19TH ANNUAL

DATA & DINE

Postdoc Research Symposium

Program & Abstract Book

April 16, 2021
9:15 A.M. – 2:00 P.M.

Program

Time	Event
09:15 – 10:00 am	**Opening Remarks** **Leslie Parent, MD** *Vice Dean, Research and Graduate Studies* *Professor, Department of Public Health Sciences, and Microbiology and Immunology, PSU* **Judith Bond, PhD** *Evan Pugh Professor Emeritus, Penn State University* *Adjunct Professor, Department of Biochemistry and Biophysics, University of North Carolina at Chapel Hill*
10:00 – 11:00 am	**Keynote I** The 3B Model of Translation: How to Create Your Own Future **Stephen Ekker, PhD** *Dean, Mayo Clinic Graduate School of Biomedical Sciences* *Director, Office of Entrepreneurship, Mayo Clinic*
11:00 – 12:00 pm	**Oral Presentation** A Review of Social Determinants of Health Questions for Use in Clinical Trials in Tobacco Users **Sophia Allen, PhD** *Public Health Science* Mapping allosteric communications within individual proteins **Jian Wang, PhD** *Pharmacology* **COVID-19 Lightning Talk** Impact of Covid-19 on the surgical management of Peripheral Artery Disease - Analysis from VQI Database **Ahsan Zil E Ali, MD, MPH** *Heart and Vascular Institute* A platform for prevention, detection, and progression of COVID-19 **Yashavantha Vishweshwaraiah, PhD** *Pharmacology* Using Virus like particles (VLPs) to understand molecular mechanism of SARS-CoV2 assembly **Rinki Kumar, PhD** *Microbiology and Immunology*

Program

Time	Event
12:00 – 01:00 pm	**Poster Session (VoiceThread)** **https://sites.psu.edu/postdocs/data-dine-presentations/**
01:00 – 01:40 pm	**Keynote II** Bottom-Up Mentoring: Getting the Most from Your Mentors to Help You Succeed **Katherine Aird, PhD** *Associate Professor, Department of Pharmacology & Chemical Biology,* *UPMC Hillman Cancer Center, University of Pittsburgh School of Medicine*
01:40 – 02:00 pm	**Award Presentation & Closing Remarks** **Gail Thomas, PhD** *Director, Office of Postdoctoral Affairs* *Professor, Medicine,, Clinical and Translational Science Institute and Heart and Vascular Institute, PSU*

WELCOME TO DATA AND DINE

The Penn State Hershey Postdoctoral Society (PSHPS) wants to thank each of you for attending this important event! We are grateful for the support of our mentors, colleagues and lab mates and are thrilled to have the opportunity to showcase our work. This is the nineteenth year of Data and Dine which has become a wonderful annual tradition at the College of Medicine.

"Data and Dine" is an annual Postdoctoral Research Symposium designed to be a semi-social scientific event to celebrate the exciting research accomplishments of the postdocs on the Penn State Hershey campus. Data and Dine was designed for Postdoctoral Scholars and Fellows as an opportunity to share their research with the entire College of Medicine community and to network with fellow postdoctoral researchers and faculty members.

This event is made possible by the active participation among postdocs throughout the campus. Due to on-going pandemic situation, we continue to have the event virtually this year too. While this limits the social events and in-person networking and interaction, we are still very pleased with the overwhelming response from the postdoctoral scholars this year. We have numerous posters, two oral presentations and a special segment of COVID19 lightning talks being presented at the event today, which showcases the rich diversity of ongoing research by the postdoc community. We encourage you to take advantage of this event and learn more about the research accomplishments of the postdocs. Take time to view your colleagues' abstracts and posters as you never know where you may find your next collaboration!

The PSHPS planning committee would like to thank you all for joining us to make this event a success.

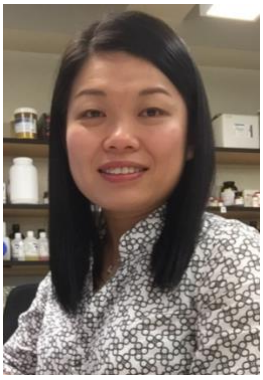
Best regards,
The PSHPS Data and Dine Planning Committee

Data and Dine Planning Committee

Gail D. Thomas, PhD
Director, Office of Postdoctoral Affairs

Khai C. Ang, PhD
Associate Director, Office of Postdoctoral Affairs

Rinki Kumar, PhD (Chair)
Microbiology and Immunology



Mee S. Ngu, PhD
Pathology

Prema Velusamy, PhD
Heart and Vascular Institute

Navaneetha Krishnan Bharathan, PhD
Dermatology

Samantha Spencer, PhD
Microbiology and Immunology

ACKNOWLEDGEMENTS

The "**Data and Dine**" Postdoc Research Symposium is co-sponsored by the Office of Postdoctoral Affairs and the Penn State Hershey Postdoctoral Society and we thank these entities for their financial support. Our deepest gratitude to **Drs. Judith S. Bond and S. Gaylen Bradley** for the annual **Bond & Bradley Award for Postdoctoral Trainees**.

We would like to thank **Drs. Leslie Parent** and **Judith Bond** for the opening remarks. We are immensely thankful to our Keynote Speakers **Drs. Stephen Ekker** and **Katherine Aird** for taking their time to share their expertise and experiences with us.

We want to acknowledge with gratitude, the guidance and support from the Director of Office of Postdoctoral Affairs, **Dr. Gail Thomas** and Associate Director **Dr. Khai Chung Ang**.

We also want to express our gratitude to **Drs. Anirban Paul, Jianfen Hu** and **Wei Li** for volunteering in the Outstanding Postdoc Award Committee, and faculty members **Drs. Andrea Hobkirk**, **Charles Lang**, **Han Chen**, **Hyun Jin Kwun**, **Prashant Nighot**, **Salvatore Stella**, **Thomas Spratt**, **Walter Vonn** and **Yongsoo Kim** for serving as judges for the Outstanding Poster Award.

Lastly, we want to thank all of you because your participation is the key towards making this symposium a success!

This event is organized for postdocs by postdocs. Your input is valuable. If you would like to serve on the postdoc society executive council or 20th Annual Postdoc Data and Dine Planning Committee, we can be reached at postdocs@pennstatehealth.psu.edu.

More information about the Penn State Hershey Postdoctoral Society can be found at https://sites.psu.edu/postdocs/.

BOND & BRADLEY AWARD FOR POSTDOCTORAL TRAINEES

The Penn State Hershey Postdoctoral Society has established the annual **Bond & Bradley Award for Postdoctoral Trainees**, which will be presented to outstanding postdoctoral fellows or scholars at Penn State Hershey at the annual Data and Dine event. The awards include Bond & Bradley Travel Award, Outstanding Postdoctoral Scholar Award and Outstanding Poster Award. These awards are made possible by a generous donation from **Drs. Judith S. Bond** and **S. Gaylen Bradley** to the Penn State Hershey Postdoctoral Society. The endowment inaugurated in 2012 through this donation has provided, in perpetuity, the financial support for these awards.

The Penn State Hershey Postdoctoral Society sincerely acknowledges the kind support from

Drs. Judith S. Bond and S. Gaylen Bradley

19th Annual Data and Dine Keynote Presentation

Stephen Ekker, Ph.D.

Dean, Mayo Clinic Graduate School of Biomedical Sciences
Director, Office of Entrepreneurship, Mayo Clinic

Dr. Stephen Ekker is the Dean of Mayo Clinic Graduate School of Biomedical Sciences and Professor of Department of Biochemistry and Molecular Biology. He is also the Director of Mayo Clinic Office of Entrepreneurship and Mayo Clinic Zebrafish Facility. Dr. Ekker has served as President of Zebrafish Disease Models Society and is currently serving as the Editor-in-Chief of the Zebrafish journal and Founding President of Genome Writers Guild genome engineering society. As a practicing entrepreneur scientist, Dr. Ekker co-founded his first biotechnology company (now Immusoft), and launched two new biotechnology companies- Lifengine Technologies and Mettaforge Therapeutics Inc.

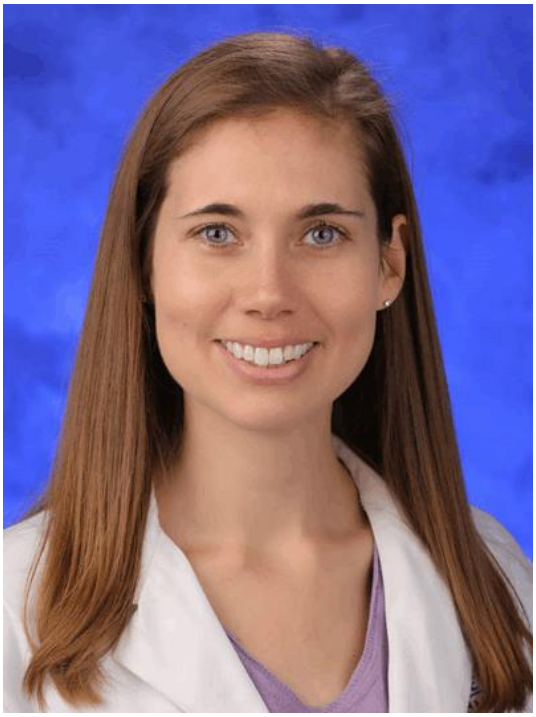
Dr. Ekker is deeply committed to the broad vision that quality science education is imperative for generating qualified scientists, engineers, and health care workers to address major concerns in the world. He also believes that a science-literate citizenry will be essential for our future. To that end, he is committed to serving as a mentor for postdocs, graduate students, MD/PhD students, post-bacs, undergraduates, and related scientists within his laboratory. He has successfully trained 14 PhD or MD/PhD students to degree completion and served on NIH study panels to review training grants. He also served as Associate Director of a T32 while he was a full-time faculty member at the University of Minnesota. Since 2013, he has been Associate Director of the Clinical and Translational Sciences PhD track.

