



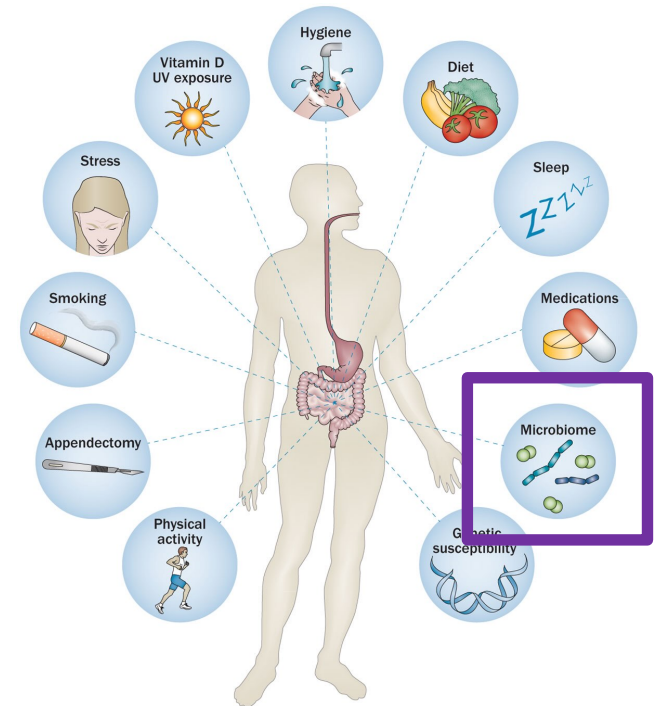
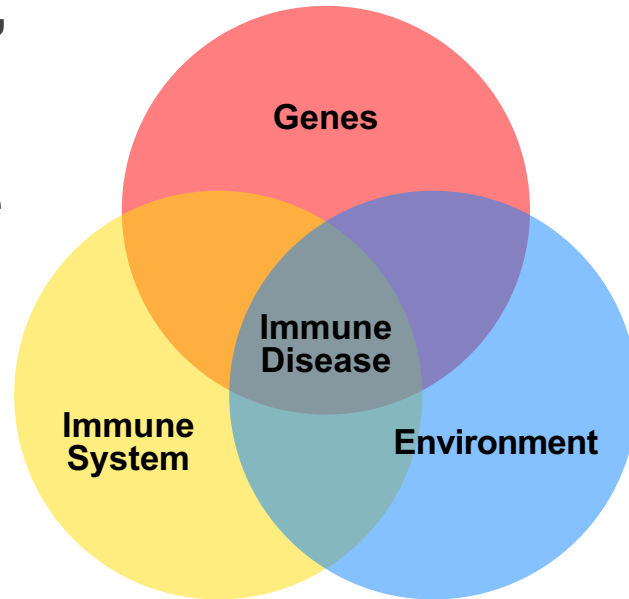
# Microbiota and Human Health: A Role in Cancer

Gretchen Diehl, PhD



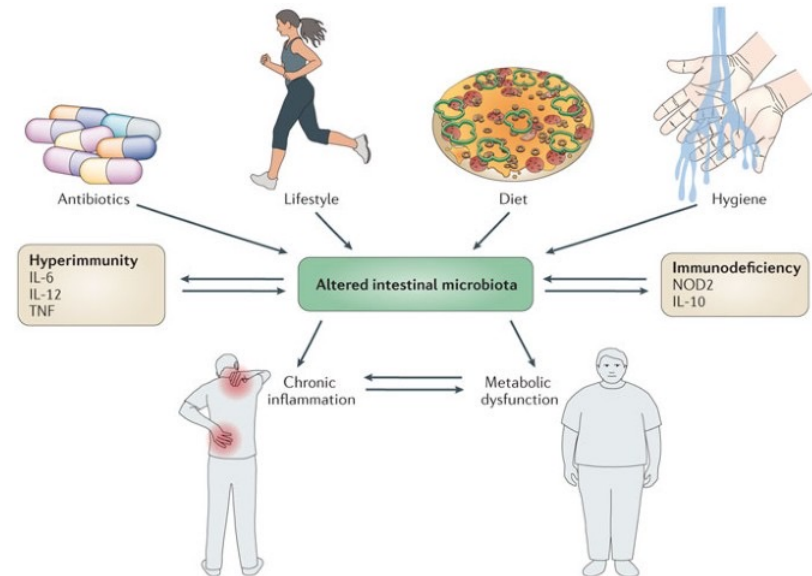
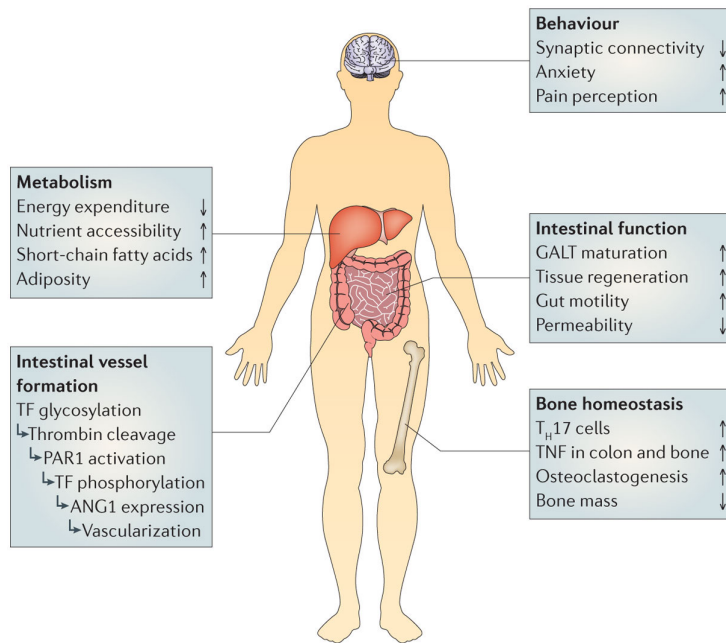
Memorial Sloan Kettering  
Cancer Center

# Interaction between genetics, immune system and environment underlies multiple diseases



Ananthakrishnan, A. N. (2015) Epidemiology and risk factors for IBD  
Nat. Rev. Gastroenterol. Hepatol. doi:10.1038/nrgastro.2015.34

# Microbiota (all the microbes)/Microbiome (all the genomes) are implicated in human disease



Sommer & Bäckhed (2013) The gut microbiota — masters of host development and physiology, *Nature Reviews Microbiology* PMID: 23435359

## Microbiota, Health, and Cancer

- How to measure the microbiota
- Describe key examples of microbiota regulation of health: colonization resistance, immune system development, hematopoiesis, vaccines, barrier repair, breakdown of dietary components and drugs
- Describe microbial metabolites (eg SCFA) that regulate host functions
- Describe contribution to disease states: alteration in community abundance in inflammatory disease
- Microbiota and tumors: drivers, dysbiosis, therapies, outcomes
- Keeping the microbiota in mind (more work to be done)



In the average adult are  
100 trillion human cells  
and 1,500 trillion  
microbes.

At best you are little  
more than 10% you.

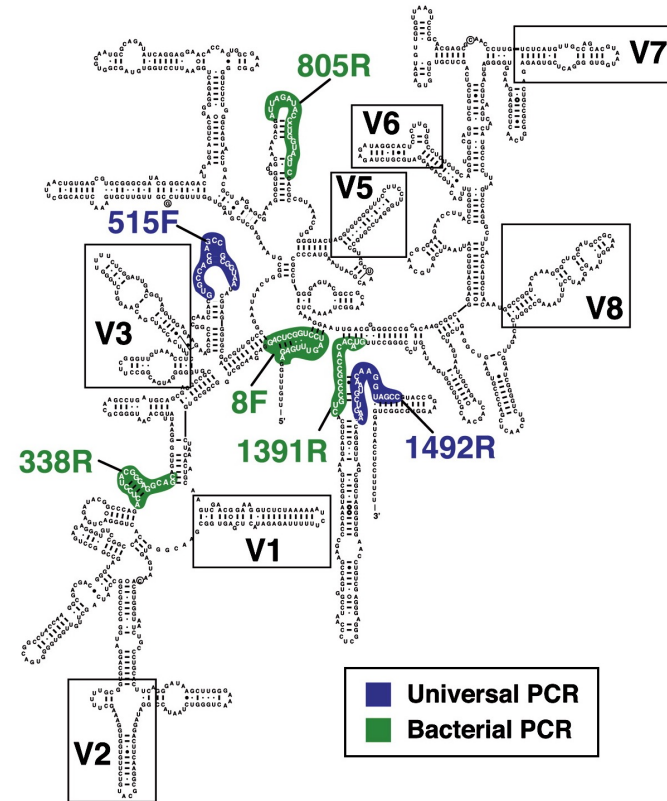
**We're all just  
petri dishes  
with shoes.**



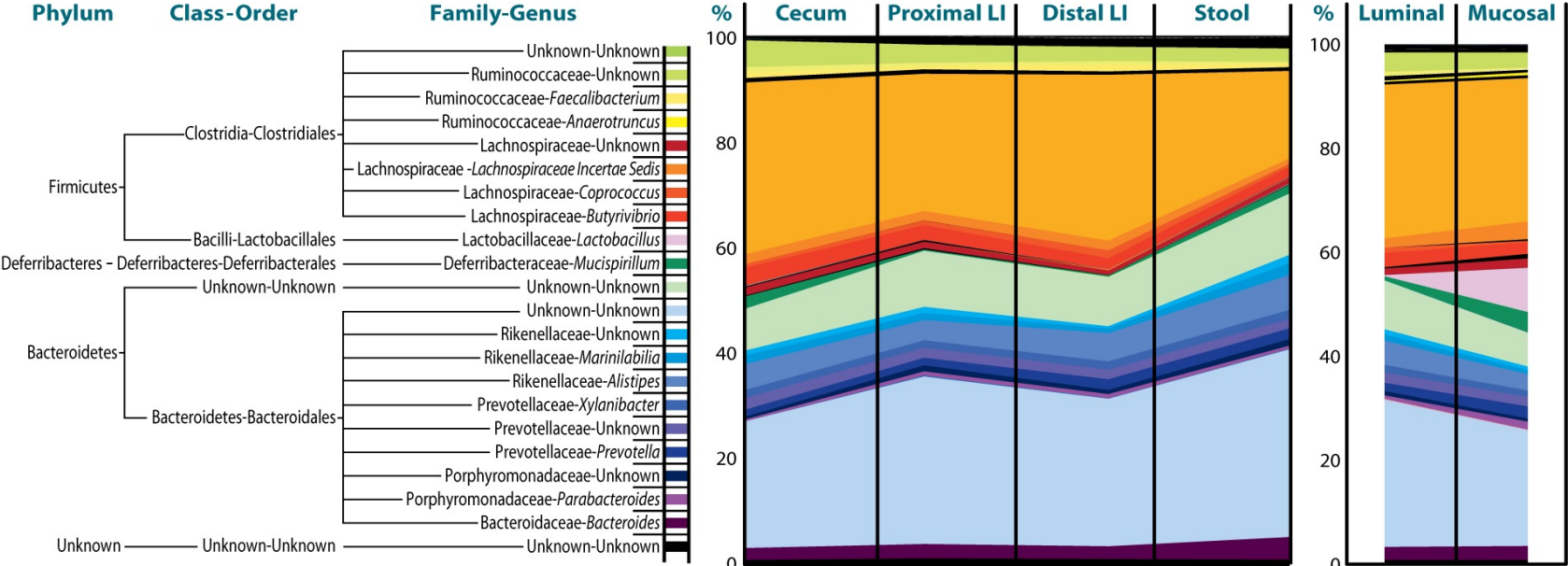
[schuhlewis.blogspot.com](http://schuhlewis.blogspot.com)

# Defining the Microbiota

- Culture-independent system based on 16S rRNA to organize bacteria species
- Metagenomics: composite genomes

