IBD and the Gut Microbiota

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Objectives

- Discuss etiologies of IBD
- Very early onset IBD
- Discuss environmental impact on the gut microbiota
- Role of gut microbiota in IBD
- Role of diet in IBD
IBD Susceptibility

Adapted from Inflamm Bowel Dis 2010:16;152
Identification of Disease Associated Pathways

<table>
<thead>
<tr>
<th>Innate immunity</th>
<th>Adaptive immunity</th>
<th>Epithelial integrity</th>
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</thead>
<tbody>
<tr>
<td>\textit{NOD2}</td>
<td>\textit{Cytokine signalling}</td>
<td>\textit{SLC22A5}</td>
</tr>
<tr>
<td>\textit{ATG16L1}</td>
<td>\textit{IL23R}</td>
<td>\textit{ZNF365}</td>
</tr>
<tr>
<td>\textit{IRGM}</td>
<td>\textit{IL12B}</td>
<td>\textit{NKX2-3}</td>
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<tr>
<td>\textit{LRRK2}</td>
<td>\textit{STAT3}</td>
<td>\textit{PTGER4}</td>
</tr>
<tr>
<td>\textit{TNFSF15}</td>
<td>\textit{JAK2}</td>
<td>\textit{MUC19}</td>
</tr>
<tr>
<td>\textit{IL12B}</td>
<td>\textit{IL17}</td>
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</tr>
<tr>
<td>\textit{ITLN1}</td>
<td>\textit{IL18RAP}</td>
<td></td>
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<tr>
<td>\textit{PTPN2}</td>
<td>\textit{CCR6}</td>
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<td></td>
<td>\textit{TNFSF15}</td>
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</table>
Very Early-Onset IBD

- Diagnosed <5
- Frequently more severe disease
- Often unresponsive to standard therapy
- Thought to have more significant genomic contribution
- Frequently missed by GWAS approaches
- Role of primary immunodeficiency
Single gene defects and VEO-IBD

- Single gene defects have been identified in the VEO IBD population
- Evidence suggests there are distinct pathways involved
- These pathways have responded to directed therapeutics
VEO-IBD Specialty Clinic

• **Multidisciplinary**: patients with VEO-IBD will be examined by GI, Immunology and nutrition

• New technology will be utilized to study and tailor therapy:

• **Genomics and Microbiome**: Samples for genomic evaluation and fecal specimens for gut microbiome studies will be obtained
Etiologic Theories in Inflammatory Bowel Disease

- Genetic Predisposition
- Mucosal Immune System (Innate/Adaptive)
- Environmental Triggers (Lumenal Bacteria, Infection)
You Are Only 10% Human

= $10^{12}$ to $10^{13}$ Cells

The Gut Microbiota
- Genome size of microbiota at least 100-fold greater than human
- Large number of species present, most uncultured

= $10^{13}$ to $10^{14}$
Intestinal Bacteria
Host-Microbial Mutualism of the Gut

**Host benefits to the bacteria**
- Provides a unique niche
- Intestinal mucus provides a source of nutrition

**Bacteria benefits to the host**
- Fermentation of indigestible carbohydrates to assist digestion
- Biotransformation of conjugated bile acids
- Urease activity participates in nitrogen balance
- Synthesis of certain vitamins
- Metabolize drugs
- Education of the mucosal immune system
Highly Parallel Pyrosequencing (454 technology)

DNA extraction of intestinal content

PCR with 16S rDNA primers

400 bp

c TAGACTCAGCTCATCCTTGAACCTGAGTACGATGACTCTAGTCAGTTCCACTG

454 sequence barcode 16S rDNA sequence

Analysis of multiple samples in the same run

454 Pyrosequencing

Gut Microbiota Development

Transition to the highly distinct, highly differentiated adult microbiota

Colonization of the gut begins at birth

Elements of Modern Lifestyle Lead to Changes in Gut Microbiota

• Improved sanitation
• Less crowded living conditions
• Decline in parasite and *H. pylori* infections
• Vaccinations

• Increased antibiotic use
• Sedentary lifestyles
• Caesarean section
• Refrigeration
• Food processing
• Diet changes
Diet and the Gut Microbiota
Bacterial Composition of the Gut Change with Long term Diet

The *Bacteroides* enterotype,
Highly associated with animal protein and saturated fats which suggests that meat consumption is associated with a Western diet

The *Prevotella* enterotype,
High values for carbohydrates and simple sugars indicating association with a carbohydrate-based diet more typical of agrarian societies
Impact of Diet in Shaping Gut Microbiota Revealed by a Comparative Study in Children from Europe and Rural Africa

African Diet: High Fiber and carbohydrate, low animal fat and protein

European Diet: High animal fat and protein, low fiber

De Filippo C, et al. PNAS 2010: 14691-96
What is the clinical relevance of the gut microbiota to disease?
## Intestinal Microbiome in Disease