Dr. Ekker's current laboratory uses the zebrafish as a rapid molecular test system to better understand our genome. Seventeen years ago, they established the use of morpholino sequence-specific knockdown technology for vertebrate functional genomics applications using the zebrafish as the pioneering model system. In parallel, they developed vertebrate transposon tools, including their protein trap gene-breaking vectors, to generate a collection of 1000+ molecularly characterized and revertible mutant zebrafish lines, the first engineered conditional alleles in any organism outside the mouse. They have deployed transposons in diverse application areas including human T cells, zebrafish, and mice. Custom restriction endonucleases offer an additional approach to targeted modification using genome editing tools. They continue to develop the science behind these new engineering toolkits, working with laboratories who study rat, pig, mouse, nematode, and fly in addition to our regular colleagues who work in human cells and zebrafish.

19th Annual Data and Dine Keynote Presentation

Katherine M. Aird, Ph.D.

Associate Professor, Department of Pharmacology & Chemical Biology,
UPMC Hillman Cancer Center, University of Pittsburgh School of Medicine



Dr. Katherine Aird is currently an Associate Professor in the Department of Pharmacology & Chemical Biology and a member of the NCI-designated UPMC Hillman Cancer Center at the University of Pittsburgh School of Medicine. She received her BA from Johns Hopkins University, PhD from Duke University, and did her postdoctoral training at The Wistar Institute in Philadelphia. She started her independent lab as an Assistant Professor at Penn State College of Medicine in 2016.

Dr. Aird's research program is focused on the metabolic and epigenetic control of senescence in the context of cancer. She has made fundamental discoveries in these fields and was one of the first to show senescence is not a irreversible process. Her lab aims to understand how senescence overcome and undergo senescence to both contribute to better understanding this process and identify new pathways as targets for therapy.

Dr. Aird enjoys working with postdoctoral fellows on both scientific and professional development, with the goal of providing critical support and training to this important group of scientists. To date, Dr. Aird has trained 3 postdoctoral fellows and 2 graduate students and has served as Assistant Director for Professional Development for the PSU College of Medicine Postdoctoral Society.

Abstracts

Abstract No.	Presenting Author	Title
1	Amanda J. Miller	Angiotensin-(1-7) increases vascular beta2-adrenergic receptor expression in aging mice
2	Amandeep Singh	Discovery of a Novel IGF2BP1 Inhibitor, Based on Lead Optimization through Structure-Activity Relationship Studies, as Metastasis-Specific Therapeutic Agent
3	Ashwinkumar Subramenium Ganapathy	AP2M1 Anchors Claudin-2 in Clathrin Mediated Claudin-2 Degradation Via Autophagy
4	Asif Raza	Stage-specific inhibition of NNK-induced lung carcinogenesis by 1,4-phenylenebis(methylene)seleno-aspirin (p-XS-Asp)
5	Dongyan Zhang	Prefusion spike protein stabilization through computational mutagenesis
6	Fang Chen	Trans-ethnic Transcriptome-wide Association Study for Smoking Addiction in 1.3 Million Individuals Yields Insights into Tobacco Use Biology and Drug Repurposing
7	Gabriela Uribe	Laboratory and Clinical Impact of Identifying and Reporting Blood Culture Isolates of Coagulase-negative Staphylococci Species in an Academic Teaching Center
8	Guifang Wang	Discovery of Small Lead Molecules for Therapeutic Applications
9	Haoyu Sun	Red Blood Cell Released ATP Regulates Systemic Immune Response in Atherosclerosis
10	Loïc Dragin	TAX1BP1 regulates Innate Immune Sensing Pathways to prevent overactivation of Macrophages
11	Marina Chulkina	MyD88 in Colonic Mesenchymal Cells Maintain Intestinal Homeostasis by Control of Macrophages Pro-inflammatory Capacity and Monocyte to Macrophages Maturation
12	Mee S. Ngu	Web-based Histology Reference Atlas for the Freshwater Crustacean *Daphnia magna*

*VoiceThread recordings available for titles highlighted in blue.

Abstracts

Abstract No.	Presenting Author	Title
13	Nicholas Streck	Multi-Site Comparative Study of Two Molecular Instruments for the Detection of Chlamydia trachomatis, Neisseria gonorrhoeae, and Trichomonas vaginalis
14	Nirupama Ramadas	Exercise Alleviates Atherosclerosis Progression through Regulation of Inflammatory Cytokines
15	Prema Velusamy	Limiting Mitochondrial Magnesium Uptake Promotes Hepatic Lipid Accumulation by Modulating the MEK/ERK pathway
16	Rebecca Kaddis Maldonado	Novel insights into how to assemble a retrovirus: beginning at the viral transcription site
17	Rinki Kumar	Investigating the relationship between UL88 and MyD88 during HCMV infection
18	Roberto E. Bruna	Unveiling the Molecular Basis of Phosphate Toxicity
19	Sophia I. Allen	A Review of Social Determinants of Health Questions for Use in Clinical Trials in Tobacco Users
20	Suchitra Mohanty	KDR/VEGFR2 Signaling Regulates Tax Expression and NF-KB Activation
21	Tatsuya Hattori	Targeting the ESCRT-III component CHMP2A for noncanonical Caspase-8 activation on autophagosomal membranes as a novel anticancer strategy
22	Upendarrao Golla	A Novel Rho-associated Protein Kinase (ROCK) Inhibitor for the Treatment of adult Acute Myeloid Leukemia (AML)

*VoiceThread recordings available for titles highlighted in blue

Abstract 1

Postdoc: Amanda J. Miller, PhD
Advisor: Amy C. Arnold, PhD
Department: Neural and Behavioral Sciences

Angiotensin-(1-7) increases vascular beta2-adrenergic receptor expression in aging mice

Amanda J. Miller, Sarah S. Bingaman, Amy C. Arnold

Department of Neural and Behavioral Sciences, Pennsylvania State University College of Medicine, Hershey, Pennsylvania, USA

Background: Aging is the largest independent risk factor for developing cardiovascular disease, with changes in cardiovascular autonomic control contributing to this increased age-related cardiovascular risk. In particular, increased sympathetic tone in aging desensitizes beta 2-adrenergic receptors (B2AR) in the heart and blood vessels to impair vasodilation and contribute to the development of hypertension. We recently demonstrated that chronic treatment with angiotensin (Ang)-(1-7), a protective hormone of the renin-angiotensin system, decreases blood pressure and cardiac sympathetic tone in aged mice. In this study, we hypothesized that sympathoinhibitory effects of Ang-(1-7) would restore vascular B2AR expression to lower blood pressure in aging.
Methods: For experiment 1, aging (16-month-old) and young (2-month-old) male C57BL/6J mice received Ang-(1-7) [400 ng/kg/min, n=4] or saline infusion (n=4) for 6 weeks via subcutaneous osmotic mini-pump. At the end of treatment, alpha- and beta-adrenergic receptor gene expression was measured in mesenteric arteries, thoracic aorta, and cardiac tissue by quantitative real-time PCR and quantified with 2-ΔΔCT methods. For experiment 2, male C57BL/6J mice received subcutaneous injection of Ang-(1-7) (2 mg/mg, n=4), saline (n=3), or the B2AR antagonist ICI 118,551 (1 mg/kg) followed by Ang-(1-7) (n=3). Blood pressure was measured via a carotid artery catheter. Data are presented as mean ± standard deviation and were analyzed by one-way ANOVA, and if significant by post hoc pairwise comparisons.

Results: Aging was associated with decreased B2AR gene expression in mesenteric vessels, which was restored by chronic Ang-(1-7) treatment (young: 1.04 ± 0.35; aged saline: 0.46 ± 0.17; aged Ang-(1-7): 1.04 ± 0.31 A.U., p=0.026). As a control, Ang-(1-7) did not induce significant changes in B2AR mRNA in thoracic aorta or cardiac tissue, or changes in other adrenergic receptor subtypes (alpha1 or beta1) in any of the tissues studied in aging mice (p>0.05). We further found that depressor effects of acute Ang-(1-7) administration in mice are attenuated by B2AR blockade (saline: Δ -11 ± 4 mmHg; Ang-(1-7): Δ -26 ± 3 mmHg; ICI 118,551+Ang-(1-7): Δ -4 ± 4 mmHg, p=0.009).
Conclusions: Ang-(1-7) can restore B2AR expression in aged mesenteric resistance vessels in mice. Further, the depressor effects of acute Ang-(1-7) are at least partially mediated by B2AR. These data support that Ang-(1-7) may decrease blood pressure in aging by restoring B2AR-mediated vasodilation. More broadly, Ang-(1-7) may provide a novel treatment target for age-related hypertension and cardiovascular disease.

Abstract 2

Postdoc: Amandeep Singh, PhD
Advisor: Arun K. Sharma, PhD
Department: Pharmacology

Discovery of a Novel IGF2BP1 Inhibitor, Based on Lead Optimization through Structure-Activity Relationship Studies, as Metastasis-Specific Therapeutic Agent

Amandeep Singh[1], Nadav Wallis[2], Vikash Singh[3], Omer Elimelech[2], Froma Oberman[2], Joel Yisraeli[2], Vladimir Spiegelman[3], Arun K. Sharma[1]

[1]The Pennsylvania State University College of Medicine, Department of Pharmacology, Penn State Cancer Institute, Hershey, PA, USA
[2]Department of Developmental Biology and Cancer Research, Hebrew University Hadassah Medical School, Jerusalem, Israel
[3]The Pennsylvania State University, Department of Pediatrics, Penn State Cancer Institute, Hershey, PA, USA