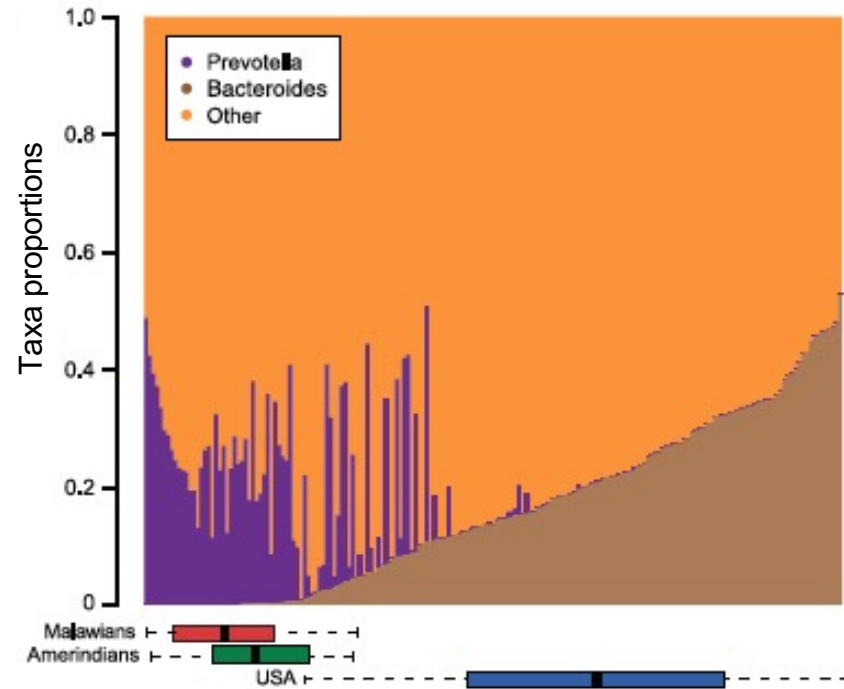


# Bacteroidetes and Firmicutes Dominate Intestinal Microbiome



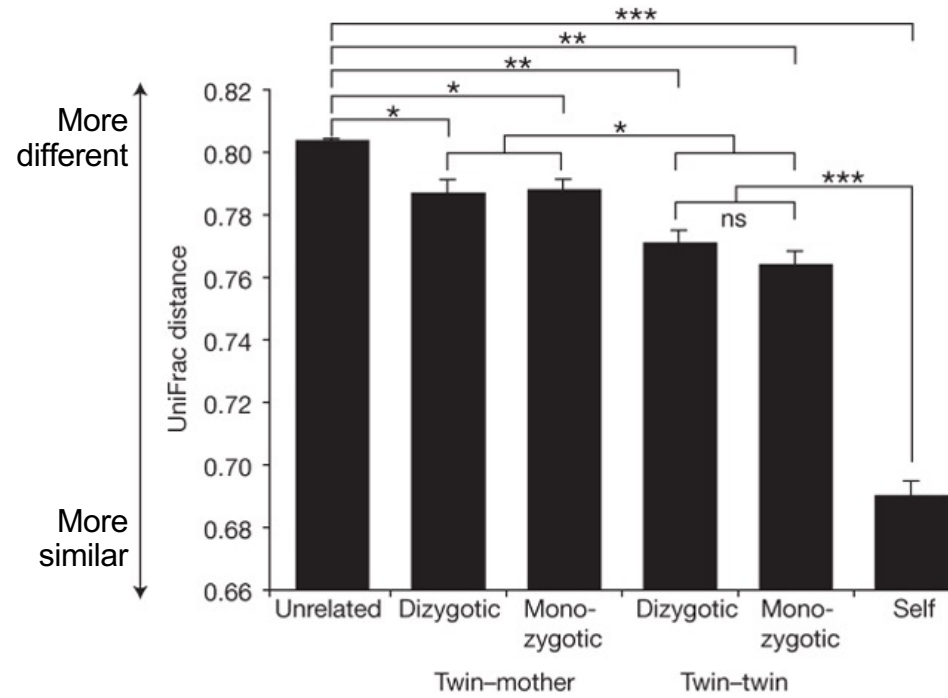
Hill DA, Artis D. 2010. Annu. Rev. Immune. 28:623-67

# Cultural/Geographic Region Affects Microbiome

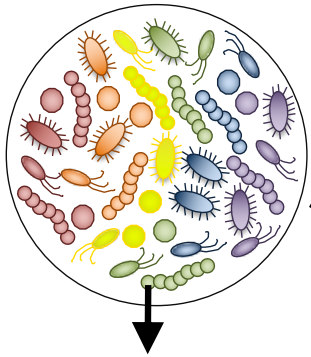


Yatsunenko, Nature 2012

# Significant Interpersonal Variation in Microbiota



## Microbiota



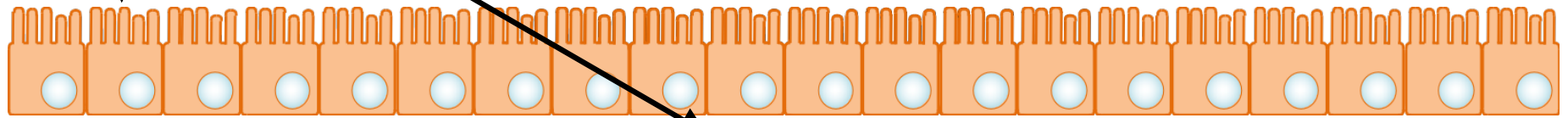
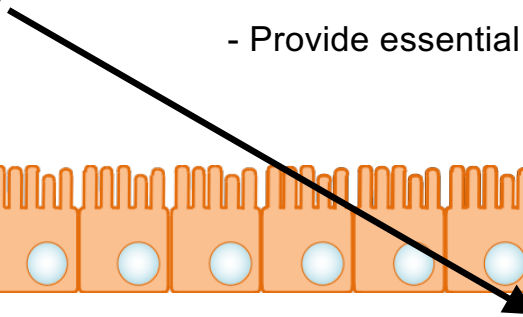
### Compete with pathogens

- Space
- Nutrients
- Secrete anti-microbial factors



### Metabolism

- Break down of indigestible carbohydrates
- Provide essential nutrients

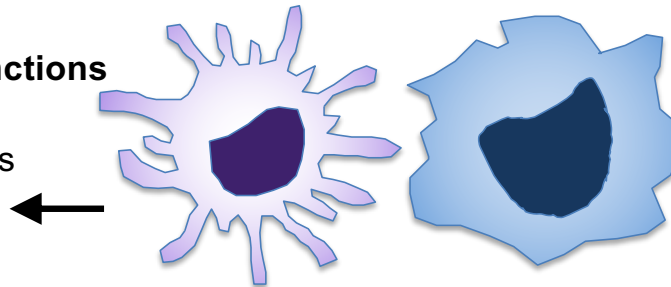


### Barrier function

- Epithelial cell differentiation and proliferation
- Intestinal repair

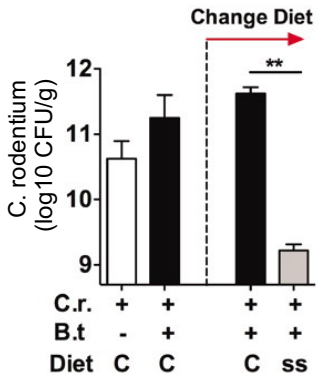
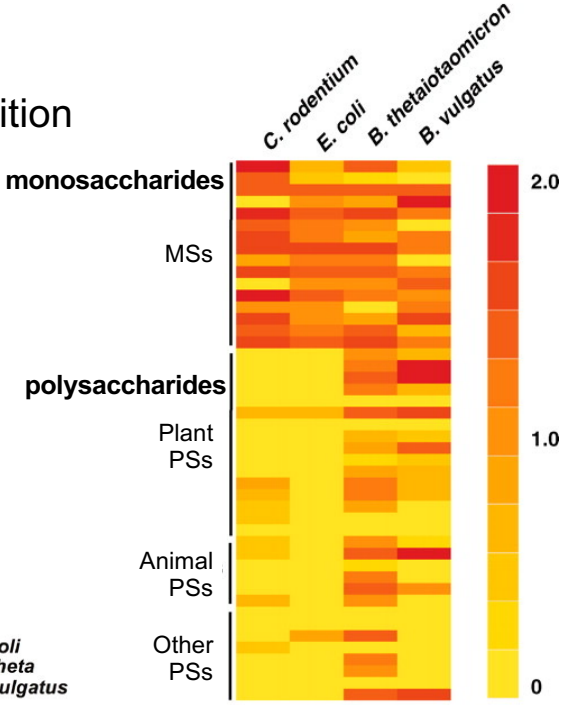
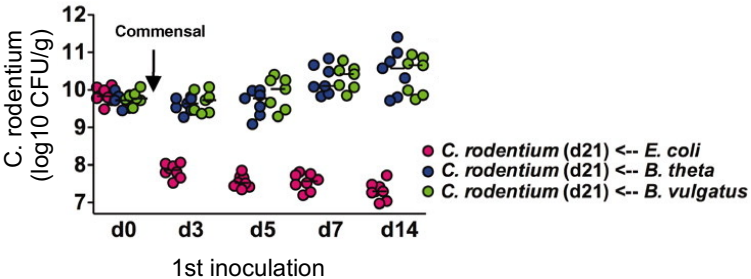
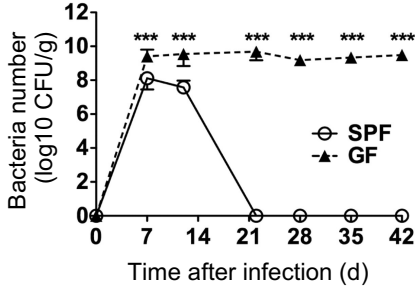
### Anti-microbial functions

