

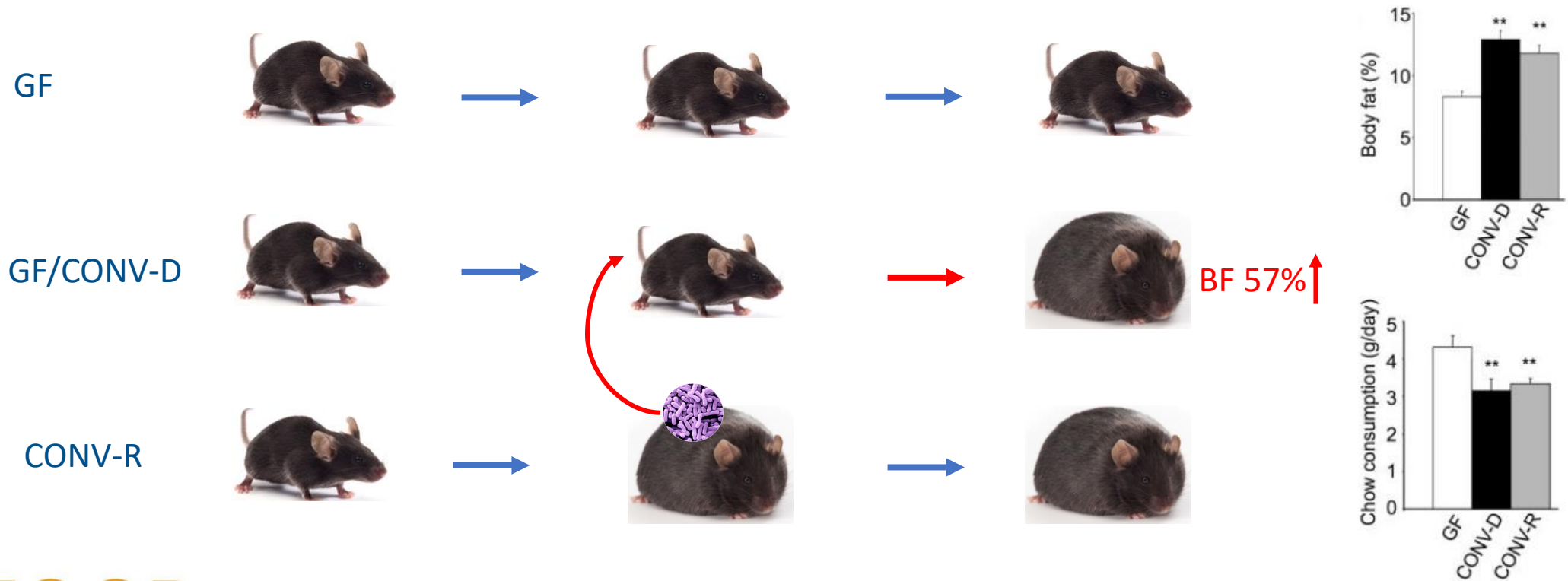
# A Forgotten Organ – The Gut Microbiome of Dogs and Cats

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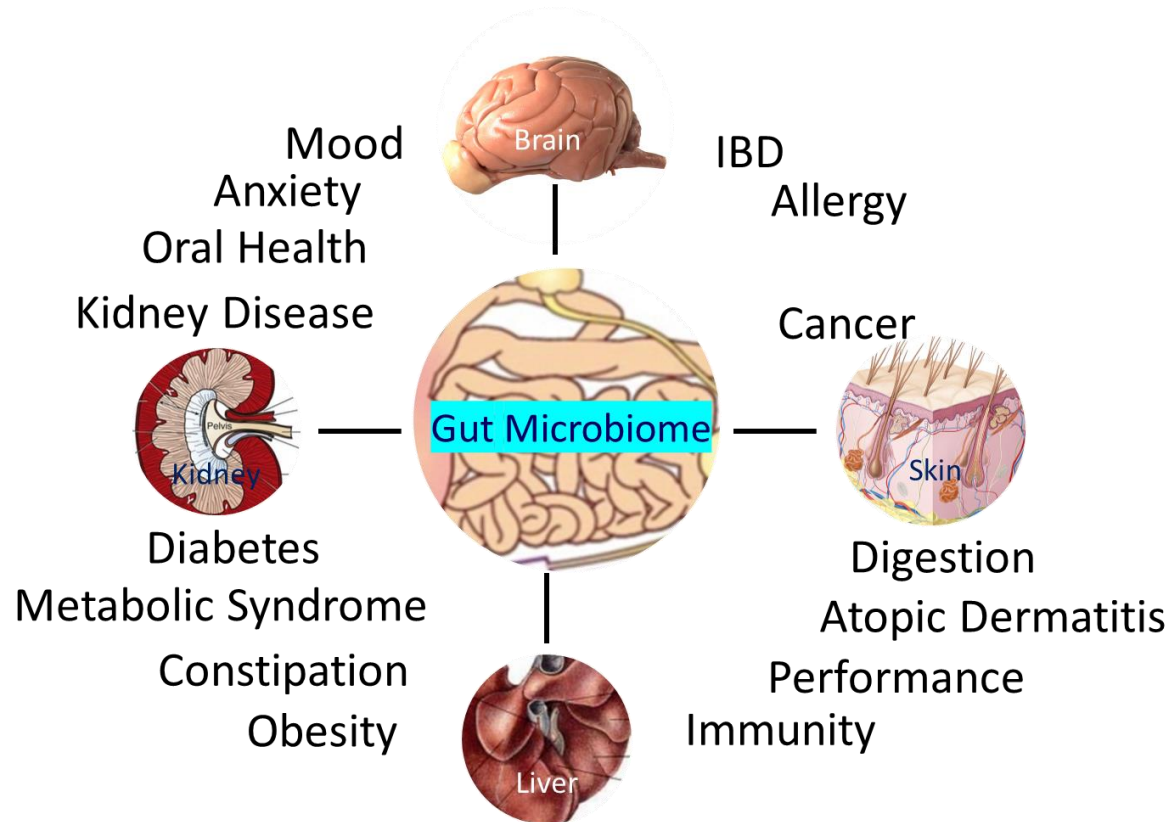
Taste, Texture & Health