Metastasis is the leading cause of death in cancer patients. However, since our knowledge of the mechanisms that govern the metastatic process is still inadequate, no effective metastasis-specific therapeutic agents to treat cancer patients exist. IGF2BP1 is a multifunctional RNA-binding protein that regulates the stability, localization and translation of its mRNA targets. High levels of IGF2BP1 expression have been shown to be associated with poor prognosis in patients with variety of malignancies including melanoma, neuroblastoma, hepatocellular carcinoma, squamous cell carcinoma, lung, ovarian, breast and colorectal cancer. Given the correlation between elevated IGF2BP1 expression and poor clinical outcomes, the specific activation of IGF2BP paralogs in a wide variety of cancers, and the effectiveness of preventing metastasis in animal models by reducing IGF2BP1 activity, therapies directed at inhibiting IGF2BP1 function constitute a potentially powerful approach for fighting cancer. In addition, IGF2BP1's pleiotropic effect on multiple pro-tumorigenic and pro-metastatic pathways make it a promising target, as this may avoid the development of resistance, associated with targeted agents in general. Furthermore, specific inhibitors of IGF2BP1would be expected to have minimal side effects since: i) IGF2BP1 is expressed at very low levels in normal adult tissues, and ii) adult mice with an inducible whole-mouse knockout of IGF2BP1 in adult animals are healthy. To identify an efficient IGF2BP1 inhibitor, a fluorescent polarization (FP)-based high throughput screen of over 100,000 small molecules was performed, and the most promising candidates were further validated in an array of *in vitro* and cell-based assays leading to the identification a lead compound, termed "7773," that was highly selective in inhibiting IGF2BP1 RNA binding and a variety of its cellular functions. To further optimize this lead compound and create more selective, effective and safe small-molecule inhibitors of IGF2BP1 that could be developed clinically as cancer therapeutics, we conducted a structure-activity relationship study based on the lead '7773'structure. Novel 27 compounds were designed and synthesized by altering/substituting rings A, B and C of the parent lead molecule 7773 (Figure 1). Since the crystal structure of IGF2BP1 is still not determined, basic skeleton and the overall length of the structure of new molecules was maintained similar to the lead compound so as to possibly not alter the active site binding while enhancing the binding potential. The new analogs generated were evaluated for IGF2BP1 inhibition using our novel cell-based split-luciferase assay, which led to the identification of six compounds that performed similar or better than the lead '7773'. Cell-based wound healing assay revealed that one of

these selected compounds (compound 16) was especially (~more than 14 times than lead "7773") effective in inhibiting cell migration in H1299 cell line that express high levels of endogenous IGF2BP1, but not in LKR-M cells that express very low levels of IGF2BP1. The specificity of compound 16 was further confirmed in LKR-M cells that ectopically express IGF2BP1 – these cells become sensitive to this compound upon overexpression of IGF2BP1 (but not GFP). Together our data provide strong evidence for identification of compound 16 that is effective and selective in inhibiting IGF2BP1 function by interfering with its ability to bind target RNAs.

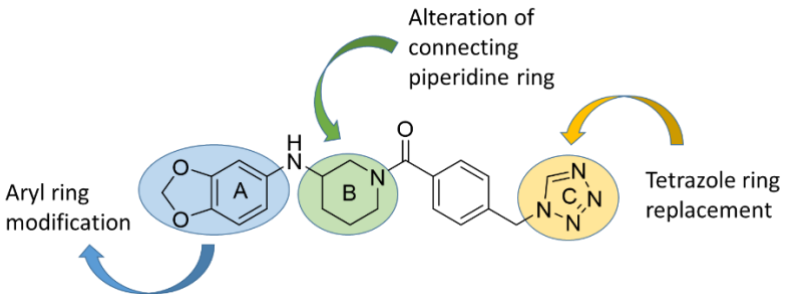


Figure 1: Structure of 7773 and sites of structural modifications

Abstract 3

Postdoc: Ashwinkumar Subramenium Ganapathy, PhD
Advisor: Prashant Nighot, MVSc PhD
Department: Medicine

AP2M1 Anchors Claudin-2 in Clathrin Mediated Claudin-2 Degradation Via Autophagy

Ashwinkumar Subramenium Ganapathy[1], Kushal Saha[1], Eric Suchanec[1], Vikash Singh[2], Thomas Ma[1] and Prashant Nighot[1]

[1]Dept of Medicine, Div of Gastroenterology and Hepatology, Penn State College of Medicine, Hershey, PA, USA.
[2]Dept of Pediatrics, Div of Hematology and Oncology Penn State College of Medicine, Hershey, PA, USA.

Autophagy, a cellular stress response, is an intracellular self-degradative process which is essential for removing misfolded aggregated proteins, and damaged organelles. Autophagy is reported to be involved in intestinal mucosal homeostasis, antimicrobial defense against pathogens and maintaining intestinal epithelial tight junctions (TJ) barrier function. Disruption in TJ barrier function is one of the important hallmarks of Inflammatory Bowel Disease (IBD). We have previously reported that, autophagy, induced by nutrient starvation, enhances intestinal epithelial TJ barrier function (increase transepithelial resistance (TER) and reduce paracellular permeability) by reducing the cation-selective, pore-forming TJ protein, claudin-2. The aim of the current study was to understand the molecular mechanism of autophagy in claudin-2 degradation and enhancing TJ barrier. The interaction of claudin-2 with autophagy and clathrin apparatus was studied using co-immunoprecipitation, microscopic, western blot analysis and further confirmed using site directed mutagenesis and gene knock out studies. Treatment with bafilomycin (autophagy inhibitor) and chlorpromazine (clathrin endocytosis inhibitor) individually, prevented starvation-induced reduction in claudin-2 levels in Caco-2 cells, suggesting an important role of autophagy and clathrin mediated endocytosis in claudin-2 reduction. Co-immunoprecipitation of claudin-2 with key proteins in clathrin apparatus revealed significant increase in association between claudin-2 and clathrin-1, AP-2 (AP2A1 and AP2M1 subunits). The AP2M1 subunit of heterotetrameric adaptor protein AP-2 plays an important role in anchoring membrane proteins into clathrin coated-pit. Autophagy also induced the protein expression and activation (phosphorylation) of AP2M1. Moreover, genetic knock out of AP2M1, and pharmacological inhibition of AP2M1 activation prevented autophagy induced increase in TER, reduction in paracellular permeability, andclaudin-2 levels. The pharmacological inhibition of AP2M1 activation in mice also showed increase in claudin-2 levels. Claudin-2 sequence analysis showed the presence of AP2M1 binding sites in it; site directed mutation of which reduced claudin-2 interaction with AP2M1. Previously, AP2M1 has been shown to interact with LC-3, which is involved in autophagosome formation. The genetic Knock out of ATG7 which plays an important role in LC-3 lipidation shows increased claudin-2 expression compared to control. Our study suggests that autophagy enhances intestinal epithelial TJ barrier by degradation of pore-forming TJ protein claudin-2 via ATG7, AP2M1 associated clathrin endocytosis.

This work was supported by NIH R01DK114024.

Abstract 4

Postdoc: Asif Raza, PhD
Advisor: Arun Sharma, PhD
Department: Pharmacology

Stage-specific inhibition of NNK-induced lung carcinogenesis by 1,4-phenylenebis(methylene)seleno-aspirin (p-XS-Asp)

Asif Raza[1], Amandeep Singh[1], Daniel Plano[2], Cesar Aliaga[1], Shantu Amin[1], Arun K. Sharma[1]

[1]Penn State University College of Medicine, Department of Pharmacology, Hershey, PA
[2]Universidad de Navarra, Spain

1,4-Phenylenebis(methylene)selenocyanate (p-XSC) was on the National Cancer Institute´s (NCI) list of chemopreventive agents but was eventually discarded due to systemic toxicity issues. This toxicity could partially be due to the release of poisonous hydrogen cyanide generated after p-XSC metabolizes to form active bis-selenol (p-XSeH). To address the said concern, we recently designed and developed p-XS-Asp, with the rationale that it would cleave in vivo to release the active p-XSeH and aspirin instead of undesired HCN, thus making the compound less toxic and possibly more potent than p-XSC. Indeed, we have shown previously that p-XS-Asp inhibits NNK-induced lung tumorigenesis in A/J mice more effectively than p-XSC in a complete A/J mouse model, and is better tolerable. Although these results were highly encouraging, this model does not reveal at what stage of carcinogenesis does p-XS-Asp acts. Therefore, in the current study, we evaluated the stage-specificity of chemopreventive efficacy of p-XS-Asp in NNK-induced lung carcinogenesis in A/J mice models.

A/J mice were divided into 7 distinct groups (n=30 per group, half male, half female; except group 1 and 2 where n=10). The mice were fed AIN-93M diet (control) or p-XS-Asp diet until they reached termination endpoint of 26 weeks (adenomas) and 40 weeks (adenocarcinomas). Two weeks after the experiment started, all the groups, except for group 1 and 2, were given a single IP injection of 10 µmol (100 mg/kg) of NNK. Group 3 and 4 were on complete control diet and p-XS-Asp diet, respectively. Group 5 (peri-initiation) was on p-XS-Asp for the first 3 weeks and then on the control diet till the end of the experiment. On the other hand, group 6 (post-initiation) was on control diet for 3 weeks and then was changed to the p-XS-Asp diet. In the progression group 7, mice were on control diet for 14 weeks and subsequently changed to p-XS-Asp diet. The Group 4 and Group 5 mice showed a remarkable decrease in the tumor multiplicity (TM) and incidence (TI) as compared to the mice on control diet at both 26 and 40 week time-points. The TM in male mice for groups 1, 2, 3, 4, 5, 6 and 7 at 26 weeks was 0.6, 0, 5.7, 1, 1.54, 4.27, and 6.27, respectively, while at 40 weeks, it was 0.2, 0, 7.7, 1, 1.8, 4.1, and 4.9, respectively. A similar trend was also observed in female mice. These data clearly demonstrated that robust inhibition at peri-initiation stage is mainly responsible for the activity observed in the complete model. The inhibition of both O6-methylguanine and pyridoxobutyl mutagenic DNA adducts by p-XS-Asp further compliments that the compound acts at initial stages of carcinogenesis. Body weights comparison and the blood and tissue analyses showed no systemic toxicity for the p-XS-Asp fed groups. RNA-seq data of the tumor tissues showed numerous signaling pathways to be affected in p-XS-Asp treated mice. The exact mechanism of action is still under investigation. In summary, our results show that p-XS-Asp may be promising candidate for future clinical evaluation as a lung cancer preventive agent.