- Induction of anti microbial peptides
- Mucus

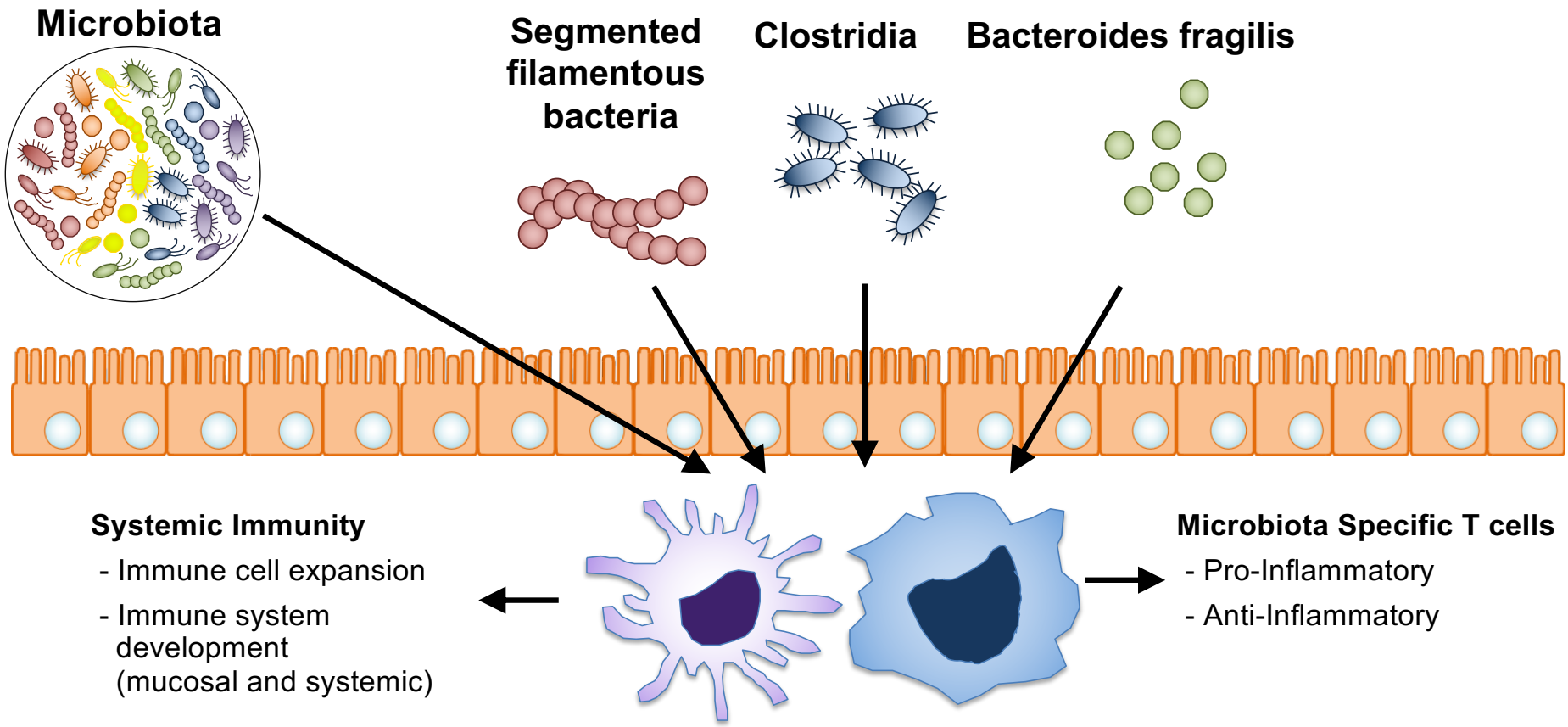


# Colonization Resistance

Microbiota protect through nutrient competition

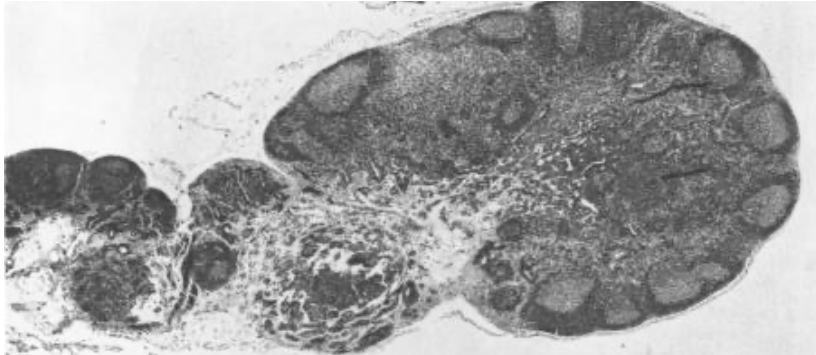


Kamada et al. Science 2012;336:1325-1329

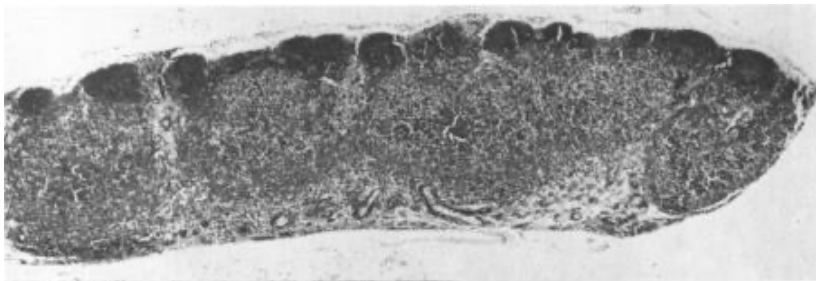




# Microbiota Regulation of Immune System Development and Function



Mesenteric lymph node  
in conventional mice:  
Numerous B cell follicles



Mesenteric lymph node  
in germ free mice:  
No B cell follicles

BAUER H, HOROWITZ  
RE, LEVENSON SM,  
POPPER H. 1963. The  
response of the lymphatic  
tissue to the microbial  
flora. *Studies on germfree  
mice. Am J Pathol.*  
42:471-83.

# Immune Defects in germfree Animals

Immunological defect	Site	Phenotype in germ-free mice compared with conventionally housed mice
Development of small intestine	Peyer's patches	Fewer and less cellular
	Lamina propria	Thinner and less cellular
	Germinal centres	Fewer plasma cells
	Isolated lymphoid follicles	Smaller and less cellular
Development of mesenteric lymph nodes	Germinal centres	Smaller, less cellular and with fewer plasma cells
CD8 <sup>+</sup> T cells	Intestinal epithelial lymphocytes	Fewer cells and with reduced cytotoxicity
CD4 <sup>+</sup> T cells	Lamina propria	Fewer cells; decreased T <sub>H</sub> 17 cells in the small intestine but increased T <sub>H</sub> 17 cells in the colon
CD4 <sup>+</sup> CD25 <sup>+</sup> T cells	Mesenteric lymph nodes	Reduced expression of FOXP3 and reduced suppressive capacity
Expression of angiogenin 4	Paneth cells	Reduced
Expression of REG3 $\gamma$	Paneth cells	Reduced
Production of secretory IgA	B cells	Reduced
Levels of ATP	Intestine	Reduced
Expression of MHC class II molecules	Intestinal epithelial cells	Reduced
Expression of TLR9	Intestinal epithelial cells	Reduced
Levels of IL-25	Intestinal epithelial cells	Reduced

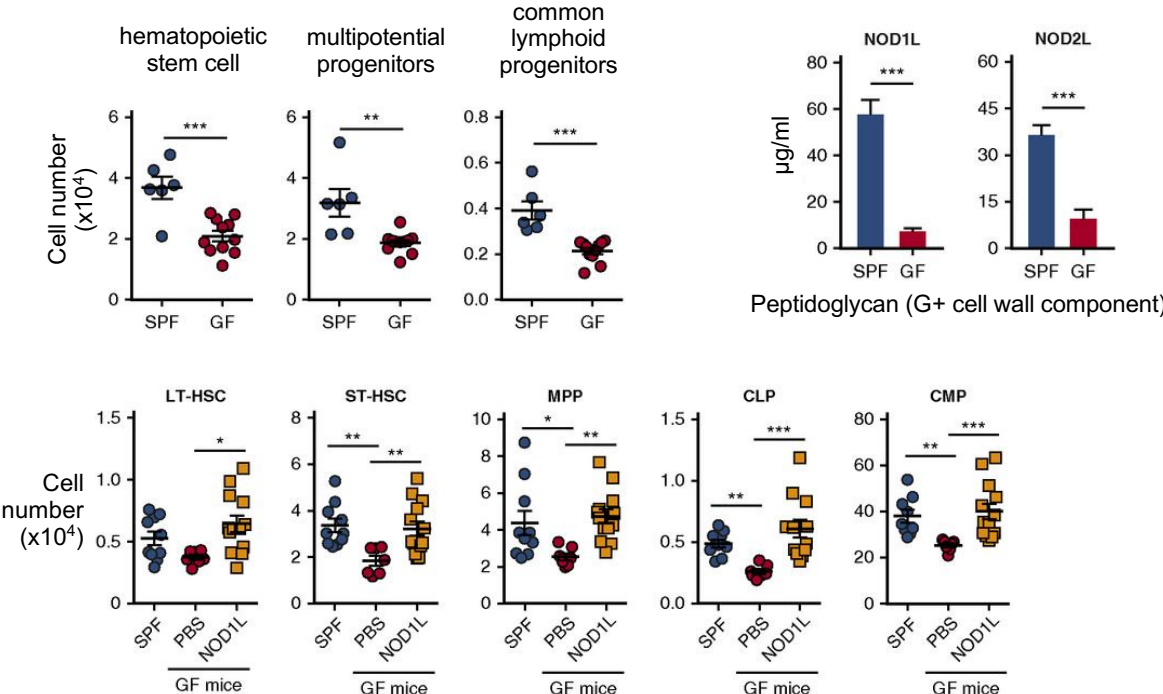
FOXP3, forkhead box P3; IL-25, interleukin 25; REG3 $\gamma$ ; regenerating islet-derived 3 $\gamma$ ; T<sub>H</sub>17, T helper 17; TLR9, Toll-like receptor 9.



Memorial Sloan Kettering  
Cancer Center

Round & Mazmanian.  
The gut microbiota shapes intestinal immune responses during health and disease. *Nature Reviews Immunology* 9, 313-323 (May 2009)  
| doi:10.1038/nri2515

# Microbiota Regulation of Hematopoiesis

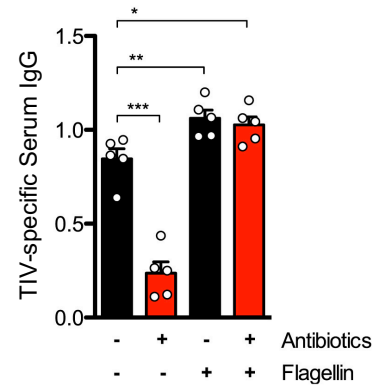
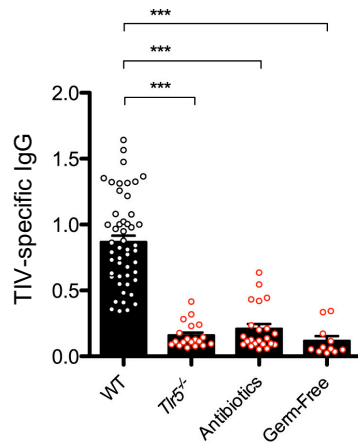


Iwamura (2017) Blood, PMCID: PMC5234217

# Microbiota as a Vaccine Adjuvant

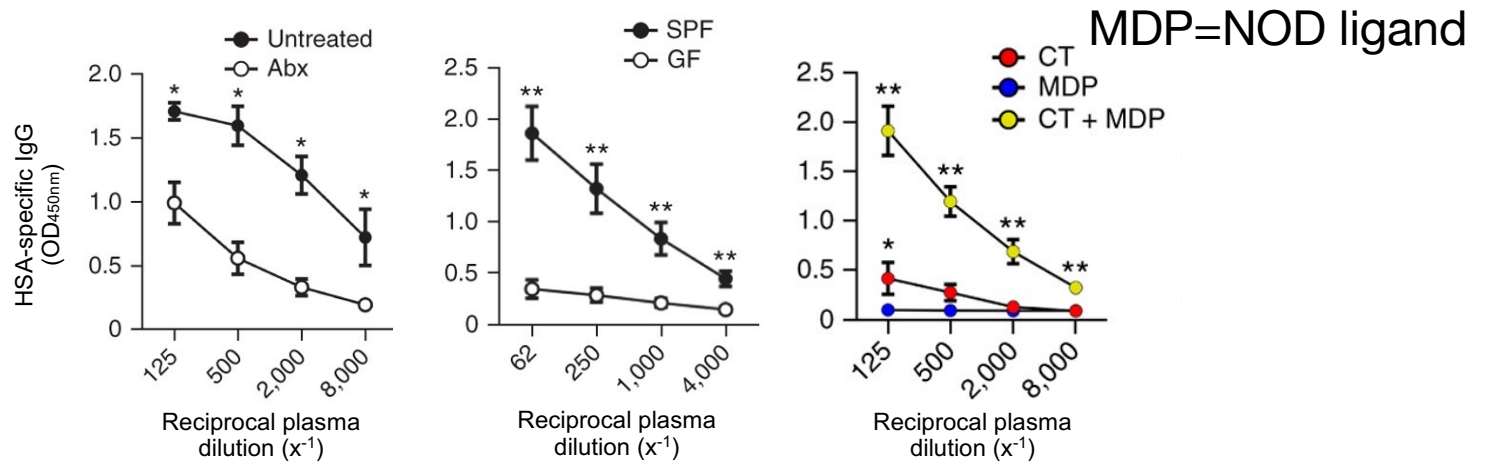
- Trivalent inactivated influenza vaccine (TIV): subunit vaccine, HA from 3 flu strains, unadjuvanted
- Earlier work: in humans found correlation between higher TLR5 expression and flu vaccine responsiveness

- Loss of responses in antibiotic treated in inactivated polio but not adjuvanted vaccines or live-attenuated yellow fever



# Microbiota as a Vaccine Adjuvant

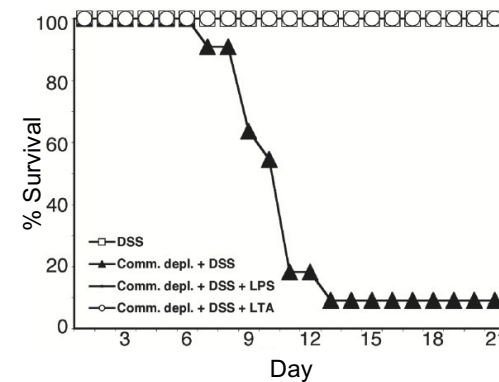
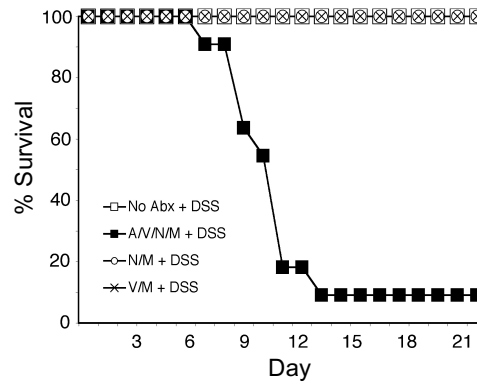
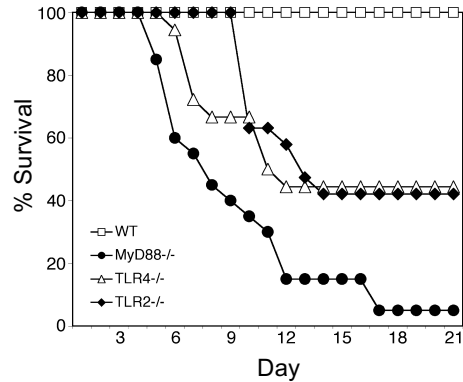
- Cholera toxin: mucosal adjuvant (responsible for diarrhea associated with cholera)



