ACS history. The mean miR let-7c level was 2-fold less in subjects with a history of ACS than in those who had never had ACS ($p<0.05$). A plasma miR let-7c level < 1 (normalized to the control subjects) had a positive predictive value of 0.78 for history of ACS and a sensitivity of 78% and specificity of 71%. Among subjects with a history of ACS, the let7c levels did not correlate with time since the last ACS event. However, among subjects who developed ACS following the baseline samples, higher miR let-7c levels correlated with increased length of time to next ACS event (R=0.8). DISCUSSION/SIGNIFICANCE: Our results in a group of subjects with SCD show that plasma miR let-7c levels are decreased in subjects with a history of ACS. They suggest that miR let-7c may be protective against development of ACS and that measurement of its levels could be a useful biomarker to assess or predict risk for this complication of SCD.

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In silico ADMET optimization and preliminary biologic activity of novel spermine oxidase inhibitors as neuroprotective agents

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OBJECTIVES/GOALS: The goal of this project was to conduct a preliminary assessment of in vivo feasibility early on in the drug-discovery process in an effort to expedite the translation of novel drug scaffolds to potential clinical candidates. The data gathered in this study will be used to direct analog synthesis of our current lead compounds through rational drug design. METHODS/STUDY POPULATION: Based on virtual and physical high-throughput screening efforts and subsequent similarity searching, we identified a set of potent and selective spermine oxidase (SMOX) inhibitors adhering to a common structural scaffold. In order to address potential barriers to in vivo use, we then conducted a robust optimization analysis in an effort to identify analogs with improved drug-like characteristics. Docking simulations to predict binding were performed and visualized using molecular modeling software (MOE and PyMol). ADMET properties were calculated using a variety of software resources including SwissADME and CDD Vault. RESULTS/ANTICIPATED RESULTS: Through these optimization efforts, we were able to successfully identify analogs with improved drug-like characteristics, including increases in predicted CNS penetration, isosteric replacement of metabolically labile functional groups, increased lipophilicity, and elimination of structural attributes suggestive of off-target activity. Analogs were ranked according to predicted binding and properties of in vivo feasibility. Compounds achieving the highest scores were then selected as scaffolds to guide analog synthesis. DISCUSSION/SIGNIFICANCE: Despite evidence implicating induction of SMOX as a mechanism contributing to neuronal pathology, the lack of potent and selective inhibitors with profiles conducive for in vivo use has significantly impeded clinical investigation of this target. In this presentation, rational drug design focusing on translational optimization will be discussed.

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Age-dependent Regulation of Follicle-Stimulating Hormone N-glycosylation in Female Gonadotrope Cells

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OBJECTIVES/GOALS: Age-specific N-glycosylation occurs on follicle- stimulating hormone (FSH) in pituitaries of post-menopausal women and results in a higher ratio of fully glycosylated to hypoglycosylated FSH. Our goal is to identify in vivo the N-glycosylation pathway enzymes and the regulatory mechanisms in gonadotropes of young and old female mice. METHODS/STUDY POPULATION: Pituitaries were isolated from female mice (at 4m and 8m; n=5 per group) carrying an Fshb-Cre transgene on a Rosa mT/mG genetic background and the GFP-tagged gonadotropes were purified by FACS. RNA-Seq analysis, and subsequent qPCR assays were performed on GFP+ cells from pituitaries of female mice at 4m (reproductively young), 8m (reproductively mid age) and â‰¥ 12m (reproductively old) of age. To identify the role of progesterone signaling in age-dependent N-glycosylation in gonadotropes, a gonadotrope-specific knockout of Pgr was achieved. Gonadotropes from these mutant mice at 4-, 8-, and â‰¥12 months of age (n=5 per group) were isolated for qPCR analysis of N-glycosylation enzyme gene expression. Predicted progesterone receptor (PR) promoter binding sequences was performed using JASPAR. RESULTS/ANTICIPATED RESULTS: RNA-seq identified 28 differentially expressed N-glycosylation enzyme-encoding mRNAs in gonadotropes of female mice at 4- and 8-months. Three genes showed significant differences between ages (Man2a1, Man1c1, and B4galt5.), and further qPCR analyses revealed six out of eight genes analyzed showed age-dependent expression, including Man2a1, Man1c1, and B4galt5. The promoters of all N-glycosylation enzyme genes showed strong predicted binding sequences for PR. Further qPCR analysis showed age-and genotype-dependent differences in N-glycosylation enzyme expression in Pgr cKO females, with the most striking differences observed at 13 months, where B4galt5, Man1a2, Mgat5, and Man2a1 were downregulated in Pgr cKO gonadotropes compared to controls. DISCUSSION/SIGNIFICANCE: We identified changes in the N-glycosylation machinery in female mouse gonadotropes and confirmed the age- and Pr-dependent regulation of the corresponding mRNAs. Our results provide insights into the mechanisms at the level of the pituitary by which old age-specific FSH glycoform regulates osteoporosis and weight gain in post-menopausal women.

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