Section 11

Bacterial Communities and Interactions
Section 11 Learning Goals

• In a hypothetical symbiotic relationship:
  – Be able to describe a potential metabolic interaction.
  – Be able to describe a potential gene regulatory event that results from the interaction.
  – Be able to describe a potential negative or positive outcome for the host.
• Identify two ways in which the relationship of the gut microbiome differs from *Agrobacterium* in terms of the symbiotic relationship with the host. Identify two ways in which the relationships are similar.
• Be able to distinguish correlation from causation in a given experiment or set of experiments.
• Explain what is meant by a metagenome.
• Be able to design an experiment to identify bacteria present in the gut.
• Be able to identify the limitations of your approach in the experiment above.
Fecal microbiota transplantation for recurrent *C. difficile* infection: Ready for prime time?

**ABSTRACT**

Recurrent *Clostridium difficile* infection has been a major challenge for patients and clinicians. Recurrence of infection after treatment with standard antibiotics is becoming more common with the emergence of more-resistant strains of *C. difficile*. Fecal microbiota transplantation is an alternative treatment for recurrent *C. difficile* infection, but it is not yet widely used.

**KEY POINTS**

Fecal microbiota transplantation involves instilling gut microbiota from a healthy donor into the diseased gut of a patient who has recurrent or recalcitrant episodes of diarrhea despite antibiotic treatment for *C. difficile* infection. The instillation can be done via nasogastric tube, endoscope, or enema.

Donor screening is necessary to prevent transmission of communicable diseases to the recipient.

Recently published studies indicate that this procedure is effective for treating recurrent *C. difficile* infection. Randomized clinical trials to assess its efficacy and safety are underway.

The field of microbiota therapy is rapidly progressing. More physicians are learning to embrace the concept of fecal microbiota transplantation, and patients are beginning to overcome the “yuck factor” and accept its benefits.

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If you had a serious disease, would you agree to an alternative treatment that was cheap, safe, and effective—but seemed disgusting? Would you recommend it to patients? Such a disease is recurrent *Clostridium difficile* infection, and such a treatment is fecal microbiota transplantation—instillation of blended feces from a healthy donor (ideally, the patient’s spouse or “significant other”) into the patient’s colon to restore a healthy population of bacteria. The rationale behind this procedure is simple: antibiotics and other factors disrupt the normal balance of the colonic flora, allowing *C. difficile* to proliferate, but the imbalance can be corrected by reintroducing the normal flora.

In this article, we will review how recurrent *C. difficile* infection occurs and the importance of the gut microbiota in resisting colonization with this pathogen. We will also describe the protocol used for fecal microbiota transplantation.

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*C. difficile* infection often recurs

*C. difficile* is the most common cause of hospital-acquired diarrhea and an important cause of morbidity and death in hospitalized patients. The cost of this infection is estimated to be more than $1.1 billion per year and its incidence is rising, partly because of the emergence of more-virulent strains that make treatment of recurrent infection more difficult.

*C. difficile* infection is characterized by diarrhea associated with findings suggestive of pseudomembranous colitis or, in fulminant cases, ileus or megacolon. Recurrent *C. difficile*...
Types of interactions

• Symbiosis – living together (Greek); often used to mean mutual benefit or dependence
• Mutualism – relationship that is beneficial to both partners
• Commensalism – relationship that is beneficial to one partner and neither beneficial nor detrimental to the other (in reality, it probably means that we simply haven’t figured out the benefit to the other one….mutualisms likely dominate in nature)
• Parasite – microbe that is detrimental to host
• Pathogen – microorganism that causes disease of host (plant or animal, sometimes other microbe)
• Host – organism that supports microbe’s lifestyle (can be beneficial or harmful relationship)
• Population—a group of members of same species; often genetically identical
• Community—multi-species assemblage
Examples of microbial interactions

Microbes of the gut (humans, termite, ruminants, etc.)
Examples of microbial interactions

Squid light organ colonized by *Vibrio fischeri*
Examples of microbial interactions

Biofilms
Examples of microbial interactions

Lichens
Examples of microbial interactions

Leaf cutter ants farm fungi and protect it by housing antibiotic-producing bacteria
Examples of microbial interactions

[Rhizobium spp. nodule formation on plants](http://en.wikipedia.org/wiki/File:Rhizobia_nodules_on_Vigna_unguiculata.jpg)
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Examples of microbial interactions

• 1. *Agrobacterium*. Injects DNA into host plant cells, resulting in proliferation of the host cells (and tumor formation).

