immunity towards a Th1(Tc1)-type response pattern that may be most preferred in promoting clinical responsiveness, regression of disease, and maintenance of durable disease-free status.

<b>Project Number</b>	<b>Title</b>			
5R01CA63613-10	Genetic Determinants of Rat Renal Carcinogenesis			
<b>Institution</b>		<b>City / State</b>		
UNIVERSITY OF TEXAS MD ANDERSON CAN CTR		HOUSTON, TX		
<b>Principal Investigator</b>		<b>Program Director</b>		
WALKER, CHERYL L		Judy Mietz		
<b>Start Date</b>	<b>End Date</b>	<b>Cancer Activity</b>	<b>Total Project \$</b>	
4/1/2004	03/31/2005	Cancer Cell Biology	DCB \$319,365	
<b>Science Area</b>		<b>Percent</b>	<b>Percent \$</b>	
KIDNEY		100.00	\$319,365	

#### Abstract

DESCRIPTION (provided by applicant): Eker rats are susceptible to spontaneous and carcinogen-induced renal cell carcinoma (RCC) due to a germline defect in the tuberous sclerosis 2 (TSC-2) tumor suppressor gene. These animals serve as useful models for studying mechanisms of renal carcinogenesis and the role of the TSC-2 gene in this process. While it is well established that loss of TSC-2 gene function contributes to tumor development in rodents and humans, the normal function(s) of tuberin, the product of the TSC-2 gene, and the mechanism by which loss of function contributes to tumorigenesis are not clear. Recently, data have emerged implicating tuberin as a negative regulator of signaling downstream of AKT. In this renewal application, we will test the hypothesis that tuberin is itself a target for AKT phosphorylation, and that this phosphorylation in conjunction with a PDZ binding domain at tuberin's carboxy terminus regulates tuberin's binding partners, subcellular localization and function within the cell. In Specific Aim 1 we will test the hypothesis that tuberin is a target for AKT phosphorylation. In Specific Aim 2 we will identify tuberin binding partners with the potential to exercise spatial control of tuberin function. In Specific Aim 3 we will determine the functional consequences of tuberin interaction with 14-3-3 or its PDZ binding partner nNOS on cell signaling downstream of AKT, RHO-mediated cell adhesion and nNOS activity. Studies such as these on the normal function of tuberin have the potential to reveal new interactions between tuberin and cell signaling pathways that can yield valuable information regarding how the TSC-2 suppressor gene participates in renal carcinogenesis.

<b>Project Number</b> 5R01CA90966-3	<b>Title</b> Biobehavioral Effects of Emotional Expression in Cancer	<b>City / State</b> HOUSTON, TX		
<b>Institution</b> UNIVERSITY OF TEXAS MD ANDERSON CAN CTR		<b>Program Director</b> Wendy Nelson		
<b>Principal Investigator</b> COHEN, LORENZO		<b>Cancer Activity</b> Basic Bio-Behavioral Research	DCCPS	<b>Total Project \$</b> \$471,684
<b>Start Date</b> 3/23/2004	<b>End Date</b> 01/31/2005			
<b>Science Area</b> KIDNEY		<b>Percent</b> 100.00		<b>Percent \$</b> \$471,684

**Abstract**

Models of cognitive processing suggest that once a traumatic event is appropriately understood and integrated the stress associated with the event will diminish. Thoughts and feelings surrounding a traumatic experience are often disorganized, yet when disclosed verbally or through writing, they can assume the form of an organized, coherent narrative resulting in improved health outcomes. This is illustrated by recent findings that indicated that a brief written emotional expression exercise was associated with improved physical health, psychological well-being, physiological functioning, and general functioning. This writing exercise was also associated with beneficial changes in immune function. The brief writing intervention is hypothesized to increase cognitive processing and foster adaptation to traumatic events. To date, however, most research examining this intervention has been conducted in healthy populations. The diagnosis and treatment of cancer are traumatic experiences associated with distress and the fear of cancer recurrence, progression, and death. The impact of stress on the immune system may be particularly detrimental to patients with renal cell cancer, as this cancer is immunogenic, meaning that the immune system regulates progression of the disease. Because emotional expression writing interventions have been shown to facilitate adaptation, reduce stress, improve psychological adjustment and QOL, and positively impact immune function, this type of intervention may be beneficial in patients with renal cancer. Pilot data from our laboratory suggest that it is feasible to conduct the emotional expression writing intervention in patients with renal cancer. Results from this study also provide initial evidence that the intervention increases cognitive processing and improves psychological well-being. The proposed study will assess the benefits of this written emotional expression exercise in patients with renal cell carcinoma. Patients in this study will be randomly assigned either to an emotional expression writing group or to a neutral writing group. This research will also evaluate the extent to which psychosocial factors mediate or moderate the effects of the intervention program and predict recovery and adjustment. The effects of the intervention should be evident throughout recovery and across indices of quality of life, mental health, subjective symptoms of stress, and immune function.

<b>Project Number</b>	<b>Title</b>			
5R01CA98897-2	Genetic Susceptibility Modifiers in Renal Cell Carcinoma			
<b>Institution</b>		<b>City / State</b>		
UNIVERSITY OF TEXAS MD ANDERSON CAN CTR		HOUSTON, TX		
<b>Principal Investigator</b>		<b>Program Director</b>		
WU, XIFENG		Kumi Iwamoto		
<b>Start Date</b>	<b>End Date</b>	<b>Cancer Activity</b>		<b>Total Project \$</b>
8/1/2004	07/31/2005	Epidemiology	DCCPS	\$511,396
<b>Science Area</b>		<b>Percent</b>		<b>Percent \$</b>
KIDNEY		100.00		\$511,396

