

Modulating the gut microbiome for health: Evidence-based testing & therapeutic strategies



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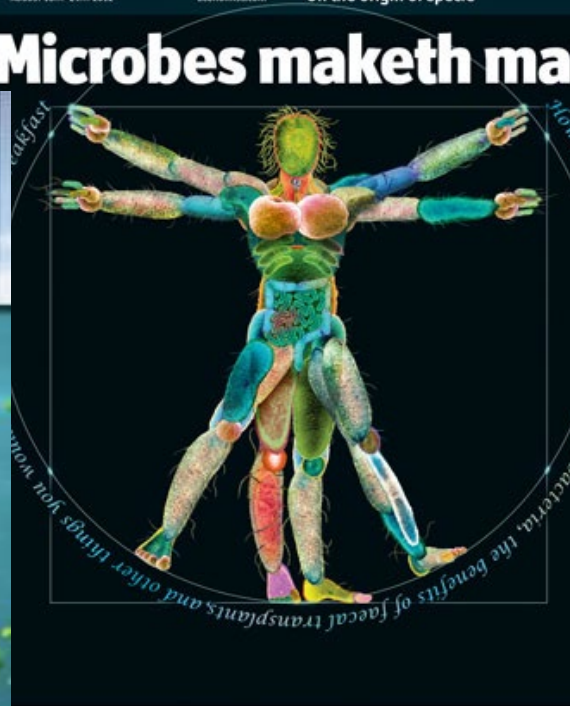
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DISCLOSURES & CONFLICTS OF INTEREST

I do not have ANY financial affiliation with any microbiome testing or supplement manufacturing companies



A microscopic view of various bacteria, primarily rod-shaped, against a blue background. The bacteria are shown in different orientations and depths, creating a sense of a dense microbial community. A semi-transparent white circle is overlaid on the left side of the image, containing text.

Roles of the gut microbiota

- **Aids in digestion**
- **Synthesizes vitamins**
- **Outcompetes pathogens**
- **Stimulates the immune system**
- **Regulates gene expression**

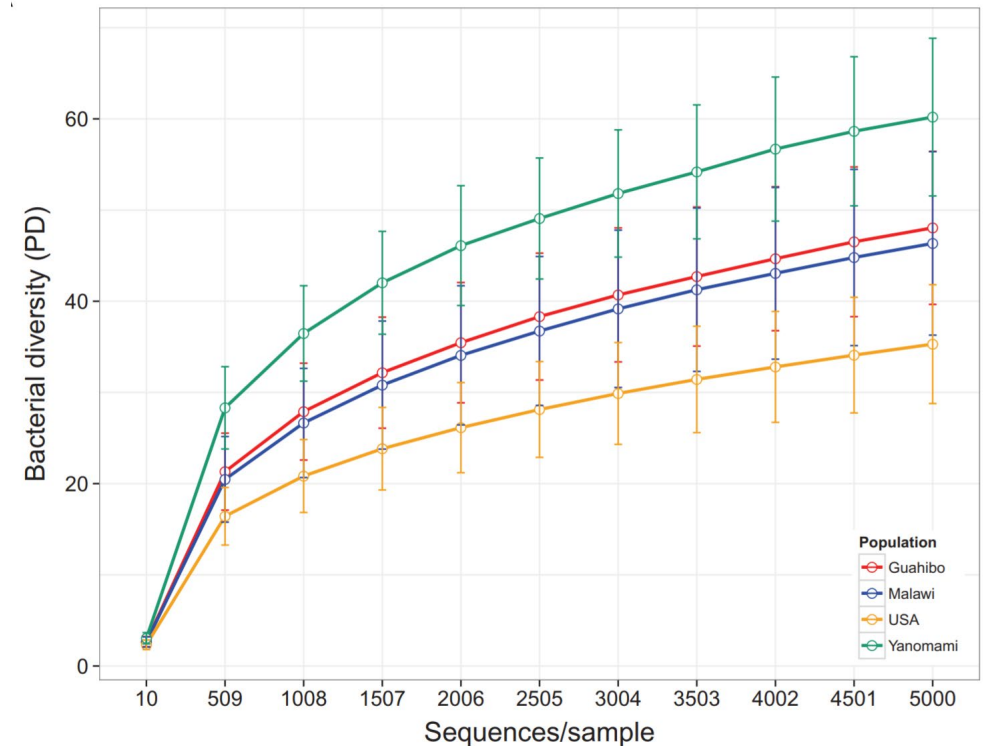
multiple sclerosis **Parkinson's** **thyroid disorders**
kidney disease **Hashimoto's** **hypertension**
obesity **liver disease** **asthma** **eczema**
sleep apnea **stroke** **psoriasis**
acne **CHRONIC DISEASE** **metabolic syndrome**
cancer **depression** **neuropathy**
anxiety **GERD** **Inflammatory bowel disease**
heart disease **diabetes** **lupus**
autoimmunity **IBS** **arthritis** **Alzheimer's**

LOST ANCESTRAL DIVERSITY: THEORY OF THE DISAPPEARING GUT MICROBIOTA

Some researchers have suggested that our lack of ancestral diversity in the gut microbiome is driving chronic disease – Dr. Martin Blaser

- Changes are cumulative across generations and associated with increasing chronic disease risk
- Some studies estimate we've already lost about 50% of our diversity

Alpha diversity of the gut microbiota in four populations



OVERUSE OF ANTIBIOTICS

Antibiotics: one of the greatest discoveries of the 20th century

- **Revolutionized medicine & saved innumerable lives**

Yet wildly overused

- **Devastate gut microbial communities**
- **Recovery is often incomplete**



NON-ANTIBIOTIC MEDICATIONS ALTER THE GUT MICROBIOME

Nearly a quarter (24%) of non-antibiotic medications alter the gut microbiome

- Pharmaceuticals may cause negative shifts in the gut microbiota that further perpetuate the cycle of dysbiosis, gut permeability, and disease.



LOSS OF SEASONALITY

Study of the microbiome of Hadza hunter-gatherers

- **Microbiota reflects the seasonal availability of food**
 - Striking differences between seasons, with many taxa dropping to undetectable levels
 - These same microbes are rare or entirely absent in industrialized populations





Lots of prevailing myths and misunderstandings

MYTH #1: CULTURE-BASED STOOL TESTS ARE ACCURATE

For decades, study of gut microbes relied on culture, staining, and microscopy.

THE PROBLEM:

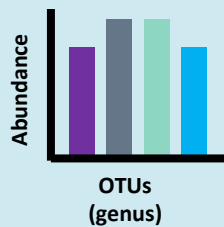
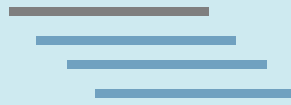
- Only a small fraction of the microbes in the gut are culturable
- Culture dramatically skews the relative abundance of microbes

Several popular stool analysis companies **STILL** using culture-based techniques.