# Microbiota Improves Barrier Repair After Intestinal Injury

Dextran sodium sulfate (DSS):

- chemical model of colitis
- causes epithelial damage

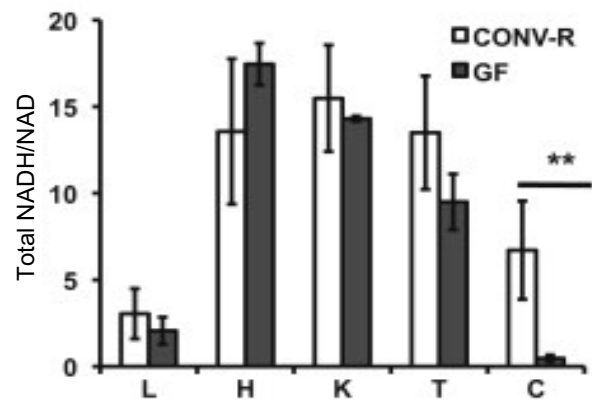


Rakoff-Nahoum (2004)  
Cell PMID: 15260992

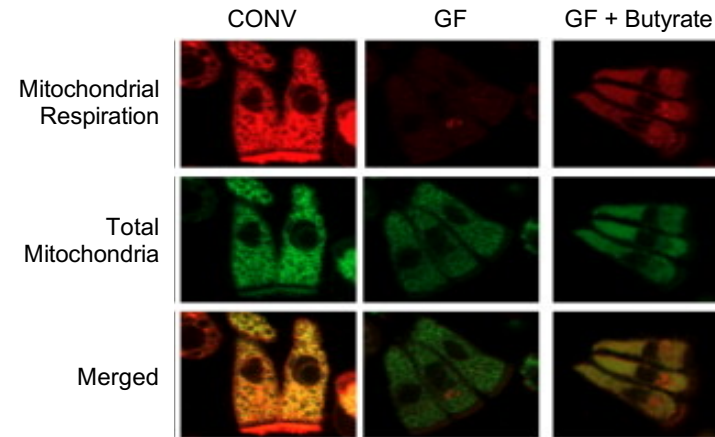
## Microbiota Directly Impacts Health of Epithelial Cells

- Short-chain fatty acids (SCFAs), primarily acetate, propionate and butyrate:  
product of bacterial fermentation by obligate anaerobes (Firmicutes, Bacteroidetes, and Clostridium)
  - Energy salvage: primarily  
breakdown of undigested dietary carbohydrates (fiber),  
mucus, and sloughed epithelial cells.  
contribute 5–15% of human total caloric requirements
  - Energy source  
Butyrate: epithelial cells,  
Acetate: muscle and adipose tissue
  - Signal through receptors: GPR109 (butyrate), GPR43 (acetate/propionate)  
Butyrate can activate transcription as a HDAC inhibitor
- \*Multiple beneficial effects: anti-inflammatory, epithelial barrier, Tregs, macrophage microbial killing, satiety (regulates GLP-1), and oxidative stress\*\*

# SCFA Butyrate is an Energy Source for Colonic Epithelial Cells



- Specific metabolic defect in defect in colon from GF mice

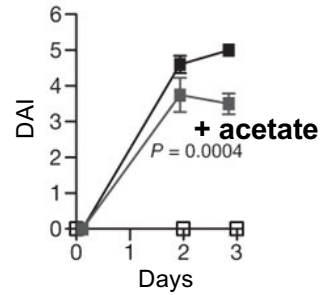
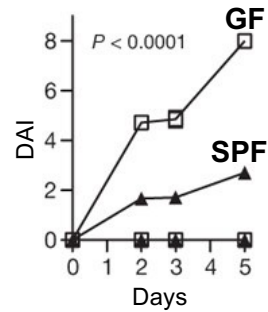


- Butyrate rescues (partially) mitochondria respiration

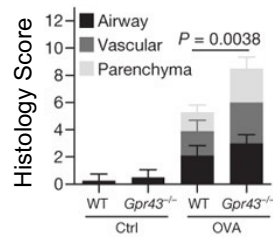
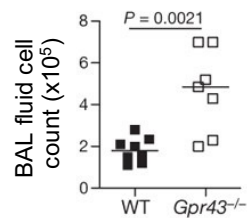


# SCFA Protect from Inflammatory Disease

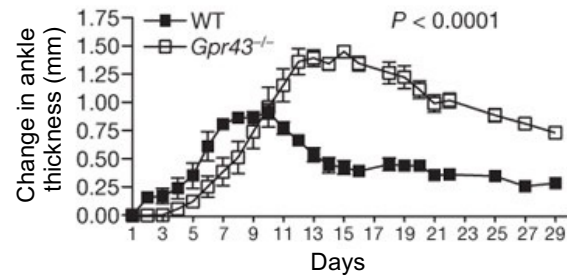
## Colitis



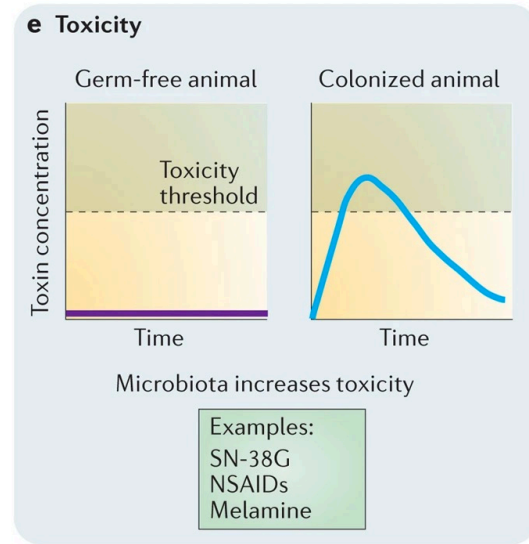
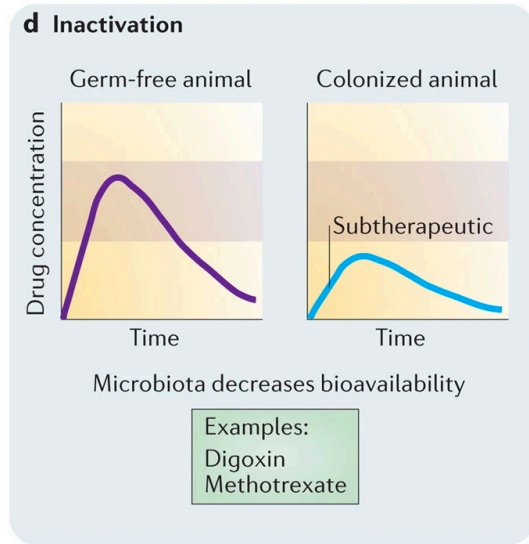
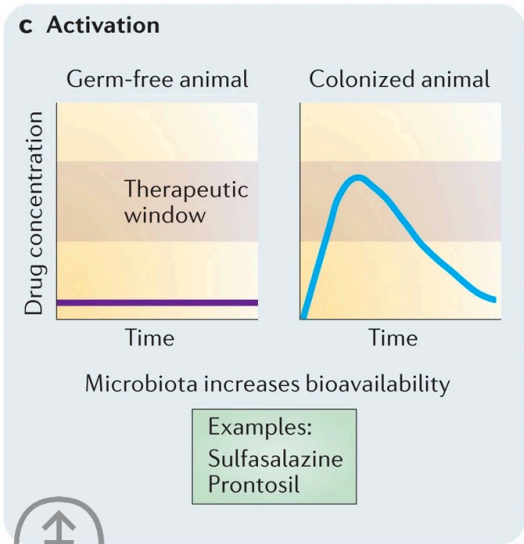
## Asthma



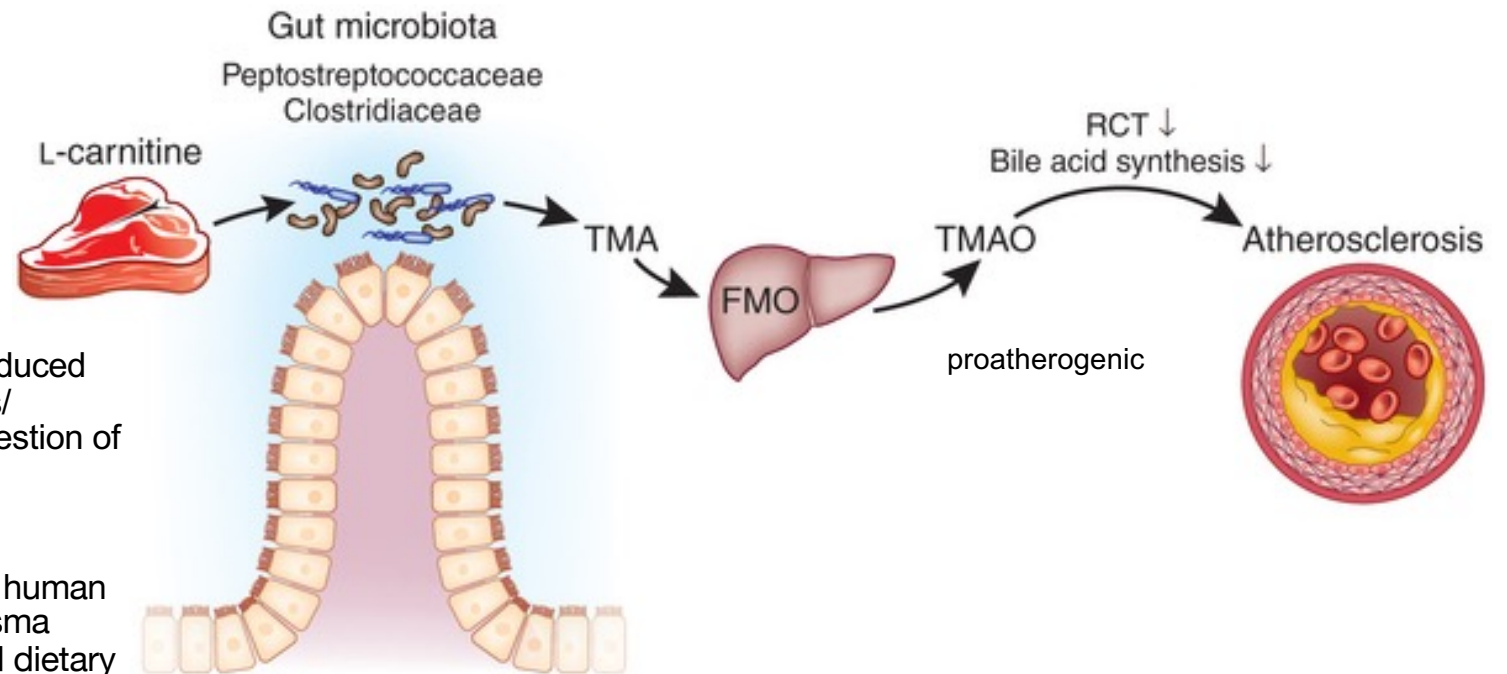
## Arthritis



# Microbiota Metabolize Xenobiotics



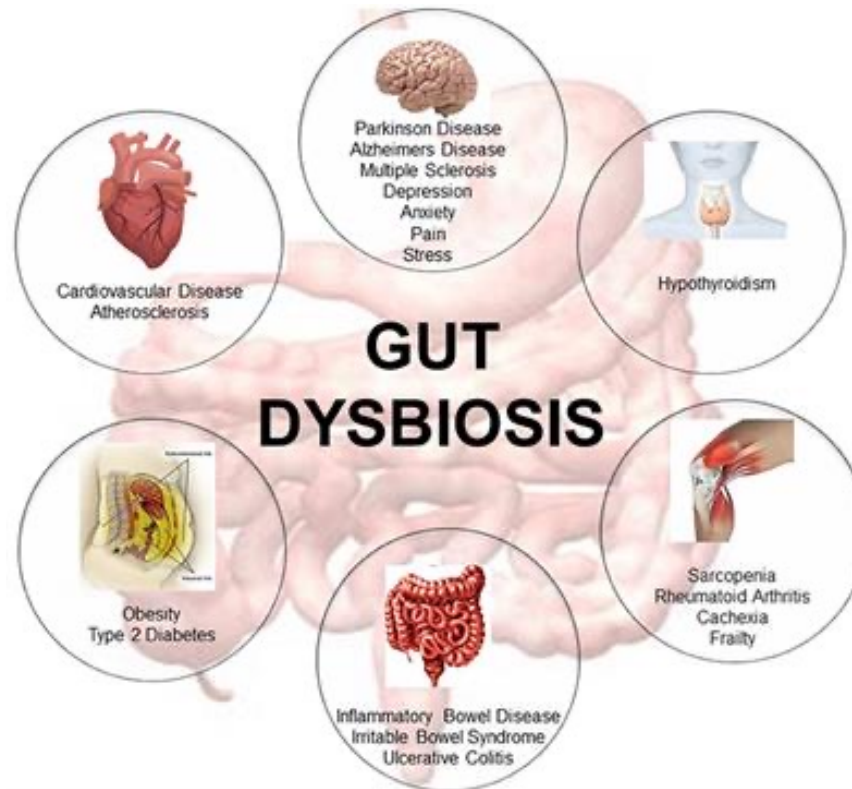
# Microbiota Metabolize Xenobiotics: Meat-metabolizing bacteria in atherosclerosis



