tissue growth and contraction, image and video analysis was performed at specific timepoints. To analyze differentiation efficiency and cell population, flow cytometry was performed using cardiac markers. To evaluate gene expression, qPCR was performed using pluripotency and cardiac specific primers. RESULTS/ANTICIPATED RESULTS: Direct cardiac differentiation of encapsulated hiPSCs resulted in synchronously contracting 3D-hECTs in both biomaterials and all tissue geometries. Spontaneous contractions started on Day 7 and increased in velocity, frequency, and synchronicity over time. 3D-hECTs had high cell viability with > 70% of cells positive for cardiac markers. Engineered tissues showed appropriate temporal changes in gene expression over time with pluripotency gene expression decreasing and cardiac gene expression increasing. DISCUSSION/SIGNIFICANCE OF IMPACT: This study shows the potential for direct differentiation of encapsulated hiPSCs to produce physiologically relevant engineered cardiac tissues. Resulting 3D-hECTS showed features of mature myocardium with appropriate cardiomyocyte populations, mechanical motion, and gene expression. Using this platform, we are able to produce engineered cardiac tissue in a variety of biomaterials and tissue geometries to study new therapeutics, mechanism of disease, and scalable tissue culture.

3300

Progesterone receptor alters lipid biology in luminal breast cancer

Ashley Vanessa Ward[1], Shawna B. Matthews[1] and Carol A. Sartorius[1]
[1]University of Colorado at Denver

OBJECTIVES/SPECIFIC AIMS: These studies seek to evaluate hormonal regulation of luminal breast cancer lipid metabolism and to identify targetable progesterone-mediated changes in lipid biology that contribute to therapeutic resistance in breast cancer. METHODS/STUDY POPULATION: Established and patient-derived luminal breast cancer cell lines, which express ER and PR, were used for this study. RNA transcript and protein expression levels were evaluated by qRT-PCR and immunoblot, respectively. Broad scale lipidomics of progesterone-treated cells was conducted via ultra-high pressure liquid chromatography-mass spectrometry (UHPLC-MS) through the UCD Skaggs School of Pharmacy Mass Spectrometry Core. RESULTS/ANTICIPATED RESULTS: Data mining of previously published microarray data of CK5+ and CK5− syngeneic cancer sublines revealed that CK5+ cells have increased expression of lipid processing genes, including LPL and PPARG. As progestin treatment induces a subpopulation of cells to turn on CK5 expression in luminal breast cancers, UHPLC-MS-based lipidomics analysis will expose whether modulation of the lipid landscape occurs in all cells with progesterone treatment, or whether this phenomenon is heightened specifically in CK5+ cells. I also expect that ER+ breast cancers with progestin induced-altered lipid content, such as lipid droplet formation, will evade therapy-induced death. DISCUSSION/SIGNIFICANCE OF IMPACT: There are numerous approved and developmental therapeutics targeting lipid biology. By determining if progestins alter lipid metabolic genes specifically in CK5+ CSCs, which are endocrine resistant, strategies may be devised to target these resistant cells using combination therapy in conjunction with existing therapies to prevent tumor recurrence.

3326

Radiofrequency Renal Denervation Prevents Further Progression of Hypertension and Decreases Renal Medullary Fibrosis in One-year-old Spontaneously Hypertensive Rats (SHR)

Juan Gao[1], Ian B Denys[1], Jane Sutphen[1], Luis Del Valle[1] and Daniel R Kapusta[1]
[1]Louisiana State University Health Sciences Center

OBJECTIVES/SPECIFIC AIMS: We have reported that radiofrequency renal denervation (RF-RDN) in SHR at 20-weeks of age, decreased blood pressure (BP) and fibrosis in kidney cortex and medulla when rats were sacrificed at 6 months. However, whether RF-RDN can have similar benefits in older rats remains unknown. This study examined whether performing RF-RDN in older rats also has a beneficial effect on BP and renal fibrosis. METHODS/STUDY POPULATION: Baseline systolic and diastolic BP (SBP/DPB) was measured (telemetry) in nine-month-old SHR and Wistar Kyoto rats (WKY). Groups of rats then received bilateral RF-RDN or Sham-RDN (SHR-RDN, n=9; SHR-Sham, n=10; WKY-RDN, n=5; WKY-Sham, n=8). Rats were then sacrificed at 12-months of age. Kidneys were harvested, sectioned, and assessed for fibrosis by Masson's trichrome stain. A pathologist, who was blinded to treatment groups, evaluated each kidney section for fibrosis. RESULTS/ANTICIPATED RESULTS: Compared to SHR with Sham-RDN, RF-RDN prevented a further increase in systolic and diastolic BP from baseline (9-month) in SHR as they aged to 12-months (SHR-Sham mmHg: 9-month 193±4/127±4; 12-month 207±3/142±5; SHR-RDN mmHg: 9-month 197±3/132±2; 12-month 197±4/132±3). RF-RDN did not alter SBP or DBP in aged WKY. One-year-old SHR with prior Sham-RDN showed extensive renal fibrosis in kidney cortex and medulla. In contrast, RF-RDN significantly decreased renal fibrosis in the medulla, but not cortex. There was no fibrosis in kidneys of age matched WKY. DISCUSSION/SIGNIFICANCE OF IMPACT: These findings suggest that RF-RDN may be a potential therapy for halting progression of hypertension and decreasing medullary fibrosis in the aged population.