## Abstract

DESCRIPTION (provided by applicant): This proposal is designed to build upon the epidemiologic and molecular genetic findings from our research on tobacco-related epithelial cancers. Tobacco exposure is an established risk factor for renal cell carcinoma (RCC). However, there are limited data on susceptibility markers and epidemiologic profiles for RCC. We therefore propose to use a molecular epidemiologic approach in a case-control study to identify interindividual differences in inherited genetic instability, focusing on assessing DNA damage/repair, telomere length, and telomerase activity as predictors of RCC risk. In addition, deletions involving 3p are the most common genetic alterations in RCC, we will also study 3p latent genetic instability. We will accrue 300 patients with RCC, who have not received chemotherapy or radiotherapy, identified from two hospitals in the Houston, Texas metropolitan area. We will also recruit 300 controls identified from population-based random digit dialing in the Houston metropolitan area. The controls will be matched to the patients by sex, age (+/- 5 years), and ethnicity. Comprehensive epidemiologic profiles will be constructed for these patients and controls. Specific goals of this project are: 1): To assess two mutagen sensitivity or DNA repair assays performed in parallel and measured by Comet 4.0 image system in patients and controls. One assay quantifies gamma-radiation-induced lymphocytic tail moment reflecting base excision repair (BER) and double strand break (DSB)/recombination repair; the other assay quantifies benzo[a]pyrene diol-epoxide (BPDE)-induced lymphocytic tail moment reflecting nucleotide excision repair (NER). Our hypothesis is that subjects who show increased gamma-radiation and BPDE sensitivity are at greater risk for RCC than are those who do not show these sensitivities. 2): To determine the telomere length in peripheral blood lymphocytes (PBLs) at baseline in patients and controls. Our hypothesis is that individuals susceptible to RCC will have shorter telomere length at baseline compared with normal individuals and that telomere length at baseline might be inversely correlated with gamma-radiation sensitivity. 3): To determine the levels of telomerase activity in PBLs at baseline and after gamma-radiation treated of patients and controls. Our hypothesis is that upon exposure to gamma-radiation, individuals susceptible to RCC will have higher telomerase activity compared with healthy individuals. 4): To determine the frequencies of BPDE-induced chromosome aberrations on 3p12.3, 3p14.2, 3p21.3, and 3p25.2 in PBLs of patients and controls. 3p is the most frequently reported abnormal region in RCC. Our hypothesis is that cases exhibit higher frequencies of BPDE-induced 3p aberrations in PBLs than do controls. These chromosomal loci may reflect genetic susceptibility of specific loci to BPDE and that individuals with aberrations at these loci are at increased risk for RCC. We also plan to conduct a substudy as a secondary aim to perform the loss of heterozygosity (LOH) analysis on 3p on the corresponding tumor tissue from Specific Aim 4. We hypothesize that there will be concordance in the severity of site-specific chromosomal lesions in target and surrogate tissues. This study will further the understanding of the genetic events leading to the development of RCC; explore the genetic basis for genetic instability and how it affects cancer risk; and eventually provide a means of identifying a subgroup who are most likely to develop RCC. Such individuals may then be targeted for intervention programs such as chemoprevention or dietary modification.

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<b>Project Number</b>	<b>Title</b>			
5R01CA61889-11	CANCER SUSCEPTIBILITY IN AN ANIMAL MODEL			
<b>Institution</b>		<b>City / State</b>		
UNIVERSITY OF WASHINGTON		SEATTLE, WA		
<b>Principal Investigator</b>		<b>Program Director</b>		
YEUNG, RAYMOND		Judy Mietz		
<b>Start Date</b>	<b>End Date</b>	<b>Cancer Activity</b>	<b>Total Project \$</b>	
1/1/2004	12/31/2004	Cancer Cell Biology	DCB	\$346,431
<b>Science Area</b>		<b>Percent</b>		<b>Percent \$</b>
KIDNEY		100.00		\$346,431

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**Abstract**

For any given cancer, there exists a wide spectrum of clinical behavior. Understanding the factors that influence tumors approach to risk assessment and the design of therapy. Inherited variation of certain genes and their interaction with the environment are thought to govern, to a large extent, tumor heterogeneity, both in terms of susceptibility and progression. This proposal makes use of hereditary cancer model in the Eker rat and a transgenic TSC2-mutant mouse model to identify post- initiation events that modulate kidney cancer phenotype. Both strains carry a germline mutation of the TSC2 gene that in humans, also predisposes the carriers to renal cell carcinoma as part of the tuberous sclerosis complex (TSC). The clinical features of the latter disease are highly variable even among family members sharing the same mutation. Those with mild forms of TSC live a normal life whereas those severely affected die prematurely of brain and kidney complications. Similarly, patients with low tumor burden fare better than those with heavy tumor load. In an attempt to understand the genetic determinants of phenotypic variation, observations in the Eker rat suggested a strain-dependent difference in renal tumor size at 18 months of age. Using the tools of quantitative biology, the investigators have demonstrated evidence for genetic factors governing tumor size that are unlinked to the TSC2 gene. The aims of this proposal are to determine the molecular identity of a dominant modifier of tumor burden and to detect and map other genetic elements that affect its phenotype (e.g., size, multiplicity, metastasis) in the two rodent models. Appreciation of these 'modifier' genes will impart new knowledge about the interplay between genes and environment during the development of complex diseases such as cancer.

<b>Project Number</b> 5R01CA92542-4	<b>Title</b> Modulation of retinoic acid action in renal cancer	<b>City / State</b> NEW YORK, NY	<b>Total Project \$</b> \$351,713
<b>Institution</b> WEILL MEDICAL COLLEGE OF CORNELL UNIV	<b>Program Director</b> Heng Xie		
<b>Principal Investigator</b> NANUS, DAVID M.	<b>Cancer Activity</b> Clinical Oncology		
<b>Start Date</b> 8/1/2004	<b>End Date</b> 07/31/2005		
<b>Science Area</b> KIDNEY	<b>Percent</b> 100.00		<b>Percent \$</b> \$351,713

**Abstract**

DESCRIPTION: (Provided by applicant) Renal cell carcinoma is the most common primary cancer arising from the kidney in adults, and is a frequent cause of cancer morbidity and mortality in the U.S., with over 12,000 deaths per year. Currently, there are no consistently effective chemotherapeutic or biologic treatment modalities for patients with advanced disease. Recent results of clinical trials in patients with advanced renal cancer (RC), and of pre-clinical studies using cell culture and human RC tissue specimens, suggest that natural and synthetic derivatives of vitamin A (retinol), a group of compounds called retinoids, play a role in the therapy of RC. We have preliminary data that intracellular levels of retinoic acid (RA) are significantly diminished in human RC cells and that retinol metabolism is aberrant in RC cells as compared to normal human kidney proximal tubule cells. Furthermore, our data indicate that combining retinoids with agents which augment retinoid actions, such as IFN (IFN) or histone deacetylase (HDAC) inhibitors, significantly increases the anti-tumor effect of RA. In this grant application, we propose to study the effects of modulating retinoid anti-tumor action by combining RA with other therapies. The specific aims are: 1) to perform a series of Phase I-II clinical trials designed to evaluate the safety and efficacy of combining liposomal all trans RA (ATRA) with modulators of RA such as interferon and HDAC inhibitors in patients with metastatic RC; 2) to collect peripheral blood samples and tumor tissues to allow laboratory analysis in order to monitor the presence and magnitude of specific therapies on retinoid metabolites and retinoid related genes; and 3) to analyze modulators of RA such as interferon and HDAC inhibitors at a molecular level in RC cell lines and xenograft models to delineate mechanisms of action, and to use this information to design strategies to test specific drugs in combination with RA in preparation for future clinical trials. These aims will allow us to perform clinical trials and laboratory studies aimed at gaining a better understanding of the involvement of retinoids in the development, progression and therapy of RC. Moreover, the experiments proposed in this application will help to clarify the potential use of retinoids as a therapeutic strategy to treat patients with RC, and may lead to the identification of new therapeutic agents which result in increased retinol actions for the treatment of various stages of renal cancer.