Abstract 5

Postdoc: Dongyan Zhang
Advisor: Nikolay V. Dokholyan
Department: Pharmacology

Prefusion spike protein stabilization through computational mutagenesis

Dongyan Zhang[1], Jian Wang[1], Nikolay V. Dokholyan[1,2]

[1]Department of Pharmacology, Penn State College of Medicine, Hershey, Pennsylvania
[2]Departments of Biochemistry & Molecular Biology, Penn State College of Medicine, Hershey, Pennsylvania

A novel severe acute respiratory syndrome (SARS)-like coronavirus (SARS-CoV-2) has emerged at the end of year 2019 as a human pathogen, causing global pandemic and resulting in over 2 million deaths worldwide. The surface spike protein of SARS-CoV-2 mediates the process of coronavirus entry into human cells by binding angiotensin-converting enzyme 2 (ACE2). Due to the critical role in viral-host interaction and the exposure of spike protein, it has been a focus of most vaccines' developments. However, the structural and biochemical studies of the spike protein are challenging because it is thermodynamically metastable. Here, we develop a computational pipeline that automatically identifies mutants that thermodynamically stabilize the spike protein. Our pipeline integrates bioinformatics analysis of conserved residues, motion dynamics from molecular dynamics simulations, and other structural analysis to identify residues that significantly contribute to the thermodynamic stability of the spike protein. We utilize our previously developed protein design tool, Eris, to predict thermodynamically stabilization mutations in proteins. We validate the ability of our pipeline to identify protein stabilization mutants through known prefusion spike protein mutants. We finally utilize the pipeline to identify new prefusion spike protein stabilization mutants.

Abstract 6

Postdoc: Fang Chen, PhD
Advisor: Dajiang Liu, PhD
Department: Public Health Science

Trans-ethnic Transcriptome-wide Association Study for Smoking Addiction in 1.3 Million Individuals Yields Insights into Tobacco Use Biology and Drug Repurposing

Fang Chen[1], Xingyan Wang[1], Seon-Kyeong Jang[2], J. Dylan Weissenkampen[1], Chachrit Khunsriraksakul[1], Dana Hancock[3], Bibo Jiang[1], Scott Vrieze[2], Dajiang J. Liu[1]

[1]Department of Public Health Sciences, Penn State College of Medicine, Hershey, PA, USA
[2] Department of Psychology, University of Minnesota, Minneapolis, MN, USA
[3]RTI International, USA

Genome-wide association studies using samples of European ancestry have discovered more than 400 loci associated with tobacco use behaviors, with a majority of the associated variants being non-coding. It remains challenging to map these non-coding variants to their target genes and translate their biological and clinical relevance. Transcriptome-wide association studies (TWAS) address this issue by integrating genetically regulated gene-expression data from ancestry-matched eQTL datasets. As genetic studies start to incorporate samples of non-European studies, it is critical to extend TWAS accordingly for trans-ethnic genetic studies.

Here we aggregated GWAS and whole genome sequencing data from TOPMed (total N = 1.3 million) and eQTL data in multiple tissues from diverse ancestries to further empower gene discovery for tobacco use behaviors. We developed a novel approach, TESLA, that optimally integrates trans-ethnic GWAS with eQTL datasets. TESLA greatly outperforms prior TWAS methods that integrate only GWAS and eQTL data with matched ancestry, and that incorporate fixed effect GWAS meta-analysis results. The advantage of TESLA is consistent regardless of the gene expression prediction models used. Applying TESLA to tobacco use phenotypes, we identified 319 novel genes that are outside 1 million base pair windows of GWAS sentinel variants, and suggested key pathways that are ubiquitous across tissues (e.g., neurotransmitter catabolic process, GABAergic and dopaminergic synapse) and pathways that are more tissue-specific. Computational drug repurposing using TESLA results also highlighted several drugs with known efficacy including dextromethorphan and galantamine, and novel drugs such as muscle relaxant that may be repurposed for treating smoking addiction.

Abstract 7

Postdoc: Gabriela Uribe, PhD
Advisor: David W. Craft, PhD
Department: Pathology

Laboratory and Clinical Impact of Identifying and Reporting Blood Culture Isolates of Coagulase-negative Staphylococci Species in an Academic Teaching Center

Gabriela Uribe, Nicholas Streck, Cindy Miller, David W. Craft
Penn State Health Milton S. Hershey Medical Center, Department of Pathology, Hershey, PA, USA

Background: Accurate interpretation of blood culture contaminants is increasingly essential for the diagnosis of bloodstream infections. Coagulase-negative staphylococci (CoNS), while a primary cause of blood culture contamination, are emerging as pathogens in certain clinical contexts. Mass spectrometry and multiplex molecular assays have increased the species-level reports of CoNS isolates from positive blood cultures. The primary aim of this study was to evaluate how reporting to species CoNS isolates from positive blood cultures impacts the workflow of the clinical microbiology laboratory and the potential to influence therapeutic management of the patient.
Methods: 9,161 positive blood cultures and associated antimicrobial susceptibility test (AST) results from 2018 to 2020 were retrospectively reviewed. We used an existing monthly data review of CoNS isolated from positive blood culture bottles to assess contamination and/or clinical relevance from concurrently drawn blood culture sets or matched with a subsequent culture within five days. AST was performed on CoNS that were considered clinically relevant. In addition, AST could be requested by a clinician when blood cultures were considered to be contaminants under our criteria.
Results: From 2018-2020, CoNS made up 72.6% (1286/1770) of blood culture contaminants, with *Staphylococcus epidermidis* making up the majority of CoNS species reported in the patient record. 68.7% (883/1286) of contaminants identified as CoNS were collected in the emergency department. Among blood cultures originally categorized as contaminants, AST was performed on 3.2% (36/1119) of CoNS without a reported species and 63.5% (106/167) of isolates reported as *S. epidermidis*. Moreover, 66.9% (95/142) of these contaminants were resistant to oxacillin. Odds ratio of oxacillin resistance in CoNS determined to be a contaminant vs. clinically relevant was 1.5 with a 95% confidence interval of 1.0 to 2.31 (p=0.05).
Conclusions: Our study indicates that AST was performed on laboratory-defined contaminants more often when CoNS was identified to the species level. The majority of *S. epidermidis*, initially regarded as a contaminant by our guidelines, were resistant to oxacillin; raising the question of appropriate therapeutic response and potential overuse of Vancomycin. This may require the re-evaluation of laboratory procedures for reporting the species of CoNS of laboratory defined blood culture contaminants.

Abstract 8

Postdoc: Guifang Wang, PhD
Advisor: Fang Tian, PhD
Department: Biochemistry and Molecular Biology

Discovery of Small Lead Molecules for Therapeutic Applications

Guifang Wang,[1] Yansheng Ye,[1] Yoshinori Takahashi,[2] Barbara Miller,[2] Hong-Gang Wang,[2] Fang Tian[1]

[1] Department of Biochemistry and Molecular Biology, The Pennsylvania State University, Hershey, PA, USA
[2] Department of Pediatrics, Pennsylvania State University College of Medicine, Hershey, PA, USA

Fragment-based library screening (FBS) is emerging as an effective alternative approach to the conventional high throughput screening (HTS) for generating a chemical starting point in drug discovery, especially when coupled with the structural-based drug design. To date, three drugs derived from FBS have been approved by the FDA. The major advantage of FBS is that despite their low affinity, the small size of hit molecules is better suited to generating lead compounds with drug-like properties. FBS is particularly powerful for challenging targets that involve protein-protein interactions, lack deep or well-defined ligand binding pockets, or are difficult to assay. Here, we will present our efforts of exploring FBS to discover lead molecules for CHMP2A and NUDT9H for therapeutic applications. CHMP2A is one of components for ESCRT-III (the endosomal sorting complex required for transport III) that was recently found to be essential for the biogenesis of autophagosome in autophagy. NUDT9H, one of C-terminal domains of Ca2+ channel TRMP2 (transient receptor potential melastatin 2), is responsible for ADPR (adenosine diphosphate-ribose) binding in the gating process. Both NUDT9H and CHMP2A are involved in neurodegenerative disease and are considered as potential drug targets, but currently no effective inhibitors are available for them. We have identified several hit molecules from NMR-based FBS, and are in the process of validating these hits and constructing their binding modes.

This work was supported by NIH R01GM127730 and PENN State Four Diamond Research Grant.

Abstract 9

Postdoc: Haoyu Sun, PhD
Advisor: Pingnian He, MD PhD
Department: Cellular and Molecular Physiology

Red Blood Cell Released ATP Regulates Systemic Immune Response in Atherosclerosis

Haoyu Sun[1], Yunpei Zhang[1], Yong Du[1], Pingnian He[1]

[1]The Pennsylvania State University College of Medicine, Department of Cellular and Molecular Physiology, Hershey, PA, USA

Our studies demonstrated that disturbed flow-induced ATP release from red blood cells (RBCs) via pannexin 1 (Panx1) channels plays a significant role in site-specific vascular inflammation and atherosclerosis. This is supported by a 40-60% reduction of atherosclerotic plaque observed in high fat diet (HFD)-fed ApoE$^{-/-}$ mice with RBC Panx1 deletion that nearly abolished the release of ATP. Given the interplay of the immune system in atherosclerosis, we hypothesize that shear stress-induced ATP release from RBCs at micromolar concentration plays a crucial role in purinergic signaling-mediated immune cell activation, cytokine production, and phenotypic transformation during atherosclerosis. To test this hypothesis, we conducted experiments in chow diet-fed wild type control, and 16 weeks HFD-fed ApoE$^{-/-}$ and ApoE$^{-/-}$RBC Panx1$^{-/-}$ mice. Plasma cytokines were measured via ELISA. Subpopulations of peripheral blood monocytes (PBMCs) were characterized by flow cytometry, and the cytokine secretion capacity of T cells was examined by intracellular interferon-gamma (IFN-γ) staining in isolated splenocytes. Results showed that plasma IFN-γ and interleukin 1 beta (IL-1β) increased 10 and 8 fold, respectively, in HFD ApoE$^{-/-}$ mice when compared to the control group, and reduced to levels close to the control in HFD ApoE$^{-/-}$RBC Panx1$^{-/-}$ mice. PBMCs analysis showed a significantly increased pro-inflammatory Ly6C^{high} monocytes in HFD ApoE$^{-/-}$ mice from 2.9 ± 0.5% (control) to 19.2 ± 2.0%, while only 7.6 ± 1.0% was found in HFD ApoE$^{-/-}$RBC Panx1$^{-/-}$ mice. Intracellular IFN-γ staining of isolated splenocytes from HFD ApoE$^{-/-}$ mice showed a 5-fold increase in IFN-γ+ T cells (%) when compared to the control. In contrast, ApoE$^{-/-}$RBC Panx1$^{-/-}$ mice had significantly reduced IFN-γ+ T cells (%) when compared with ApoE mice (0.12 ± 0.03% vs 1.5 ± 0.48%, $p < 0.05$), especially in CD4+ T cells (0.06 ± 0.01% vs 0.79 ± 0.21%, $p < 0.05$). After stimulation with phorbol 12-myristate 13-acetate and Ionomycin, these cells yielded a markedly higher percentage of IFN-γ+ T cells in the HFD ApoE$^{-/-}$ group (40.4 ± 3.3%) than control (22.7 ± 1.8%) and ApoE$^{-/-}$RBC Panx1$^{-/-}$ group (26.6 ± 3.2%), and more prominent differences were found in CD4+ subgroup (29.0 ± 2.5%, 11.1 ± 1.7%, and 14.5 ± 2.9%, respectively)($p<0.05$). In conclusion, our results indicate that shear stress-induced release of ATP from RBCs via Panx1 plays important roles in the immune cell activation and cytokine production, contributing to immune system-mediated vascular inflammation and formation of atherosclerosis. Our findings provide novel mechanistic insight into systemic risk factor-mediated vascular pathogenesis and disease progression.

