I FEEL IT IN MY GUT: THE BRAIN AND MICROBIOME
Learning Objectives

• Provide an overview of the origins and importance of the gut-brain connection

• Examine the relationship between the gut-brain-microbiome axis and chronic inflammation

• Consider current evidence of the gut-brain connection’s impact on mental health

• Consider potential implications of the gut-brain connection for the treatment of mental illness
Origins and Importance of the Gut-Brain Connection
The Gut-Brain Connection: A Topic of Much Interest

In 2014 and 2015, NIMH offered a fund of $1 million to study the gut microbiota-brain axis.

In 2015, the US Navy Institute announced a fund of $14.5 million over the next 6–7 years to research the role of the gut in cognitive and stress disorders.

The European Union launched a 5-year “MyNewGut” project ($10.1 million US dollars) for research on brain development and related disorders.

In 2014, >$1 million was transferred to the Human Microbiome Project, supported by the NIH, which aims to understand microbiome diversity and determine roles of microorganisms in health and disease.

The gut and brain originate from the same tissue during embryogenesis, the neural crest, and influence each other during development.

Bacteria, mostly residing in the gut, outnumber the body’s cells 10:1 and are referred to as a microbiome.

A microbiome comprises the entirety of microscopic organisms and their genetic material that inhabit a particular environment.

Gut microbiota begin to colonize the gastrointestinal tract at birth; a complex microbiome forms after the first year.

Bacteria in the gut is estimated to consist of >1000 species and >7,000 subspecies.

Microbiome bacteria have co-evolved with us and interact with our body in a bidirectional manner.

Communication Along the Microbiota-Gut-Brain Axis

Diseases Associated with Dysbiosis of Gut Microbiota

Neurodegenerative disorders
- Multiple sclerosis
- Parkinson’s disease

Cardiovascular disease
- Coronary artery disease
- Hypertension

Intestinal disorders
- Inflammatory bowel disease
  - Crohn’s disease
  - Ulcerative colitis
- Irritable bowel syndrome
- Celiac disease

Psychiatric and mood disorders
- Anxiety
- Depression
- Autism spectrum disorder
- Bipolar disorder
- Schizophrenia

Metabolic disorders
- Metabolic syndrome
- Obesity
- Types 1 and 2 diabetes

Pregnancy-related conditions
- Gestational hypertension
- Gestational diabetes

• Preclinical data and early clinical research support the gut-brain connection

• Too soon to make therapeutic recommendations based on randomized clinical trials and best practices

• Interesting and well-funded avenue of research with promising future
The Gut-Brain Connection and Inflammation
Early Life Adversity and the Microbiome

- Negative in-utero and early life experiences alter initial gut colonization, which predisposes an individual to stress-induced inflammation later in life by:
  - Establishing a hyperactive hypothalamic-pituitary-adrenal (HPA) axis
  - Altering immune system activity
  - Increasing intestinal permeability
  - Influencing epigenetic regulation

- Early life experiences, through modification of the gut ecology, magnify stressors, amplify the stress response, and inhibit corrective homeostatic mechanisms resulting in chronic sustained inflammation

Sudo N et al. J Physiol 2004;558(Pt 1):263-75;
A Healthy Microbiome

• Normal gut microbiota
• Normal behavior and cognition
• Healthy levels of inflammatory cells/mediators
• Normal intestinal permeability

Causes and Consequences of Dysbiosis

*Examples of potential causes of dysbiosis. Not an exhaustive list.

- Increased inflammation
- Altered behavior and cognition
- Increased gut permeability

Impact of Ketogenic Diet on the Gut Microbiota is Distinct From a High-Fat Diet

Fold change from baseline of genera whose abundances were significantly different between high-fat diet and ketogenic diet (false discovery rate < 0.1, DESeq2), in mice fed ketogenic diet for 3 weeks (n = 6 mice). Data are presented as mean ± standard error of the mean. Each data point represents an individual, singly housed animal.