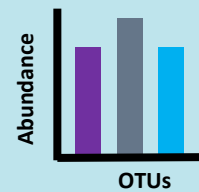
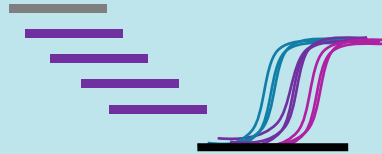
WHAT WE SHOULD BE USING INSTEAD: SEQUENCE-BASED TESTS

16S rRNA gene sequencing



- All bacteria at genus level
- No species/strain info
- No archaea, fungi, or eukaryotes
- Subject to primer bias

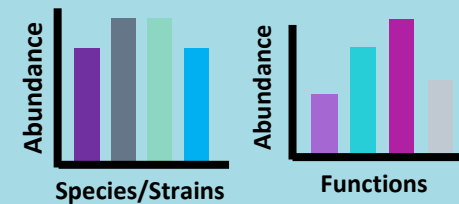
Targeted qPCR



- Targeted probes
- Improved sensitivity, accuracy, & speed of results
- Need probe for each microbe of interest

Metagenomics

```
ATACGTATGCATAGCA
TGACATGCGATCGGA
TTATAGCGAAGCTATA
ATACGTATGCATAGCA
```



- Captures ALL microbes at species/strain level
- Also tells function
- Expensive (but rapidly dropping in cost)

EXAMPLE: CULTURE VS. SEQUENCING FOR ASSESSING BACTERIAL ABUNDANCE

Comprehensive Stool Analysis / Parasitology x3

BACTERIOLOGY CULTURE		
Expected/Beneficial flora	Commensal (Imbalanced) flora	Dysbiotic flora
4+ Bacteroides fragilis group 1+ Bifidobacterium spp. 3+ Escherichia coli 1+ Lactobacillus spp. 4+ Enterococcus spp. 3+ Clostridium spp. NG = No Growth	3+ Bacillus spp, not cereus or anthracis	

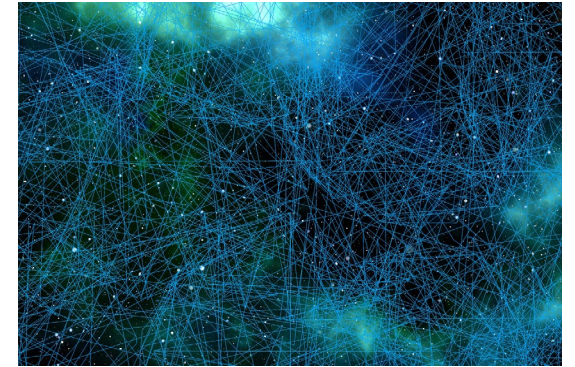
Same bacteria by 16S sequencing	Relative Abundance
<i>Bacteroides fragilis</i>	2.26%
<i>Bifidobacterium</i> spp.	0.00%
<i>Escherichia coli</i>	0.02%
<i>Lactobacillus</i> spp.	0.02%
<i>Enterococcus</i> spp.	0.00%
<i>Clostridium</i> spp.	1.09%
<i>Bacillus</i> spp.	0.00%
TOTAL	3.39%

LESS THAN <4% of human gut bacteria captured by culture!

Skewed bacterial abundance

MYTH #2: WE KNOW EXACTLY WHAT A HEALTHY MICROBIOME LOOKS LIKE

- Generally believed that diversity and community stability are key components of a healthy gut ecosystem -> but even these can be associated with disease states
- Keystone “beneficial” microbes recognized to be crucial for microbiome health, such as *Bifidobacterium*, are absent from the guts of traditional cultures like the Hadza
- In general, the gut has a high degree of interindividual variability, and is a complex network
- Abundance of a few microbes does not tell use the health of the network interactions.



AN INDEPENDENT ANALYSIS OF POPULAR STOOL/MICROBIOME TESTS

Bacterial abundances = uBiome

Bacterial pathogen detection = GI-MAP

Fungal detection = GI-MAP

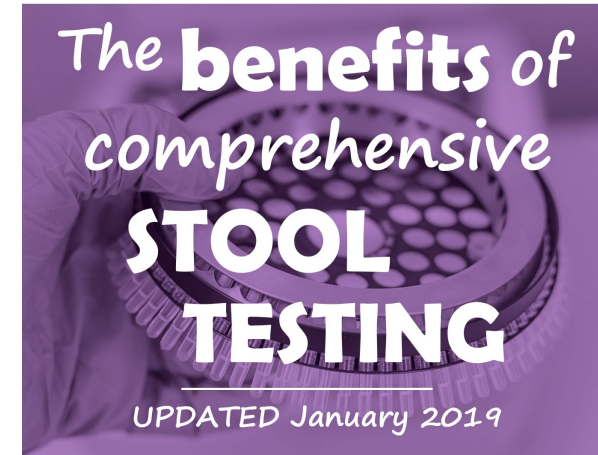
Parasite detection = GI-MAP

Virus detection = GI-MAP

Digestion/absorption markers = Doctor's Data or GI Effects

Inflammation/immunology markers = Doctor's Data or GI Effects

Gut environment = Doctor's Data, GI-MAP, or GI-Effects



WHAT I'M USING IN PRACTICE TO TEST AND TRACK GUT HEALTH

GI-MAP™

DNA Stool Analysis

- Bacterial pathogens
- Yeast, parasites
- Gut health markers

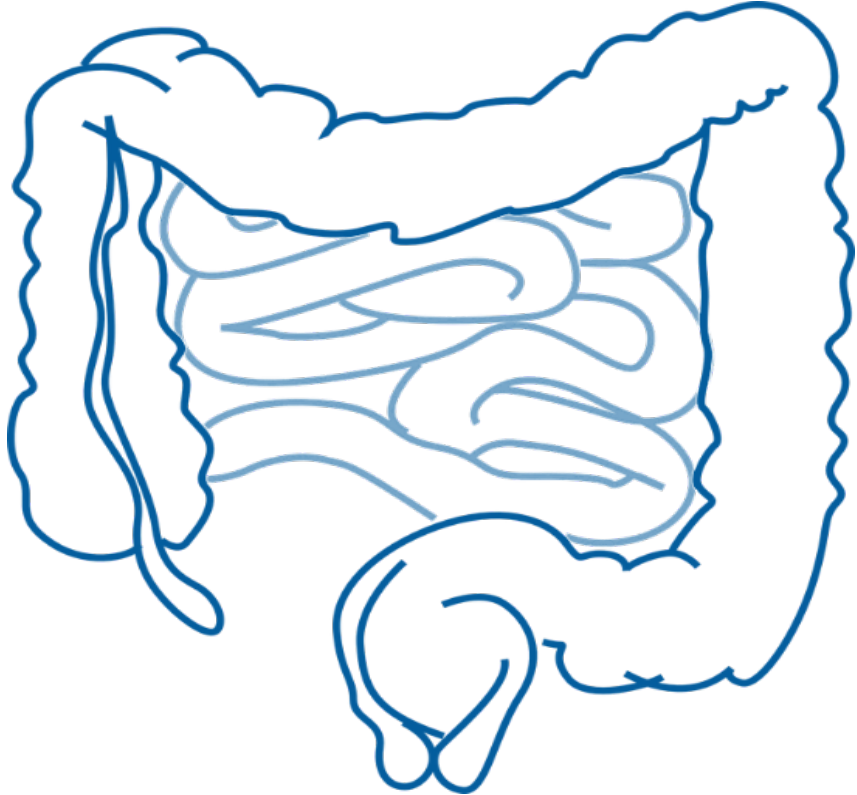
+

uBiome

- Bacterial abundance
& relative balance
“Red flags”

