Maternal obesity increases offspring risk for obesity and metabolic disease
What We Don’t Know!

- What are the consequences of exposure to maternal obesity during pregnancy and lactation on fetal metabolic systems and neonatal adiposity? Liver, WAT, bone marrow, appetite control.
- What are the potential mediators of these effects? Maternal lipids, inflammation, breast milk? Microbiome?
- Is there a role for maternal nutrition in triggering epigenetic factors leading to NASH? DNA methylation/acetylation, in early infancy.
- What are the public health consequences of exposure to maternal obesity on the childhood obesity epidemic and the evolution of pediatric NASH?

Collaborative Research
Oregon National Primate Research Center,
University of Colorado

Long-Term Goal:
- To develop a Non-Human Primate Model to study the effects of Maternal Diet, Obesity and GDM on the development of metabolic systems (liver, muscle, fat, heart, brain) in utero and the effects on infant behavior and post-natal disease pathways.

Maternal Obesity is Non-Alcoholic Fat
These metabolic abnormalities will persist despite switching to a healthy diet up to 1 year of age.

**Juvenile Hypotheses**

1. Maternal high fat diet exposure will result in hepatic steatosis, inflammation and insulin resistance in juvenile offspring at 1 yr of age.

2. These metabolic abnormalities will persist despite switching to a healthy diet up to 1 year of age.

3. Mechanism may involve persistent activation of inflammatory pathways in liver.
Programmed effects in juvenile

- Liver
- Lipid export
- Gluconeogenesis
- Lipid oxidation
- Mitochondrial dysfunction
- Inflammatory mediators
- Kupffer cell activation
- ROS
- Mitochondria (CS activity)
- Hypoxia, dyskinesias
- ILS/ROS
- Triglycerides
- NO/ROS
- Insulin Action

Does hepatic steatosis occur in infants of obese-GDM Mothers? Infant “Papoose” for whole body MRI

MRI for Neonatal Fat Measurement
- Magnetic Resonance Spectroscopy (MRS)
- Cohort of 25 infants
  - 13 born to normal weight controls
  - 12 born to obese mothers with GDM
68% Increase in Hepatic Fat in Neonates Born to Obese GDM mothers

- Can Excess Maternal Fat Delivery to the Fetal-Placental Interface Result in the Genesis of NAFLD?

Visceral fat = less than 0.1% of total fat and independent of subcutaneous fat.

Consequences of Maternal Overfeeding on Fetal Liver

- Fetal Hepatic Fat Accumulation
- Oxidative Stress
- Inflammation
- Gluconeogenesis

- Extraction of Excess FFA/TG Delivery
- Placental Inflammation
- Placental Nutrient Transfer

- Recruitment and Activation of Bone Marrow WBC Precursors
- Lifelong Increased Risk of a Proinflammatory Response to Overnutrition

US adults: 20-30%
Obese adults: 60%
US kids 9-19*: 17%
Obese kids: 55%

Host remodeling of the gut microbiome and metabolic changes during pregnancy.

Microbiome

- Best studied is gut microbiome
  - 100 trillion microorganisms (10x more than the total of all other cells in the whole body)
  - Using 16S rRNA sequences, composition of microbiomes can be determined
  - >90% from 2 phyla: Firmicutes and Bacteriodetes
  - Human gut microbiome established by 1 year of age and depends on genetics, mode of delivery (vaginal vs C-section), and form of infant feeding
  - Later influenced by demographics, diet, and lifestyle
  - Evidence supports a role in inflammation, intestinal cell health and hormone production, efficiency of energy harvest from food, and appetite.

Gut microflora may stimulate hepatic fat deposition and promote NASH through several mechanisms:

1. It promotes obesity by improving energy yield from food.
2. It regulates gut permeability, low-grade inflammation and immune balance.
3. It modulates metabolism-genesis directly in the liver.
4. It regulates bile acid metabolism.
5. It increases ethanol production by bacteria—ROS and mitochondrial function.
Maternal Phenotype
• Normal weight
• Overweight/obese
• Type 2 Diabetes
• Gestational Diabetes

Maternal Microbiome

Infant Microbiome

Infant Adiposity

4 groups of subjects

4-6 weeks prior to delivery
2 weeks after birth
4 months after birth
1 year post-partum

Specimen Processing

• Bacterial DNA extraction
• 16s rRNA (V1V2) amplification and sequence analysis (Illumina Miseq) of all samples
• Sequence sorting by OTU (phyla, etc) and relative abundance calculated (Explicet)
• 2 week infant stool: Shotgun sequencing with sequence classification/analysis using MG-RAST.
• SCFA analysis (Acetate, Butyrate, Propionate by MS-MS.)
**Importance of γ-Proteobacteria**

- “Pioneering Bacteria”, marker of transition to mature MB. Low abundance found in infant stool in Premature neonates. Increases on HFD.
- May have disproportionate impact on intestinal microenvironment (LPS, oxygen tension, intestinal mucous production).
- Microenvironment changes may create conditions for critical establishing TLR4 based immunity, that persists into childhood?

**Question:**

Are these changes meaningful biologically?

**From Bedside (or toilet) to Bench: Germ Free Mice**

Hypothesis: the reduction in the early pioneering γ-proteobacteria in obese offspring may allow a more pro-inflammatory gut to develop and SCFA may influence body fat in GF mice
Increased Pro-inflammatory shift in plasma cytokines –hepatic portal vein.

ER stress and heightened susceptibility for inflammatory processes

Treg cells reduced in Liver in mice with Ob infant microbiome
Surprise: Bile acid enzymes and BA receptor FXR are induced in mice with MB from Ob infants.

Figure 1. Relationship between a high-fat diet, imbalanced gut microbiota, and host pathophysiology.

Future Risks for NAFLD

Genetic Risk
- PNPLA3

Gestational Risk

Lifestyle Risk
- Excess Adiposity
- Metabolic syndrome
- Diet (Fructose, n-3 PUFA)

“First Hit”
- Hepatic Lipid Accumulation

“Second Hit”
- Oxidative Stress
- Hepatocyte Injury
- Inflammation
- Fibrosis

Maternal Prebiotic and Probiotic Oligosaccharide Formulations feeding Cesarean delivery Perinatal antibiotics Altered SCFA - Pathway and Metabolites Immune Cell Recognition Increased energy extraction Altered BA pathway Inflammation, Appetite, Excess adiposity BMI GWG Diet GDM Gammaproteobacteria Bacteroidetes Bifidobacteria Lactobacilli Maternal Dysbiosis Infant Dysbiosis Altered SCFA Inflammation Increased energy extraction Increased risk of: Later life obesity Immunological dysfunction NAFLD → NASH

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