“Classic” vs. Chronic Inflammation

Classical inflammation
- Described ~2000 years ago
  - Pain (dolor)
  - Redness (rubor)
  - Heat (calor)
  - Swelling (tumor)
- Short-term response to infection and injury
- Aims to remove infective stimulus and allow repair of damaged tissue

Chronic inflammation (“metaflammation”)
- Described in 1993
  - Low-grade, causing only a small rise in immune system markers
  - Persistent, resulting in chronic, rather than acute, allostasis
  - Has systemic, rather than local, effects
  - Has antigens that are less apparent as foreign (“inducers”)
  - Appears to perpetuate, rather than resolve, disease
  - Is associated with reduced, rather than increased, metabolism
Leaky Gut, Inflammation, and the Brain

- Leaky Gut
- Inflammation
- Brain


- Prebiotics
- Probiotics
- Good diet

- Disrupted intestinal barrier “leaky gut”
- Bacteria translocation
- Infection
- Antibiotics
- Poor diet

- HPA activation

- Immune activation
- Proinflammatory cytokines
- Neuroinflammation

- Stress

- Intact intestinal barrier

- Cognitive dysfunction
  - Depression
  - Anxiety
  - Decreased social function

- Proper diet

- Infection

- Antidepressants

- Antipsychotics

- Anxiolytics

- Decreased social function
Proinflammatory Cytokine Entry Pathways From the Periphery Into the Central Nervous System

Inflammatory Signals (PAMPs and DAMPs) → Activation of Peripheral Proinflammatory Cytokines (i.e. IL-1β, IL-6, TNF-α, IFN-γ)

Cytokine Entry (Humoral Pathway):  
(I) Cytokines gain entry through leaky regions of the blood brain barrier (BBB)  
(II) Endothelial cells and macrophages in cerebral vasculature activated to produce local inflammatory mediators  
(III, VI) Inflammatory signals recruit activated immune cells to CNS, induce local cytokine production.

Cytokine Entry (Transport Pathway):  
(IV) Saturable active cytokine transporters participate in carrier-mediated transport of cytokines across the BBB

Cytokine Entry (Neural Pathway):  
(V) Activation of peripheral nerve afferents (i.e. trigeminal, vagus) relay cytokine signals to the nucleus of the solitary tract (NTS) and the hypothalamus.

CNS=central nervous system  
DAMP=damage-associated molecular patterns  
IFN=interferon  
IL=interleukin  
PAMP=pathogen-associated molecular patterns  
TNF=tumor necrosis factor

The Role of the Gut-Brain Connection in Mental Health
Clinical Observations Suggestive of Gut-Brain Connection

• Common co-occurrence of gastrointestinal (GI)-related comorbidities with a variety of psychiatric conditions (e.g., anxiety, depression, schizophrenia)

• GI problems can create stress and anxiety; conversely, stress and anxiety can make GI problems worse

• Experiencing emotion, such as excitement or nervousness, may impact the digestive system, causing the “butterflies in the stomach” feeling

Prinsloo S, Lyle RR. NeuroRegulation 2015;2(4):158-61;
The Cleveland Clinic 2017;https://my.clevelandclinic.org/health/treatments/16358-gut-brain-connection.
## Evidence of the Gut-Brain Connection

<table>
<thead>
<tr>
<th>Preclinical</th>
<th>Clinical (limited)</th>
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<tr>
<td>• Stress in rats has been reported to lead to a reduction in diversity of the gastrointestinal microbiome</td>
<td>• A double-blind, placebo-controlled trial of 66 subjects who ingested a fruit bar containing a probiotic formula for 30 days reported significantly lower levels of anxiety, anger, depression, and somatization, as well as lower cortisol levels</td>
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<td>• The naturally exaggerated physiological response to stress in germ-free mice was reversed when they were colonized with a particular strain of bacteria</td>
<td>• A placebo-controlled study assessing the effect of a fermented milk product containing probiotics on the brain activity of healthy women reported significant impact on the regions controlling emotional and sensory processing</td>
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<td>• Rats given a strain of bacteria for 28 days showed a decline in both anxiety and depression scores</td>
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Kelly et al. demonstrated a connection between depressive behavior and the microbiome:

