and serum antibody titers were similar between severe and non-severe groups. However, we observed that individuals recovered from severe COVID-19 have a significantly reduced frequency of spike specific IgG + memory B cells expressing Tbet and FcRL5 (markers associated with long lived immunity). In the non-severe patients, we observed IgG +Tbet+ B cells targeting the spike protein peak at 2-3 weeks post-symptom onset, decrease by almost fifty percent 4-5 weeks post-symptom onset, and return to baseline 5 months post-symptom onset. Our study also validated previous findings of a short-lived primary response of IgM+ B cells targeting the spike protein. DISCUSSION/ SIGNIFICANCE: Our findings highlight potential implications for long-term immunity against re-infection or severity of the resulting disease in patients with severe COVID-19. Further investigation will be necessary to determine whether the maintenance of immunological protection is hindered in patients who overcame severe COVID-19.

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Post-translational role of RNA modifications in sRNA chaperone Hfq

Jalisa Nurse[2], Joseph[l], Aubee[2] and Karl M Thompson[1]
[1]Department of Microbiology, Howard University College of Medicine, Washington DC and [2]Howard University

OBJECTIVES/GOALS: The goal of this study is to determine the role of the tRNA modifications in the translation of Hfq. Hfq is an RNA chaperone that acts as a co-factor for the action of the largest class of small RNAs in E. coli. RNA modifications have been known to play critical roles in the translational fidelity of many cellular proteins in bacteria. METHODS/STUDY POPULATION: In this study, we used an hfq-lacZ translation fusion to screen several RNA modification mutant genes to uncover additional RNA modifications that may play a role in Hfq translation. We measured hfq-lacZ activity in genetic backgrounds mutated for several additional RNA modification enzymes previously untested for Hfq effects. RESULTS/ANTICIPATED RESULTS: We identified 5 RNA modification genes that were defective for hfq-lacZ fusion activity, and we subsequently performed western blot analysis on the Hfq protein in the absence of these modification mutant genes to determine the effect of these mutants more directly on Hfq protein levels. We identified 2 out of these 5 RNA modification mutants that also affect Hfq protein levels. DISCUSSION/ SIGNIFICANCE: Since Hfq is critically important for small RNA function is a wide range of bacteria, it is possible tRNA modifications regulate Hfq expression in other bacteria. These processes, when further investigated, could provide us with the basic information to develop new antibiotics needed to address emerging antibiotic resistance.

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Inhibition of GPR30 Reveals Putative Genes Involved in the Pathogenesis of Inflammatory Breast Cancer

Melanie Carrion-Laureano[1], Melvyn Acosta-Montalvo[1,2], Xavier Bittman-Soto[2], Esther Peterson-Peguero[2,3] and Josué Pérez-Santiago[3,4]
[1]University of Puerto Rico Comprehensive Cancer Center, [2]University of Puerto Rico Rio Piedras, Department of Biology, [3] University of Puerto Rico Comprehensive Cancer Center, Division of Cancer Biology and [4]University of Puerto Rico Medical Sciences Campus, School of Dental Medicine

OBJECTIVES/GOALS: Inflammatory Breast Cancer (IBC) is the most aggressive form of breast cancer and does not have targeted therapy. GPR30, a 7-transmembrane estrogen receptor, may play a role in regulating cell growth and proliferation of cancerous cells. Here, we evaluated changes in gene expression while inhibiting GPR30 to determine putative targets to treat IBC. METHODS/ STUDY POPULATION: IBC cell lines (SUM149PT) were cultured in medium with serum stripped from growth factor and hormones for 48 hours. Cells were then exposed to either G15 (GPR30 inhibitor) at a concentration of $1 \hat{A} \mu M$ or ETOH (vehicle negative control) 3 hours in triplicates. After exposure, total RNA was extracted using the Qiagen RNAeasy Mini kit and RNA was sequenced using the Illumina NextSeq (2 X 75bp). The higher-quality reads were aligned, annotated, and quantified to the human genome (HG38) using STAR and RSEM softwares. Gene expression analysis was performed in R statistical software (packages tximport and DESeq2). Functional and enrichment analyses were performed using Metascape and STRING database, respectively. RESULTS/ANTICIPATED RESULTS: There were 656 significantly expressed genes ($p < 0.05$) between groups (G15 vs. ETOH). The top 5 significant genes include: SMIM7, FANCG, ARID1A, MAML2, and ATF3. Significantly impacted biological processes and pathways include: electron transport chain, mitotic cell cycle process, microtubule cytoskeleton organization, cellular component morphogenesis and DNA-dependent DNA replication (adj $p < 0.05$). Additionally, physical and functional interaction networks showed 3 major clusters (â‰¥ 12 genes), which contained several gene hubs including BRCA1, BRCA2, FOS (proto-oncogene), PLK1 and PAK1 (both serine/threonine-protein kinases), among others. Interestingly, the network analysis showed the previously known interaction between FANCG and BRCA2, which were both dysregulated by GPR30 inhibition. DISCUSSION/SIGNIFICANCE: Through gene expression, functional and enrichment analyses we found several targets genes that could be associated with the pathogenesis of IBC. Validation of candidates genes (qRT-PCR and Western blot), and functional assays (cell proliferation, motility, and invasion) will be performed to understand the potential of these genes in treating IBC.