SOON → Metagenomics

MODULATING GUT HEALTH: MICROBIAL THERAPEUTICS

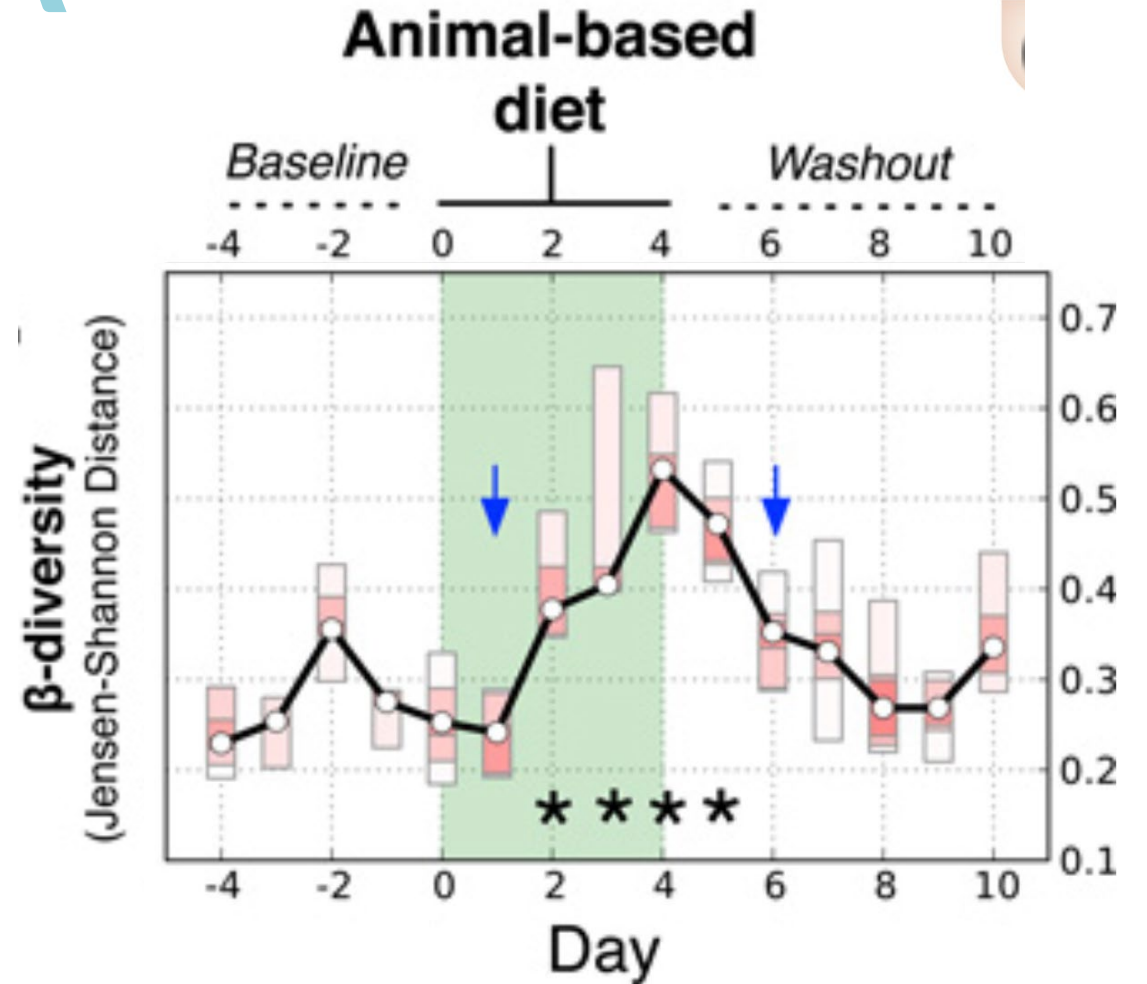


- Antibiotics
- Diet
- Exercise
- Herbal antimicrobials
- Probiotics
- Prebiotics
- Fecal microbiota transplant

DIETARY CHANGES RAPIDLY SHIFT THE GUT MICROBIOTA

Diet rapidly & reproducibly alters the gut microbiome

- Diet altered gut microbiota composition **within 48 hrs**
- Diet also alters microbial gene expression
- HIGHLY cited paper (>2,700 times)



MYTH #3: A HIGH-FAT DIET IS BAD FOR THE GUT MICROBIOTA

Animal studies that use a “high fat diet” are misleading

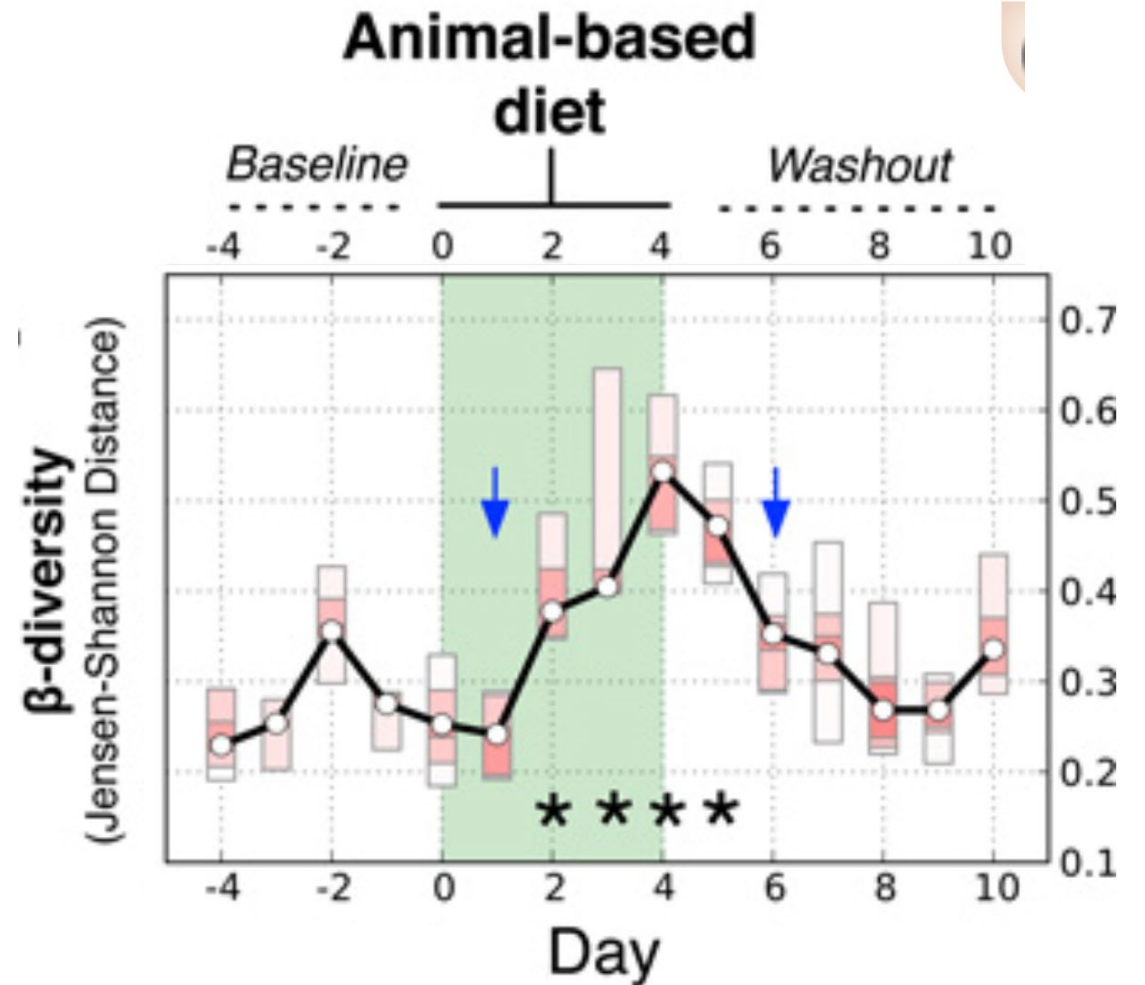
- Rodent “high-fat diet” = diet high in refined soybean oil, lard, and refined sugar, low in fiber
- Natural diet of a mouse is low in fat and high in carbohydrates
- Lab mice of choice genetically selected for weight gain in response to high-fat diet



MYTH #3: A HIGH-FAT DIET IS BAD FOR THE GUT MICROBIOTA

Supplementary data: animal-based diet was ketogenic!