Omnivorous humans produced more TMAO than vegans/vegetarians following ingestion of L-carnitine

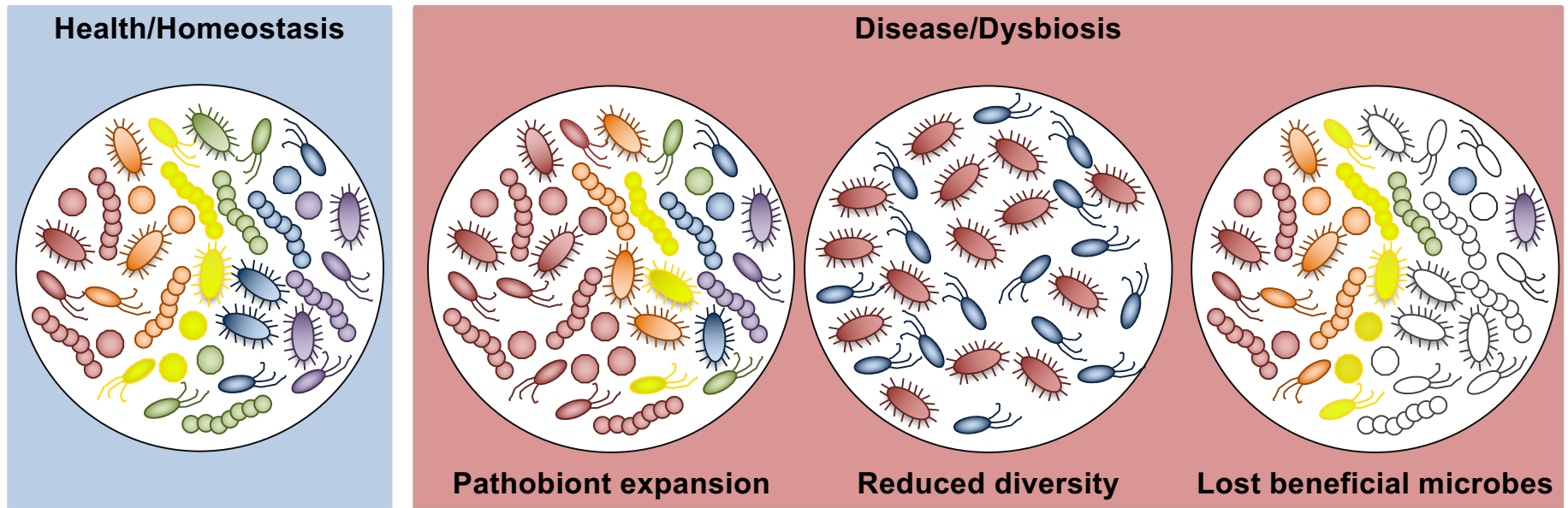
Specific bacterial taxa in human feces associate with plasma TMAO concentration and dietary status.

# Changes in Microbiota Composition are Associated with Human disease



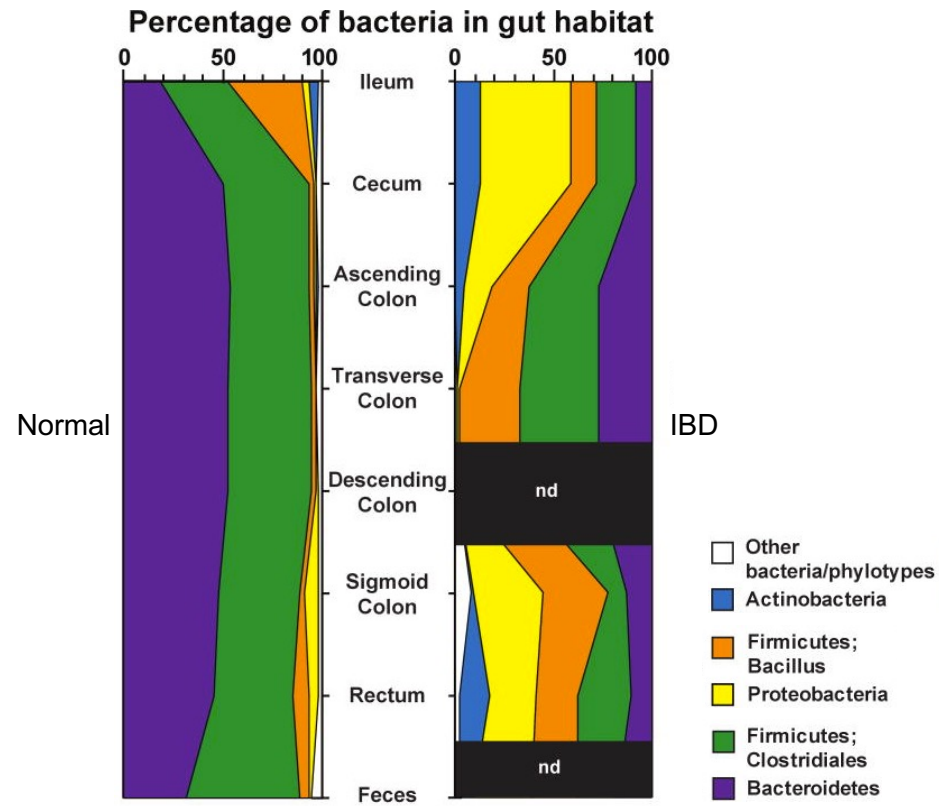
Baptista et al (2020)  
Frontiers in Nutrition.  
<https://doi.org/10.3389/fnut.2020.00017>

# Changes in Microbiota Composition are Associated with Disease



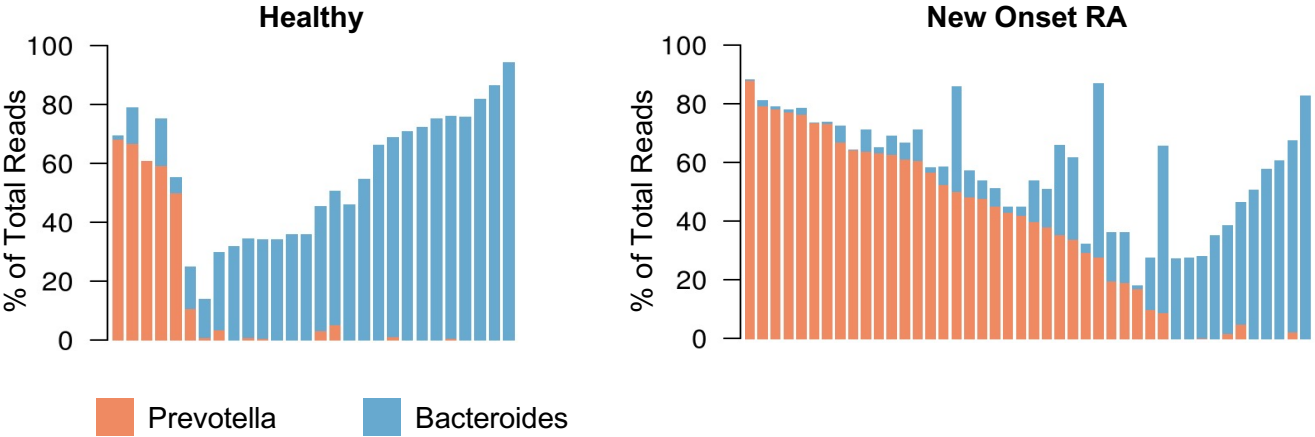
Adapted from Peterson & Round 2014. Cell Microbiol.

# Dysbiosis is Associated With Human Disease: IBD

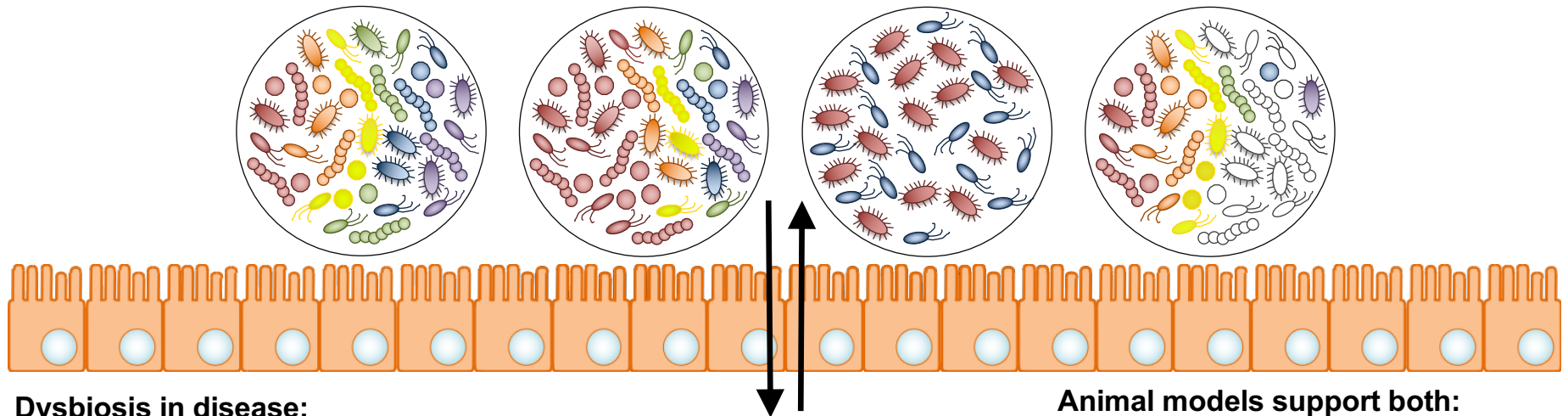


# Dysbiosis is Associated With Human Disease: RA

Prevotella dominates the intestinal microbiota in Rheumatoid Arthritis (RA)



Scher\*, Sczesnak\*, Longman\* et al eLife 2013



**Dysbiosis in disease:**

- Increased mucosal-associated bacteria
- Outgrowth of pathobionts eg proteobacteria
- Loss of anti-inflammatory microbes  
(Bacteroidetes, Lachnospiraceae and Faecalibacterium prausnitzii)

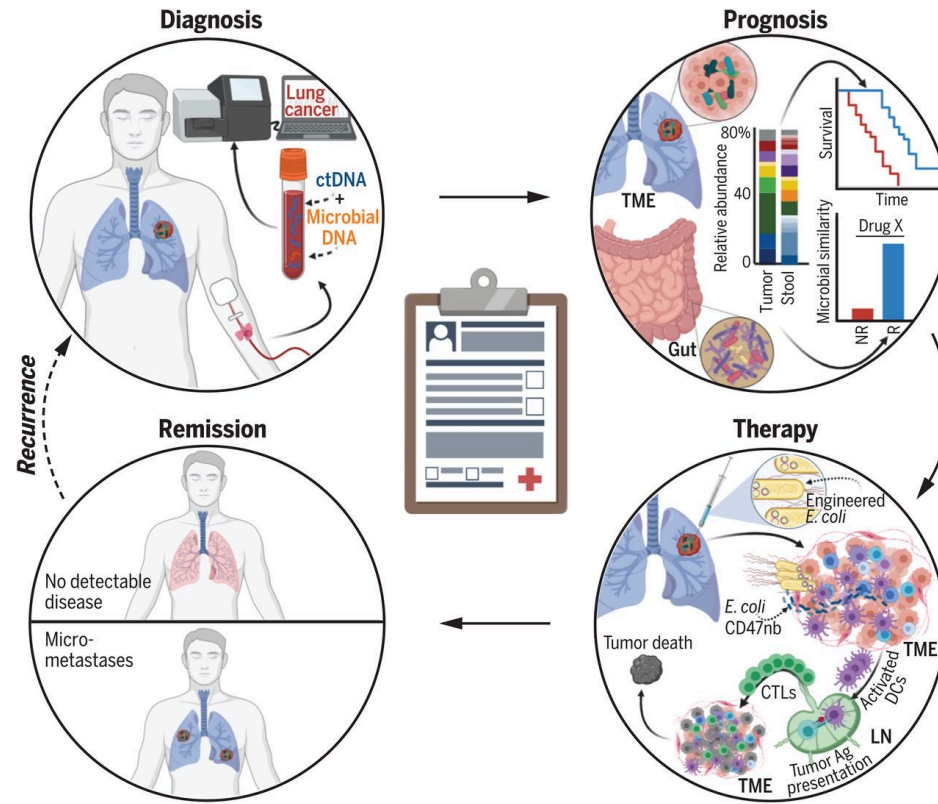
**Does dysbiosis support disease or does disease support dysbiosis?**