# Gut microbiota increases the body fat in germ free mice



Backhel et al., 2004

# Gut microbiome organ axis



# Microbiome



Bacteria



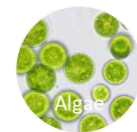
Archaea



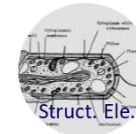
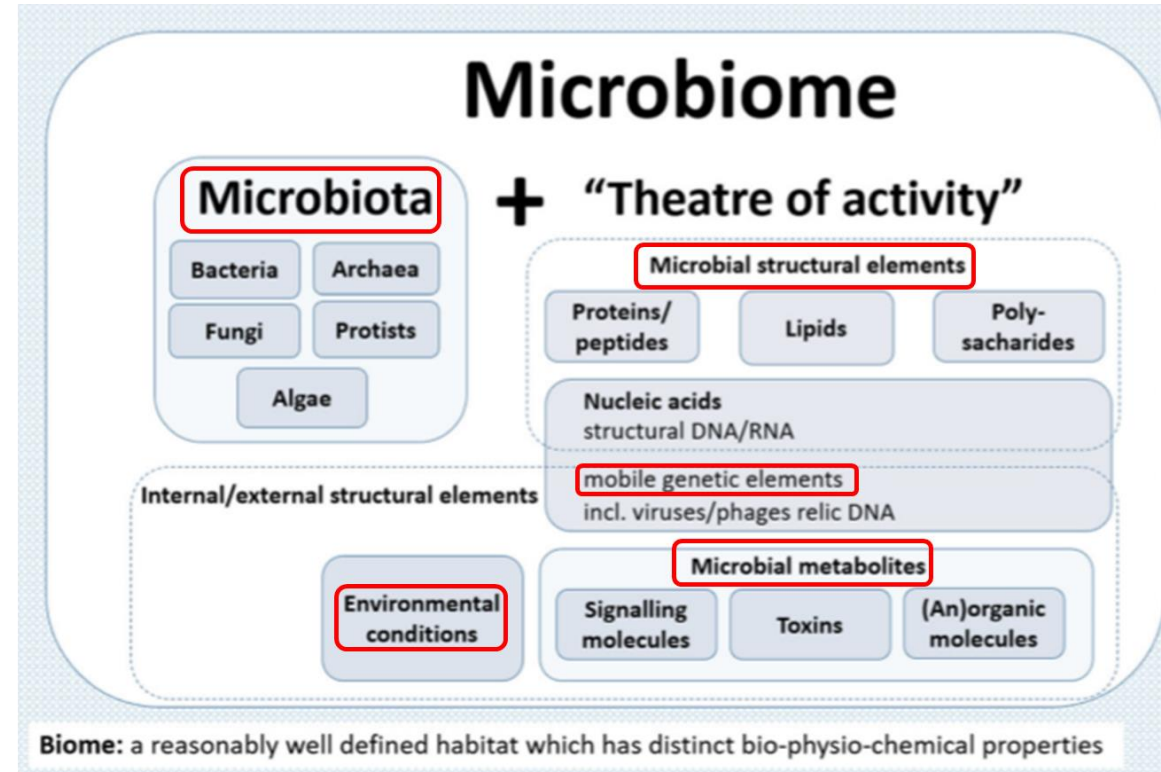
Fungi



Protists



Algae



Struct. Ele.