- Increased beta diversity
- “Metabolic flexibility” of the gut microbiome
- Back to seasonality



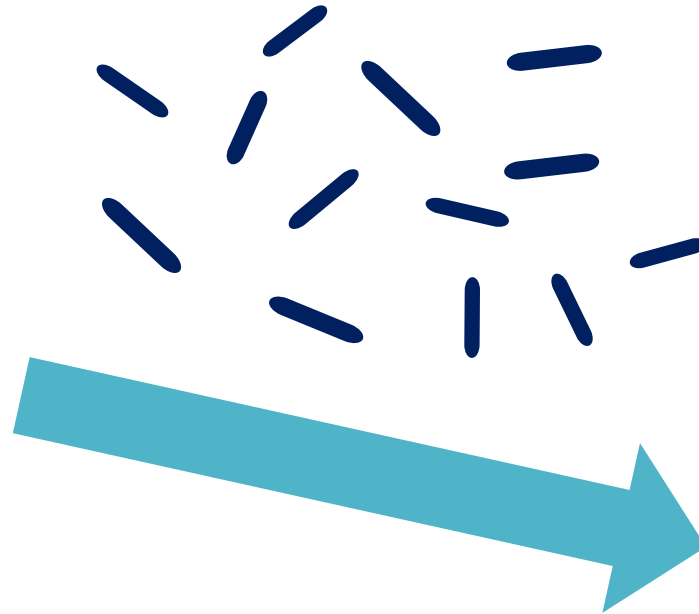
AN EVOLUTIONARY PERSPECTIVE:

“Our findings that the human gut microbiome can rapidly switch between herbivorous and carnivorous functional profiles may reflect past selective pressures during human evolution. Consumption of animal foods by our ancestors was likely volatile, depending on season and stochastic foraging success, with readily available plant foods offering a fallback source of calories and nutrients. Microbial communities that could quickly, and appropriately, shift their functional repertoire in response to diet change would have subsequently enhanced human dietary flexibility.”

DIETARY FIBER IS FERMENTED INTO SHORT-CHAIN FATTY ACIDS



DIETARY FIBER



Microbial
fermentation



Acetate

Propionate



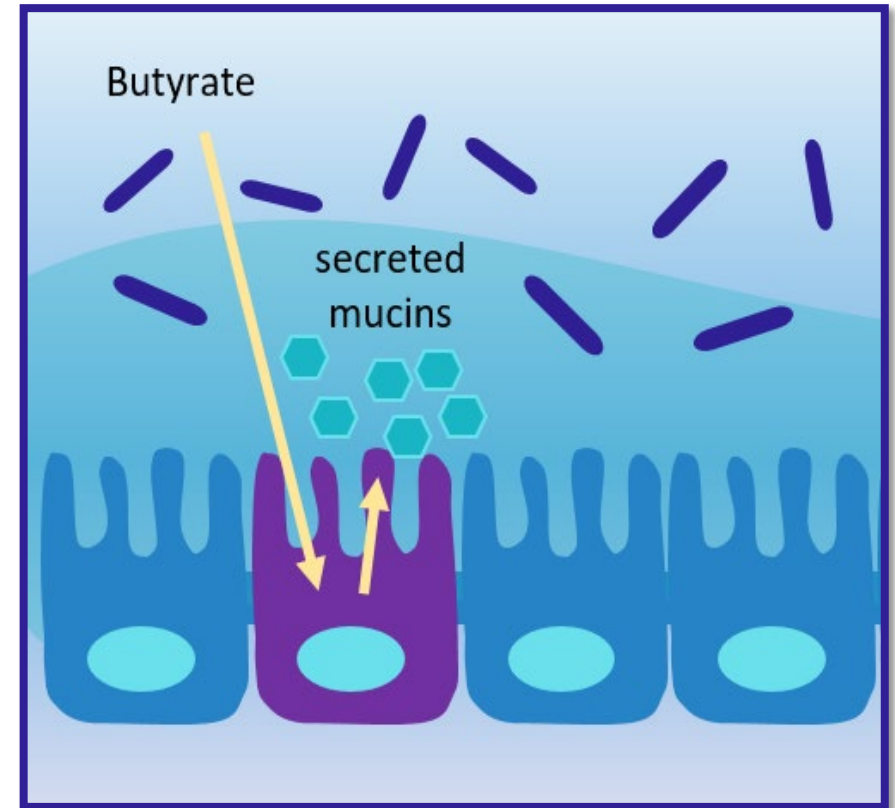
Butyrate

SHORT-CHAIN FATTY ACIDS

BUTYRATE HELPS MAINTAIN GUT BARRIER FUNCTION

Gut-derived butyrate:

- Increases mucus secretion, epithelial proliferation & turnover
- Promotes release of antimicrobial peptides
- Maintains “physiologic hypoxia” and gut homeostasis



Won't keto reduce your production of butyrate?