**Animal models support both:**

- Inflammatory environment supports microbiota shift
- Microbial shifts can cause disease

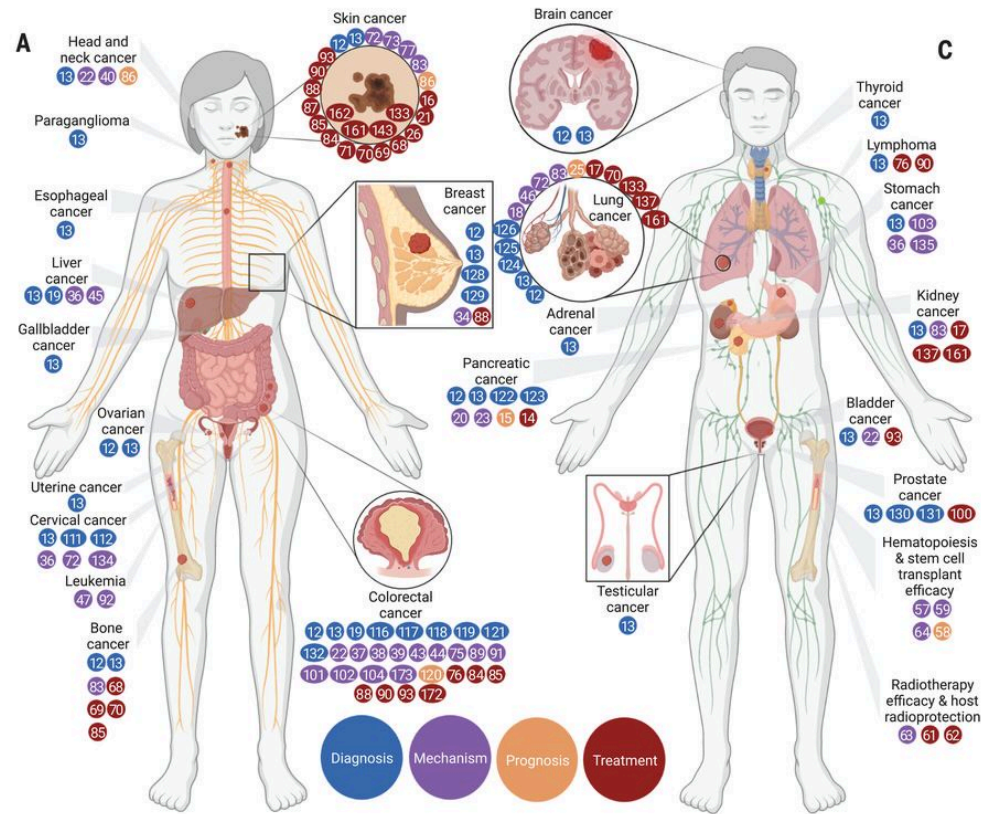


# What About Cancer?



Sepich-Poore (2121)  
Science. PMID: 33766858

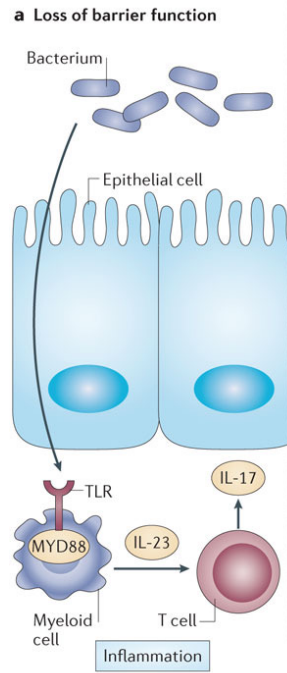
# Microbiota Association with Multiple Types of Human Cancers



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Sepich-Poore (2121)  
Science. PMID: 33766858

# Microbiota Associated with Colorectal Cancer



Elinav (2013)  
Nat Rev Cancer  
PMID: 24154716

## Microbial Drivers of Cancer

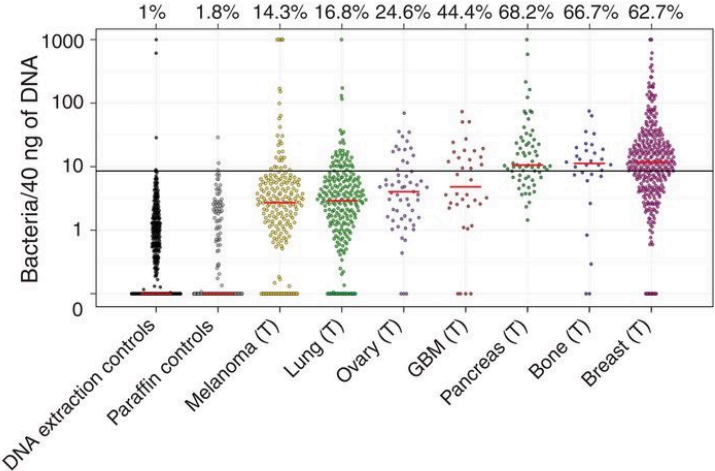
**11 microbes are identified by the International Association for Cancer Registries as human carcinogens**

Epstein Barr virus	Human Papillomaviruses
Hepatitis B virus	Human T-cell Lymphotropic virus type 1
Hepatitis C virus	Opisthorchis viverrini and Clonorchis sinensis (flatworms)
Kaposi Sarcoma herpesvirus	Schistosoma haematobium
HIV-1	Helicobacter pylori

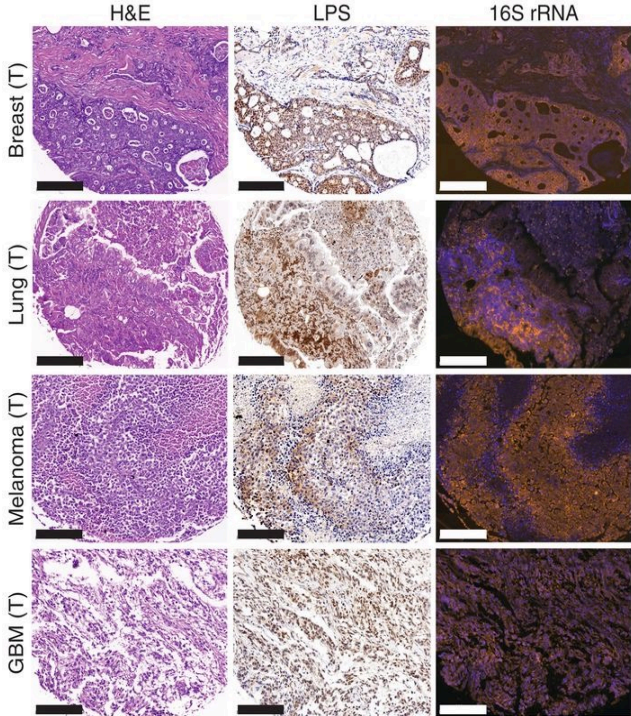
**These account for ~13% of global cancer cases**

# Intra-Tumoral Microbes

Tissue	Normal: # samples (# centers)	Tumor: # samples (# centers)
Breast	256 (3)	355 (3)
Lung	231(3)	245 (3)
Melanoma	—	206 (3)
Pancreas	—	67 (2)
Ovary	29 (2)	58 (2)
Bone	—	39 (2)
GBM	—	40 (2)
Total	1526	
DNA extraction controls	437	
16S 5R PCR controls	206	
Paraffin Controls	168 (4)	

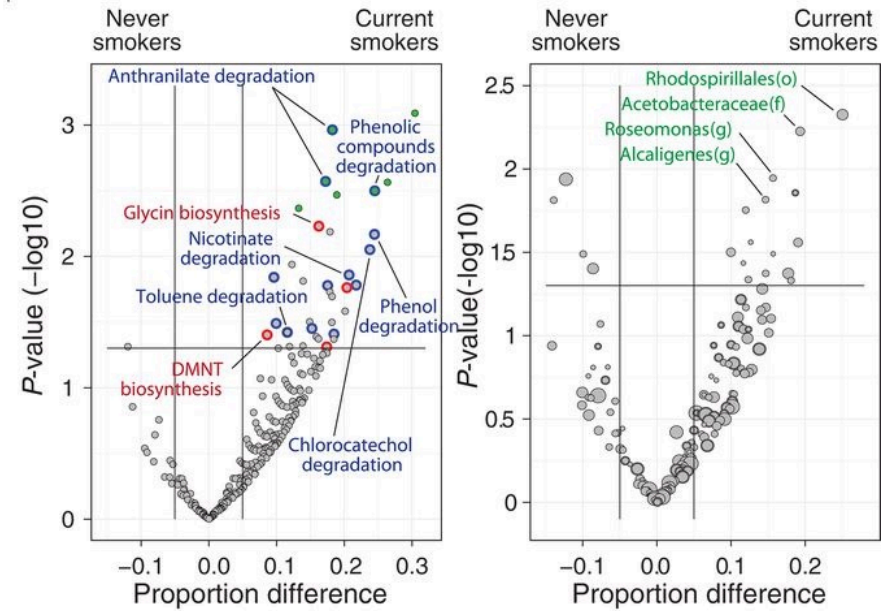
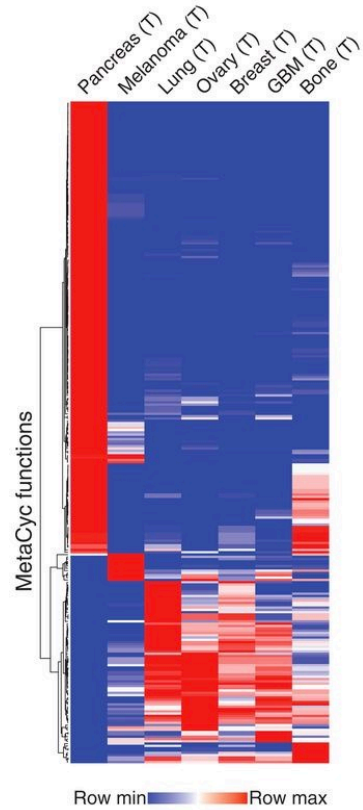
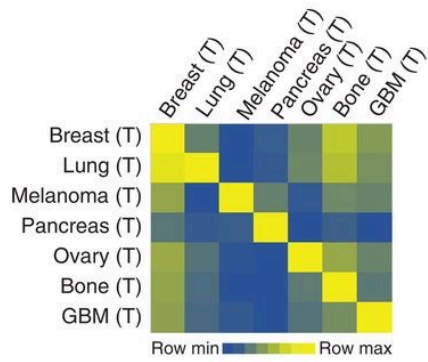


Bacterial DNA found in normal as well



Nejman (2020) Science PMID: 32467386

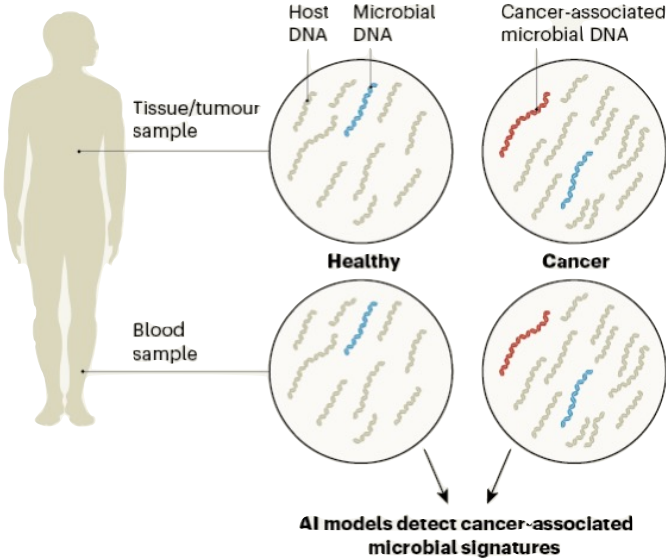
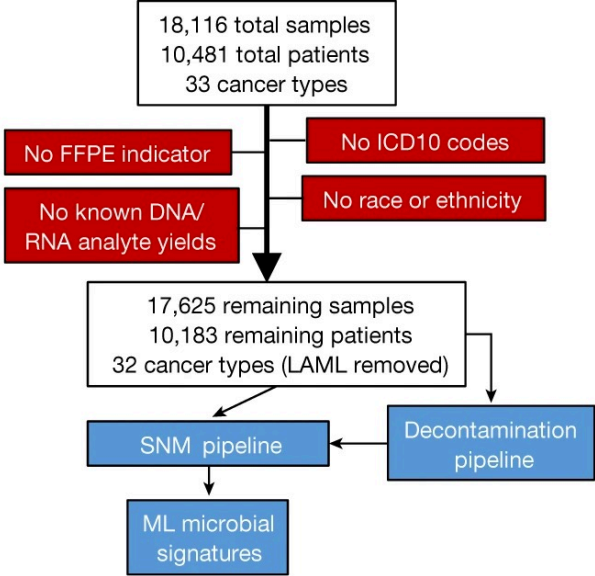
# Intra-Tumoral Microbes