Metabolites



Virus/Relic DNA

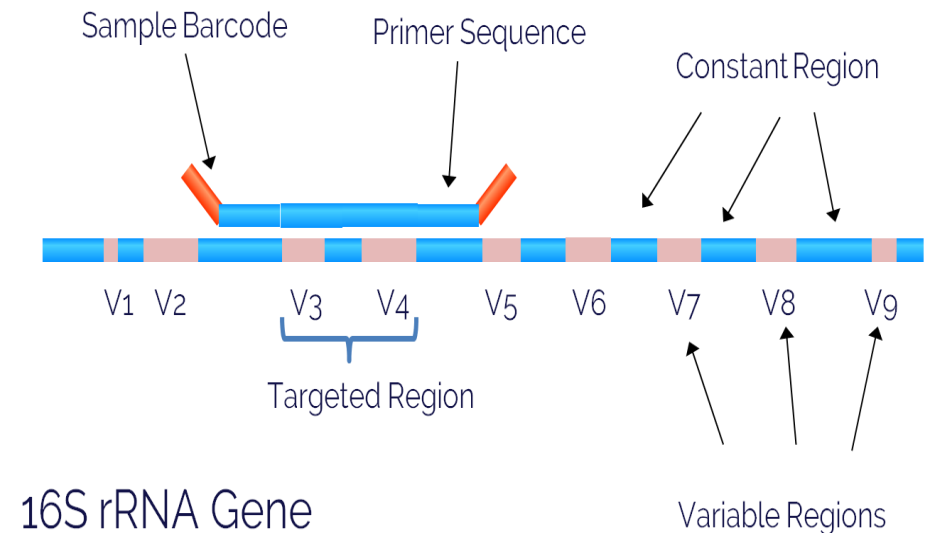


Environment

Berg et al. 2020

# Microbiome assessment

- **16S rRNA gene sequencing**
- Metagenomics (shotgun sequencing)
- Metatranscriptomics
- Metabolomics
- Quantitative real-time PCR
- Fluorescence in situ hybridization (FISH)
- Bacterial culture