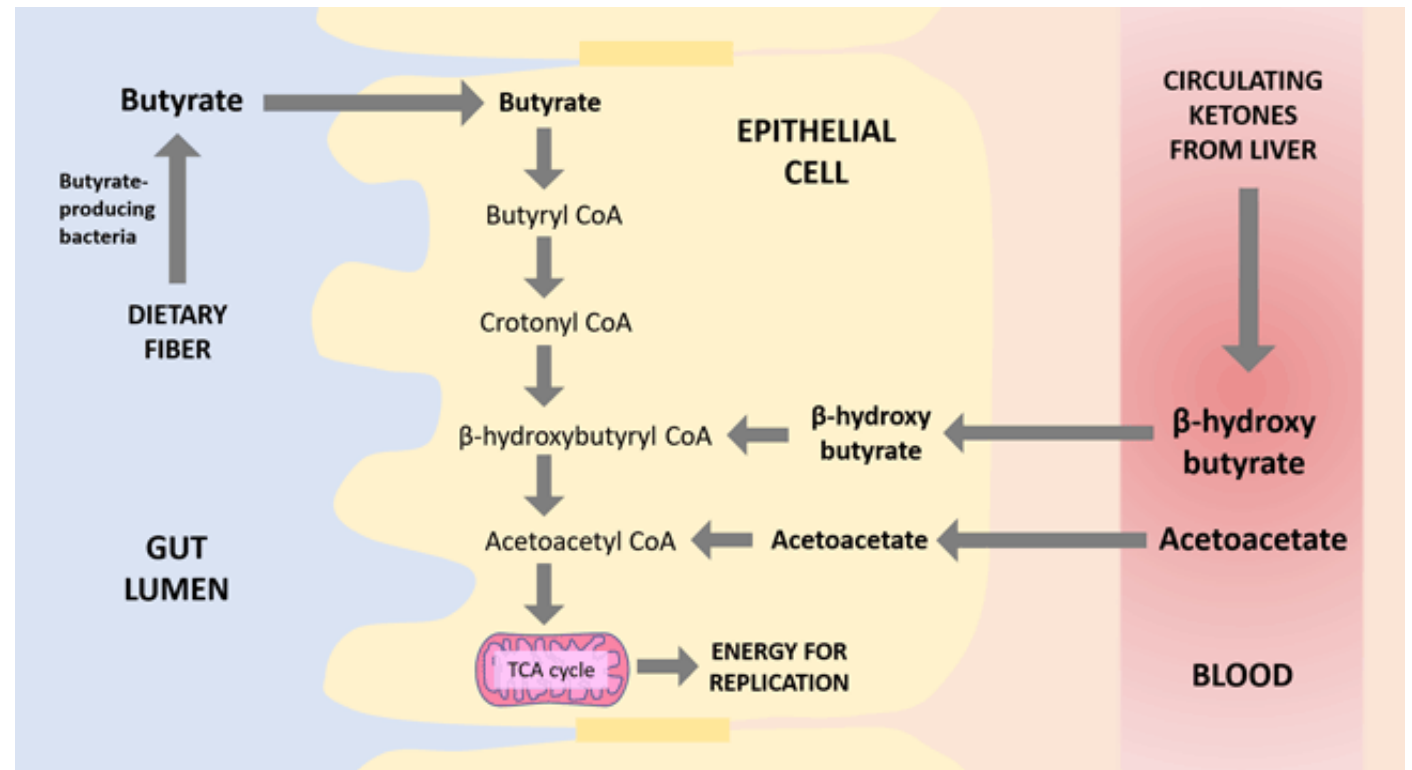
KETONE BODIES & ISOBUTYRATE CAN MAKE UP FOR LOW BUTYRATE

Isobutyrate = metabolite of protein fermentation

- Stimulates same receptors as butyrate (FFAR2, FFAR3, GPR109a)

Acetoacetate and β HB from the blood can supplement colonocyte energy pathways

- Inflamed gut has impaired butyrate uptake (e.g. IBD)



NO EVIDENCE THAT A WELL-DESIGNED KETO DIET IS DETRIMENTAL TO GUT HEALTH

Keto-induced changes in the gut microbiota and gut barrier may even be the reason we see so many benefits from ketosis

However, ketogenic diet is not right for everyone

- Keto is not necessarily the appropriate intervention for everyone with a gut issue

Can definitely do keto in a way that supports gut health

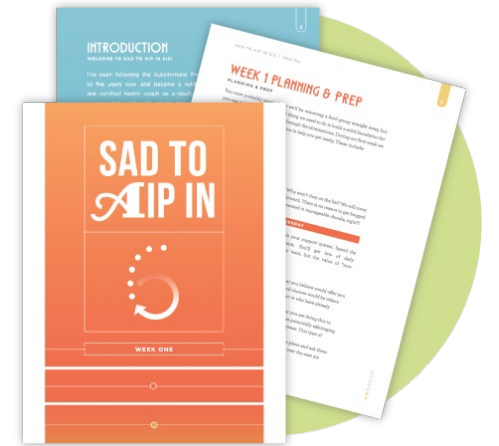
- Non-starchy vegetables, and quality meats & fats



UPCOMING STUDY!

AIP FOR ECZEMA & PSORIASIS

- Determine how the autoimmune protocol (AIP) impacts the gut microbiota and SCFAs
- Directly measure gut barrier function *in vivo* with the double sugar lactulose-mannitol test



EXERCISE ALTERS GUT MICROBIOTA COMPOSITION & FUNCTION

Animal models

- ↑ microbial diversity
- ↑ butyrate-producers
- ↑ SCFA production
- ↑ *Lactobacillus* and *Bifidobacterium*



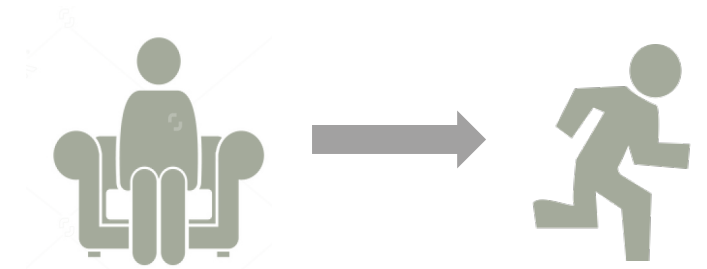
Cross-sectional studies

- ↑ Microbial diversity
- ↑ *Faecalibacterium prausnitzii*
- ↑ *Akkermansia muciniphila*
- ↑ Carbohydrate turnover and SCFA production



Longitudinal studies

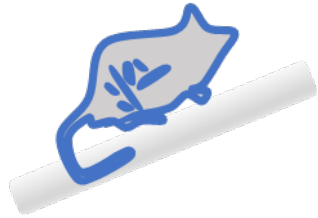
- ↑ Fecal butyrate
- ↑ Butyrate-producers (in lean individuals)



MYTH #4: “NO PAIN, NO GAIN”