Nejman (2020) Science PMID: 32467386



# Microbial Signatures in Blood May Reveal Cancer Presence



Poore (2020). Nature. PMID: 32214244

Ajami (2020) Nature. PMID: 32161344

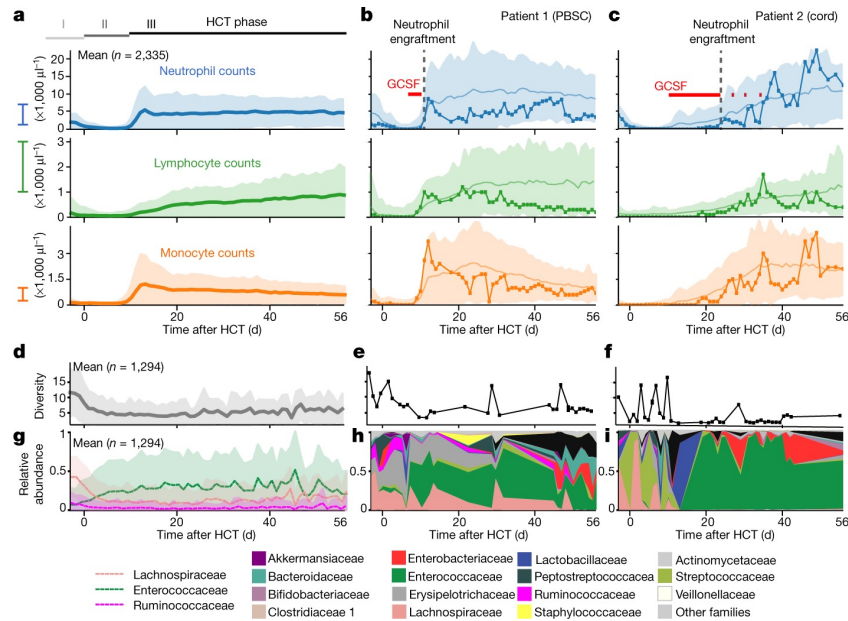
## Intratumoral microbes

- Genotoxins increase mutations
- Activate pro-tumor survival or proliferation
- Suppress anti-tumor immunity
- Drug resistance through microbial metabolism
- As therapies:
  - Can we selectively target intratumor specific microbes?
  - if there are specific tumor homing microbes, can we target them to tumors to deliver drugs (anti-cd47) to avoid systemic toxicity?



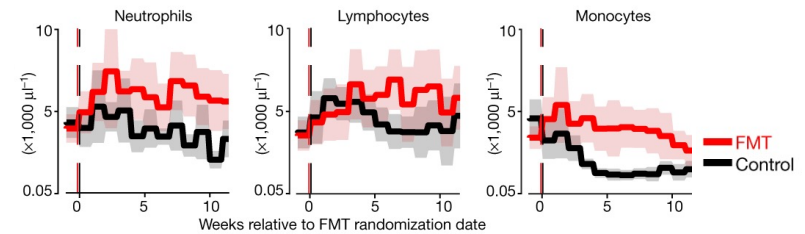


# Better Outcomes: Microbiota Diversity Supports HSCT



- Diversity is good

-Allo-Fecal microbiota transplant (FMT) improves engraftment

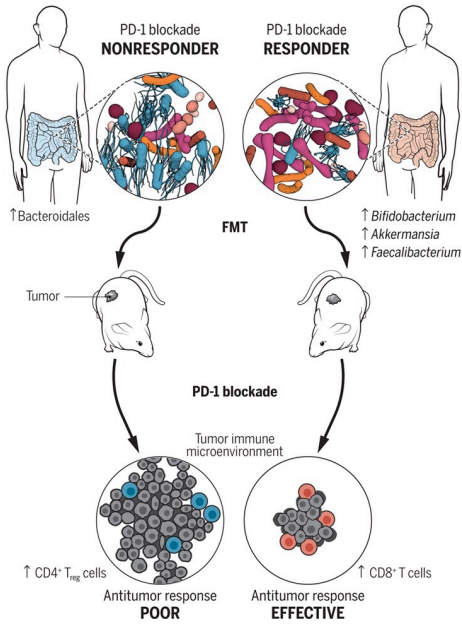


## How does a diverse microbiota help?

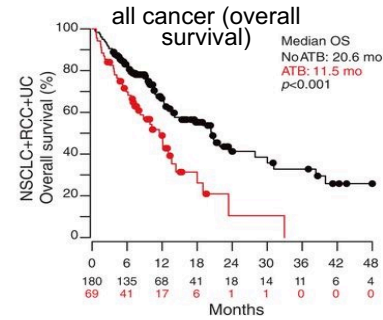
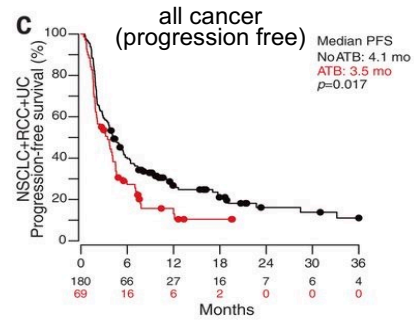
- Immune stimulatory (including mylo/lymphopoiesis)
- Nutritional benefits
- Radiation resistance
- etc...

Schluter (2020) Nature  
PMID: 33239790

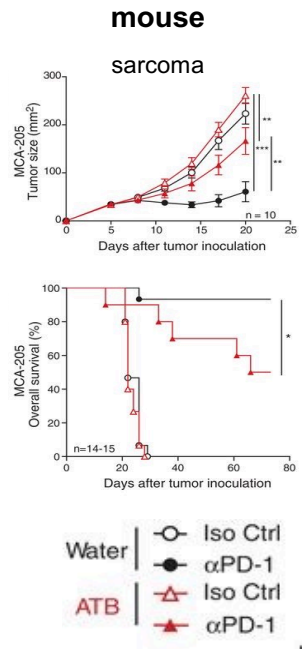
# Predicting Outcomes: Microbiota and Checkpoint Therapy



Jobin (2018) Science, PMID: 29302001

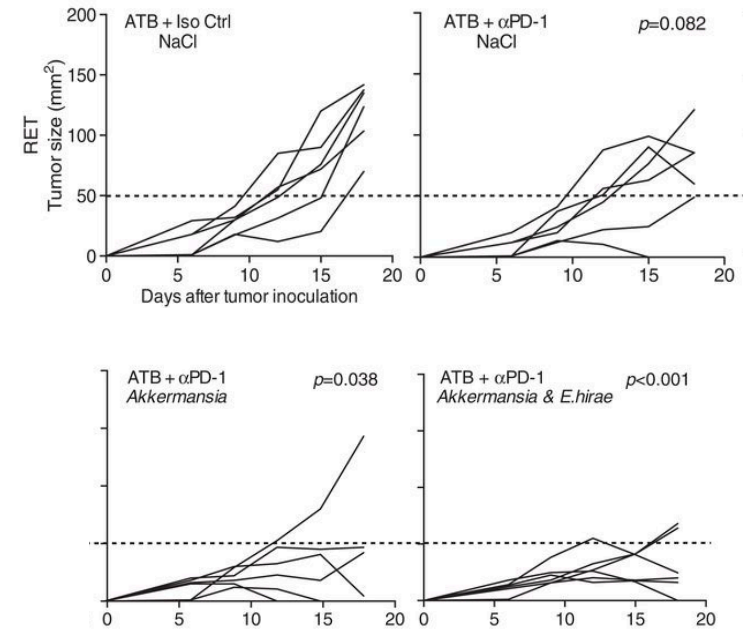
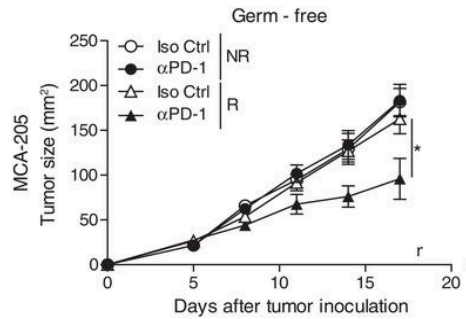
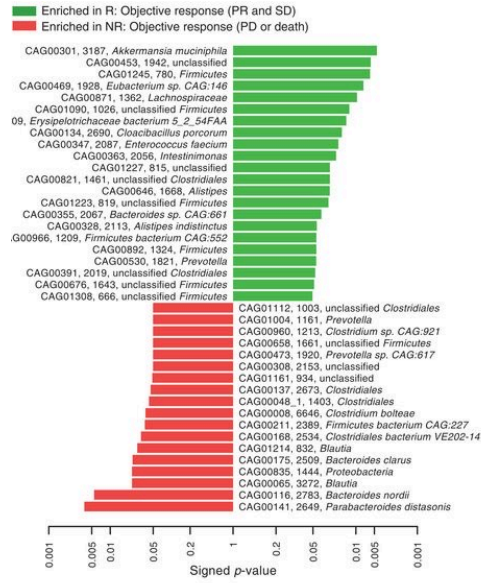


— No ABX — No ABX

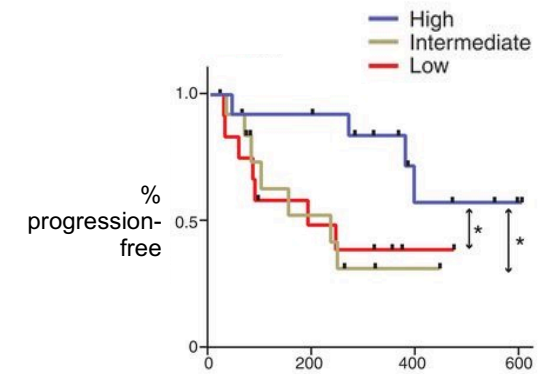
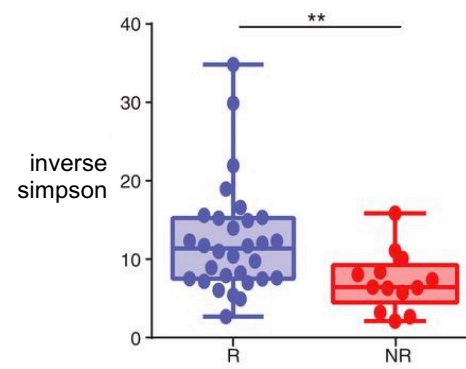
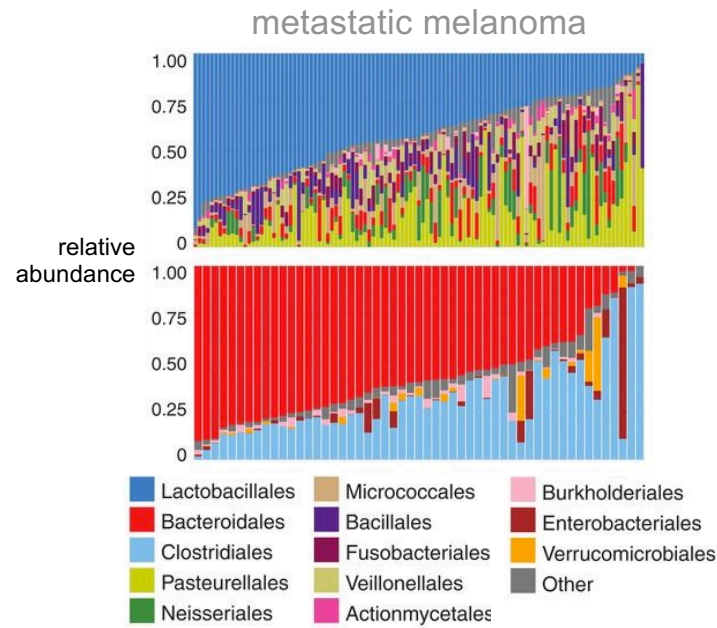


