Persistent Epigenetic Effects of Dietary Imbalance

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Acknowledgements

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Sources of Interindividual Epigenetic Variation

Genetic & Epigenetic Inheritance

Developmental Stochasticity

Environmental Influences

Age
Methyl Supplementation Changes Coat Color by Increasing $A^{vy}$ Methylation

Wolff et al 1998 *FASEB J*
Waterland & Jirtle 2003 *Mol Cell Biol*
Significance of $A^{vy}$ Study

• Demonstrated causal pathway:
  maternal nutrition – epigenetic change – phenotypic change

• Highlighted the potential involvement of metastable epialleles
Metastable Epiallele (ME)

“An allele at which the epigenetic state can switch and establishment is a probabilistic event. Once established, the state is mitotically inherited.”

Rakyan et al *Trends in Genetics* 2002

*Agouti viable yellow*  
*Axin fused*
At MEs, Maternal Environment Affects Establishment of Epigenotype

• Maternal methyl supplementation increases DNA methylation
  - At $A^{vy}$ (Waterland and Jirtle 2003 Mol Cell Biol)
  - At $Axin^{Fu}$ (Waterland et al 2006 Genesis)

• Maternal bisphenol A exposure decreases DNA methylation at $Cabp^{IAP}$
  - Dolinoy et al. 2007 PNAS
Does epigenetic variation at MEs contribute to human phenotypic variation?
Characteristics of Murine Metastable Epialleles

- Interindivudal variation in DNA methylation that is
  - Systemic (not tissue-specific)
  - Stochastic (not due to genetic variation)

- Establishment of DNA methylation affected by maternal nutrition before/during pregnancy
Seasonal Variation in Rural Gambia
Seasonal Variation in Rural Gambia
Seasonal Variation in Rural Gambia

Rainy Season - Hungry

Dry Season - Harvest
Season of Conception in Rural Gambia Affects DNA Methylation at Putative Human Metastable Epialleles

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Maternal nutrition at conception modulates DNA methylation of human metastable epialleles

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Genome-Wide Screen for Human MEs

- Studied peripheral blood lymphocytes (PBL) and hair follicle (HF) from each of two Caucasian males

- Genome-wide bisulfite sequencing (Bisulfite-seq)
  - 26x average coverage per sample
  - Estimated % methylation in 6.2 million 200bp bins containing at least 2 CpG sites (85% of all CpGs)

Kunde-Ramamoorthy, et al 2014 *Nucleic Acids Res*
109 Top Candidate MEs Show No Evidence of Genetic Variation, and High CpG Density
We looked for genes associated with multiple ME bins. *VTRNA2-1* was a top hit.
What is known about VTRNA2-1? (aka pre-miR-886, aka nc886)

- 102-nucleotide, ubiquitously expressed, cytoplasmic RNA
- Neither a vault RNA nor a canonical pre-microRNA
- Genomically imprinted
- Inhibits cell proliferation by inhibiting phosphorylation of protein-kinase – RNA inducible (PKR)

Lee et al. 2011 RNA
Paliwal et al. 2013 PLoS Genetics
Methylation of *VTRNA2-1* Predicts Survival in Acute Myeloid Leukemia Patients

Treppendahl et al 2012 *Blood*
Validation of VTRNA2-1 Metastable Epiallele

• Systemic interindividual variation confirmed in post-mortem liver, kidney, brain

• No effects of genetic variation on DNA methylation at VTRNA2-1 (Zhang et al 2014 Hum Mol Genet)

• Effects of periconceptional nutrition confirmed in rural Gambians
In an Independent Genome-Scale Screen (450k array) for Differentially Methylated Regions (DMRs) According to Season of Conception, *VTRNA2-1* was THE Top Hit!

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PBL DNA from 120 children studied by Illumina 450k arrays

Silver & Kessler, et al Genome Biol 2015
450k Data – Season of Conception Effect Spans the Entire VTRNA2-1 Imprinted DMR

PBL DNA from 120 children studied by Illumina 450k arrays

Silver & Kessler, et al Genome Biol
Individual Methylation at \( VTRNA2-1 \) is Highly Stable

\[ y = 0.91x + 3.6 \]
\[ R^2 = 0.83 \]

\( VTRNA2-1 \) methylation in PBL by pyrosequencing; \( n=55 \) Gambian children.
Maternal Periconceptional Nutrition Predicts VTRNA2-1 Hypomethylation in Her Infant

Silver & Kessler, et al Genome Biol 2015
Maternal Periconceptional Nutrition Predicts VTRNA2-1 Hypomethylation in Her Infant

Hypomethylation defined as <40% methylation by pyrosequencing. Complete data on ~120 infants.

Silver & Kessler, et al Genome Biol 2015
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