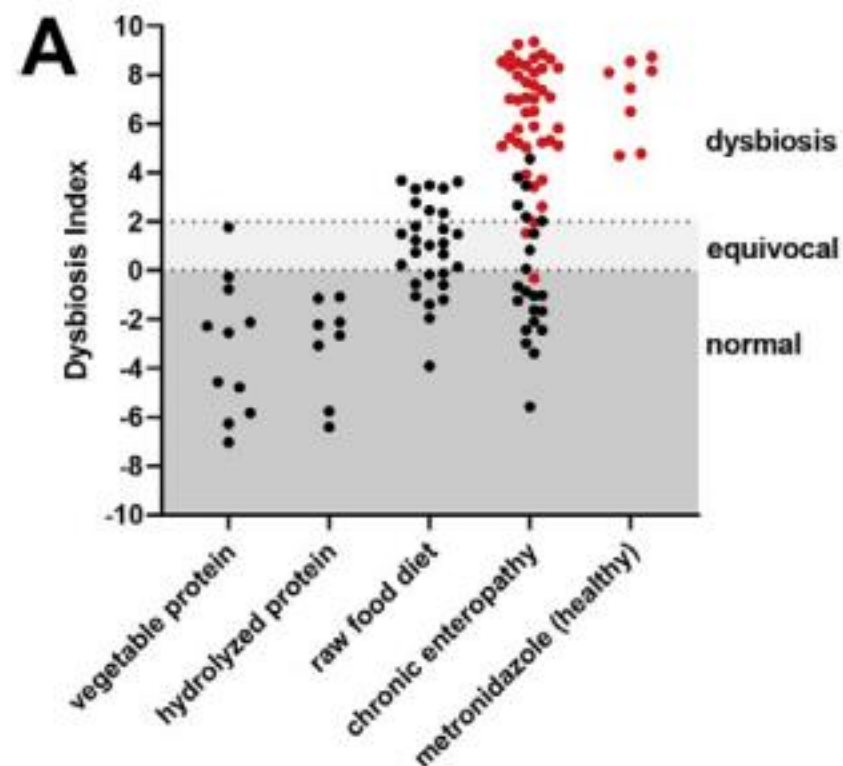


16S rRNA Gene

<https://www.lcsciences.com>

# What is a healthy microbiome?

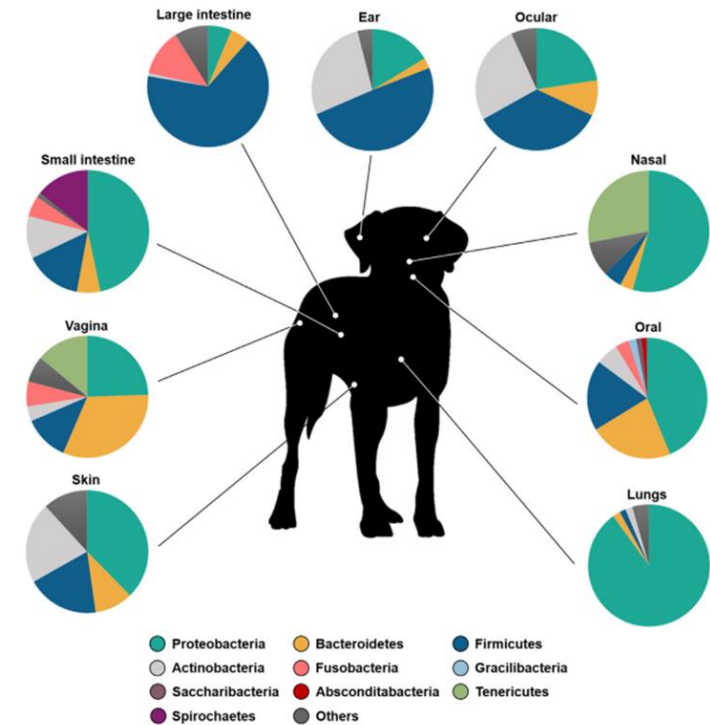
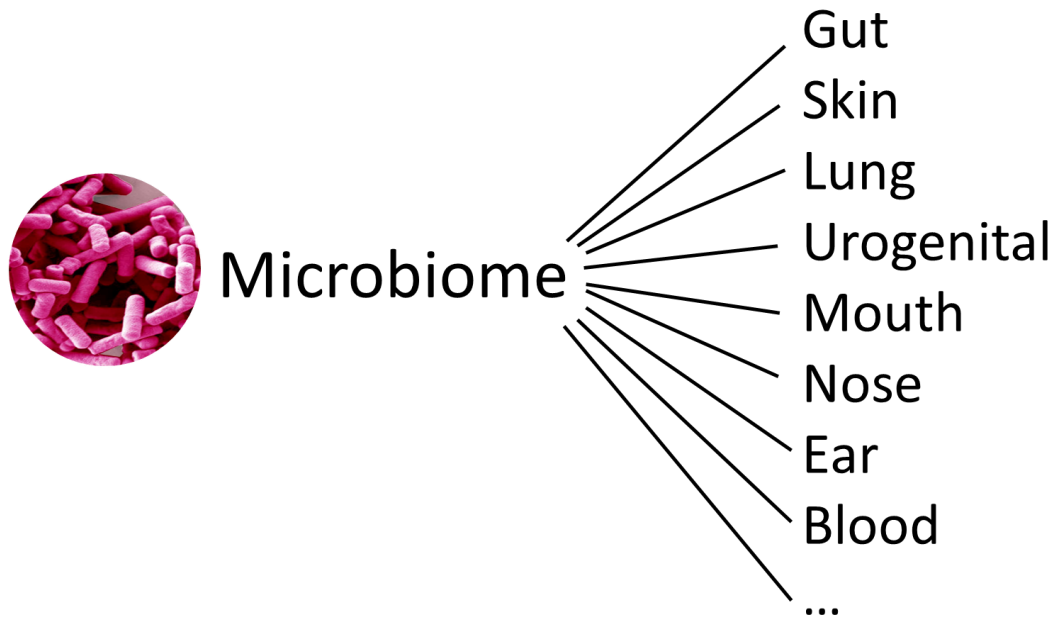
- $\alpha$ -diversity: within a sample
- $\beta$ -diversity: between samples
- Observed OTU
- Loss of beneficial bacteria
- Overgrowth of pathogenic bacteria
- Dysbiosis: imbalance of microorganisms
  - Canine Dysbiosis Index (< 2)
    - Faecalibacterium
    - Fusobacterium
    - C. hiranonis
    - Blautia
    - uricbacter,
    - Streptococcus
    - E. coli