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Isolation and identification of bioprospects capable of metabolizing 17-beta-estradiol and 17-alpha-ethinylestradiol using metagenomics and culture-dependent techniques in Puerto Rico

Carlos RÃos-VelÃ¡zquez[1], Melissa M. Cabrera-RuÃz[1], Kristal M. Delgado-Serrano[1] and Hailey N. Morales-Lajara[2]
[1]University of Puerto Rico at Mayaguez, Biology Department and [2]University of Puerto Rico at Mayaguez, Chemical Engineering Department

OBJECTIVES/GOALS: This research project aims to isolate and identify bioprospects capable of metabolizing estrogen using culture-dependent, culture independent methods and the identification of the gene/genes responsible for the metabolization of estrogen by the bioprospects. METHODS/STUDY POPULATION: For the culture dependent technique, samples were collected from the water treatment plant in MayagÃ¼ez, cultivated on TSA medium and selected specific and diverse colonies were patched on M-9NC (no carbon sources), M-9-glucose (M9G) and M-9-hormone mixture (M9H: 17-beta-Estradiol and 17-alpha-Ethynylestradiol). After the 48hrs incubation at 25 and 37 Celsius, growth was scored on the different media, to choose those potential bioprospects that use the hormones as the sole carbon source. For the culture independent approach, metagenomic clones from libraries generated from the

Guajatacas water reservoir and Cabo Rojos Saltern microbial mats were cultivated on the appropriate M9NC, M-9G and M-9H, supplemented with Leucine and Thiamine for 48 hours at 37 Celsius. RESULTS/ANTICIPATED RESULTS: The culture-dependent approach showed a total of 6 potential bioprospects capable of growing on hormones as a sole carbon source on M9H at 25 and 37 Celsius and no-growth on the M-9 media used. Currently, we are in the process of determining the identities of the cultivable isolates genetically, any preference on hormone metabolization, monitoring a larger number of metagenomic clones and optimizing the selection conditions. DISCUSSION/SIGNIFICANCE: The identification of bioprospects capable of metabolizing hormones could potentially lead to the generation of new water treatments that could help remove unwanted chemicals, such as high concentrations of estrogens.

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Gender Differences in the Association of Impulsive Behavior and Susceptibility to E-cigarette Use among Adolescents with Congenital Heart Defects

Kristen R. Fox[1], Amy K. Ferketich[2], Judith A. Groner[3], Joseph R. Rausch[1], Vidu Garg[3], Steven P. Neville[1], Victoria R. Grant[1] and Jamie L. Jackson[1]
[1]The Abigail Wexner Research Institute at Nationwide Children's Hospital, [2]The Ohio State University and [3]Nationwide Children's Hospital