- **Depressed patients**
  - ↓ microbiota richness and diversity
  - ↑ inflammatory markers
  - ↑ cortisol levels

- **Healthy controls**

**Microbiota-depleted rats**

- Compared to healthy controls:
  - ↑ Anhedonia-like behavior
  - ↑ Anxiety-like behaviors
  - ↑ Intestinal transit time
  - Trend toward increased levels of plasma C-reactive protein

Possible Role of Gut Microbiota-Brain Axis in Antidepressant Effects of (R)-Ketamine in a Social Defeat Stress Model

Figure. 1 Antidepressant effects of ketamine enantiomers in susceptible mice after CSDS. (c–e) Behavioral tests including locomotion test (F(3,20)=0.159, P=0.923), TST (F(3,20)=18.362, P<0.001) and FST (F(3,20)=15.107, P<0.001) were performed after treatment. (f) Sucrose preference test was performed 3 days after treatment (F(3,20)=20.287, P<0.001). Data are shown as mean ± standard error of the mean (n=6). *P<0.05, **P<0.01, ***P<0.001.

NS: not significant; ket: ketamine; CSDS: chronic social defeat stress; TST: tail suspension test; FST: forced swim test.
The family Christensenellaceae is thought to be a highly heritable taxon and is associated with beneficial metabolic effects on its host.

Flavonifractor, a bacterial genus that may induce oxidative stress and inflammation in its host, was associated with bipolar disorder.

BD: bipolar disorder (N=113)
UR: unaffected relatives (N=39)
HC: healthy controls (N=77)

Characterizing the Gut Microbiota in Adults With Bipolar Disorder


*\( p < 0.05 \), Bonferroni correction

**BD**: bipolar disorder (N=23)

**HC**: healthy controls (N=23)

**OTU**: operational taxonomical unit
Differential Effects of Diet on Gut Microbiome Genes Associated With Alzheimer’s Disease in Older Adults

Six weeks of Mediterranean-style ketogenic diet (MMKD), but not American Heart Association Diet (AHAD) decreased abundance of gene families annotated to Alzheimer’s disease.

The abundance of gene pathways associated with type 1 diabetes, type 2 diabetes, and bacterial toxins was also decreased following MMKD, but not AHAD.

N=17 (11 mild cognitive impairment, 6 cognitively normal)

*p<0.05

Unique Microbial Profile in Co-Occurring Irritable Bowel Syndrome (IBS) and High Anxiety/Depression

- High Anxiety/Depression > Low Anxiety/Depression
- Greater abundance of bacterial phyla, Bacteroidetes and Proteobacteria
- Lower alpha diversity

Phylum Proteobacteria
Phylum Bacteriodetes
FamilyPrevotellaceae
GenusAnaerotruncus
FamilyLachnospiraceae

Effects of Probiotic Supplementation on CRP Levels in Adults With Mental Illness or Symptoms

Study ID                  |   WMD (95% CI)   | Weight |
--------------------------|------------------|--------|
Akkasheh G (2016) a      | -1.33 (-2.51, -0.14) | 15.56  |
Akbari E (2016) b         | -3.30 (-5.12, -1.48) | 8.81   |
Pinto-Sanchez MI (2017) c | -0.94 (-1.30, -0.58) | 31.54  |
Tamtaji OR (2018) b       | -1.70 (-2.26, -1.14) | 27.42  |
Ghaderi A (2019) d        | -2.00 (-3.11, -0.89) | 16.67  |
Overall (I² = 66.4%, p = 0.018) | -1.59 (-2.22, -0.97) | 100.00 |