STRENUOUS EXERCISE IS ALWAYS BETTER

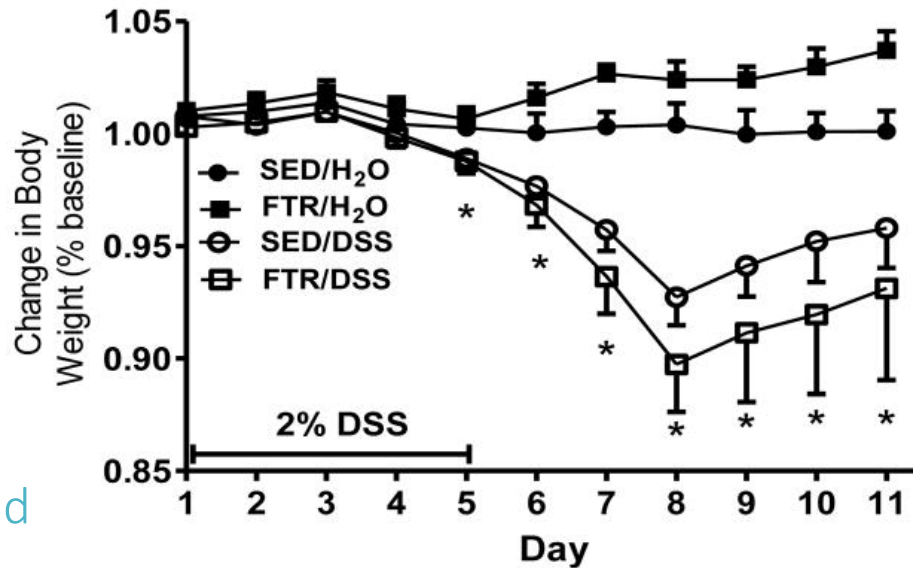
FTR



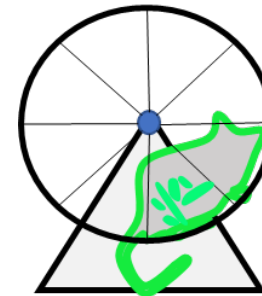
6 Weeks

3/10 mice died

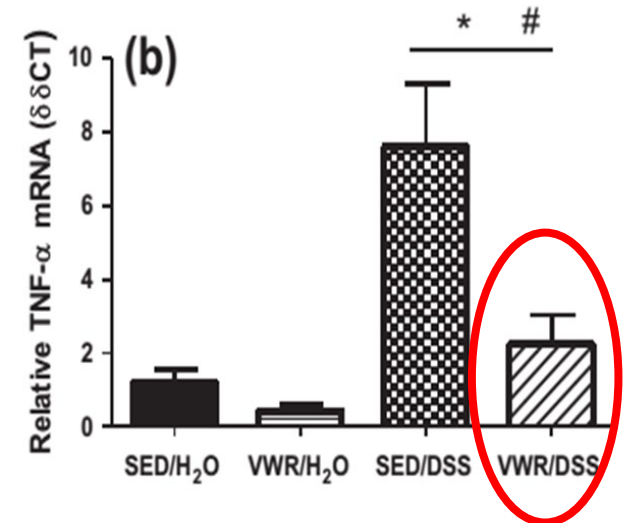
Figure 1a



VWR



6 Weeks



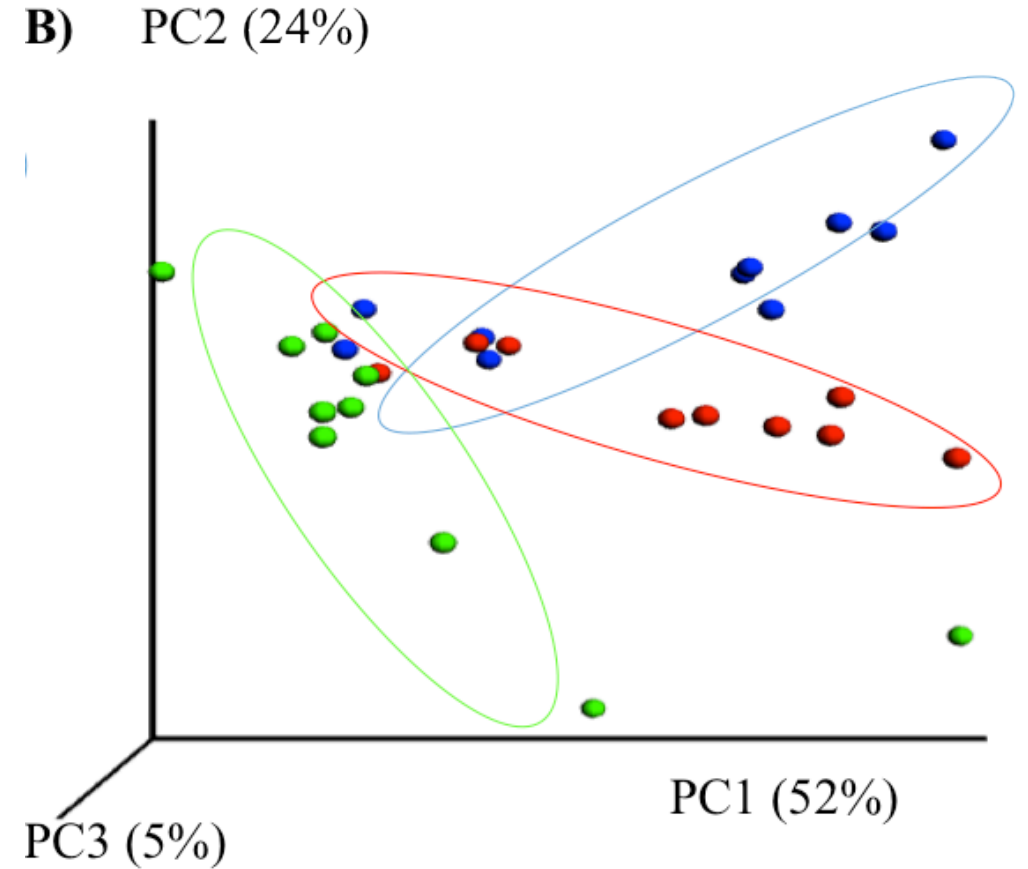
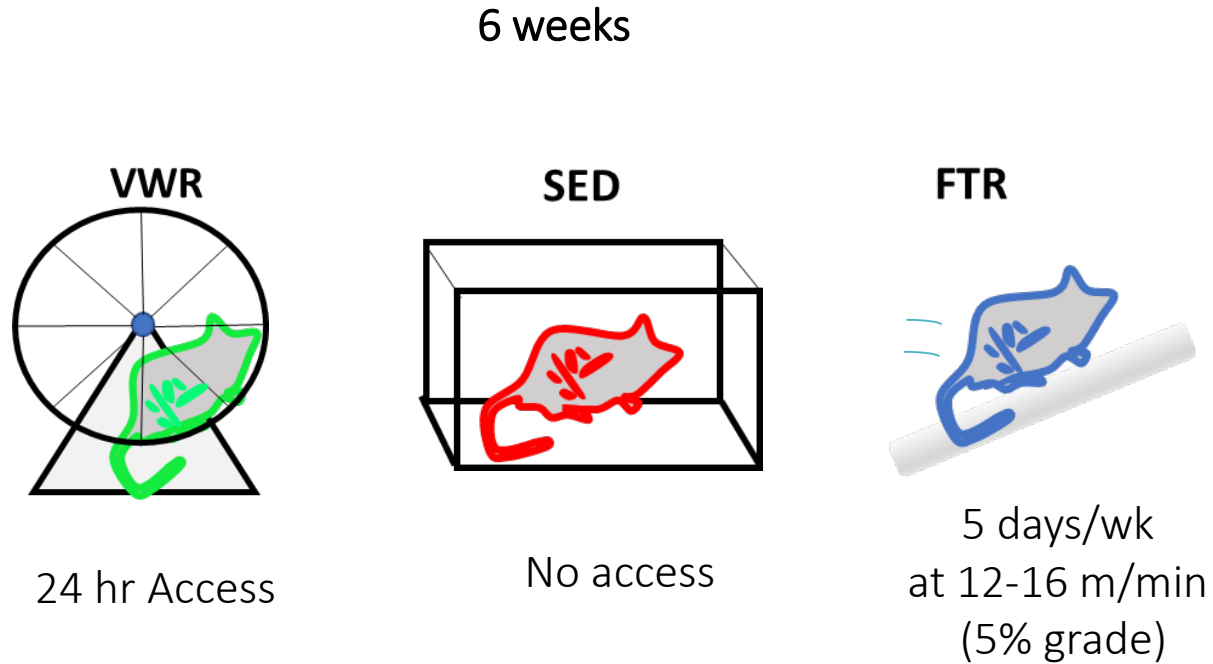
FORCED EXERCISE TRAINING:

