Neuroendocrinology and Pituitary

TOOLS AND MECHANISMS OF REGULATION IN THE ANTERIOR PITUITARY

The Musashi1 RNA-Binding Protein Functions as a Leptin-Regulated Enforcer of Pituitary Cell Fate and Hormone Production

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The pituitary gland is the major endocrine organ that produces and secretes hormones in response to hypothalamic signals to regulate important processes like growth, reproduction, and stress. The anterior pituitary adapts to metabolic and reproductive needs by exhibiting cellular plasticity, resulting in altered hormone production and secretion. The adipokine, leptin, serves a critical role to couple energy status to pituitary function. We have recently reported that the cell fate determinant, Musashi, functions as a post-transcriptional regulator of target mRNA translation in the mouse pituitary and have speculated that Musashi may modulate pituitary cell plasticity. However, the underlying mechanisms governing such pituitary plasticity are not fully understood. Musashi is an mRNA binding protein that is required for self-renewal, proliferation, and to control the differentiation of stem and progenitor cells. We have recently shown that Musashi is expressed in Sox2+ pituitary stem cells and surprisingly, we also found Musashi expression in all differentiated hormone expressing cell lineages in the adult anterior pituitary. The role of Musashi in these mature differentiated cells is unknown. We have observed that a range of critical pituitary mRNAs, including the lineage specification transcription factors *Prop1* and *Pou1f1,* as well as hormone mRNAs including *Tshb, Prl,* and *Gnrhr,* all contain consensus Musashi binding elements (MBEs) in their 3' untranslated regions (3' UTRs). Using RNA electrophoretic mobility shift assays (EMSAs) and luciferase mRNA translation reporter assays we show that Musashi binds to these mRNAs and exerts inhibitory control of mRNA translation. Moreover, we determined that leptin stimulation opposes the ability of Musashi to exert translational repression of the *Pou1f1* and *Gnrhr* 3' UTRs. This de-repression does not require regulatory phosphorylation of Musashi on two conserved C-terminal serine residues. Interestingly in the same cell assay system, Musashi exerts translational activation of the *Prop1* 3' UTR. We observed that this translational activation requires Musashi phosphorylation on the two regulatory C-terminal serine residues, consistent with the requirement for regulatory phosphorylation to drive translational activation of Musashi target mRNAs during *Xenopus* oocyte cell maturation. The distinction between MBEs in 3' UTRs that exert repression (*Pou1f1, Prl, Tshb,* and *Gnrhr*) and the *Prop1* 3' UTR that directs translational activation is under investigation. We propose that Musashi acts as a bifunctional regulator of pituitary hormone production and lineage specification and may function to maintain pituitary hormone plasticity in response to changing organismal needs.

Non-Steroid Hormone Signaling

NOVEL MOLECULAR INSIGHTS IN TO CARDIOVASCULAR AND METABOLIC DISEASE

An Epigenomic Signature in Children Born Small for Gestational Age (SGA) With "Catch-up" Growth Is Present From Birth and Predicts Early Adulthood Pre-Hypertension

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Background: Cardiometabolic conditions in adulthood are more common in children born SGA1. The relationship of the transcriptome (gene expression) and epigenome (DNA methylation) to birth size and the future development of cardiometabolic disease has not been characterized. **Aims:** To identify **I)** the relationship between epigenome at age 0, 7 and 17 years, transcriptome at age 9 years and birth size in a normal population; **II)** links between the transcriptome and epigenome in childhood and adult cardiometabolic risk. **Study Design:** Normal children (n=6487) from the Avon Longitudinal Study of Parents and Children were assigned to groups based on birth size using bodyweight (BW) and gestation and divided into groups using the population 10^{th} centile. Adverse cardiometabolic risk at age 17 years was defined by the National Heart Lung and Blood Institute criteria of prehypertension using systolic and diastolic blood pressure as well as HDL and LDL^2. Blood transcriptome at age 9 and blood epigenome at age 0, 7, and 17 years and were available from 980 and 947 children, respectively. Hypernetworks were used to integrate differentially expressed genes in the transcriptome (DEGs) and differentially methylated points (DMPs) in the epigenome, identifying functional links. Random Forest, a machine learning approach, was used to determine the predictive value of 'omic data presented as the area under the curve (AUC) of the receiver operating characteristic. **Results:** Pre-hypertensive participants at age 17 years were distinguished from normotensive participants and this group was enriched for children born small who caught up by age 7 years (155/611 unhealthy/healthy SGA compared to 1979/12746 in all other BW groups; 1.6-fold, $p<1x10^{-5}$). This group had a greater height velocity during their catch-up period than the normotensive participants (1.2-fold, p=0.027). Hypernetwork integration of 'omic data identified a functional relationship between 55 DEGs at age 9 years and DMPs at age 7 years. Random forest analysis was able to accurately predict the presence of pre-hypertensive young adults from the age 9 transcriptome (AUC: 0.973). Using a gene-level contraction of DMPs which map to the 55 DEGs (i.e. cis-DMPs), we demonstrated accurate classification of pre-hypertensive young adults from their blood methylome at age 0 (AUC [95% CI]: 0.92 [0.89-0.95]), 7 (0.90 [0.87-0.93]), and 17 (0.91 [0.88-0.94]) years. **Conclusions:** Through the integration of transcriptome and epigenome, we have identified a set of genes with an epigenomic and transcriptomic signature which predict pre-hypertension in children born SGA who catch up. Specifically, we have shown that the associated epigenomic signature tracks from birth to early adulthood, indicating the possibility of