<b>Project Number</b> 5R01CA85412-3	<b>Title</b> Functional Analysis of the VHL Tumor Suppressor Gene			
<b>Institution</b> YESHIVA UNIVERSITY		<b>City / State</b> NEW YORK, NY		
<b>Principal Investigator</b> BURK, ROBERT D.		<b>Program Director</b> Grace Ault		
<b>Start Date</b> 2/1/2004	<b>End Date</b> 01/31/2005	<b>Cancer Activity</b> Tumor Biology	<b>Total Project \$</b> DCB	\$296,131
<b>Science Area</b> KIDNEY		<b>Percent</b> 100.00	<b>Percent \$</b>	\$296,131

#### Abstract

DESCRIPTION: (provided by applicant) Approximately 30,000 Americans will develop renal cancer this year and nearly 12,000 will die from the disease. Mutation and/or inactivation of the von Hippel Lindau (VHL) gene occurs as an early, initiating event, promoting the development of most renal cell carcinoma (RCC). We have demonstrated that the binding of Elongins B & C protects VHL proteins from proteasomal degradation, in a manner similar to SOCS (suppressor of cytokine signaling) protein/elongin complexes. Expression of exogenous VHL gene product(s) in a renal cancer cell line (lacking wild type VHL) influences the growth and differentiation properties of confluent cells grown on extracellular matrix (ECM). In addition, VHL protects these cells from apoptosis. In accord with the function of VHL to inhibit apoptosis and suppress stress-related signaling pathways (e.g., hypoxia), our observation that VHL utilizes internal translation to synthesize a functional protein suggests a mechanism to assure adequate levels of VHL protein under conditions of suppressed cap-dependent translation. Taken together, these findings suggest the hypothesis that VHL may function as a tumor suppressor by suppression of stress signals (SOSS) through modification (e.g., ubiquitination) of key regulators of pathways involved in cell-cell, cell-ECM and stress-related signaling. The aims of this project are: (1) To determine the mechanism of VHL-mediated protection from apoptosis by assaying specific regulators of apoptosis common to UV irradiation, serum starvation, and glucose deprivation. We will also evaluate the pro-apoptotic potential of specific mutations in VHL and investigate whether these mutations drive apoptosis and provide selective pressure toward cells that are able to escape apoptosis; (2) To investigate the mechanism and functional importance of translational and post-translational regulation of VHL; (3) To identify potential targets of VHL activity, VHL-associating proteins will be compared under conditions of cell stress, low and high cell density and on plastic and ECM.

<b>Project Number</b>	<b>Title</b>			
1R21CA99069-1	Real-Time Imaging of Hypoxia based on VHL Activity			
<b>Institution</b>		<b>City / State</b>		
DANA-FARBER CANCER INSTITUTE		BOSTON, MA		
<b>Principal Investigator</b>		<b>Program Director</b>		
KAELIN, WILLIAM G		Houston Baker		
<b>Start Date</b>	<b>End Date</b>	<b>Cancer Activity</b>	<b>Total Project \$</b>	
6/10/2004	04/30/2005	Diagnostic Imaging	DCTD	\$186,788
<b>Science Area</b>		<b>Percent</b>		<b>Percent \$</b>
KIDNEY		75.00		\$140,091

## Abstract

DESCRIPTION (provided by applicant):

Von Hippel-Lindau (VHL) disease is a hereditary cancer syndrome caused by germline mutation of the VHL tumor suppressor gene. The VHL gene product, pVHL, is part of a multiprotein complex that polyubiquitinates the alpha subunits of the heterodimeric transcription factor called HIF (hypoxia-inducible factor) in the presence of oxygen, which results in their destruction by the proteasome. pVHL binds directly to a colinear 20 amino acid residue sequence located within a region of HIF called the Oxygen-Dependent Degradation Domain (ODD). This interaction is strictly regulated by oxygen-dependent, enzymatic hydroxylation of a conserved proline residue within this HIF 20 mer. I fused this 20 mer to firefly luciferase (ODD-Luciferase). In pilot experiments I established that ODD-luciferase binds to pVHL in vitro in a hydroxylation-dependent manner. In transfection experiments the ODD-luciferase chimera, but not wild-type luciferase, was induced following treatment with hypoxia mimetics. Finally, my preliminary data with tumor xenografts indicate that ODD-luciferase activity can be detected with a Xenogen camera in living animals and is restricted to the central regions of tumors, which are known to be hypoxic. I propose to use ODD-luciferase or an analogous fusion protein, ODD-GFP, as oxygen biosensors. Using these biosensors, I plan to develop cell based assays that might be used to identify compounds that either directly or indirectly disrupt the VHL/HIF interaction. Furthermore, I will develop a transgenic mouse expressing ODD-luciferase. Such mice could be used to study diseases characterized by the presence of hypoxic cells, such as cancer, and could also be used to monitor pharmacodynamic effects of agents that alter tissue oxygenation and/or modulate HIF.

<b>Project Number</b>	<b>Title</b>			
IR01CA112533-1	Enhanced RF Tumor Ablation with Liposomal Chemotherapy			
<b>Institution</b>		<b>City / State</b>		
BETH ISRAEL DEACONESS MEDICAL CENTER		BOSTON, MA		
<b>Principal Investigator</b>		<b>Program Director</b>		
GOLDBERG, S NAHUM		Keyvan Farahani		
<b>Start Date</b>	<b>End Date</b>	<b>Cancer Activity</b>	<b>Total Project \$</b>	
8/1/2004	07/31/2005	Diagnostic Imaging	DCTD	\$410,762
<b>Science Area</b>		<b>Percent</b>	<b>Percent \$</b>	
KIDNEY		50.00		\$205,381