↑ Colitis outcomes ↑ Gut inflammation

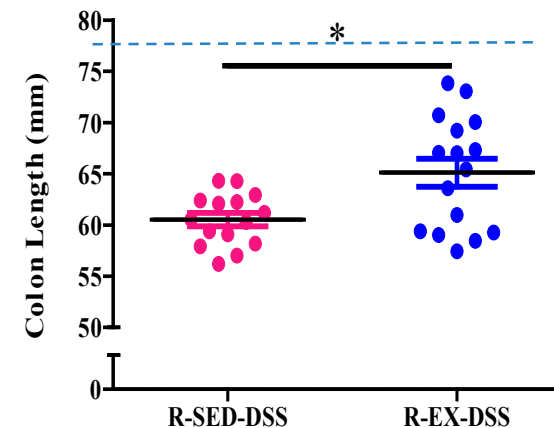
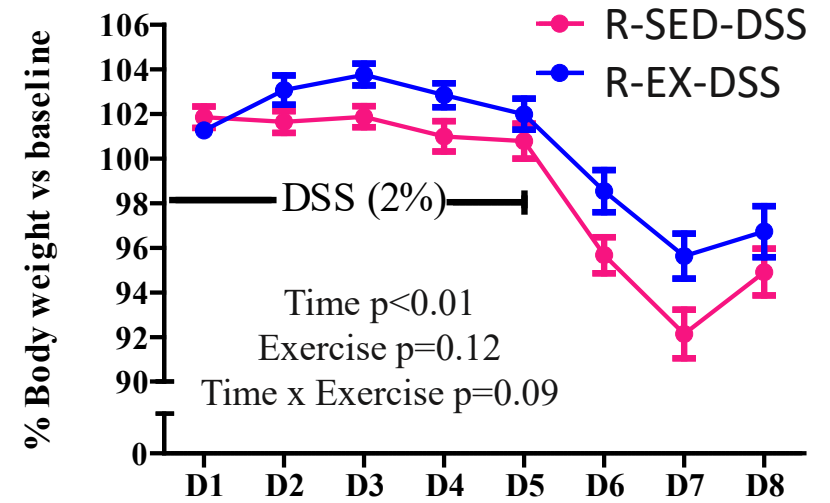
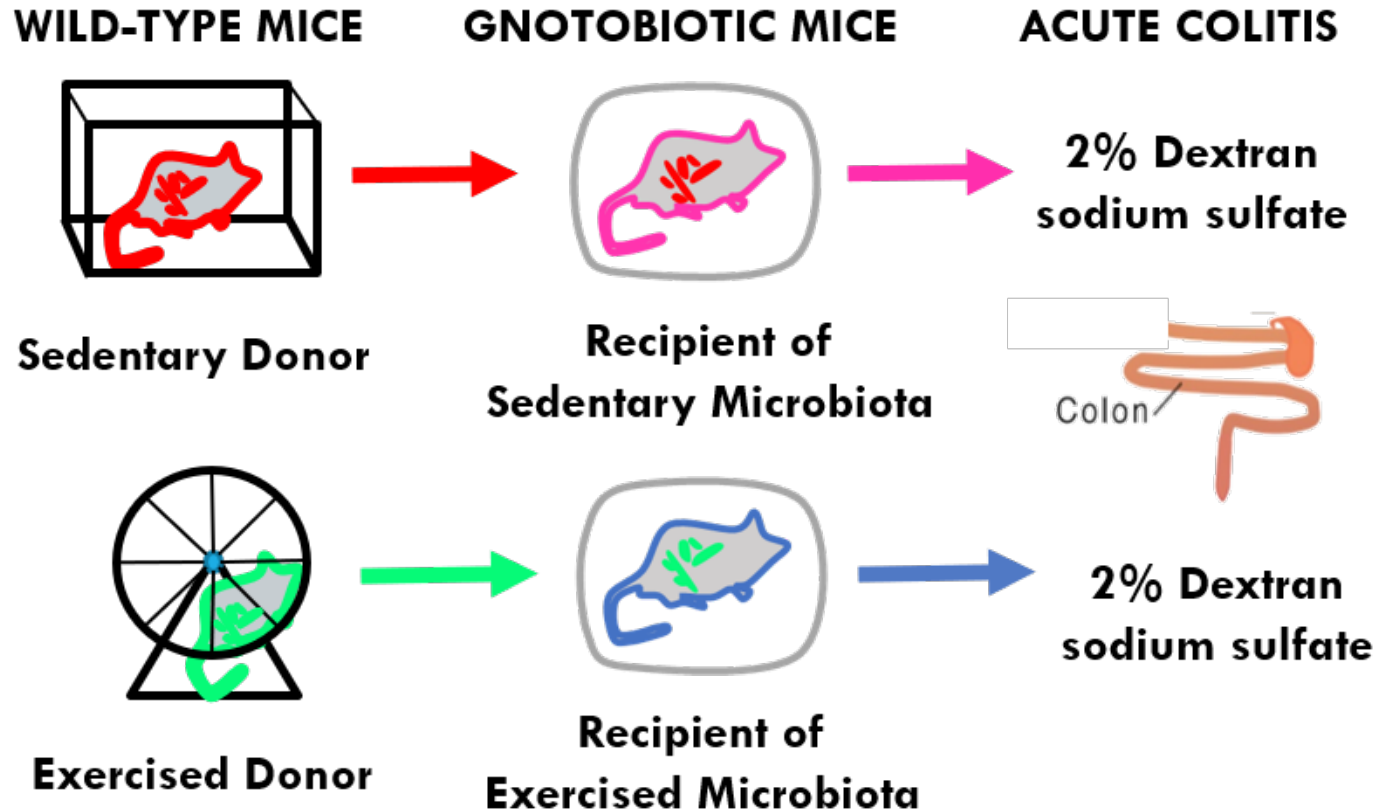
VOLUNTARY EXERCISE TRAINING:

↓ Colitis outcomes ↓ Gut inflammation

VOLUNTARY AND FORCED EXERCISE DIFFERENTIALLY ALTER THE GUT MICROBIOTA



AN "EXERCISED" MICROBIOTA TRANSPLANT ATTENUATES SYMPTOMS OF COLITIS



MYTH #5: ALL HERBAL SUPPLEMENTS USED TO TREAT DYSBIOSIS ARE SAFE & EFFECTIVE

Herbs have great potential to beneficially modulate the gut microbiota

- Inhibit pathogenic overgrowth
- Act as prebiotics, stimulating growth of beneficial bacteria

But, also great potential to do harm.

- *In vitro*, grape seed extract found to be worse than the antibiotic clindamycin!
 - Even appeared to have greater inhibition of beneficial bacteria



Most studies done using culture inoculated with human fecal material → NEED for more human clinical trials!



PROBIOTICS:

“Live microorganisms which when administered in adequate amounts confer a health benefit on the host”

– Hill et al. 2014 --

MYTH #6: STRAIN DOESN'T MATTER IF YOU KNOW THE PROBIOTIC SPECIES

Genus	Species	Strain
Lactobacillus	rhamnosus	GG

Two strains of the same species can have VERY different characteristics!!

- Most probiotic formulations do not provide information about their strains or have “secret strains” they are not willing to share

I only recommend probiotics that provide strain information AND have demonstrated efficacy and safety in human clinical trials

MYTH #7: FERMENTED FOODS HAVE ALL OF THE HEALTH BENEFITS OF PROBIOTICS

Fermented foods include kombucha, sauerkraut, kimchi, yogurt, and kefir

- May contain a diverse community of microbes that are not well-defined
- Strains may differ from batch to batch and lack specific therapeutic qualities

Wild ferments are typically not harmful, BUT can't be relied upon for therapeutic effects

Not the same as products containing standardized, thoroughly-researched strains.



MYTH #8: PROBIOTICS HELP “RE-SEED” THE GUT MICROBIOTA

Most probiotics do not permanently colonize the gut

- Some appear to transiently colonize
- Some do not colonize at all

However, probiotics still confer benefits while in transit through the GI tract

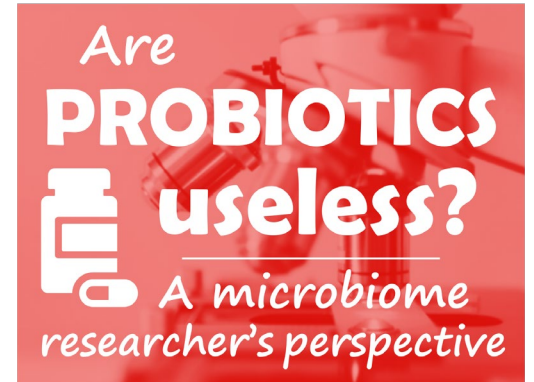
- Alter gene expression
- Modulate the immune system
- May outcompete pathogens



MYTH #9: YOU SHOULD ALWAYS TAKE PROBIOTICS AFTER ANTIBIOTICS

Latest evidence suggests that probiotics taken after antibiotics may delay the return of the native microbiota