This work was supported by HL130363 and HL144620.

Abstract 10

Postdoc: Loïc Dragin, PhD
Advisor: Edward W. Harhaj PhD
Department: Microbiology and Immunology

TAX1BP1 regulates Innate Immune Sensing Pathways to prevent overactivation of Macrophages

Loïc Dragin[1], Sarah McCormick[2], Young-Bong Choi[2], Edward W. Harhaj[1]

[1]Penn State University College of Medicine, Department of Microbiology, Hershey, PA, USA
[2]Department of Oncology, Johns Hopkins School of Medicine, Baltimore, MD, USA

Innate immunity constitutes the first line of host defense against invading pathogens. The cyclic GMP-AMP (cGAMP) Synthase (cGAS) senses invading DNA genomes and synthesizes the second messenger cGAMP that activates Stimulator of interferon genes (STING). Once activated, STING allows for the activation of the transcription factors IRF3 and NF-κB, responsible for the expression of both type I IFN (IFN-I) and proinflammatory cytokines, respectively, orchestrators of the innate immune response. The NLRP3-induced inflammasome is another pathogen-driven inflammatory response that consists of the activation of an aggregated complex of proteins that promote caspase-1-mediated cleavage of pro-IL-1β into its secreted active form that drives inflammation. This is accompanied by an inflammatory type of cell death known as pyroptosis. Despite the need for an efficient innate immune response, negative regulation of the signaling pathways involved is crucial to avert overactivation of the cells and subsequent inflammation and tissue damage; however, the mechanisms involved in this regulation remain unclear.
TAX1BP1 is a ubiquitin binding, selective autophagy receptor that has been shown to negatively regulate immune signaling pathways such as the RNA sensing pathway or the TNF-induced NF-κB pathway. We hypothesized that TAX1BP1 also regulates the cGAS-STING DNA sensing pathway as well as the NLRP3 inflammasome. To this end, we generated TAX1BP1 knockout (TAX1BP1-KO) monocytic cells and mouse bone-marrow-derived macrophages (BMDMs), and found that TAX1BP1-KO cells were more resistant to infection with the DNA virus Herpes Simplex Virus 1 (HSV-1) compared to wild-type (WT) cells. The increased resistance correlated with elevated expression of antiviral IFN-I molecules in response to infection, or stimulation with the STING ligand cGAMP. Interestingly, TAX1BP1 co-immunoprecipitated and colocalized with STING, and regulated STING degradation after activation, suggesting that TAX1BP1 perturbs the STING turnover necessary for IFN-I production. Finally, we found that pyroptosis was significantly increased in TAX1BP1-KO macrophages treated with NLRP3 agonists, which correlated with increased IL-1β secretion in the supernatant. Growing evidence shows that TAX1BP1 promotes aggrephagy, the clearance of aggregated proteins by autophagy. We propose that TAX1BP1 functions as an aggrephagy receptor in innate immune signaling pathways by targeting aggregates of adaptor proteins for autophagy to prevent excessive inflammation and promote homeostasis.

This work is supported by funds from the Penn State Cancer Institute. HSV-YFP (OK-12 strain 17) was generously provided by Dr. John Wills.

Abstract 11

Postdoc: Marina Chulkina, PhD
Advisor: Iryna V. Pinchuk, PhD
Department: Medicine

MyD88 in Colonic Mesenchymal Cells Maintain Intestinal Homeostasis by Control of Macrophages Pro-inflammatory Capacity and Monocyte to Macrophages Maturation

Marina Chulkina,[1] Gabriela Uribe,[1] Steven McAninch,[1] Kamil Khanipov,[2] George Golovko,[2] Jing He,[2] Beswick, Ellen;[3] Don W. Powell,[2] Iryna V. Pinchuk[1]

[1]The Pennsylvania State University College of Medicine, Department of Medicine, Hershey, PA, USA
[2] The University of Texas Medical Branch, Galveston, TX, USA
[3] University of Utah School of Medicine, Department of Internal Medicine, Salt Lake City, UT, USA

Background & Aims: Colonic mesenchymal cells are essential to mucosal tolerance in the intestine. These processes require signaling through MyD88 the major adaptor of TLR-signaling pathways. The genetic deletion of MyD88 promoted the silencing of many key signaling pathways, which most likely aggravated the inflammatory process. However, the role of MyD88-dependent signaling by MFs in the regulation of macrophages under mucosal tolerance in the gut is unknown. Herein, we evaluate the hypothesis that MyD88 signaling in MFs contributes to the control macrophage fates in the colonic mucosa under homeostasis.

Methods: Deletion of MyD88 in B6Col1a2CreMyD88$^{fl/fl}$ mice (MyD88KO-fib) was achieved by tamoxifen. Colon tissue was collected and RNA-seq, qRT-PCR, and FACS were used to evaluate changes in the tolerogenic and inflammatory immune responses. 16s rRNA sequencing was used to access changes in the microbiota. Primary mouse colonic fibroblasts were used to analyze the direct impact of MyD88KO fibroblasts on monocytes.

Results: Fibroblast-specific deletion of MyD88 *in vivo* resulted in the initiation of microcolitis and aggravation of DSS colitis. RNAseq analysis demonstrated dramatic changes in the expression of monocytes/macrophage gene cluster, and changes in signaling pathways associated with myeloid cell activity. Increase of colonic Ly6C$^+$ cells consisting of inflammatory monocytes/macrophages producing inflammatory mediators TNF and iNOS were observed. Depletion of macrophages using clodronate resulted in the decrease of CCR2$^+$ cell number and TNF levels in the colonic tissue. Using the fibroblast-monocyte co-culture system we demonstrated the essential role of MyD88 signaling in fibroblasts on macrophage maturation and suppression of the myeloid cell inflammatory capacity. Inflammatory changes in the colonic mucosa induced by MyD88 deficiency within fibroblasts involved dysbiosis with the increase of Gram-negative bacteria and decrease in Firmicutes to Bacteroidetes ratio (F/B ratio), which known to be a hallmark of intestinal inflammation. Treatment with the antibiotic mix of MyD88KO-fib - mice resulted in the significant reduction of mucosal inflammation.

Conclusion: Intrinsic MyD88 signaling within colonic mesenchymal cells is critical to these cells mediated regulation of innate immune cells under homeostasis through defining Ly6C$^+$monocytes differentiation and suppress of pro-inflammatory responses of macrophages; thus, preventing microbiota induced immunopathological damage to the colonic mucosa.

This work was supported by CTSA/TL1 5TL1TR001440-04 UTMB Institute for Translational Science and R01 DK103150 National Institute of Diabetes and Digestive and Kidney Diseases.

Abstract 12

Postdoc: Mee S. Ngu, PhD
Advisor: Keith C. Cheng, MD PhD
Department: Pathology

Web-based Histology Reference Atlas for the Freshwater Crustacean *Daphnia magna*

Mee S. Ngu[1,2], Daniel J. Vanselow[1,2], Jean E. Copper[1,2], Chadwick Harris[3], Debra A. Shearer[1,2] Alex Lin[1,2], Keith C. Cheng[1,2,4], Khai C. Ang[1,2,4]

[1]Pennsylvania State University College of Medicine, Department of Pathology, Hershey, PA, USA
[2]Pennsylvania State University College of Medicine, Jake Gittlen Laboratories for Cancer Research, Hershey, PA, USA
[3]Pennsylvania State University College of Medicine, Department of Cytology, Hershey, PA, USA
[4]Pennsylvania State University College of Medicine, Zebrafish Functional Genomics Core, Hershey, PA, USA

The assessment of pathological change in tissues requires prior knowledge of normal tissue structure, making microanatomical atlases useful for ecotoxicological and environmental monitoring studies. *Daphnia magna* has been widely used as an invertebrate model in aquatic toxicology but lacks a reference atlas. To promote whole-organism phenotyping for toxicology, we have created a web-based histology reference atlas for *D. magna* (https://bio-atlas.com/Daphnia). Here, we report our protocols towards creation of the atlas, including optimized fixation protocol and designing a casting mold to improve sample orientation and consistency of sectioning plane. This atlas includes 40X digital scans (at 0.25 micron per pixel) of serial sections in 3 orthogonal planes. A carousel-like tool is used to navigate through the sections. Interactive highlighting and labelling of cell and tissue types facilitates the identification of structures and potential referencing to the literature. This atlas is intended to be a community effort, serving as a visualization platform of high-resolution images, a tool for education and research collaboration, and a platform for integration across imaging modalities and potentially -omic data. The features and functionalities of this atlas can serve as a model for other organism atlases.