### Abstract

DESCRIPTION (provided by applicant): Image-guided radiofrequency (RF) tumor ablation is being used to treat focal liver, renal, and other tumors. Yet, complete ablation is often difficult to achieve, particularly at tumor margins, presenting substantial barriers toward clinical efficacy. Our preliminary studies take advantage of complementary interactions between RF ablation and liposomal doxorubicin, and demonstrate that this combination may markedly increase tumor destruction in animals and patients, and animal survival compared to RF alone or RF combined with free doxorubicin. RF ablation also increased intratumoral liposomal doxorubicin 7 fold compared to controls, particularly in a peripheral region immediately adjacent to the zone of RF induced coagulation. Thus, this combined treatment paradigm has the unique potential both to potentiate preferential delivery of cytotoxic agents in liposomal vehicles, and to maximize the completeness of ablation of a treated tumor, in humans. Our immediate goals are to address several key issues that require further knowledge prior to initiation of Phase II or larger multicenter clinical trials. 1) Our data indicate that gains in both drug uptake and tumor destruction can be further increased through the systematic optimization of the relevant treatment parameters. 2) The mechanisms driving this interaction particularly the hyperthermic dosimetry of increased tissue destruction and the causes for increased tumor destruction seen when empty liposomes are given without doxorubicin need to be better understood, as this could lead to developing clinical strategies having even greater efficacy. 3) Local and systemic toxicities need to be better defined. This will be accomplished over 3 aims. 1) We will improve tumor destruction of combined therapy using simplex optimization in canine venereal sarcoma. The relationship between intratumoral thermal dosimetry to increased doxorubicin uptake and increased tumor destruction will be characterized to elucidate the role of hyperthermia, threshold thermal dosage, and the conditions under which combined therapy is most effective. 2) We will test specific hypotheses about the possible role of the liposome carrier components. RF and empty liposomes will be studied to define the relative contributions of the lipids and of the doxorubicin to the mechanisms of cell killing. We will further determine whether increased tumor destruction can be ascribed to enhanced oxidative stress and lipid peroxidation and whether manipulation of the substrate upon which lipid peroxidation occurs can further potentiate the antitumor effect of thermal ablation. 3) We will characterize potential local and systemic treatment toxicity including determining the thermal dosimetry at which tissue effects are seen in normal canine liver and kidney and in a one-month survival study in renal tumors.

<b>Project Number</b> 5R21CA101629-2	<b>Title</b> Innovative Toxicology Models for Drug Evaluation	<b>City / State</b> DUARTE, CA		
<b>Institution</b> CITY OF HOPE/BECKMAN RESEARCH INSTITUTE		<b>Program Director</b> George Johnson		
<b>Principal Investigator</b> YEE, JIING-KUAN		<b>Cancer Activity</b> Biochemistry and Pharmacology		
<b>Start Date</b> 6/1/2004	<b>End Date</b> 05/31/2005		<b>Total Project \$</b> DCTD	\$175,000
<b>Science Area</b> KIDNEY		<b>Percent</b> 50.00	<b>Percent \$</b>	\$87,500

**Abstract**

DESCRIPTION (provided by applicant): A. Specific Aims: The overall goal of this proposal is to use reversible immortalization approach to establish cell lines derived from primary human renal proximal tubule epithelial cells (RPTECs) and hepatocytes. Liver and kidney are two critical organs for drug metabolism and excretion, and are frequently affected by drug-induced toxicity. Currently, no suitable human kidney cell line is available for drug screening in the early stages of drug discovery and development. While primary human hepatocytes are available, the high cost to culture such cells precludes their use in the drug screening process. If immortalized cell lines that are phenotypically stable and retain all or most of the primary cell functions can be established, they can be used for high-throughput screening of potential drugs. There are three specific aims in this proposal. I. Characterization of TH1 and TH7 cells for kidney functions: In our preliminary study, we used human immunodeficiency virus (HIV)-based vectors containing the cDNAs encoding the SV40 T antigen (Tag) and the catalytic subunit of human telomerase (hTERT) to establish several immortalized cell lines derived from primary human RPTECs. We propose to characterize the biochemical and functional activities in two of these cell lines, TH1 and TH7. We will determine the basal expression and the activity of xenobiotic metabolizing enzymes in these two cell lines. We will evaluate the activity of drug transport proteins, drug uptake and efflux. We also propose to use Cre-LoxP mediated recombination system to remove the introduced Tag and the hTERT cDNAs from these two cell lines and examine its effect on cell proliferation, differentiation and biochemical functions. II. Establishment and characterization of immortalized human hepatocytes: We propose to use the same strategy as described in specific aim I to establish and characterize immortalized human hepatocytes. III. Evaluation of drug toxicity in the immortalized cell lines: We will determine the drug sensitivity of the established cell lines, including methotrexate (MTX), cisplatin, cyclophosphamide and paclitaxel. The toxicity induced by these drugs in primary human cells and in cell lines derived from hepatocytes and RPTECs will also be examined and the results will be compared with those from the immortalized cell lines established in this current study.

<b>Project Number</b> 7R01CA89344-4	<b>Title</b> Clinical and Cellular Effects of Interferon alfa 1	<b>City / State</b> CLEVELAND, OH		
<b>Institution</b> CLEVELAND CLINIC LERNER COL/MED-CWRU		<b>Program Director</b> Heng Xie		
<b>Principal Investigator</b> BORDEN, ERNEST C.		<b>Cancer Activity</b> Clinical Oncology	<b>Total Project \$</b> DCTD	\$317,475
<b>Start Date</b> 9/19/2004	<b>End Date</b> 06/30/2005			
<b>Science Area</b> KIDNEY		<b>Percent</b> 50.00	<b>Percent \$</b>	\$158,738

### Abstract

Using a preparation that was in limited supply, we previously assessed Interferon-alpha 1 (IFN-alpha 1) clinically in a randomized, double blind comparison to IFN-alpha 2. The frequency of side effects was much less with IFN-alpha1 (p less than 0,01). Yet effects of IFN-alpha 1 on granulocyte counts, NK cell cytotoxicity, and an ISG were comparable to IFN-alpha2. These findings suggest that IFN-alpha 1 might be given with better tolerance and or higher doses than IFN-alpha 2. Recombinant DNA produced IFN-alpha 1 has now come available in adequate supply through a collaboration with the Ministry of Public Health in Shanghai. In randomized, multi-center clinical trials in China, IFN-alpha 1 has proven effective for chronic hepatitis B and C. Data from China also suggests IFN-alpha 1 is better tolerated than IFN-alpha 2. IFN-alpha 1, together with IFN-alpha 2, is a predominant IFN-alpha species generally characteristic of an IFN. However, IFN-alpha 1 has differing receptor binding affinities and differing cross species antiviral activity differences in the transcription factor complexes activated in response to IFN-alpha 1 as compared to IFN-alpha 2. In addition to expected IFN antiproliferative and gene stimulatory (ISG) effects, we have also identified an increase in STAT1 protein expression and induction of apoptosis in myeloma cells by IFN-alpha 1. Studies of IFN-alpha have been designed with the following goals: a) confirm good clinical tolerance and define the dose response characteristics of ISG induction in patients, b) compare the signal transducing and gene modulatory activity to those of IFN-alpha 2 by oligonucleotide gene array and assessment of transcription activating factors, c) study apoptosis and gene induction in myeloma and renal carcinoma to confirm clinical antitumor activity. Since expanded clinical use of IFN- alpha 2 has increasingly been limited by side effects, IFN-alpha 1 may be particularly important in extending clinical leads provided by LFN-alpha 2. Finally, results will further confirm the postulate that antiviral specific activity in vitro does not predict for other biological and clinical effects of IFN. In addition, if the pattern of novel gene induction and cellular effects are expanded, IFN-alpha 1 could have a broader spectrum of responsive clinical tumor types.

