**Health Equity & Community Engagement**

**The Bench Tutorials Program: An Essential Educational Pivot in response to COVID-19**
Chantele Singleton, MS, MBA, Sharon A. Croisant, MS, PhD, Lance Hallberg, PhD, John Prochaska, DrPH, Krista Bohn, MPH, Michelle Puig, MEd and Cornelis Elferink, PhD

1The University of Texas Medical Branch and 2Galveston Independent School District

ABSTRACT IMPACT: The Bench Tutorials Program is an independent study course in biomedical research in which high school students are paired with graduate and post-doctoral students during the academic year. The purpose is to enhance the rigor of high school science education and build the pipeline of tomorrow’s researchers. OBJECTIVES/GOALS: The Bench Tutorials Program: o Proficiency in research design, implementation, and presentation; o Acquisition of hands-on laboratory skills; o Increase in scientific literacy; o Increase in analytical skills and critical thinking; o Career in science; o Build the pipeline of tomorrow’s biomedical researchers

METHODS/STUDY POPULATION: High School seniors are paired with graduate and postdoc mentors through a matching process. Students spend approximately four hours/week in supervised instruction and research from a participating laboratory in addition to classroom experience at their High School. Mentors design research projects relating to the larger research framework of their laboratories. In light of COVID-19, approaches have been adjusted to maintain the program safely through the program under COVID-19 restraints without putting anyone in harms way. Go-Pros have been essential for our program to maintain continuity so even in the absence of COVID-19 in the future, the continued use of these devices will still be of great value. DISCUSSION/SIGNIFICANCE OF FINDINGS: We previously reported a role for DNA-PK(cs) in immunosuppression. We now have evidence that this occurs in part through stabilization of Egr1 that was barely detectable. In contrast, expression of the transgene factor Egr1 was highly induced after activation but attenuated after treatment with NU7441 in both Jurkat T cells and mouse splenocytes. Phosphorylated serine 301 of Egr1 was identified by mass spectrometry in stimulated cells and fits the kinase consensus sequence for DNA-PK(cs). Both an endogenous CRISPR-generated sequence 301 to alanine mutant and expression of a plasmid-based S301A mutant resulted in an unstable form of Egr1 that was barely detectable.

**Mechanistic Basic to Clinical**

**DNA-PK(cs) Regulates Stability of Egr1 During T Cell Activation**
Zachary Waldrip, David Harrison, Marie Burdine and Lyle Burdine
University of Arkansas for Medical Sciences, Arkansas Children’s Research Institute

ABSTRACT IMPACT: This work provides supporting evidence for the development of a novel immunosuppression therapy for transplant patients. OBJECTIVES/GOALS: Our laboratory reported that inhibition of the kinase DNA-PK(cs) in mice delays allogeneic graft rejection in part by mitigating the induction of certain cytokines. We hypothesized that this was due to an inhibition of intracellular signaling programs in T cells and designed studies to identify the mechanism(s) by which this occurs. METHODS/STUDY POPULATION: The immortalized Jurkat T cell line was used to evaluate the effect of the DNA-PK(cs) inhibitor NU7441 on T cell activation by PMA/Ionomycin or PMA/PHA. Mouse primary splenocytes also were used to demonstrate the universality and reproducibility of our observations. Initially, protein mass spectrometry of lysates from untreated and NU7441-treated Jurkat cells identified proteins of interest regulated by DNA-PK(cs) that play a role in T cell activation and cytokine production. CRISPR genome editing was used to validate a potential downstream target of DNA-PK(cs). Western blot, ELISA, and flow cytometry were used to document changes in protein levels with respect to treatments. RESULTS/ANTICIPATED RESULTS: We observed that expression of the transcription factor Egr1 was highly induced after activation but attenuated after treatment with NU7441 in both Jurkat T cells and mouse splenocytes. Phosphorylated serine 301 of Egr1 was identified by mass spectrometry in stimulated cells and fits the kinase consensus sequence for DNA-PK(cs). Both an endogenous CRISPR-generated sequence 301 to alanine mutant and expression of a plasmid-based S301A mutant resulted in an unstable form of Egr1 that was barely detectable. In contrast, expression of the transgene factor Egr1 was highly induced after activation but attenuated after treatment with NU7441 in both Jurkat T cells and mouse splenocytes. Phosphorylated serine 301 of Egr1 was identified by mass spectrometry in stimulated cells and fits the kinase consensus sequence for DNA-PK(cs). Both an endogenous CRISPR-generated sequence 301 to alanine mutant and expression of a plasmid-based S301A mutant resulted in an unstable form of Egr1 that was barely detectable.

**Circadian Disruption in Pancreatic Cancer Carcinogenesis**
Patrick B. Schwartz, MD, Morgan T. Walcheck, BS, Kristina A. Matkowskyj, MD, PhD, Christopher A. Bradfield, PhD and Sean M. Ronneklev-Kelly, MD
1Department of Surgery, University of Wisconsin School of Medicine and Public Health, 2Department of Pathology and Laboratory Medicine, University of Wisconsin School of Medicine and Public Health, 3Department of Oncology, University of Wisconsin School of Medicine and Public Health and 4Department of Surgery, Division of Surgical Oncology, University of Wisconsin School of Medicine and Public Health

ABSTRACT IMPACT: Circadian disruption is known to cause significant human pathology but has not been evaluated in pancreas cancer carcinogenesis; through understanding how disruption of circadian rhythms can lead to pancreatic cancer development and spread, preventive and therapeutic strategies can be devised. OBJECTIVES/GOALS: Pancreatic ductal adenocarcinoma (PDAC) is a lethal cancer due to early spread and poor response to therapy. Identifying factors driving PDAC growth could lead to new therapeutic strategies. Thus, we evaluated the extent to which circadian rhythm disruption, a factor strongly associated with cancer formation, contributes to PDAC pathogenesis. METHODS/STUDY