This project is funded by NIH (R24OD018559-06), Jake Gittlen Laboratories for Cancer Research, and under a grant with the Pennsylvania Department of Health using Tobacco CURE Funds for Human Health and Environment. The Pennsylvania Department of Health specifically disclaims responsibility for any analysis, interpretations or conclusions.

Abstract 13

Postdoc: Nicholas Streck, PhD
Advisors: Wallace Greene, PhD, D(ABMM) and David Craft, PhD, D(ABMM)
Department: Pathology

Multi-Site Comparative Study of Two Molecular Instruments for the Detection of *Chlamydia trachomatis*, *Neisseria gonorrhoeae*, and *Trichomonas vaginalis*

N. Streck[1], G. Uribe[1], W. Greene[1], S.N. Taylor[2], C.L. Cammarata[2], D. Diodene[2], J. Galbraith[3], M. Rose[3], E. Lockamy[3]

[1]Department of Pathology, Penn State College of Medicine, Hershey, PA, USA
[2]Louisiana State University Health Sciences Center, New Orleans, LA, USA
[3]Becton, Dickinson and Company, BD Life Sciences, Diagnostic Systems, Sparks, MD, USA

Background: *Chlamydia trachomatis* (CT), *Neisseria gonorrhoeae* (GC), and *Trichomonas vaginalis* (TV) are among the most common sexually transmitted infections worldwide that cause significant health and economic burdens if left untreated. Screening of high-risk populations could reduce transmission and health risks associated with undiagnosed infections. The BD COR™ PX/MX System* is a fully automated molecular platform intended to reduce workflow complexities and provide high throughput testing in clinical laboratories. The modular system is designed to offer a variety of molecular assays to detect multiple pathogens. One such assay, BD CTGCTV2, currently cleared on the BD MAX System, is a DNA extraction and real-time polymerase chain reaction test for the simultaneous detection of CT, GC, and/or TV DNA in patient- or clinician-collected vaginal swab specimens (in a clinical setting), and male and female urine specimens. The assay may also be used for the detection of CT and GC DNA in endocervical swab and specimens in PreservCyt® Solution. The objective of this study was to determine the agreement of the BD CTGCTV2 assay between the BD COR™ PX/MX* and BD MAX™ Systems, using urine panels.

Methods: 1,299 paired urine specimens were tested using the BD CTGCTV2 assay at three testing sites. Specimens were tested on both the BD COR™ PX/MX* and BD MAX™ Systems. Panel members for the clinical comparison study were created by pooling negative clinical specimens and, where necessary, spiking in a high positive clinical specimen or utilizing individual positive specimens at specific Ct scores to reach the necessary analyte level(s) for each target (CT, GC, TV). Assay results from the BD MAX™ System served as the reference test for this study. The overall BD CTGCTV2 positive and negative percent agreement between BD COR™ PX/MX* and BD MAX™ Systems was determined.

Results: Positive and negative percent agreement between the BD COR™ PX/MX* and BD MAX™ Systems was 99.7% and 99.1% for CT, 97.6% and 98.5% for GC, and 99.4% and 98.8% for TV respectively.

Conclusions: The results of the BD CTGCTV2 assay demonstrated equivalent and high levels of positive and negative agreement between the BD COR™ PX/MX System* and BD MAX™ System. The BD COR™ PX/MX System* provides a high throughput testing and screening solution for the detection of CT, GC, and TV.

*The BD COR™ PX/MX System and its associated assays are not currently cleared for use in the US.

Abstract 14

Postdoc: Nirupama Ramadas, PhD
Advisor: Pingnian He, MD PhD
Department: Cellular and Molecular Physiology

Exercise Alleviates Atherosclerosis Progression through Regulation of Inflammatory Cytokines

Nirupama Ramadas, Yong Du, Haoyu Sun, Hope Uwase, Pingnian He

Pennsylvania State University College of Medicine, Department of Cellular and Molecular Physiology, Hershey, PA, USA

Atherosclerosis is one of the leading causes of mortality accounting for 30% of deaths worldwide. It is a disease of multifactorial etiology exemplified by lipid-driven inflammatory disease of the arterial intima. The atherosclerotic progression appears to be due to defective regulation of inflammatory events. Though exercise is conceived as a beneficial factor to combat many cardiovascular diseases, the effects of exercise on atherosclerosis progression under hyperlipidemic conditions and the precise mechanisms of exercise-induced plaque regression and stability remains unclear. Therefore, this study is to investigate the impact of exercise training on plaque formation and stability and regulation of inflammation in hyperlipidemia-induced atherosclerosis in ApoE mice. Male ApoE mice were randomly assigned to 3 groups (n = 6/group): Sedentary controls; pre-exercise and exercise groups and all fed with high fat diet (HFD) for 16 weeks. Pre-exercise group had exercise for 4 weeks with normal diet prior to HFD. Animals in pre-exercise and exercise groups were subjected to treadmill exercise for 60 min/day, 5 days/week at 15 m/min intensity with 5˚ slope for 16 weeks. After the scheduled exercise regime, the mice were sacrificed. Blood samples were collected for cytokine and lipid profile analyses, and aorta was harvested for *en face* Sudan IV staining. Image J software was used for lesion quantification. All groups of mice had total cholesterol levels over 400 mg/dl after 16 weeks of HFD (no significant difference among groups). Sudan IV staining of whole aorta revealed significant reductions of lesion in both exercise groups compared with sedentary controls. In sedentary controls, the total aorta lesion area was 33 ± 2.2 % after 16 weeks of HFD, while in pre-exercise and exercise groups, the mean lesion area was 24 ± 0.9 % and 27 ± 1.5 % of total aorta area, respectively, representing 28 and 19 % of lesion reduction relative to sedentary controls. The most lesion reduction occurred at arch and abdominal aorta and the reductions in pre-exercise group are more significant. This reduction in lesion is strikingly accompanied by reduction in major cytokines associated with atherosclerosis i.e. TNF-α, IL-1β, CRP and IFN-γ in pre-exercise and exercise groups. Though we see reduced cytokines in both exercise groups, preexercise showed more prominent response, indicating routine exercise prior to hyperlipidemia to have preventive effect on HFD-induced cytokine secretion. Pearson correlation analysis showed a significant positive correlation of reduction of lesion with IL-1β ($r = 0.610$, $p < 0.01$); TNF-α ($r = 0.550$, $p<0.05$) and IFN-γ ($r = 0.470$, $p<0.05$). Overall, our results indicated that exercise can significantly suppress the production of inflammatory cytokines and reduce lesion formation despite hyperlipidemic conditions. This study revealed a positive correlation between plasma pro-inflammatory cytokine levels and aorta lesion formation and provided underlying mechanisms by which sustained exercise training alleviates atherosclerosis progression under hyperlipidemic conditions.

Supported by HL130363 and HL144620

Abstract 15

Postdoc: Prema Velusamy
Advisor: Shanmughapriya Santhanam
Department: Heart and Vascular Institute

Limiting Mitochondrial Magnesium Uptake Promotes Hepatic Lipid Accumulation by Modulating the MEK/ERK pathway

Prema Velusamy[1], Natarajaseenivasan Kalimuthusamy[2], Shanmughapriya Santhanam[1,3,4]

[1]Heart and Vascular Institute, [3]Department of Medicine, [4]Department of Cellular and Molecular Physiology, Pennsylvania State University College of Medicine, Hershey – 17033
[2]Department of Neuroscience, Lewis Katz School of Medicine, Temple University, Philadelphia, PA - 19140

Background: Magnesium (Mg^{2+}) is an abundant intracellular divalent cation and is an important co-factor in the machineries that replicate, transcribe and translate genomic information. Like calcium, Mg^{2+} is also compartmentalized to mitochondria and Mrs2 is the only known molecular machinery associated with mitochondrial Mg^{2+} (mMg^{2+}) influx. Although we know that Mg^{2+} plays a crucial role in cellular homeostasis, our understanding of how mMg^{2+} homeostasis alters the metabolic state of the cell remains incomplete. Mg^{2+} is essential for mitochondrial oxidative function and low mMg^{2+} is known to impair mitochondrial fatty acid oxidation (FAO) and TCA cycle. Recent studies also elucidated, lower Mg^{2+} content in liver to disrupt mitochondrial function and to elicit inflammatory responses and oxidative stress. Because liver is the central hub of lipid metabolism, we hypothesized loss of mMg^{2+} uptake in the hepatocytes to alter lipid homeostasis and accumulation of lipid droplets (LDs) in the hepatocytes. **Methods:** The ubiquitous expression and localization of Mrs2 to the inner mitochondrial membrane were analyzed using protein flux and Western blot analysis. In order to elucidate the impact of mMg^{2+} on the metabolic state of the cell, we made a liver-specific Mrs2 knockout mice using CRISPR-Cas9 based Cre-loxP system. The gene targeting was confirmed by genotyping and the loss of Mrs2 in the liver was confirmed using qPCR and Western blot. Control and KO hepatocytes were isolated and infected with adenovirus expressing Mario or Mito-Mario and FRET measurement was performed after stimulating the cells with glucagon. Also, the impact of Mrs2 KO on mitochondrial function, cellular bioenergetics, LD accumulation, oxidative stress, and lipid peroxidation was studied using electron microscopy, confocal microscopy, spectrofluorometer, and seahorse XFe24 flux analyzer. **Results:** Preliminary data using isolated hepatocytes from control and Mrs2 KO mice showed a substantial decrease in mMg^{2+} uptake and ablated Mrs2 current validating Mrs2 as an authentic mammalian mitochondrial Mg^{2+} channel. The hepatocytes from the KO mice showed shorter and more circular mitochondria with a compromised FAO coupled oxygen consumption rate (OCR). We also found a considerable increase in the number and size of lipid droplets together with an increase in lipid peroxidation in KO hepatocytes when compared to the control. In spite of increased LD load in the KO hepatocytes we did not notice any change in the fatty acid synthesis. In this regard, we postulate that compromised FAO in KO hepatocytes could redirect free fatty acids back to lipid droplets (LDs) resulting in higher hepatic LD load. We also observed an increase in the mROS levels and the BODIPY 581/591 (ratiometric lipid peroxidation sensor) green to red ratio in the Mrs2 KO indicating increased mROS and subsequent lipid peroxidation. Additionally, mMg^{2+} homeostasis significantly upregulated the apoB levels and decreased the ERK1/2 phosphorylation.