<b>Project Number</b>	<b>Title</b>			
1R01CA102309-1	The MiT Transcription Factor in Pediatric Malignancies			
<b>Institution</b>		<b>City / State</b>		
DANA-FARBER CANCER INSTITUTE		BOSTON, MA		
<b>Principal Investigator</b>		<b>Program Director</b>		
FISHER, DAVID E		Judy Mietz		
<b>Start Date</b>	<b>End Date</b>	<b>Cancer Activity</b>	<b>Total Project \$</b>	
4/1/2004	03/31/2005	DNA Chromosome Aberrations	DCB	\$315,495
<b>Science Area</b>		<b>Percent</b>		<b>Percent \$</b>
KIDNEY		50.00		\$157,748

### Abstract

DESCRIPTION (provided by applicant): The MiT transcription factor family contains four bHLHZip proteins which are critical for development of several cell lineages and exhibit overlapping DNA recognition with the Myc family. One member, Miff, is regulated by Melanocyte Stimulating Hormone (MSH) to stimulate pigment cell growth and differentiation. Another, TFE3, modulates osteoclast development together with Mitf. We recently cloned a recurring translocation in Papillary Renal Cell Carcinoma (PRCC), and identified the MiT family member TFEB as a new fusion oncogene, which places full length TFEB under the transcriptional regulation of a ubiquitous and abundant gene of unknown function reminiscent of c-Myc in Burkitt's lymphoma. Translocations which fuse another MiT factor, TFE3, also occur in PRCC as well as Alveolar Soft Part Sarcomas. The oncogenicity of transcriptionally dysregulated MiT factors sparked investigation of Clear Cell Sarcoma (CCS), an EWS-ATF1 translocated tumor which inexplicably expresses melanoma markers. EWS-ATF1 was seen to constitutively upregulate Miff via mimicking Melanocyte Stimulating Hormone signaling in melanocytes. Dysregulated Mitf, in turn, drives CCS growth and survival. All known MiT-dysregulated tumors are uniformly resistant to conventional chemotherapy. This proposal examines this growing family of malignancies to dissect the pathways through which they transform and identify diagnostic markers and therapeutic targets using this information. In Specific Aim 1 we employ disruption-rescue assays we developed to determine structural and post-translation requirements of MiT factors for specific oncogenic behaviors. In Specific Aim 2 we ask whether the overlapping DNA recognition properties of the MiT and Myc families reflect overlapping transformation mechanisms. Specific Aim 3 uses candidate- and microarray approaches to identify transcriptional targets of the MiT family in these cancers, and scrutinizes their functional importance in tumorigenicity. One such newly identified MiT target gene is c-MET, a finding of particular interest because human germline c-MET mutations produce hereditary PRCC-- the same malignancy in which TFEB or TFE3 translocations occur for sporadic tumors, c-MET was seen to be superactivated in all 6 MiT-associated tumor lines examined, and will be studied in primary tumors. Small molecule inhibitors of c-MET exist and will be examined (together with molecular controls) as potential targeted therapy for these incurable cancers.

<b>Project Number</b> 5R37CA54358-14	<b>Title</b> BWS and Embryonal Tumor Suppressor Genes on 11p15	<b>City / State</b> BALTIMORE, MD	
<b>Institution</b> JOHNS HOPKINS UNIVERSITY		<b>Program Director</b> Paul Okano	
<b>Principal Investigator</b> FEINBERG, ANDREW P		<b>Cancer Activity</b> DNA Chromosome Aberrations	<b>Total Project \$</b> DCB \$405,419
<b>Start Date</b> 5/1/2004	<b>End Date</b> 04/30/2005		
<b>Science Area</b> KIDNEY		<b>Percent</b> 50.00	<b>Percent \$</b> \$202,710

### Abstract

DESCRIPTION: Beckwith-Wiedemann Syndrome (BWS) causes prenatal overgrowth, midline birth defects, and a wide variety of embryonal tumors. Our laboratory previously mapped BWS to 11p15 by genetic linkage analysis and also demonstrated frequent loss of heterozygosity (LOH) of the same region in embryonal tumors. In the past grant period, in order to identify the genes involved in BWS and LOH, we molecularly cloned genes within and surrounding a cluster of balanced germline chromosomal rearrangement breakpoints from BWS patients termed BWSCRJ. Surprisingly, within this region we identified at least 8 genes which are imprinted, i.e., show preferential expression of a specific parental allele. Several of these genes, which span 1 Mb of 11p15, show genetic or epigenetic alterations in BWS patients. These include p57/KIP2, KvLQT1, H19, IGF2, and LIT1, a novel antisense orientation untranslated RNA that we found is within, and imprinted and transcribed oppositely to KvLQT1. This multigene domain was itself divided into two separate imprinted subdomains, with nonimprinted genes between them. Genetic complementation experiments mapped an embryonal tumor suppressor gene to this nonimprinted interval, although rare mutations were also found as well in a novel imprinted gene, TSSC5. Based on our identification of specific genetic alterations that cause BWS, we will now determine the relationship between genotype and phenotype in BWS, and the genetics of transmission of BWS in families. With the assistance of the MIT/Whitehead Genome Center, we will obtain sequence of the entire 1.2 Mb homologous region in mouse, and identify the conserved genes, CpG islands, and other potential intergenic regulatory elements within it. We will determine the functional role of these sequences in normal cells, as well as alterations in BWS patients, including those who appear to show altered imprinting affecting the entire imprinted gene domain. We will identify the gene(s) that suppress the growth of embryonal tumors and explore the mechanism of their alteration, including the possibility that aberrant imprinting leads to inactivation of one copy. Finally, we will determine the normal function of these genes and regulatory sequences using transgenic mice. These studies should continue to provide novel insights into the role of these genes in birth defects and cancer, as well as an exciting species comparative approach to understanding the regulation of multiple genes within a large imprinted domain.