3293

Region Specific Dysregulation of Dopaminergic Signaling in Mice Displaying Excessive Over-Grooming

Daniel Foster[1], Samantha Yohn, Muhammad Mahmood, Madigan Lavery, Daniel O'Brien, Weimin Peng and P. Jeffrey Conn
[1]Vanderbilt University Medical Center

OBJECTIVES/SPECIFIC AIMS: The objective of this study was to determine if dopamine signaling is altered in a mouse model displaying excessive self-grooming and further elucidate the potential utility of compounds targeting the striatal DA system in modulating repetitive behaviors. METHODS/STUDY POPULATION: Here, we report studies using fast-scan cyclic voltammetry (FSCV) in mice lacking the postsynaptic protein SAP90/PSD95-associated protein (SAPAP3 KO mice) as well as control littermates. Rodent self-grooming provides a behavioral output with which one can monitor repetitive, self-directed, patterned behavior that has great translational value to OCD-like disorders. Total time spent grooming was monitored in SAPAP3KO mice and control littermates. To further examine the role of DA in regulating repetitive grooming behaviors the magnitude and kinetics of DA transients were assessed using

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FSCV in ex vivo slice preparations as well as in anesthetized mice in vivo. DA transients were elicited in the dorsolateral striatum (DLS), dorsomedial striatum (DMS); and nucelus accumbens core (NAcc). In some experiments mice were crossed with DAT-Cre animals and channelrhodopsin 2 (ChR2) was virally expressed in DA neurons to allow optical stimulation of DA transients. RESULTS/ANTICIPATED RESULTS: As previously reported, SAPAP3 KO mice showed excessive grooming compared to control littermates at the age assessed (4-5 months). DA transients evoked by a single electrical pulse in slices from SAPAP3 KO mice were not significantly different from those observed in slices from control littermates in any of the regions tested including the DLS, DMS and NAcc. However, when four electrical pulses were applied at a frequency of 10Hz to mimic DA neuron bursting, the magnitude of DA transients observed in the DMS and NAcc of SAPAP3 mice were greater than those evoked in control littermates.Interestingly, phasic stimulation produced similar DA transients in the DLS of both genotypes suggesting that phasic DA signaling was not globally altered. To confirm this finding we crossed SAPAP3 KO mice with DAT-Cre mice and injected ChR2 containing virus into the midbrain to selectively express ChR2 in DA neurons. Transients were then optically evoked resulting in selective activation of DA neurons. Optical stimulation produced a pronounced enhancement of DA release in SAPAP3 KO mice specifically in the DMS and only following phasic-like stimulation. DISCUSSION/SIGNIFICANCE OF IMPACT: These exciting findings suggest that DA signaling in SAPAP3KO mice is dysregulated in a very precise manner that is sub-region specific as well as dependent on the pattern of stimulation. These results suggest that targeted therapies that can modulate these specific modes of dopaminergic signaling in these distinct striatal subregions could provide improved efficacy in OCD patients that are resistant to SSRI treatment.