Conclusion: Because we saw mMg^{2+} to modulate LD accumulation, ApoB and ERK1/2 phosphorylation levels, our future work will verify whether the increased accumulation of triglyceride (TG)-rich LDs can combine with LDL precursors to form mature and secretion-competent VLDL, thus subsequently altering the plasma LDL cholesterol (LDL-C) and triglyceride (TG) levels through the MEK-ERK/ApoB pathway axis. Also, because elevated plasma LDL-C and TG levels are major risk factors for atherosclerosis, we will also investigate whether limiting mMg^{2+} uptake promotes atherosclerotic plaque formation. Our results from the study will help us understand how Mg^{2+} regulates lipid metabolism and contributes to cardiovascular health.

Abstract 16

Postdoc: Rebecca Kaddis Maldonado, PhD
Advisor: Leslie Parent, MD
Department: Medicine

Novel insights into how to assemble a retrovirus: beginning at the viral transcription site

Rebecca Kaddis Maldonado[1] and Leslie J. Parent[1,2]

Penn State College of Medicine, [1]Departments of Medicine, [2]Microbiology & Immunology, Hershey, PA, USA

HIV, the retrovirus that causes AIDS, remains a major global health crisis. Although an HIV diagnosis is no longer considered a death sentence as it was in the 1980s due to advancements in treatment, an ongoing challenge to ending the pandemic is the high level of viral mutations leading to drug resistance. To develop better drugs to treat HIV-infected patients, it is imperative to better understand how the virus assembles, and to determine which host factors and processes are involved in this process.

Retrovirus assembly is nucleated when the viral Gag structural protein binds unspliced viral RNA (vRNA) and selects it as genome (gRNA) that will be packaged into new virus particles. It was previously thought that the initial Gag-vRNA interaction occurred in the cytoplasm or at the plasma membrane. However, our lab showed that the Gag proteins of HIV-1 and the chicken retrovirus Rous sarcoma virus (RSV) form nuclear foci that co-localize with their respective unspliced vRNA in a co-transcriptional manner. Furthermore, RSV Gag nuclear trafficking is linked to efficient gRNA packaging. Together these data suggest that the Gag proteins of multiple retroviruses enter the nucleus of infected cells and traffic to sites of vRNA transcription to select unspliced vRNA for packaging.

Unspliced retroviral RNA comprises less than 1% of the total mRNA in an infected cell; therefore it would be beneficial for Gag to traffic to the site of the highest concentration of unspliced vRNA, the site of viral transcription in the nucleus. Unspliced vRNA also serves as mRNA for the translation of Gag and Gag-Pol, and it is currently unknown how Gag distinguishes unspliced vRNA destined for packaging from that which will serve as mRNA. Because the cytoplasmic fate of cellular RNAs is thought to be determine by the binding of various factors to nascent RNAs co-transcriptionally, we hypothesize that Gag binds to unspliced vRNA as it's being synthesized to "mark" it as gRNA. Experiments are underway to examine whether Gag uses host factors in actively-transcribing euchromatin domains to target viral transcription sites. If so, disrupting Gag-euchromatin interactions is a potential novel antiviral strategy.

Abstract 17

Postdoc: Rinki Kumar, PhD
Advisor: Nick Buchkovich, PhD
Department: Microbiology and Immunology

Investigating the relationship between UL88 and MyD88 during HCMV infection

Rinki Kumar[1] and Nicholas Buchkovich[1]

[1]The Pennsylvania State University College of Medicine, Department of Microbiology and Immunology, Hershey, PA, USA

HCMV infection is largely asymptomatic in healthy individuals but may cause serious and even fatal disease in immunocompromised individuals and newborns. HCMV is the largest of the human herpesviruses with a 230 kilobase double-stranded DNA genome encoding at least 200 open reading frames (ORF). UL88 is a tegument protein that is dispensable for replication, but has critical roles in tegumentation of the virion. We are currently exploring a potential role for UL88 in immune evasion, which has not previously been explored. We have shown that deletion of UL88 from the viral genome does not reduce infectious virus titers when grown in fibroblasts, epithelial cells or differentiated cells of the myeloid lineage (Kumar et al., 2020. However, UL88 is essential for HCMV spread in the presence of activated myeloid cells. We observed an increase in MyD88 levels during infection with UL88-deficient HCMV, and a significant reduction when UL88 was overexpressed. Thus, we hypothesize that UL88 antagonizes the MyD88 signaling pathway, which is activated by cytokines (IL-1/IL-18) produced by activated myeloid cells. We are studying the relationship between UL88 and MyD88 during HCMV infection and want to determine the underlying mechanism of UL88-mediated antagonization of MyD88.

This work was supported by NIH/NIAID 1R01AI130156-01.
Reagents were generously provided by Dr. Wade Gibson, Dr. Eain Murphy, Dr. Jens von Einem and Dr. John Purdy

Abstract 18

Postdoc: Roberto E. Bruna, PhD
Advisor: Mauricio H. Pontes, PhD Assistant Professor
Department: Pathology and Laboratory Medicine

Unveiling the Molecular Basis of Phosphate Toxicity

Roberto E. Bruna[1], Christopher G. Kendra[1], Mauricio H. Pontes[1,2]

[1]The Pennsylvania State University College of Medicine, Department of Pathology and Laboratory Medicine, Hershey, PA, USA. [2]The Pennsylvania State University College of Medicine, Department of Microbiology and Immunology, Hershey, PA, USA

Background: Phosphorus (P) is essential for life. In bacteria, P is acquired mainly as inorganic orthophosphate (Pi) and assimilated into adenosine triphosphate (ATP) –the main cellular P-carrier molecule– in the cytoplasm[1]. Cells must tightly regulate Pi acquisition and utilization because excessive cytoplasmic Pi is toxic[1,2]. For instance, mutations that increase Pi uptake via the PstSCAB transport system (e.g., in *phoU*) inhibit growth in many bacterial species[3-7]. However, the underlying molecular basis for this cytotoxicity has remained elusive. Notably, assimilation of Pi into ATP is coupled with Pi charge neutralization by cytoplasmic Mg^{2+} and the formation of ATP:Mg^{2+}, the substrate for the majority of ATP-dependent enzymes[8]. The transfer of Pi from ATP into ribosomal RNAs (rRNAs) is also coupled with the sequestration of Mg^{2+}. Negatively charged Pi-residues in the rRNA chelate large amounts of Mg^{2+}, reducing electrostatic repulsion to enable the folding and assembly of functional ribosomes[8,9]. Consequently, ATP and rRNA constitute the largest cytoplasmic reservoirs of Pi and Mg^{2+}. Given this inherent connection between Pi and Mg^{2+}, we hypothesized that the cytotoxic effects of excessive Pi uptake result from uncontrolled Pi assimilation into ATP, chelation of free cytoplasmic Mg^{2+} and subsequent disruption of downstream core processes, such as translation.

Objective: To define the molecular basis of phosphate cytotoxicity, using *Salmonella enterica* serovar Typhimurium as a model.

Results: Increased Pi uptake via PstSCAB was achieved either by PstSCAB overexpression or deletion of *phoU* regulatory gene. In both cases, this resulted in a rise of ATP intracellular concentrations along with reductions in growth rate and growth yield, in comparison with the control strains. As predicted, these phenotypes were alleviated through the provision of excess Mg^{2+} in the growth medium, or ATP hydrolysis. In addition, PstSCAB overexpression provoked a reduction in translation rates, and precipitated *mgtC* transcription, indicative of translational disruption caused by cytoplasmic Mg^{2+} starvation.

Conclusions: Our findings indicate that excess Pi imported into the cytoplasm is rapidly assimilated into ATP, decreasing levels of free cytoplasmic Mg^{2+}. The reduction in cytoplasmic Mg^{2+} lowers the rate of protein synthesis, and ultimately inhibits growth. These findings provide a framework to understand the molecular basis for Pi toxicity, based on its intimate connection with cytoplasmic Mg^{2+} homeostasis[10].

Abstract 19

Postdoc: Sophia I. Allen, PhD
Advisor: Jonathan Foulds, PhD
Department: Public Health Sciences

A Review of Social Determinants of Health Questions for Use in Clinical Trials in Tobacco Users

Sophia I. Allen[1,2], Susan Veldheer[1,3], Andrea Hobkirk[1,4], Jessica Yingst[1,2], Nicolle Krebs[1,2], Jonathan Foulds[1,2]

[1]The Pennsylvania State University College of Medicine, Hershey, PA, USA
[2]Department of Public Health Sciences, Hershey, PA, USA
[3]Department of Family and Community Medicine, Hershey, PA, USA
[4]Department of Psychiatry and Behavioral Health, Hershey, PA, USA

Introduction: Disparities in smoking behaviors and tobacco-related harms to health persist among adults in the U.S. The disproportionate impact to health from tobacco use is influenced, in part, by social determinants of health (SDoH). Here we describe the selection of items included on a SDoH assessment developed for use in clinical trials including adult smokers.

Methods: Questions were adapted from the Protocol for Responding to and Assessing Patients' Assets, Risks, and Experiences (PRAPARE) and the Adverse Childhood Experience (ACE) surveys. The questions align with recommendations for the inclusion of SDoH measures from the National Academies of Sciences, Engineering and Medicine. Questions included housing stability (worried about losing housing), household size, material security (e.g., food, clothing, utilities, etc.), transportation, adverse childhood experience, social connection, and neighborhood safety. Other standard questions on race, ethnicity, education, employment, health insurance, income, and mental health/stress were also included. Questions chosen for inclusion in the final survey were categorized within the five Healthy People 2020 domains.

Results: A comprehensive assessment of SDoH-related risk included 17 questions from the PRAPARE and the ACE surveys. These items provide a score between 0 and 17 for measures within the SDoH domains. A response of "yes" to a question on the ACE survey counts as only one tally.