<b>Project Number</b> 5R01CA59998-11	<b>Title</b> SPROUTY, A WT1 TARGET FOR GROWTH AND DEVELOPMENT		
<b>Institution</b> MOUNT SINAI SCHOOL OF MEDICINE OF NYU	<b>City / State</b> NEW YORK, NY		
<b>Principal Investigator</b> LICHT, JONATHAN D.	<b>Program Director</b> Judy Mietz		
<b>Start Date</b> 1/1/2004	<b>End Date</b> 12/31/2004	<b>Cancer Activity</b> Cancer Cell Biology	<b>Total Project \$</b> DCB \$322,262
<b>Science Area</b> KIDNEY		<b>Percent</b> 50.00	<b>Percent \$</b> \$161,131

#### Abstract

DESCRIPTION: (Adapted from the applicant's abstract) In this proposal we will characterize the expression of sprouty in renal development, murine development in general and in Wilm's tumors. We will characterize the mechanism of action of sprouty. Hence we will determine if sprouty is a growth inhibitor which may mediate some of the effects of WT1. We will determine at which step in signaling through receptor tyrosine kinase sprouty acts. We then isolate partner proteins of spry which will lead to better idea of the molecular mechanism of sprouty. In order to determine the role of spry in normal and aberrant development we will determine in cell culture models and transgenic animals if engineered expression of spry interferes with normal renal morphogenesis. Finally to provide a critical role of sprouty as a target of WT1 important for renal development we will create knockout animals for sprouty one using conditional cre-lox technology. Through these studies we will characterize an exciting molecule involved in signal transduction and development. Spry may present a target for future strategies against Wilm's tumor, other malignancies and renal disorder such as polycystic kidney disease.

<b>Project Number</b>	<b>Title</b>			
5R03CA106006-2	Discovery of Genotoxic Biomarkers in Urine for Cancer			
<b>Institution</b>		<b>City / State</b>		
NORTHEASTERN UNIVERSITY		BOSTON, MA		
<b>Principal Investigator</b>		<b>Program Director</b>		
GIESE, ROGER W		Kumi Iwamoto		
<b>Start Date</b>	<b>End Date</b>	<b>Cancer Activity</b>		<b>Total Project \$</b>
9/1/2004	08/31/2005	Epidemiology	DCCPS	\$78,625
<b>Science Area</b>		<b>Percent</b>		<b>Percent \$</b>
KIDNEY		50.00		\$39,313

### Abstract

DESCRIPTION (provided by applicant): This project seeks to develop a new methodological procedure that has the potential for improving the quality of cancer epidemiological research. The new procedure involves the use of mass spectrometry (MS) to analyze DNA adducts inherent to or derived from urine. The "inherent DNA adducts" will be analyzed for two purposes: (1) as potential biomarkers for human exposure to carcinogens, and (2) as potential biomarkers for the presence and tissue location of cancer. This latter hypothesis is suggested by bringing together the following observations by others: (1) each tissue has a unique pattern of lipophilic, apparently endogenous DNA adducts (as detected by 32P-post-labeling/HPLC); (2) some tissue DNA is spilled into the blood (increasingly so in cancer), and (3) some of the free DNA in the blood ends up in the urine. The "derived DNA adducts" will be obtained by following a published method in which genotoxic chemicals are extracted from smoker's urine and reacted in the presence of an S9 metabolic activation system with calf thymus DNA. Previous investigators have considered the genotoxic chemicals in smoker's urine to be the same as the mutagens, which are present. Detection in this project of the three types of DNA adducts (inherent/exogenous, inherent/endogenous and derived) will be accomplished by using a method that we have recently developed in which the adducts, after isolation, are labeled with an imidazole substituted mass tag followed by use of matrix assisted laser desorption ionization time-of-flight mass spectrometry (MALDI-TOFMS). Our new technique is very sensitive and measures the exact masses of a broad range of known and unknown DNA adducts simultaneously, unlike any prior analytical method for DNA adducts. Along with urine samples from control subjects and smokers, urine from patients with renal and bladder cancer will be tested. Urine is an attractive sample for epidemiological studies. The project initiates a collaboration between an analytical chemist and a molecular biologist.

<b>Project Number</b> 5R01CA95785-3	<b>Title</b> ASPL TFE3 fusion in human cancers	<b>City / State</b> NEW YORK, NY	
<b>Institution</b> SLOAN-KETTERING INSTITUTE FOR CANCER RES		<b>Program Director</b> Judy Mietz	
<b>Principal Investigator</b> LADANYI, MARC		<b>Cancer Activity</b> Cancer Cell Biology	<b>Total Project \$</b> DCB \$282,130
<b>Start Date</b> 4/1/2004	<b>End Date</b> 03/31/2005		
<b>Science Area</b> KIDNEY		<b>Percent</b> 50.00	<b>Percent \$</b> \$141,065

### Abstract

**DESCRIPTION:** (provided by applicant) The novel ASPL-TFE3 fusion arises from a t(X;17)(q25.3;p11.2) in two distinct human cancers, alveolar soft part sarcoma (ASPS), a lethal sarcoma of uncertain lineage, and a unique subset of pediatric renal adenocarcinomas. In these two tumor types, the gene fusions occur respectively through an unbalanced or balanced t(X;17). ASPL is a novel ubiquitously expressed gene that encodes a protein with no recognizable complex motifs, but is similar to unknown predicted proteins in several model organisms. The ASPL-TFE3 fusion replaces the N-terminal portion of TFE3 by the fused ASPL sequences, while retaining the DNA-binding domain of the TFE3 transcription factor, implicating transcriptional deregulation in the pathogenesis of this tumor. Preliminary transactivation and subcellular localization data support the function of ASPL-TFE3 as a transcription factor. ASPL-TFE3 is of special interest as the only chimeric transcription factor associated with malignancies of both mesenchymal and epithelial derivation. The overall goal of this proposal is to extend the understanding of the biology of these genes and tumors through functional studies, gene expression profiling, and molecular genetic analyses. In Aim 1, the investigators will map the putative ASPL activation domain and perform a transactivation analysis of the reciprocal TFE3-ASPL product; examine transactivation of reporters driven by portions of endogenous TFE3 target promoters where cooperative interactions with other transcription factors are critical; and analyze specific ASPL-TFE3 protein-protein interactions. In Aim 2, they will use microarray hybridizations to examine genes induced by TFE3 and ASPL-TFE3 in heterologous human mesenchymal and renal cell lines stably transfected with tetracycline-regulated TFE3 and ASPL-TFE3 constructs, and to establish and compare expression profiles of ASPs and pediatric renal adenocarcinomas containing ASPL-TFE3, and other major types of renal adenocarcinomas. Finally, in Aim 3, to address the hypothesis that the unbalanced structure of the t(X;17) in ASPs is driven by a growth advantage conferred to ASPs cells by the functional loss of a gene on 17q25.3 telomeric to ASFL, the investigators will examine polymorphic loci in Xp11.2->qter in ASPs from women to establish whether the t(X;17) of ASPs forms only in G2 or shows no intrinsic cell cycle preference; close sequence gaps in the ASFL-containing BAC and orient and order the sequence fragments, to determine which genes are telomeric to ASPL; analyze exceptional ASPs cases with a reciprocal t(X;17) for smaller deletions of 17q25.3->qter; and finally analyze selected candidate genes in the same region for inactivating mutations.

