The role of the microbiota in Inflammatory Bowel Disease
The Bugs within Us

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Outline

• Background on the human microbiome
• Review what we know about the gut microbiome
• The relationship between IBD and the gut microbiome
• Microbial based treatments of IBD
Biology 101

- **Cells** are fundamental working unit of living organisms
- **DNA** is the instruction manual for all cellular activities
- The **genome** is an organism’s complete set of DNA
- **Genes** carry specific instructions on how to make proteins
- **Proteins** perform most of life functions

Definitions

- **Microflora/ Microbiota**
  - Group of microorganisms localized to specific location
  - ~80% are uncultured
- **Microbiome**
  - The microbiota including its genes
- **Phylotype**
  - Characterization of bacteria based on its evolutionary history

http://www.rsc.org/images/b786657e-ga_tcm18-97409.jpg
The Human Genome Project, 1990-2003

- Ultimate goal to generate a reference DNA sequence for the human genome and to identify all human genes
The Human Microbiome Project, 2007 -

21st century – human microbiome project

Human Microbiome Project

- Characterize microbial communities at different sites on the human body
- Understand the role of microbes in human health and disease
Number of publications related to the intestinal microbiota in the last two decades, per year.

Sekirov I et al. Physiol Rev 2010;90:859-904

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Bacterial Identification

http://www.bio.ku.dk/nuf/images/anaerobic_incubator.jpg
http://phil.cdc.gov/phil_images/20021108/7/PHIL_2297_lores.jpg
You are more microbial than “you”!

- The human body is comprised of 10 trillion cells
- There are 10 times that many microbes within the human body

The human genome itself is currently estimated to encode for only 23,000 genes in comparison
A Partnership

- We have co-evolved with microorganisms
- Our genetic make-up is a combination of human and microbial DNA -SUPER-ORGANISM

Human Bacteria Phylotypes

Firmicutes
- Listeria
- Lactobacillus
- Enterococcus
- Clostridium

Bacteroidetes
- Bacteroides
- Flavobacteria
Gut Bacteria Composition

- Approximately 500 bacterial species in the gut
- 90% of bacteria come from 30-40 species
- Human gut microbiota dominated by 2 phyla
  - Bacteroidetes
  - Firmicutes

Varied habitat for bacteria in gut

B. Sartor Gastroenterology 2008
Individualized Microbial Landscape

- We are born germ-free

- The microbes that live in our gut come from the environment
  - Infant diet, hygiene, medications, breast-fed vs. formula-fed

- Genetic influence
  - Identical twins have similar bacterial communities

The Human Microbiome

Nature 2007; 449: 804-810
LER1 would be nice to have figure showing development of microbiota and contrib factors. Baby to adult with factors vag vs c sect, host genotype, host lifestyle, abic, diet, disease, etc.

Laura E Raffals, 2/24/2012
Enterotypes

- Correlation found between enterotypes & host age and BMI

Artistic impression of the three human gut types. (EMBL/P. Riedinger)

Bacteria have important role in the GI ecosystem

- Extract calories and nutrients from indigestible complex carbohydrates
- Synthesize vitamins and short chain fatty acids
- Protect against injury to the cells lining the colon
- Instruct our immune system
- Regulate fat storage
- Stimulate intestinal angiogenesis

Diet & Microbiota

Complex plant polysaccharides

Diet

Gut microbiota

SCFA

Colonic epithelium

Main energy source
Less oxidative DNA damage
Regulation of proliferation
Maintenance of barrier function
Tumor suppression
Cytokine production

Receptors and mechanisms

GPR43
MCT1
Histone demethylase inhibition
GPR109A
GPR41

Immune system

Enhanced ROS burst
More phagocytosis
Induction of apoptosis
Modulation of recruitment
Cytokine production

Nature Immunology 12, 5-9, 2011

We are not at war with our Bugs!
Human-Microbe Balancing Act


Microbiota & Disease
Pathogenesis of IBD

- Immune Response
- Genetic Background
- Environment

Microbes
Clinical Arguments for Role of Bacteria in IBD

- Diversion of the fecal stream diminishes severity of disease
- Therapeutic benefit of antibiotics
- Antibodies against micro-organisms in patients with IBD (ASCA, anti-OmpC, Anti-I2, Anti-CBir1)
- Efficacy of probiotics in pouchitis and UC

Role of commensal enteric bacteria in experimental intestinal inflammation

IBD & the Microbiota

Crucial groups of bacteria missing

Predominance of bad bacteria

Diet, microbial composition and regulation of the immune system

- Host genetics
- Maternal transfer and early colonization
- Antibiotics and medications
- Infection
- Inflammation
- Stress
- Hygiene
- Age

Microbial composition

Symbiosis
- SCFA
- PSA
- PTOH
(and so on)

Dysbiosis

Immune regulation

Homeostasis

Immune dysregulation

Virulence factors

Inflammation

Nature Immunology 12, 5-9, 2011
LER2  make slide with various theories of link of microbes and ibd
causative (cause vs effect)
dysbiosis

then another slide on treatment of ibd
probiotics, prebiotics, symbiotics, antibiotics

also need slide on enterotypes
Laura E Raffals, 2/24/2012
Microbiota and IBD

- The fecal microbiota in patients with IBD differs from that seen in healthy individuals
- Decreased diversity of fecal microbiota seen in IBD patients
- Increase in mucosally-adherent bacteria in IBD patients
- IBD associated genes (NOD2, ATG16L1, and OCTN2) have important roles in the human interaction with bacteria