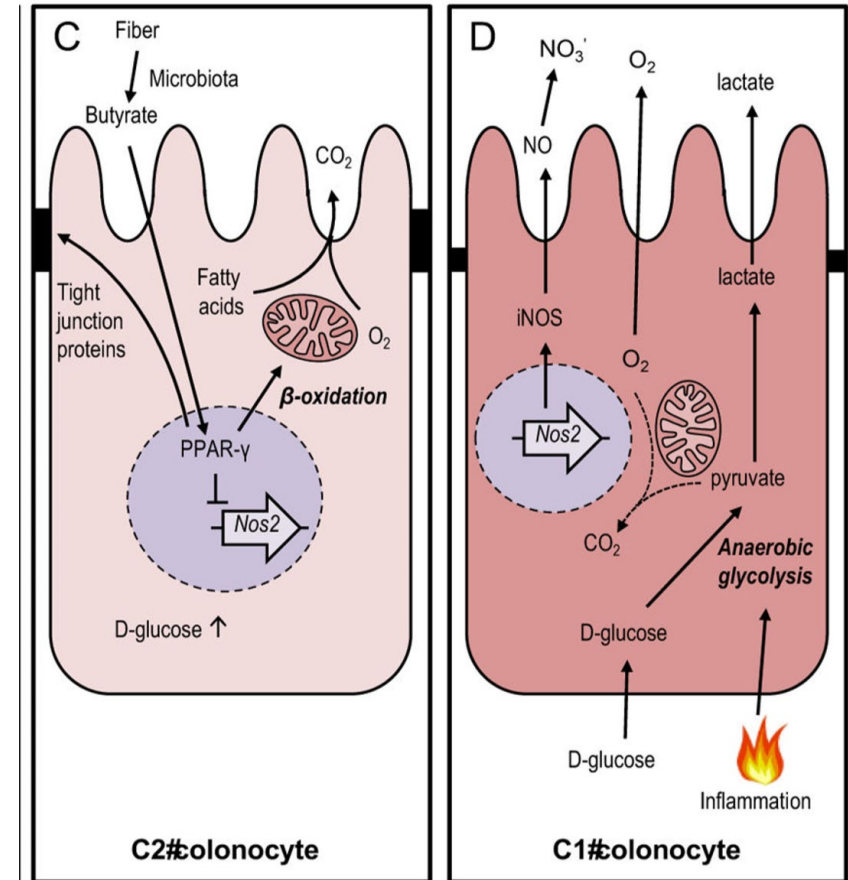
- Particularly delayed return of butyrate-producing microbes
- Possible that *Saccharomyces cerevisiae* var. *boulardii* or other probiotic formulations may be effective, but more research needs to be done.



THE OXYGEN-DYSBIOSIS CONNECTION: COULD BUTYRATE PREVENT ABX-INDUCED DYSBIOSIS?

Depletion of colonic butyrate by antibiotics or a low fiber diet results in oxygenation of the colonic mucosa

- Downregulation of PPAR γ and a shift towards an inflammatory colonocyte profile.
- Possible that taking butyrate during and after antibiotics may prevent this switch in colonocyte metabolism and attenuate dysbiosis.





PREBIOTIC:

“A substrate that is selectively utilized by host microorganisms conferring a health benefit”

– Gibson et al. 2017 --

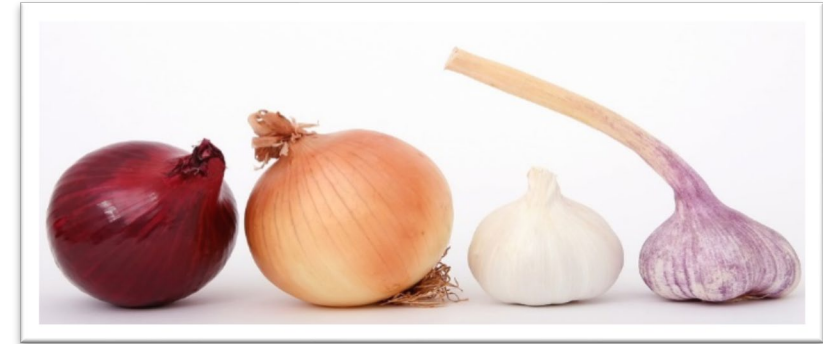
MYTH #10: PREBIOTIC SUPPLEMENTS WORK THE SAME FOR EVERYONE

3rd most common and fastest-growing non-vitamin dietary supplement in the U.S

- Lots of people testing the gut microbiome, and then looking to find a prebiotic that will boost particular microbes

But response to prebiotic supplementation is **HIGHLY** individualized

- For instance, resistant starch is supposed to increase production of butyrate
- On average, it does. However, some people actually go down in butyrate production with RS supplementation



In the future, in vitro diagnostic tools could be used to predict prebiotic response and personalize prebiotic therapy

MYTH #11: FECAL MICROBIOTA TRANSPLANT IS SAFE TO DO AT HOME

FMT: excellent efficacy for recurrent C. diff.

Currently being explored for a wide range of other conditions

Still a lot we are missing with current screening techniques: viruses, uncharacterized microbes

- Recent FDA report of two patients that contacted severe infections, one of whom died, from an FMT that contained a multi-drug resistant organism

Always best to find an experienced clinic that is carefully screening donors

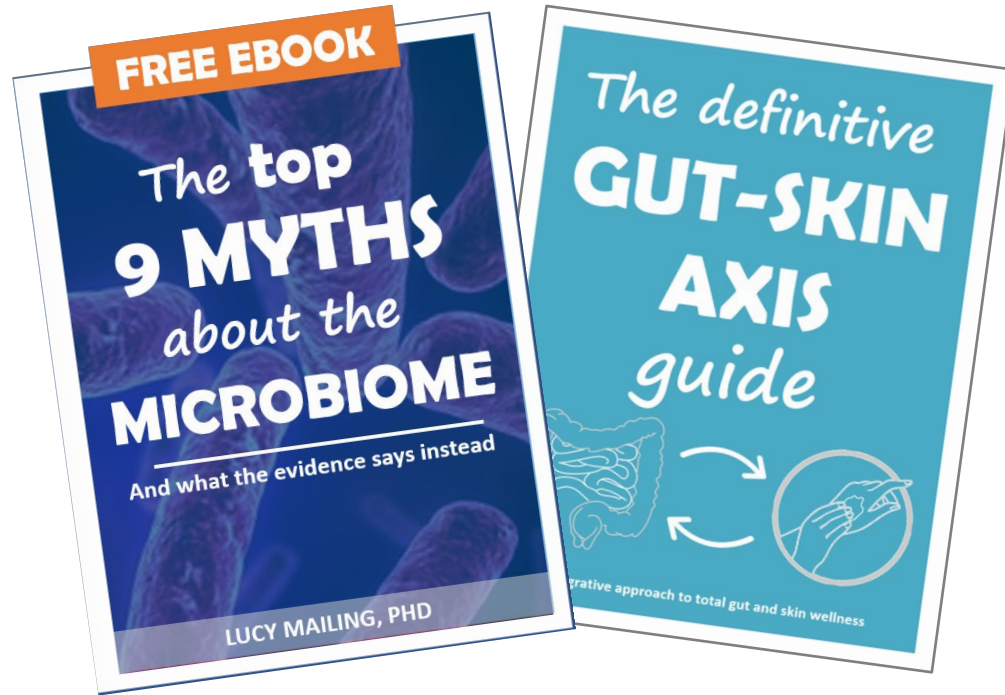


SUMMARY

- **Microbiome modulation has enormous therapeutic potential, but we need to follow the evidence**
- **Be wary of tests and supplements that lack evidence**
- **Many companies taking advantage of hype without any understanding of the literature**
- **Lots yet to be understood and discovered!**

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