N=251

WMD = weighted mean difference

NOTE: Weights are from random effects analysis

a Major depressive disorder
b Alzheimer’s disease
c Irritable bowel syndrome with mild to moderate anxiety and/or depression
d Schizophrenia

The Role of the Gut-Brain Connection in Mental Health Treatment
Gut-Brain Axis and Microbiota in Mood Disorders


BDNF = brain-derived neurotrophic factor
HPA = hypothalamic-pituitary-adrenal
IL = interleukin
TNF = tumor necrosis factor
**Probiotics and Prebiotics**

- **Probiotics**: “Live microorganisms which, when administered in adequate amounts, confer a health benefit on the host” (World Health Organization)
  - Found in fermented foods
  - Use can result in both beneficial and detrimental effects

- **Prebiotics**: “A nonviable food component that confers a health benefit on the host associated with modulation of the microbiota” (Food and Agriculture Organization of the United Nations)
  - Nutritional substances required by probiotics for survival
  - The modern Western diet often fails to provide prebiotics
Probiotic Supplementation Improves Psychological Symptoms Among Healthy Volunteers

Meta-analysis of randomized, placebo-controlled trials of healthy volunteers pre- and post-supplementation with a probiotic (21–56 days) measuring effects on symptoms of depression, anxiety, and perceived stress (7 studies; n=374)

Probiotics Reduce Depressive Symptoms in Adults With Mild to Moderate Depression, Not Healthy Controls

No evidence of publication bias (Egger test, \(p=0.550\))

No significant effect of probiotic treatment on depressive symptoms across all clinical trials (\(p=0.059\))

Positive effect of probiotic treatment on depressive symptoms across clinical trials of MDD or mild to moderate anxiety and/or depression (\(p=0.029\))

No significant effect of probiotic treatment on depressive symptoms across clinical trials of healthy controls (\(p=0.146\))

No Difference in Depression Following Probiotic Treatment Versus Placebo in Depressed Adults

Meta-analysis of randomized clinical trials of participants with depression (major depressive disorder or self-referrals with at least moderate depression score) where probiotics were used as supplementary (2 studies) or stand-alone (1 study) treatment versus placebo for 8 weeks (N=229).

Probiotics, Not Prebiotics, Associated with Reduction in Depression

A larger effect of probiotics on depression was observed for clinical/medical samples (d=-.45, p<.001) than community samples (d=-.09, p=.09)

Probiotics, Not Prebiotics, Associated with Reduction in Anxiety

Probiotic Treatment Improves Depression in Adults With Major Depressive Disorder (MDD)

Results from meta-analysis of 4 double-blind, randomized control trials and 2 open-label trials (n=302, MDD) comparing effects of probiotic intervention (6-8 weeks) and placebo on depressive symptoms

Significantly favors treatment

Results from meta-analysis of 4 cross-sectional observational studies comparing gut bacterial diversity of participants with MDD (n=156) versus non-depressed controls (n=131)