3439

Role of the Airway Microbiome in Viral Bronchiolitis Associated Respiratory Failure

Emily Wasserman[1], Stefan Worgall, M.D., Ph.D.[1] and Anurag Sharma, Ph.D.[1]
[1]Weill Cornell

OBJECTIVES/SPECIFIC AIMS: This study aims to determine if a bronchiolitis specific microbiome exists and how it evolves through disease course. Objective 1. Determine the microbiome profile of the airway in virus induced bronchiolitis-associated respiratory failure. Objective 2. Identify changes in the airway microbiome profile through the course of virus induced bronchiolitis associated respiratory failure, and the relationship between microbiome composition and clinical respiratory status. Objective 3. Determine the impact of rhinovirus infection on the lung and stool microbiome in a murine asthma model. METHODS/STUDY POPULATION: Objectives 1 &2: We are conducting a single-center prospective case-control study of patients admitted to the Komansky - Weill Cornell Pediatric Critical Care Unit. Infants less than two years of age with a diagnosis of bronchiolitis requiring intubation and mechanical ventilation are enrolled as subjects. Infants less than two years of age intubated and requiring mechanical ventilation without primary lung pathology are enrolled as controls. To evaluate our primary objective, tracheal aspirates will be collected from both subjects and controls on the day of intubation. We will perform 16s RNA sequencing on the tracheal aspirate samples and compare the resulting microbiomes. To evaluate secondary objective, we will collect tracheal aspirates of our study population on a daily basis and map the microbiome in parallel with objective measures of respiratory status including oxygen index and successful extubation. Both subjects and controls are being enrolled as a convenience sample. Objective 3: Mice, heterozygous for the sptlc2 gene (Spltc2 +/−) demonstrate reduced de-novo sphingolipids and increased airway hyperresponsiveness with methacholine challenge. Airway hyperresponsiveness is a cardinal feature of asthma. This airway hyperresponsiveness is exacerbated in the setting of rhinovirus (Figure 1). Using 16s sequencing, we will examine the lung microbiome of Sptlc2 +/− ad Sptlc2 +/+ at 1- and 7-days following rhinovirus infection. RESULTS/ANTICIPATED RESULTS: This clinical study is currently IRB approved and enrollment is ongoing. We have enrolled 12 subjects and 5 controls. Sample analysis will begin following the 2018-2019 respiratory season, with an anticipated cohort of 20 subjects and 20 controls. With regards to the murine studies, we have demonstrated that the lung microbiome of Sptlc2 +/− and Sptlc2 +/+ mice is similar at baseline (Figure 2) and remains similar following 1-day infection with rhinovirus. We do however, see a distinct change in the microbiome profile of the stool of Sptlc2 +/− mice following rhinovirus infection (Figure 3). Lung analysis at day 7 post infection is pending. DISCUSSION/SIGNIFICANCE OF IMPACT: These studies will lay the groundwork for detailing the functional role of the airway microbiome in bronchiolitis, with the objective of developing new modalities for disease treatment and prevention. In addition our murine studies allow us to view the microbiome in the context of sphingolipid deficiency, providing a potential mechanistic link to rhinovirus and ORMDL3 associated asthma.

3582

Scavenger Receptor Expression is Differentially Affected by DNAzyme-Gold Nanoparticle Conjugates

Cory Sylber[1], Jessica Petree[1], Nusaiba Baker[1], Khalid Salaita[1] and Cherry Wongtrakool[1]
[1]Emory University

OBJECTIVES/SPECIFIC AIMS: Scavenger receptor (SR) surface proteins are highly conserved motifs and are implicated in the uptake of nanotherapies. Gold nanoparticles functionalized with DNAzymes (DzNP) represent a promising novel nanotherapy for lung diseases such as asthma, particularly because they can be delivered directly to the lung. Our lab has been studying the therapeutic potential of a DzNP targeting GATA-3, a master transcription factor regulating Th2 inflammation. Although nanoparticle uptake through scavenger receptors has been described in macrophages in other models, the role of SRs in DzNP uptake in the lung is poorly understood. We hypothesize that scavenger receptors mediate DzNP uptake in alveolar macrophages. To begin examining this hypothesis, we examined whether DzNP exposure and uptake regulates gene expression of MARCO and MSR1, two class A scavenger receptors. METHODS/STUDY POPULATION: Using a silver stain, we measured dose dependent DzNP uptake in murine alveolar macrophages (MH-S). Using qRT-PCR, we measured gene expression of scavenger receptors MSR1 and MARCO in murine alveolar macrophages (MH-S) and after 24 hour exposure to 2251 DzNP, a DzNP targeting GATA-3, and dextran sulfate sodium (DSS), a known SR-A blocker. RESULTS/ANTICIPATED RESULTS: 2251 DzNP uptake in alveolar macrophages is dose dependent. MARCO gene expression levels significantly increase in murine alveolar macrophages when cultured with increasing concentrations of 2251 DzNP (10 pM-2 nM) or DSS 25-200 ug/ml) for 24 hours. However, MSR1 gene expression

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