Pilla & Suchodolski, 2021



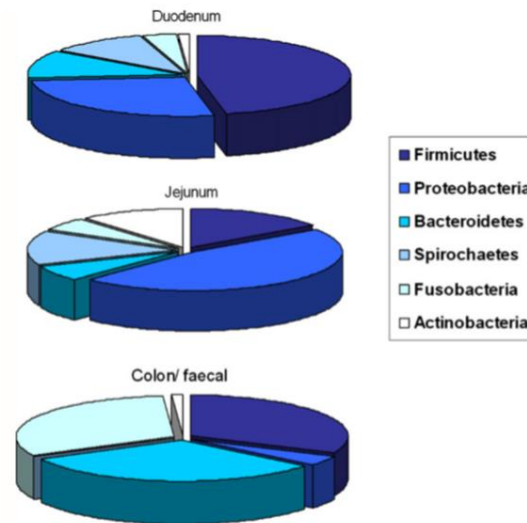
# Various microbiomes



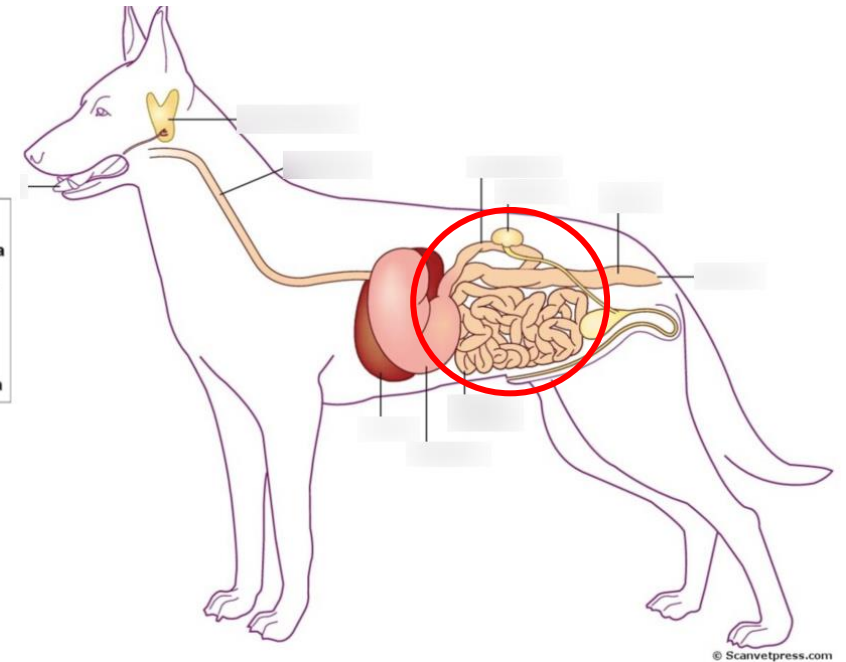
Pereira & Clemente, 2021

# Gut microbiome

- Different compartments
- Large intestine
- Fecal sample
- Bacteria



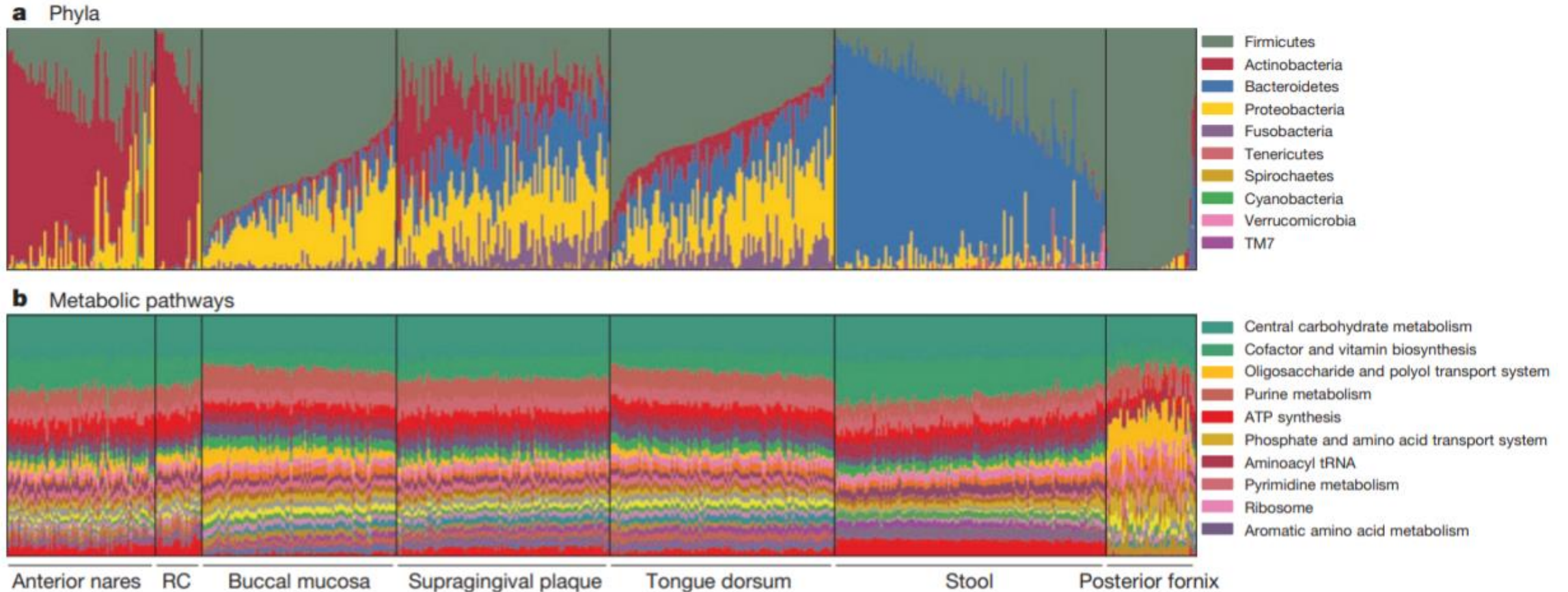
Schmitz & Suchodolski 2016



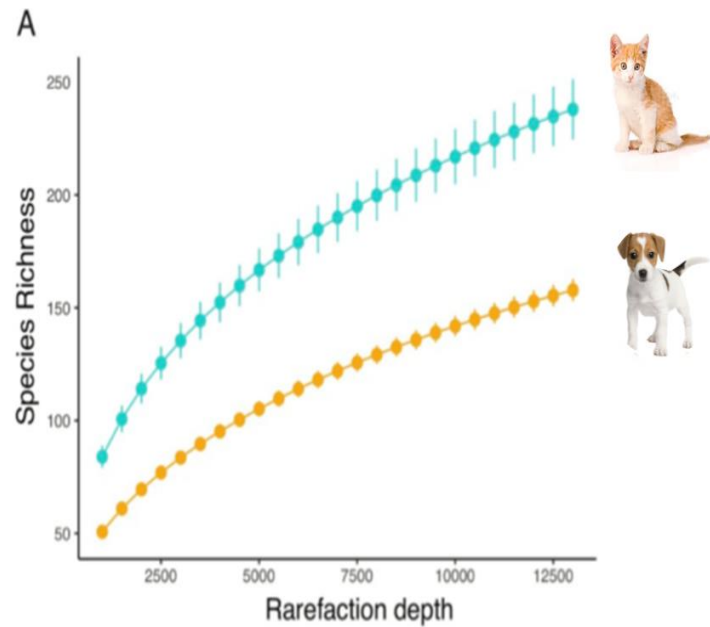
<https://quizlet.com/252065925/dog-digestive-system-diagram/>