**Microbiota and Probiotics in Major Depressive Disorder, Bipolar Disorder, and Schizophrenia**

<table>
<thead>
<tr>
<th>Study</th>
<th>Participants and Treatment</th>
<th>Outcomes</th>
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<tr>
<td><strong>Major Depressive Disorder (MDD)</strong></td>
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<tr>
<td>Akkasheh et al., 2016</td>
<td>MDD, probiotic (n=20), or placebo (n=20) for 8 weeks</td>
<td>Probiotic&gt;Placebo: decreased depressive symptoms, serum insulin, HOMA-IR, and serum hs-CRP; increased plasma total GSH.</td>
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<tr>
<td>Romijn et al., 2017</td>
<td>Self-reported depressed, probiotic (n=40), or placebo (n=39) for 8 weeks</td>
<td>Probiotic&gt;Placebo: N.S.</td>
</tr>
<tr>
<td>Ghorbani et al., 2018</td>
<td>Moderate depression, Familact H + fluoxetine (n=20), or placebo (n=20) for 4 weeks</td>
<td>Familact H + fluoxetine&gt;placebo: decreased depression symptoms.</td>
</tr>
<tr>
<td>Kazemi et al., 2019</td>
<td>MDD, probiotic (n=28), prebiotic (n=27), or placebo (n=26) for 8 weeks</td>
<td>Probiotic&gt;prebiotic or placebo: decrease depression symptoms.</td>
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<td><strong>Bipolar Disorder (BD)</strong></td>
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<td>Evans et al., 2017</td>
<td>115 BD and 64 controls</td>
<td>BD&gt;Controls: lower <em>Faecalibacterium</em> and <em>Ruminococcaceae</em> genera. <em>Faecalibacterium</em> levels associated with better health and mood in BD.</td>
</tr>
<tr>
<td>Dickerson et al., 2018</td>
<td>BD hospitalized for mania, adj. probiotic (n=33), or adj. placebo (n=33) for 24 weeks</td>
<td>Probiotic&gt;Placebo: reduced number of re-hospitalizations (greater effect in those with elevated systemic inflammation at baseline) and days hospitalized.</td>
</tr>
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<td>Reininghaus et al., 2018</td>
<td>20 BD euthymic, OMNiBiOTic Stress Repair formula for 3 months</td>
<td>OMNiBiOTic improved attention, psychomotor processing speed, and executive function.</td>
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<td><strong>SCZ</strong></td>
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<tr>
<td>Dickerson et al., 2014</td>
<td>SCZ, adj. probiotic (n=33), or placebo (n=32) for 14 weeks</td>
<td>Probiotic&gt;Placebo: no difference in SCZ symptoms; less likely to develop severe bowel difficulty.</td>
</tr>
</tbody>
</table>

Adj=adjunctive; GSH=glutathione; hs-CRP=high sensitivity C-reactive protein; HOMA-IR=homeostasis model assessment of insulin resistance; SCZ=schizophrenia.
Future Considerations for the Gut-Brain Connection in the Treatment of Mental Illness

• Mental health disorders are unlikely to be treated with probiotics and prebiotics alone

• While mental health requires specialized care provided by mental health professionals, complementary therapeutic strategies should also be utilized
  - Sufficient rest, digestion, exercise, mindful relaxation, and eating a nutritionally rich and varied diet may be considered

• Additional studies are required to identify strains of bacteria which may confer mental health benefits, as well as those associated with specific symptoms

• Additional research may lead to nutritionally based and psychobiotic treatments that can be combined with traditional psychological and psychotropic approaches

Summary

• The gut-brain connection is facilitated by systemic (i.e., immune and endocrine) and neural (i.e., vagus nerve and sympathetic nervous system) communication

• Dysbiosis leads to increased inflammation, altered behavior and cognition and increased gut permeability

• Individuals with mental illness may display reduced microbiota richness and diversity

• There is mixed evidence that probiotics improve mood symptoms; mental health disorders are unlikely to be treated with probiotics alone
Posttest Question 1

Negative in-utero and early life experiences alter initial gut colonization, which predisposes an individual to stress-induced inflammation later in life by:

1. Altering immune system activity
2. Establishing a hypoactive HPA axis
3. Decreasing intestinal permeability
4. All of the above
5. None of the above
Posttest Question 2

Tony is a 34-year-old patient with major depressive disorder. Laboratory examination of this patient would likely show:

1. Increased microbiota diversity
2. Increased inflammatory markers
3. Decreased cortisol levels
4. All of the above
5. None of the above
Posttest Question 3

Treatment of dysbiosis in mood disorders is purported to improve mood symptoms. Meta-analyses of clinical trials largely show:

1. Probiotics improve mood symptoms
2. Prebiotics improve mood symptoms
3. Probiotics and prebiotics improve mood symptoms
4. The findings are mixed