Routy 2018 Science PMID: 29097494

# Predicting Outcomes: Microbiota and Checkpoint Therapy

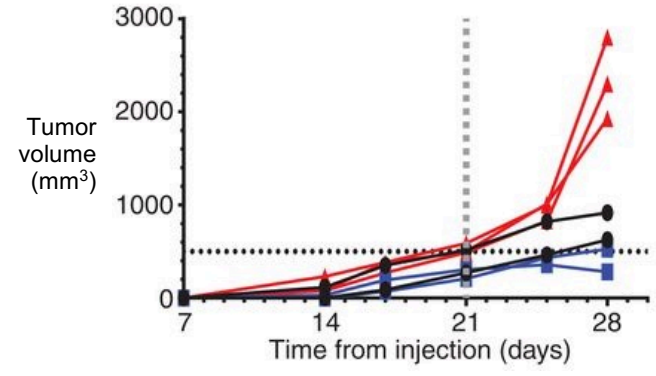
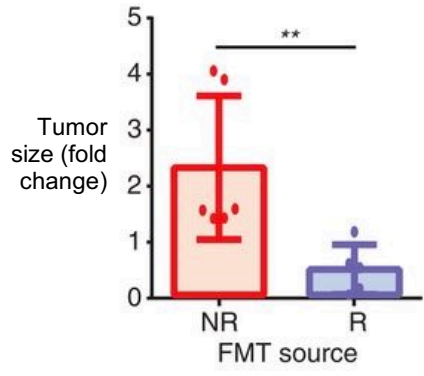
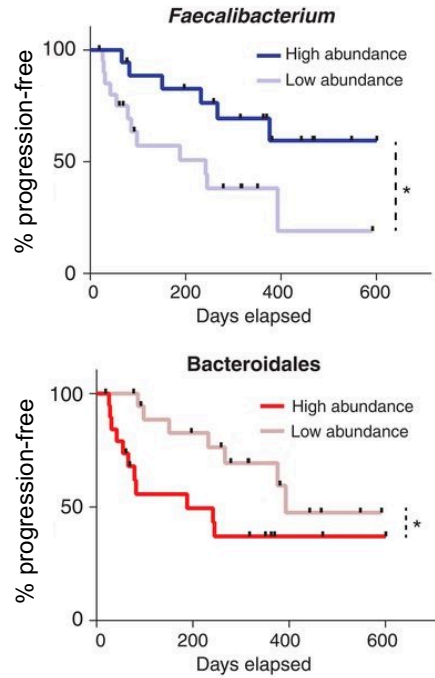


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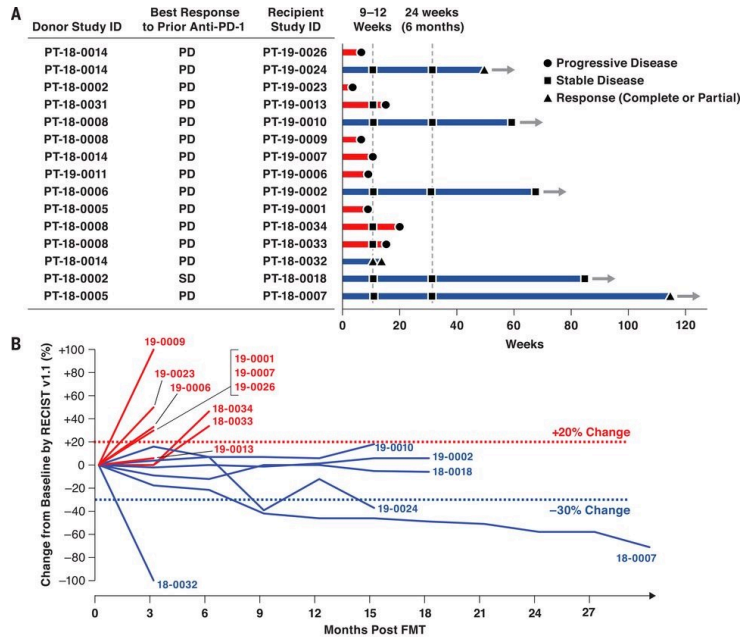
Gopalakrishnan (2018) Science PMID: 29097493

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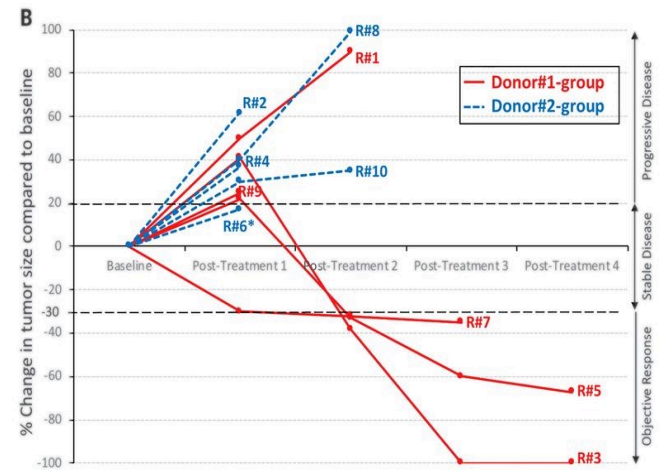
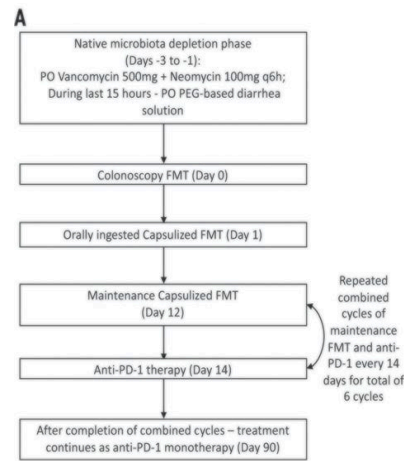


Gopalakrishnan (2018) Science PMID: 29097493

# Fecal Microbiota Transplant Improves Outcomes in Anti-PD1 Resistant Melanoma Patients



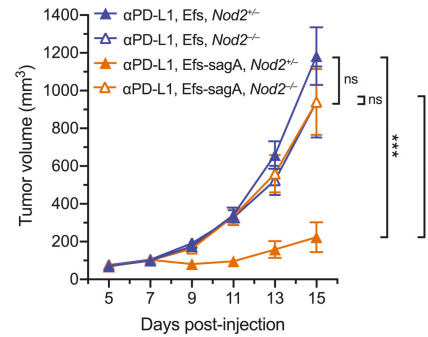
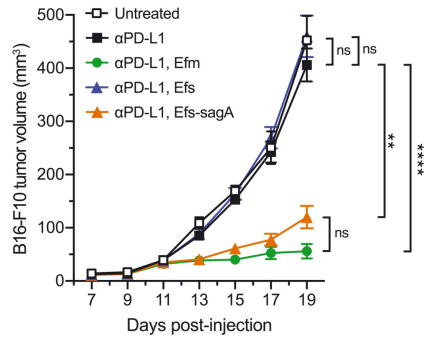
Davar, Dzutsev (2021) Science PMID: 33542131



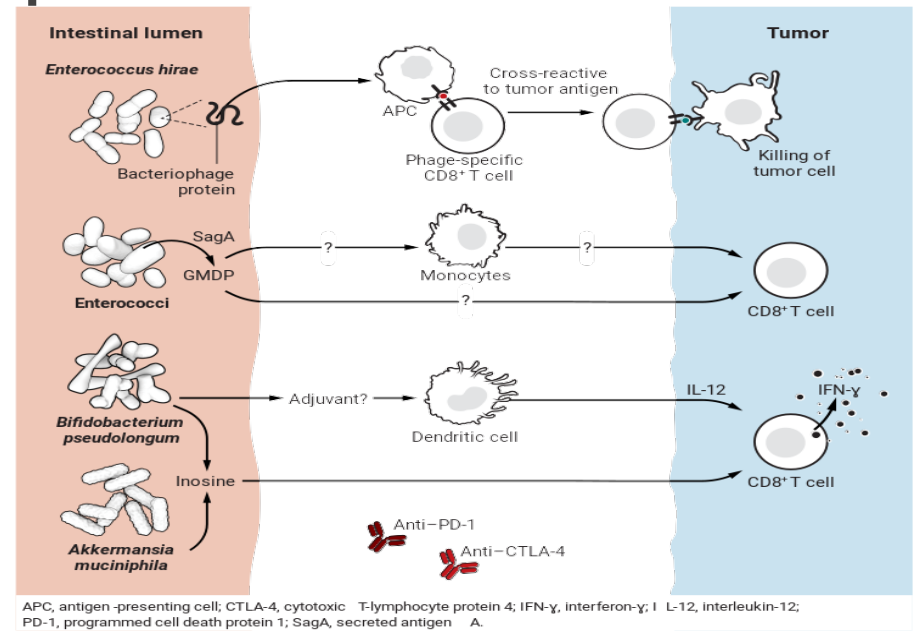
Baruch (2021) Science PMID: 33303685



# How Do Microbes Improve Checkpoint Blockade?



- Identified protective and non-protective enterocci (mice)
- Protective have distinct peptidoglycan with many small non-crosslinked fragments
- Identified conserved hydrolase (SagA)
- Overexpress SagA, bacteria now improves outcome (nod2/MDP)

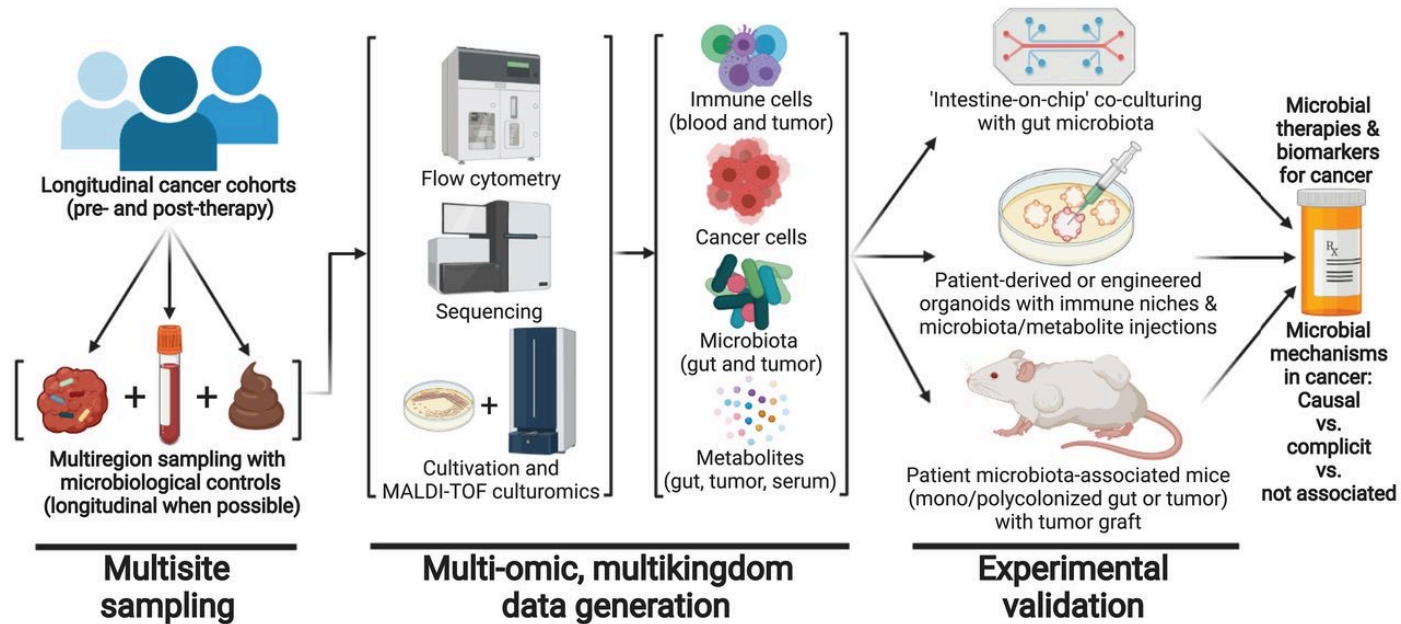


Ansaldo 2021. Science DOI: (10.1126/science.abl3656)

Griffin 2021 Science PMID: 34446607



# So How Do We Use the Microbiota to Improve Outcomes?



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PMID: 33766858



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NCI Awardee Skills Development Consortium

# Microbiota and Human Health: A Role in Cancer

## Questions