# Gut microbiome: composition vs. function

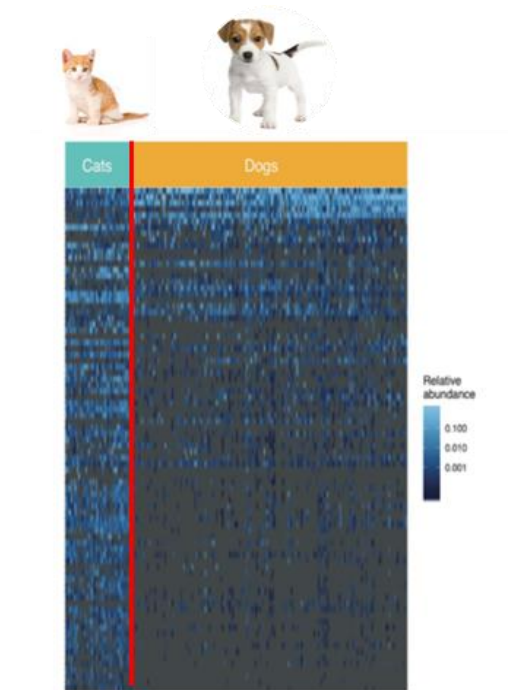


# Gut microbiome: dog vs. cat



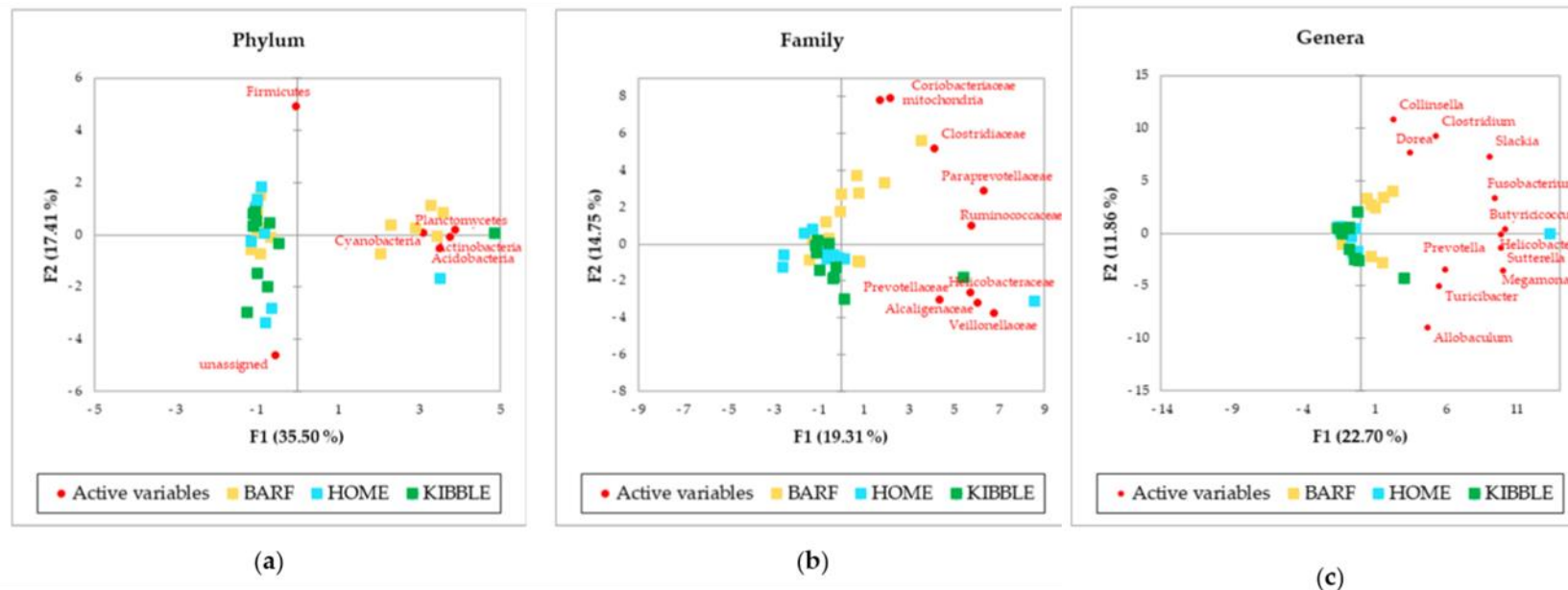
Main phyla

- ❖ Firmicutes
- ❖ Bacteroidetes
- ❖ Proteobacteria
- ❖ Fusobacteria
- ❖ Actinobacteria



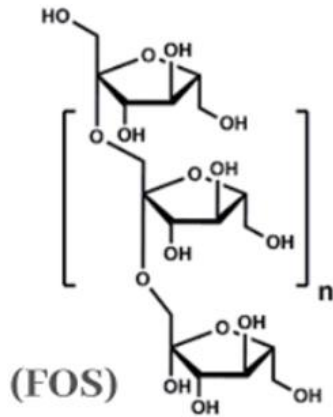
Jha et al., 2020

# Gut microbiome: diet effect



**Figure 2.** Principal Component Analysis (PCA) of bacteria Absolute Abundances (AA) regarding (a) phylum level, (b) family level and (c) genus level on fecal samples of dogs fed with a raw-meat-based diet (BARF), a homemade based diet (HOME) and a commercial complete extruded diet (KIBBLE).

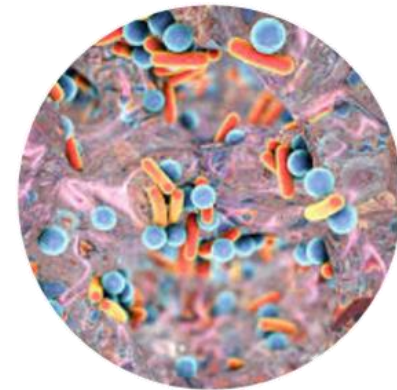
## Dietary modulation of gut microbiome: -biotics



Prebiotics



Probiotics



Postbiotics



# ISAPP definitions

## Probiotics

**Live microorganisms** that, when administered in adequate amounts, confer a health benefits on the host (2014)

## Prebiotics

**A substrate** that is selectively utilized by host microorganisms conferring a health benefit (2017)

## Synbiotics

**A mixture** comprising live microorganisms and substrate(s) selectively utilized by host microorganisms that confers a health benefit on the host (2020)

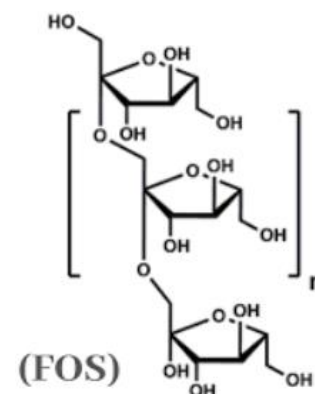