Packey D, Sartor B. Curr Opin Infect Dis 2009; 22(3):292-301

IBD Patients Have Alterations in Their Microbiota

- Study of mucosal bacteria:
  - 20 CD and 15 UC
  - 14 healthy controls (HC)
  - 3 bx from each site
  - Inflamed (I) vs. non-inflamed (NI)
- Bacteria profiles stable w/in individual
- UC have more bacteria than CD and HC
- Inflamed tissue have more bacteria
- CD and UC have different microbial populations
- CD have more abundant unclassified Bacteroidetes

Gut bacteria composition & IBD

- **Cause or Effect?**
  - Blooms of Proteobacteria
  - Decrease in Firmicutes
  - Decrease in Bacteroidetes
  - *Similar patterns seen in other non-IBD inflammatory states.*

Infectious agents linked to IBD

- **No individual microbe has been definitely linked to IBD**

- **Organisms of interest**
  - *Mycobacterium avium paratuberculosis*
  - Adherent-invasive *Escherichia coli*
  - *Pseudomonas*
  - *Listeria monocytogenes*
  - *Mycoplasma*
  - *Chlamydia*
  - *Coxiella*
  - *Streptococci*
  - *Helicobacter pylori*
  - *Yersinia pseudotuberculosis*
  - *Saccharomyces cerevisiae*
  - *Measles virus*

*Eckburg PB., Relman, DA. CID. 2007; 44*
**Mycobacterium avium paratuberculosis**

- Causative organism of Johne disease, a chronic inflammation of the ileum occurring in mostly in domestic livestock
- Similarities between Johne disease and Crohn’s disease have raised concern that MAP is a cause of Crohn’s
- MAP is present in our food chain and this organism can be found in intestine of CD patients (causal vs coincidental???)
- Anti-MAP not effective for treatment of CD

**Adherent invasive Escherichia coli**

- AIEC have been isolated from ileal biopsies in patients with ileal Crohn’s disease
- AIEC adhere to the intestinal epithelial cells and invade the cells
- AIEC may be involved in formation of granulomas
- Unclear if this infection is the cause of CD or secondary to CD
Challenges of Studying Microbiota & IBD

• Most studies performed after the development of IBD
  – What comes first?
• Diet and the microbiota
• It is difficult to control for disease activity, disease behavior, genotype, medications
• Inherent differences in microbial diversity among individuals

Can “bugs” treat IBD?

• Probiotics
• Prebiotics
• T. suis (whipworm)
• Fecal transplant
Probiotics

• Live organisms, usually bacteria, that are similar to beneficial organisms in the gut

• First probiotic food was acidophilus milk, sold in 1920s as supplement for digestive problems

• Foods containing probiotics
  – Yogurt, fermented and unfermented milk, miso, natto, kimchi, tempeh, sauerkraut, kefir

Probiotics

• Despite popularity, few have been proven effective treatment for IBD
• Escherichia coli Nissle 1917
  – Maintains remission in adults with UC\textsuperscript{1,2}
• Lactobacillus GG
  – Maintains remission in adults with UC\textsuperscript{3}
• VSL#3
  – Prophylactic against onset of pouchitis\textsuperscript{4}
  – Prevents recurrence of pouchitis\textsuperscript{5,6}

*Studies in CD have shown no benefit of probiotics over placebo.

Prebiotics
(non-digestible carbohydrates)

• Indigestible nutrients used as an energy source by “good” bacteria in the gut
• Foods containing prebiotics includes
  — Artichokes, bananas, barley, berries, chicory, dairy, flax, garlic, honey, leeks, legumes, onions, wheat and whole grains
• On their own, can only enhance growth of bacteria already present in gut

Fecal Bacteriotherapy (FB)

• Aka:
  — Fecal Microbiota Transplantation (FMT)
  — Human probiotic infusion
  — Stool transplant
  — Fecal transfer
• Ultimate probiotic
• Identify “healthy donor”
• Collect stool from donor
• Screen donor and stool for infections
• Transfer fecal matter by enema, colonoscopy, or nasogastric tube to patient

http://www.cafecolon.jp/coffee_enemaimg/good_colon.gif
Fecal Microbiota Transplant

• Stool transplant shown effective as treatment for recurrent C. difficile
  – Found effective in 3 studies, including a long-term follow-up of 77 patients
• Case reports of FMT for treatment of IBD in Australia
• FMT may help re-establish depleted bacterial species

Trichuris suis
pig whipworm

• a roundworm, similar to human whipworm
• common mild intestinal pathogen of pigs
• cleared rapidly from human host, making infection unlikely
**T. suis**

- rationale for effectiveness based on hygiene theory
- in animal models, helminths can suppress disease in asthma, multiple sclerosis, type 1 DM, and arthritis
- ingestion of live *T. suis* eggs effective in treatment of IBD in small clinical trials
- Ingestion of *T. suis* may cause moderate to severe gastrointestinal reaction, likely reflecting immune response to the parasite

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**Conclusions**

- Microbes play a crucial role in health and disease
- Microbes likely are important in the pathogenesis of IBD, although this relationship is still not clear
- Probiotics have a limited role in the treatment of IBD
- Hope of future therapies for IBD aiming to manipulate the human gut microbiota
Should we be at war with our bacteria?

SUPPORT BACTERIA!

it's the only culture some people have