<b>Project Number</b>	<b>Title</b>		
5R01CA76035-6	Molecular-Genetic Analysis of 3p14 Genomic Stability		
<b>Institution</b>	<b>City / State</b>		
UNIVERSITY OF COLORADO HLTH SCIENCES CTR	AURORA, CO		
<b>Principal Investigator</b>	<b>Program Director</b>		
DRABKIN, HARRY A	Judy Mietz		
<b>Start Date</b>	<b>End Date</b>	<b>Cancer Activity</b>	<b>Total Project \$</b>
5/1/2004	04/30/2005	Cancer Cell Biology	DCB \$325,358
<b>Science Area</b>		<b>Percent</b>	<b>Percent \$</b>
KIDNEY		50.00	\$162,679

## Abstract

DESCRIPTION (provided by applicant): TRC8 represents the first rearrangement of any protein that interacts with VHL and its function may be germane to the development of renal cell carcinoma (RCC). The TRC8 gene was identified by positional cloning of a t(3;8) translocation in a family with hereditary RCC and non-medullary thyroid cancer. Affected individuals lacked germline VHL mutations although the morphology of the RCCs was identical. VHL functions as an F-box protein in an SCF-related (VCB) ligase complex that targets HIFalpha for ubiquitination. TRC8 is located in the ER and contains a RING-H2 finger with E3-ubiquitin ligase activity. At least part of the function of VHL is believed to involve the ER in an alternative (non-VCB) ubiquitin ligase complex. To identify a TRC8 function, we isolated the Drosophila homologue, DTrc8, and exploited genetic approaches in flies. Loss of DTrc8 and DVhl result in identical midline phenotypes, and Trc8 physically and genetically interacts with Vhl. From a 2-hybrid screen, we found that human and Drosophila TRC8 interact with the MPN domain of JAB1/CSNS, a component of the COP9 Signalosome which regulates SCF function by removing NEDD8 from Cullins. Jab1/Csn5 has deneddylase activity, and JAB 1 has also been shown to regulate nuclear-cytoplasmic transport and proteasome-mediated degradation of p27 kip1, which is characteristically abnormal in RCCs and which has been linked to VHL mutations. Overexpression of DTrc8 is growth suppressive with several distinct phenotypes, and overexpression of Trc8 and Vhl together produces a unique wing phenotype. Genetic crosses in flies demonstrate that the effects of Trc8 overexpression are corrected by cyclin E and exacerbated by the p27kip1-like molecule, Dacapo. Thus, we hypothesize that the Trc8 overexpression phenotypes are dependent upon the interactions of Trc8 with Vhl and Jab1/ Csn5. We also hypothesize that overexpression of human TRC8 will have similar consequences in mammalian cells. Our overall strategy is to define the interaction domains and introduce point mutations that disrupt them. Mutant proteins will be tested in transgenic flies for: 1) alterations in phenotypes resulting from Trc8 overexpression; 2) biochemical properties of the Signalosome (deneddylation and protein complexes) and, 3) effects on defined Jab1/Csn5 mutant phenotypes. Naturally occurring VHL mutations from RCCs will be tested for their effects on TRC8 binding, ER localization and function, and the assembly of VHL complexes. Adenoviruses expressing wt and mutant TRC8 will be analyzed in mammalian cells for effects on Cullin deneddylation, ubiquitination of a VHL-GFP fusion, cell-cycle parameters and levels of regulatory factors. In our final Aim, mitotic recombinant clones expressing wt or mutant DTrc8 will be used for an independent analysis of Trc8 function (and results integrated into the mammalian analysis). Lastly, because wt DTrc8 induces Minute-like phenotypes in adult sensory hairs, the MPN domains of eIF3h and eIF3f will be tested for interactions and further characterized if positive.

<b>Project Number</b>	<b>Title</b>			
5R01CA89655-4	Combining Anti Angiogenesis Strategies and Radiotherapy			
<b>Institution</b>		<b>City / State</b>		
UNIVERSITY OF FLORIDA		GAINESVILLE, FL		
<b>Principal Investigator</b>		<b>Program Director</b>		
SIEMANN, DIETMAR W		Francis Mahoney		
<b>Start Date</b>	<b>End Date</b>	<b>Cancer Activity</b>	<b>Total Project \$</b>	
7/1/2004	06/30/2005	Radiotherapy	DCTD	\$237,793
<b>Science Area</b>		<b>Percent</b>		<b>Percent \$</b>
KIDNEY		50.00		\$118,897

**Abstract**

DESCRIPTION (provided by applicant):Radiotherapy is the most important non-surgical treatment for cancer. Even so, large numbers of patients still fail at the local treatment site. New treatment strategies aimed at improving tumor response are therefore of high interest and the tumor vasculature, which is critical for the growth and survival of the neoplastic cell population, offers an attractive target. The goal of this grant proposal is to investigate approaches for optimally combining antiangiogenic strategies with localized radiation therapy in order to improve overall tumor response. Investigations are focused on interfering with two activators of angiogenesis (vascular endothelial growth factor (VEGF), basic fibroblast growth factor (bFGF)) and amplification of an endogenous suppressor (endostatin). The approach will be the delivery and transfer of cDNA coding for antisense VEGF/bFGF or endostatin using adeno-associated virus (AAV) or cationic liposomes. Experiments are designed to first characterize and optimize the proposed antiangiogenic approaches in tumor cells, fibroblasts and endothelial cells in vitro. These studies will include the examination of the effect of angiosuppression strategies on the paracrine communication between the various cell types and the determination of whether direct interactions occur between such strategies and radiation treatment. The in vivo efficacy of antiangiogenic treatments used in conjunction with radiation therapy will then be evaluated in xenograft models of AIDS associated Kaposi's Sarcoma and clear cell renal cell carcinoma. These models were chosen because we believe that these neoplasms' typical manifestation of extensive vascularization, coupled with their lack of satisfactory responses to traditional therapeutic interventions, make them excellent candidates for new therapeutic strategies such as antiangiogenic approaches. Both tumor response and critical normal tissue toxicities will be assessed to determine whether a therapeutic benefit can be achieved when angiosuppressive treatments and radiation are combined. We believe that these studies will provide essential insights into the therapeutic utility of employing antiangiogenic treatment strategies as adjuvants to radiotherapy.