## Postbiotics

A preparation of **inanimate microorganisms** and/or their components that confers a health benefit on the host; **It must contain inactivated microbial cells or cell components**, with or without metabolites, that contribute to observed health benefits. (2021)

# Gut microbiome modulation: Prebiotics

**Table 5.** Concentrations of fecal short-chain (SCFA) and branched-chain fatty acids (BCFA;  $\mu\text{mol/g}$  of fecal DM) for cats fed select fibers

	Treatment				
Item	Cellulose	FOS <sup>1</sup>	Pectin	PSEM <sup>2</sup>	<i>P</i> -value <sup>3</sup>
SCFA					
Acetate	172.9 <sup>a</sup>	217.9 <sup>ab</sup>	291.8 <sup>b</sup>	30.0	0.030
Propionate	58.3 <sup>a</sup>	61.1 <sup>a</sup>	109.0 <sup>b</sup>	14.9	0.035
Butyrate	39.2 <sup>a</sup>	97.3 <sup>b</sup>	94.3 <sup>b</sup>	13.6	0.010
Total SCFA	270.3 <sup>a</sup>	376.3 <sup>ab</sup>	494.0 <sup>b</sup>	48.7	0.016
BCFA + valerate					
Isobutyrate	8.2 <sup>a</sup>	12.6 <sup>b</sup>	12.6 <sup>b</sup>	1.2	0.011
Isovalerate	13.3 <sup>a</sup>	21.0 <sup>b</sup>	21.1 <sup>b</sup>	2.2	0.012
Valerate	22.5 <sup>a</sup>	29.8 <sup>b</sup>	30.1 <sup>b</sup>	2.5	0.026
Total BCFA + valerate	44.0 <sup>a</sup>	63.3 <sup>b</sup>	63.9 <sup>b</sup>	5.4	0.008