OBJECTIVES/GOALS: Adolescents with congenital heart defects (CHD) have an elevated risk for future cardiovascular events, but information about their risk for e-cigarette use ("vaping") is unknown. This study aims to present preliminary findings on gender differences in the association of impulsive behavior and vaping susceptibility from an ongoing investigation. METHODS/STUDY POPULATION: Adolescents with CHD (12-18 years; N=63) reported their vaping susceptibility and completed subjective (UPPS-P)/objective (Iowa Gambling Task; IGT) assessments of impulsive behavior previously associated with tobacco use. The UPPS-P includes 5 facets: 1) negative urgency (impulsivity under negative emotions), 2) positive urgency (impulsivity under positive emotions), 3) lack of premeditation (acting without thinking), 4) lack of perseverance (inability to focus), and 5) sensation seeking (seeking thrilling experiences). The IGT is a computerized task that creates conflict between immediate reward and delayed punishment via selections from advantageous/disadvantageous card decks. Linear regressions stratified by gender determined associations between vaping susceptibility and impulsivity. RESULTS/ANTICIPATED RESULTS: Nearly 30% (29.7%) of adolescents with CHD were susceptible to vaping. Negative urgency was associated with vaping susceptibility among females ($\hat{\beta} = 0.44$, p = .035) but not males ($\hat{\beta} = 0.25$, p = .128). Positive urgency was associated with vaping susceptibility among males ($\hat{\beta} = 0.37$, p = .021) and trended toward significance among females ($\hat{\beta} = 0.40$, p = .058). Lack of premeditation was associated with vaping susceptibility among males ($\hat{\beta} = 0.36$, p = .025) but not females ($\hat{\beta} = 0.15$, p = .490). The association between lack of perseverance and vaping susceptibility trended toward significance among males ($\hat{\beta} = 0.30$, p = .064) but not females ($\hat{\beta} = -0.18$, p = .413). IGT performance was not associated with susceptibility to vaping among either gender. UPPS-P facets and IGT performance were not significantly correlated. DISCUSSION/SIGNIFICANCE: The association of impulsivity and vaping susceptibility appears to be characterized by emotion-based rash action (positive/negative urgency) for females and by decreased conscientiousness (lack of premeditation/perseverance) for males. If replicated, the findings have implications for assessment of vaping risk and tailored intervention.

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Unique Gray Matter Volume Differences in Bilingual Children with Reading Disability*

Alison Schug[1] and Guinevere F. Eden[2]
[1]Georgetown Howard Universities Center for Clinical and Translational Science and [2]Center for the Study of Learning, Department of Pediatrics, Georgetown University Medical Center, Washington, DC, USA

OBJECTIVES/GOALS: Developmental dyslexia is a common reading disability (RD) which negatively impacts academic success. To address the role of early language experience on RD, we tested if the reported differences in gray matter volume (GMV) in RD also manifests in poor readers with a bilingual language background. METHODS/STUDY POPULATION: We studied 54 Spanish-English Bilingual Typical Readers with Oral Reading Recognition Test (ORRT) scores above 100 (avg. =113 ± 10), 51 Spanish-English Bilinguals with RD with ORRT scores below 92 (avg. =84 ± 7), 54 English Monolingual Typical Readers with ORRT scores above 100 (avg. =113 ± 10) and 51 English Monolinguals with RD with ORRT scores below 92 (avg. =84 ± 7) from the Adolescent Brain & Cognitive Development Study. All groups had an average age of 12 ± 0.7 years and were matched for sex and self-ratings of English ability. Structural magnetic resonance images were analyzed using Voxel-Based Morphometry and the bilingual and monolingual groups were separately compared in two-sample t-tests (p < 0.05). RESULTS/ANTICIPATED RESULTS: Monolinguals with RD had less GMV than the Monolingual Typical Readers in the right supramarginal gyrus (Brodmanns Area (BA) 40; MNI Coordinates: 69, -27, 39) (p=0.011) similar to the right superior temporal finding reported in the existing literature. However, a comparison of Bilinguals with RD and Bilingual Typical Readers did not show any GMV differences in superior temporal regions. Instead, our Bilinguals with RD had less GMV compared to the Spanish-English Bilingual Typical Readers in the right superior frontal gyrus (BA 11; MNI Coordinates: 21, 44, -24) extending to the middle frontal gyrus (BA 10) (p = 0.014). DISCUSSION/SIGNIFICANCE: Our findings suggest that the neuroanatomical bases of RD in Spanish-English Bilingual children are not the same as those observed for monolinguals, and biological models developed in monolinguals cannot be generalized. This has implications for diagnoses and treatment of RD in bilinguals.

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An Intrinsic Pathway in the Brain Underlying the Relationship Between Pain Catastrophizing and Chronic Pain in Temporomandibular Disorders

Rachel L. Cundiff-O'Sullivan[1], Rachel Massalee[1], Yang Wang[1] and Luana Colloca[1]
[1]University of Maryland, Baltimore

OBJECTIVES/GOALS: Pain Catastrophizing is a negative coping mechanism involving rumination, magnification, and helplessness and is associated with worse chronic pain. The neurobiological mechanisms underlying this relationship are poorly understood.