<table>
<thead>
<tr>
<th>Aberration</th>
<th>Most relevant observations and potential correlation</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>Crohn’s disease</td>
<td>Diversity decrease – reduced <em>F. prausnitzii</em></td>
<td>Kaser et al. 2010(^51); Sokol et al. 2009(^52); Willing et al. 2010(^53)</td>
</tr>
<tr>
<td>Ulcerative colitis</td>
<td>Diversity decrease – reduced <em>A. muciniphila</em></td>
<td>Png et al. 2010(^54); Kaser et al. 2010(^51); Lepage et al. 2011(^55)</td>
</tr>
<tr>
<td>Irritable bowel syndrome</td>
<td>Global signatures – increased <em>Dorea</em> and <em>Ruminococcus</em></td>
<td>Salonen et al. 2010(^36); Saulnier et al. 2011(^56); Rajilić-Stojanović et al. 2011(^13)</td>
</tr>
<tr>
<td><em>Clostridium difficile</em></td>
<td>Strong diversity decrease – presence of <em>C. difficile</em></td>
<td>Grehan et al. 2010(^57); Khoruts et al. 2010(^58)</td>
</tr>
<tr>
<td>infection</td>
<td>Variation in <em>Bacteroides</em> spp. – increased fusobacteria</td>
<td>Sobhani et al. 2011(^59); Wang et al. 2012(^60); Marchesi et al. 2011(^61)</td>
</tr>
<tr>
<td>Colorectal cancer</td>
<td>Altered diversity – specific signatures</td>
<td>Stsepetova et al. 2007(^62); Bisgaard et al. 2011(^63); Storrø et al. 2011(^64)</td>
</tr>
<tr>
<td>Allergy/atopy</td>
<td>Altered composition, notably in small intestine</td>
<td>Nistal et al. 2012(^65); Di Cagno et al. 2011(^66); Kalliomäki et al. 2012(^67)</td>
</tr>
<tr>
<td>Celiac disease</td>
<td>Signature differences</td>
<td>Vaarela 2011(^68); Giongo et al. 2011(^69); Brown et al. 2011(^70)</td>
</tr>
<tr>
<td>Type 1 diabetes</td>
<td>Signature differences</td>
<td>Larssen et al. 2010(^71); Wu et al. 2010(^72); Kooette et al. 2012(^73)</td>
</tr>
<tr>
<td>Type 2 diabetes</td>
<td>Specific bacterial ratios (<em>Bacteroidetes</em>/<em>Firmicutes</em>)</td>
<td>Ley et al. 2006(^74); Turnbaugh et al. 2009(^10); Musso et al. 2011(^75)</td>
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<tr>
<td>Obesity</td>
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The Impact of Environment vs. Host Genotype on the Gut Microbiota Associated with Disease

• Increased Incidence

• Geographic distribution
  – Clustering in industrialized nations

• Immigration studies
  – Adoption of disease risk of the host country within 1 or 2 generations

• Genomic advances
  – Contribution of host genetics to the risk of disease development is significantly less than 50%
Trends in Incidence of IBD

N. America/Europe

Trend not explained by genetics, but environmental changes (Westernized diet, gut microbiome)
What is the Clinical Evidence that bacteria plays a role in IBD

• Inflammation occurs predominantly in the terminal ileum and colon, where the greatest concentrations of bacteria are found

• Antibiotics can be used as a treatment for IBD

• Surgical diversion of the fecal stream is an effective treatment for Crohn disease
  • Inflammation is known to recur upon restoration of the fecal flow
Associations of Environmental Factors with New Onset IBD

• Infectious gastroenteritis
• Early antibiotic use has been associated with IBD
  – Tetracycline
  – Oral antibiotics for ear infection
Increase in IBD Incidence for Patients Exposed to Salmonella/Campylobacter
“Bacterially”-Generated Phenotypes

IL-10−/− → Germ-Free

IL-10−/− → Commensal Bacteria

IL-10−/− → E. faecalis

IL-10−/− → E. coli

Dysbiosis of Gut Microbiota

Potentially injurious species in susceptible hosts

- *Bacteroides vulgatus*, *B. thetaiotaomicron*
- *Escherichia coli* (adherent/invasive)
- *Enterococcus faecalis* (nonpathogenic)
- *Klebsiella pneumoniae*
- *Fusobacterium varium*
- *Helicobacter hepaticus* and other intestinal species

Protective species

- *Lactobacillus* species
- *Bifidobacterium* species
- *Escherichia coli*
- *Bacteroides thetaiotaomicron*
- *Faecalibacterium prausnitzii*
- *Bifidobacterium animalis*
- *Myobacterium Avium* subspecies *Paratuberculosis* (MAP)

Determinants of Gut Microbiota

- Environment
- Host Phenotype
- Genetics
- Antibiotics
- Inflammation
- Diet
Role of Nutrition in Pathogenesis of IBD

• Increased risk of IBD
  – Diet high in refined sugars and fat
  – High ratio n-6 to n-3 polyunsaturated fatty acids
  – Lack of breastfeeding

• Decreased risk of IBD
  – Diet high in fruits, vegetables, and fiber

Bacterial populations in pediatric IBD subjects on semi-elemental diet (16S rDNA sequencing)

Conclusion:
Rapid change in gut bacterial populations upon initiating diet
Fecal Transplant-The Future?

- Used as therapy for C. difficile Colitis
- More research needed for IBD

Microbes play crucial role in human health, researchers discovering

Scientists are becoming more convinced that modern trends — diet, antibiotics, obsession with cleanliness— are disrupting this delicate balance, contributing to disease.
Summary

• VEO-IBD is a distinct population
  – requires careful evaluation and perhaps different therapy

• Dramatic rise in common diseases
  – associated with both genetic and environmental factors

• Effect of the gut microbiota in development of IBD

• Effect of diet in development of IBD, and potential therapy
Thank You