• 2. Gut microbiome. All humans contain a community of microbes in their guts. Current research indicates that the community composition can affect the health of the host.
Example 1: *Agrobacterium* induces tumors in plant hosts

Illustration: Margaret Senior

Photo: University of California Statewide IPM Project, J.K. Clark, photographer
• Bacteria enter at plant wound site; they sense compounds released by the plant upon injury and respond by initiating transcription of genes required for tumor formation in the plant.

Expression of *vir* genes leads to transfer and integration of T-DNA into plant genome. T-DNA encoded genes for plant growth and opine production are expressed within the plant cell.

Plant-synthesized opines are secreted and taken up by bacteria; only bacteria of the infecting strain can metabolize these opines.
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Example 2: The community of microbes that inhabit the gut
The gut “microbiome”
• What is the role of the microbes in the gut?
• Is their relationship mutualistic, parasitic or commensal?
• How can we learn what they are doing in our gut?
Understanding the Human Gut microbiome

• Identify the gut metagenome of one healthy individual
• Identify the gut metagenomes of several healthy individuals

Is there a core gut microbiome?

• Identify the gut metagenomes of individuals with health conditions

Does the core gut metagenome vary according to the health of the individual human host?
Initial metagenomic analyses of the human GI tract (16S):

• Demonstrated that 80% of organisms hadn’t been cultured and 62% were previously unknown

• Revealed that two bacterial phyla (Bacteroidetes and Firmicutes) dominate the gut microbiome in mice and in humans.

And...

Recent work has shown obesity to be associated with a shift in the representation of the dominant phyla of bacteria in the gut, both in humans and animal models.
The proportion of Bacteroidetes is decreased in obese people

Before diet, obese had fewer Bacteroidetes and more Firmicutes

With either fat-restricted or carb-restricted diet, over time Bacteroidetes increases and Firmicutes decreases

More body weight lost, larger the increase in Bacteroidetes (data not shown here)


What conclusions would you draw from this data?
Questions to consider:

Did the change in diet affect the microbiome which then led to weight loss?

Or did the change in diet lead to weight loss which then led to alteration of the microbiome?

How can you answer these questions?

Are there variables other than diet that could be affecting the outcome?
There is a correlation between gut bacteria and obesity in different mouse model systems.

Relative abundance of *Firmicutes* increases in obese mice while *Bacteroidetes* decreases.

**A**

Diet-induced obesity

Mice with “humanized” gut microbiota also showed the same result.

*Turnbaugh et al. (2008) Cell Host and Microbe 3:213*
There is a correlation between gut bacteria and obesity in different mouse model systems.

Relative abundance of Firmicutes increases in obese mice while Bacteroidetes decreases.

Ley R E et al. PNAS 2005;102:11070-11075
Analyze the following data and construct a model to explain the functional difference between the different microbiomes (obese vs. lean)

wt germ-free mice colonized with microbes from ob/ob (genetically-induced obese) mice or +/- (lean) donors.

There was no difference in chow consumption

After two weeks, mice were sacrificed and gut microbiomes assessed

Colonization of germ-free mice with an 'obese microbiota' results in a significantly greater increase in total body fat than colonization with a 'lean microbiota'. Why?

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Energy present in fecal contents of obese mice is less than that of lean mice

In experiments designed to measure the calorie content of fecal matter, obese mice had significantly less energy remaining in their feces than lean mice.

Are these data consistent or in conflict with your model?

It can be difficult to demonstrate causality between correlated observations.

Controlled studies are usually necessary to establish causality.

For human data, scientists often accumulate lots of correlated observations then use the power of statistics to establish *significance*. 
What is the functional difference between the two distinct microbiomes of obese vs. lean individuals? What are the bacteria doing?

• Obese microbiome has an increased capacity to harvest energy from the diet (in mice).
• ob/ob microbiome is enriched for genes necessary for initial breakdown of otherwise indigestible polysaccharides and for proteins that import and metabolize the products of these glycoside hydrolases.
• ob/ob mice guts contain more fermentation end products (e.g. butyrate and acetate)

Too early to make broad claims; the first comprehensive metagenomic studies about the healthy microbiome are just coming out
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