<b>Project Number</b> IR21CA105293-1	<b>Title</b> Dietary affects upon genomic methylation in Trout cancer	<b>City / State</b> Pullman, WA		
<b>Institution</b> WASHINGTON STATE UNIVERSITY		<b>Program Director</b> Sharon Ross		
<b>Principal Investigator</b> BRUNELLI, JOSEPH P		<b>Cancer Activity</b> Nutrition (DCP)	<b>Total Project \$</b> DCP	\$128,329
<b>Start Date</b> 5/28/2004	<b>End Date</b> 04/30/2005			
<b>Science Area</b> KIDNEY		<b>Percent</b> 50.00	<b>Percent \$</b>	\$64,165

### Abstract

DESCRIPTION (provided by applicant): The goal of this project is to characterize the effect of dietary supplements expected to impact epigenetic methylation of genomic regions, and associate the variability in methylation to development of nephroblastomas and hepatocarcinomas through exposure to mutagens in clonal lines of rainbow trout. Trout are uniquely suited for this study because of their large size, their sensitivity to mutagens, the well developed research program exploring dietary impacts upon cancer formation, the availability of clonal lines and the ongoing development of a good comparative map in this species. Extensive evidence from studies in mammals demonstrates that the development of cancer involves epigenetic mechanisms affecting gene expression through aberrant genomic methylation. These changes to the genomic methylation status can also develop in response to dietary regimens affecting availability of biochemical substrates to metabolic pathways controlling CpG methylation. Furthermore, this process appears to include a heritable influence through a dietary impact upon the imprinting of gametes. Trout offer an opportunity to analyze, at low cost, candidate dietary elements capable of impacting cancer formation by experimentally controlling dietary variables and tumor development, in a genetically defined background of clonally derived isogenic populations. Specifically, this investigation is designed to reveal correlations between cancer incidence and dietary influences upon aberrant epigenetic CpG methylation causing gene silencing. Project objectives include: 1) Conducting genome-wide screens of CpG methylation patterns characterizing differentiated tissues recovered from isogenic juvenile trout, using the Methylation Sensitive-Amplified Fragment Length Polymorphism (MS-AFLP) technique to determine if genomic methylation patterns are influenced by dietary regimens which manipulate the availability of (folate, methionine, choline, vitamins B12, betaine and zinc). 2) Characterize nephroblastoma and hepatocarcinoma incidence and methylation polymorphisms in isogenic populations exposed to carcinogens and grown under dietary regimens demonstrated to alter methylation. 3) Analyze genome sequences of common MS-AFLP polymorphisms found in cancers, verify tumor associated methylation polymorphisms reflect physiologically relevant epigenetic events, by methylation-specific PCR and bisulfite sequencing to reveal normal tissue methylation patterns, compared to affected loci. Compare expression of genes found to be linked to tumor associated MS-AFLP sites, between normal and tumor tissues, using RT-PCR. 4) Integrate mapable diet associated MS-AFLP polymorphisms into the existing OSU x Arlee map. This investigation will provide fundamental insights into toxicological epigenetic influences through altered genomic methylation and gene expression, and allow the development of a genome-wide tissue-specific methylome profile using an experimentally tractable lower vertebrate model system. Pullman, WA 99164-4236.

<b>Project Number</b>	<b>Title</b>			
5R01CA91048-3	Role of hsnf5/BAF47 Loss in Human Cancer Development			
<b>Institution</b>		<b>City / State</b>		
UNIVERSITY OF NORTH CAROLINA CHAPEL HILL		CHAPEL HILL, NC		
<b>Principal Investigator</b>		<b>Program Director</b>		
WEISSMAN, BERNARD E.		Judy Mietz		
<b>Start Date</b>	<b>End Date</b>	<b>Cancer Activity</b>		<b>Total Project \$</b>
3/1/2004	02/28/2005	Cancer Cell Biology	DCB	\$256,674
<b>Science Area</b>		<b>Percent</b>		<b>Percent \$</b>
KIDNEY		50.00		\$128,337

## Abstract

The identification of human tumor suppressor genes has led to new insights into the mechanisms of human cancer development. Isolation of the first tumor suppressor genes resulted from studies of pediatric malignancies including the RB and WT1 genes. In the case of rhabdoid tumors, frequent LOH on chromosome 22 has led to the discovery of a novel tumor suppressor gene designated INI1/hSNF5/BAF47. This gene codes for the human homolog of the yeast SNF5 gene, a member of the SWI/SNF chromatin remodeling complex. The SWI/SNF complex acts as a global transcriptional activator that alters nucleosome positioning on DNA in an energy-dependent manner. The role of altered chromatin remodeling during neoplastic progression has gained increasing recognition over the last several years. Recent reports strongly support the notion that INI1/hSNF5/BAF47 acts as a prototypical tumor suppressor gene. These include demonstrations that mutations and deletions occur in rhabdoid tumors, choroid plexus tumors and rhabdomyosarcomas, that LOH drives the removal of the remaining wild-type allele, that families carrying germline mutations develop these tumors at a high frequency and that germline inactivation in mice leads to the development of rhabdoid-like tumors. We have found that re-expression of SNF5 in rhabdoid tumor cell lines causes growth inhibition accompanied by a dramatic rise in p16INK4A protein levels. Based on these preliminary studies as well as the known functions of the SWI/SNF complex and other relevant scientific literature, we hypothesize that alterations in the INI1/SNF5 component of the hSWI/SNF complex contribute to human tumor development by blocking the induction of p16INK4A and disrupting normal cell cycle control. In this application, we will test this hypothesis by determining the mechanism by which loss of activity of this gene affects p16INK4A protein levels using biochemical, biological and animal model assays. In Specific Aim number 1, we will determine the cell cycle control pathways regulated by INI1/SNF5 and the relevant domains for these activities. In Specific Aim number 2, we will ascertain the biochemical effects of INI1/SNF5 loss on SWI/SNF function and whether the protein directly interacts with the p16INK4A promoter. In Specific Aim number 3, we will develop a mouse model for choroid plexus carcinomas by crossing TAg transgenic mice to SNF5<sup>+/-</sup> mice. The characterization of the INI1/SNF5 gene role in regulation of gene expression will broaden our understanding of tumor suppressor gene functions, provide important clues about the role of chromatin remodeling complexes in normal and neoplastic development and impact upon treatment and detection of these devastating pediatric cancers.

END OF ABSTRACTS