Conclusion: A comprehensive assessment of SDoH-related risk should be considered in future clinical trials of tobacco users and other contexts of health as they impact health care outcomes. The collection of SDoH-related risk data can be achieved without excessively lengthy questionnaires.

This work was supported by the National Institutes of Health (NIH), National Institute on Drug Abuse (NIDA) U01-DA-04551701S1 (PI: JF). The content is solely the responsibility of the authors and does not necessarily represent the official views of the NI

Abstract 20

Postdoc: Suchitra Mohanty, PhD
Advisor: Edward W. Harhaj, PhD
Department: Microbiology and Immunology

KDR/VEGFR2 Signaling Regulates Tax Expression and NF-κB Activation

Suchitra Mohanty[1], Alfonso Lavorgna[2] and Edward W. Harhaj[1, 2*]

[1]Penn State University College of Medicine, Department of Microbiology and Immunology, Hershey, PA, USA
[2]Sidney Kimmel Comprehensive Cancer Center, Johns Hopkins School of Medicine, Baltimore, MD, USA

The human T-cell leukemia virus type 1 (HTLV-1) is an enveloped retrovirus, and the causative agent of adult T-cell leukemia/lymphoma (ATLL), an aggressive neoplasm of CD4+CD25+ T cells that occurs in 2–5% of infected individuals after decades of asymptomatic latent infection. HTLV-1 infection is also associated with inflammatory diseases including HTLV-1-associated myelopathy/tropical spastic paraparesis (HAM/TSP). The HTLV-1 genome encodes a regulatory trans-activating protein, Tax that regulates viral gene expression by coordinating the recruitment of the CREB transcription factor and CBP/p300 co-activators to the 5' viral long terminal repeat (LTR). HTLV-1 Tax also acts as a potent oncogene that interacts with and modulates a plethora of host cellular proteins and triggers the aberrant activation of signaling pathways, most notably, NF-κB to drive clonal proliferation and survival of T cells bearing the HTLV-1 provirus. Accumulating evidence has revealed the tight regulation of Tax expression by other HTLV-1 regulatory proteins for successful viral immune evasion and persistence; however, the role of host cellular proteins in the regulation of Tax expression and protein stability is largely unknown. The identification of host proteins that control Tax expression and stability could potentially yield targets for the suppression of HTLV-1 proviral load or novel therapeutic approaches for ATLL. We have conducted a kinome-wide shRNA screen to identify host factors crucial for the survival of an HTLV-1-transformed T cell line, MT-2. The top hit in the screen was the tyrosine kinase receptor KDR/VEGFR2, which we have validated as a critical survival factor in a subset of HTLV-1-transformed T cell lines. Inhibition of KDR with shRNAs or small molecule inhibitors induced caspase-dependent apoptotic cell death selectively in KDR+ HTLV-1-transformed T cells. Furthermore, inhibition of KDR elicited autophagic degradation of Tax and diminished the chronic activation of NF-κB. Collectively, our results have revealed an oncogenic role of KDR/VEGFR2 signaling in HTLV-1-transformed cells and ATLL, and have further implicated KDR/VEGFR2 as a novel regulator of Tax protein stability which could be exploited as a strategy to decrease proviral loads in HTLV-1-infected individuals.

Abstract 21

Postdoc: Tatsuya Hattori, PhD
Advisor: Hong-Gang Wang, PhD
Department: Pediatrics

Targeting the ESCRT-III component CHMP2A for noncanonical Caspase-8 activation on autophagosomal membranes as a novel anticancer strategy

Tatsuya Hattori[1], Yoshinori Takahashi[1], Longgui Chen[1], Zhenyuan Tang[1], Carson A. Wills[1], Xinwen Liang[1], and Hong-Gang Wang[1]

[1]The Pennsylvania State University College of Medicine, Department of Pediatrics, Hershey, PA, USA

Autophagosomal membranes can serve as activation platforms for intracellular death-inducing signaling complexes (iDISCs) to initiate Caspase-8-dependent apoptosis. In this study, we explore the impact of ESCRT-III-dependent phagophore closure on iDISC assemblies and cell death in osteosarcoma and neuroblastoma cells. Inhibition of phagophore closure by conditional depletion of CHMP2A, an ESCRT-III component, stabilizes iDISCs on immature autophagosomal membranes and induces Caspase-8-dependent cell death. Importantly, suppression of the iDISC formation via deletion of ATG7, an E1 enzyme for ubiquitin-like autophagy-related proteins, blocks Caspase-8 activation and cell death following CHMP2A depletion. Although DR5 expression and TRAIL-induced apoptosis are enhanced in CHMP2A-depleted cells, the canonical extrinsic pathway of apoptosis is not responsible for the initiation of cell death by CHMP2A depletion. Furthermore, the loss of CHMP2A impairs neuroblastoma tumor growth associated with decreased autophagy and increased apoptosis in vivo. Together, these findings indicate that inhibition of the ESCRT-III-dependent autophagosome sealing process triggers noncanonical Caspase-8 activation and apoptosis, which may open new avenues for therapeutic targeting of autophagy in cancer.

This work was supported by NIH grants GM127954 and CA222349, the Lois High Berstler Research Endowment Fund, and the Four Diamonds Fund. Confocal images were generated using the Leica SP8 microscope (NIH Shared Instrumentation grant S10OD010756-01A1) located in the Penn State College of Medicine Microscopy Imaging Core Facility.

Abstract 22

Postdoc: Upendarrao Golla, PhD
Advisor: David F. Claxton, MD and Arati Sharma, PhD
Department: Medicine, Division of Hematology and Oncology

A Novel Rho-associated Protein Kinase (ROCK) Inhibitor for the Treatment of adult Acute Myeloid Leukemia (AML)

Upendarrao Golla[1], Charyguly Annageldiyev[1], Diwakar B. Tukaramrao[2], Zheng Zeng[3], Melanie Ehudin[2], Sinisa Dovat[2], Shantu Amin[3], Dhimant Desai[3], David Claxton[1,4], and Arati Sharma[2,3,4]

[1]Department of Medicine; [2]Department of Pediatrics, Division of Hematology and Oncology; [3]Department of Pharmacology; [4]Penn State Cancer Institute, Penn State College of Medicine, Hershey, PA, USA

Acute myeloid leukemia (AML) is a rare but severe form of hematological malignancy with a heterogeneous genetic landscape. AML accounts for 80% of acute leukemia cases in adults, with a percent remission of 35-40% for younger patients and 5-15% for older patients. Despite the advancement in understanding of AML pathogenesis, the standard treatment regimen largely unchanged over the past decade with limited clinical impact on the survival of AML patients. Therefore, there is an urgent need for novel therapies for effective treatment of AML. A Rho-associated coiled-coil kinase (ROCK) is a serine-threonine kinase regulated by small GTPase RhoA and modulates several cellular functions including malignant transformation, metastasis, and cell death. RhoA is frequently dysregulated in hematological disorders and recent RNAi profiling identified ROCK1 as a potential therapeutic target for AML treatment. It was shown that the oncogenes such as KIT, BCR-ABL, and FLT3 induce activation of ROCK and led to the progression of leukemia. Therefore, current study was designed to assess the efficacy and safety of novel ROCK kinase inhibitor DJ4, [(5Z)-2--5-(1H-pyrrolo [2,3-b]pyridine-3-ylmethylene)-1,3-thiazol-4(5H)-one] against AML *in vitro* and *in vivo* disease models. DJ4 found to be effective against multiple cancer cell lines by selectively inhibiting the activities of ROCK and myotonic dystrophy kinase-related Cdc42-binding kinases (MRCK) kinases. In the present study, DJ4 exhibited dose-dependent cytotoxicity in AML cell lines at low micromolar concentration (IC_{50}: 0.05-2 μM). Similarly, DJ4 effectively induced apoptosis and inhibited colony-forming ability of primary AML patient cells. DJ4 treatment led to decreased phosphorylation of ROCK1 downstream effectors such as myosin phosphatase (MYPT1) and myosin light chain (MLC) in AML cell lines indicating ROCK pathway inhibition. Furthermore, DJ4 exhibited anti-leukemic activity against both disseminated and subcutaneous AML in human xenograft mouse models (OCI-ALM3/MV4-11) by extending the survival, impeding tumor growth and leukemia progression significantly ($p<0.01$) without systemic toxicity. Altogether, our data indicate that novel ROCK inhibitor DJ4 could be a potential therapeutic agent for the treatment of AML and warrants further studies to compare DJ4 with other ROCK inhibitors and evaluate its therapeutic efficacy in combination with standard AML drugs such as Ara-C/Daunorubicin.

This work was supported by the Kenneth F. Noel Memorial Fund (DC), Delbert J. McQuaide Cancer Research Fund (AS), Austin R. Orwan Memorial Research Fund (AS), The Penn State Cancer Institute (PSCI), and the NIH/NCI P01CA171983 (AS & DC).

Follow Postdoc Society on Social Media

Twitter: @PSUCoMPostdoc https://twitter.com/PSUCoMPostdoc
Instagram: psucom_postdoc https://www.instagram.com/psucom_postdoc/
Facebook group: Hershey Postdocs https://www.facebook.com/groups/439767336548787/about/

Our goal is to give Hershey Postdocs as much visibility as possible. Thus, if you have news that you want to share (eg. publications, awards, grants etc), please feel free to tell us and we will post it!

"Connect" is the quarterly newsletter of the Post-Doctoral Society at the College of Medicine, PSU, Hershey. It brings to the community a glimpse into the activities, events and achievements of the post-doctoral fellows. From warm welcomes to new fellows to "Connect" them with their colleagues, to notifications about opportunities for professional growth and enrichment in upcoming quarters.

"Connect" is a professional platform that aims to bring you the recognition you deserve and an opportunity to showcase your work. Let us know about your publications, grants, awards that you receive or personal milestones, and we will highlight it in our spotlight section. We are keen to hear from you about your work. Write to us for a chance of an interview and let fellow colleagues learn from your experiences.

We invite everyone to write about what matters to you- arts, science, society, philosophy or life in general, or express yourself though photography or cartoons. We call for interesting science images from your research, to be featured on the cover page. So, keep connected and look out for the next issue.

Rinki Kumar
Editor