**Table 7.** Fecal microbial genera ( $\log_{10}$  cfu/g of fecal DM) of cats fed select fibers

Item	Treatment				<i>P</i> -value <sup>3</sup>
	Cellulose	FOS <sup>1</sup>	Pectin	PSEM <sup>2</sup>	
<i>Bifidobacterium</i> spp.	10.4 <sup>a</sup>	11.6 <sup>b</sup>	10.7 <sup>a</sup>	0.3	0.006
<i>Clostridium perfringens</i>	10.3 <sup>a</sup>	10.1 <sup>a</sup>	11.5 <sup>b</sup>	0.3	<0.001
<i>Escherichia coli</i>	9.3 <sup>b</sup>	8.4 <sup>a</sup>	11.1 <sup>c</sup>	0.3	<0.001
<i>Lactobacillus</i> spp.	10.9 <sup>a</sup>	11.0 <sup>a</sup>	11.2 <sup>b</sup>	0.1	0.030

<sup>a-c</sup>Different superscript letters in the same row denote differences ( $P < 0.05$ ) among treatments.

<sup>1</sup>Fructooligosaccharides.

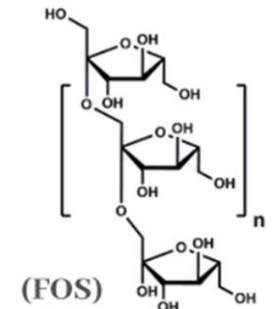
<sup>2</sup>Pooled SEM.

<sup>3</sup>Indicates type 3 fixed effect of diet.

Barry et al, 2010

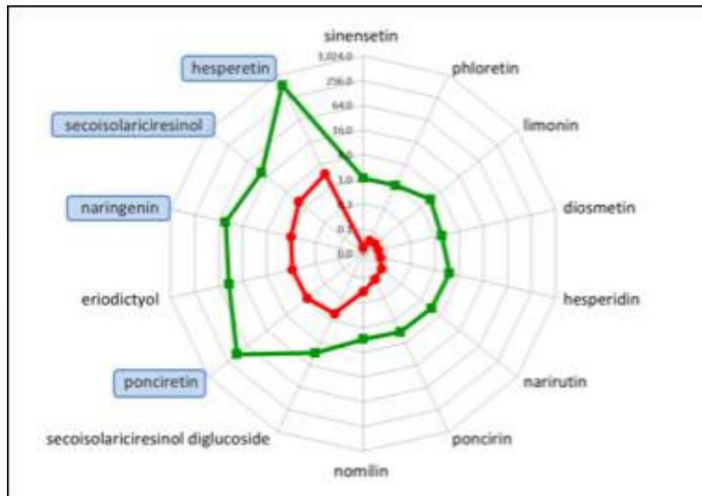


# Gut microbiome modulation: Prebiotics

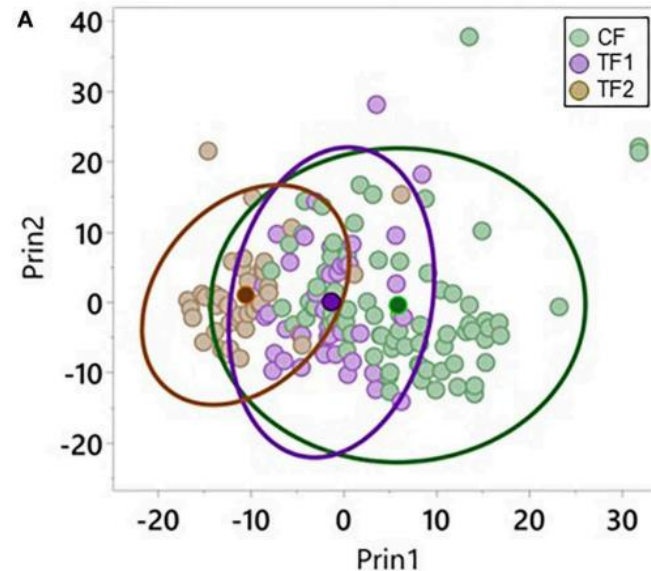


TF1: Ground pecan shells, Flaxseed, Dried beet pulp, Dried citrus pulp, Pressed cranberries and Psyllium seed husk

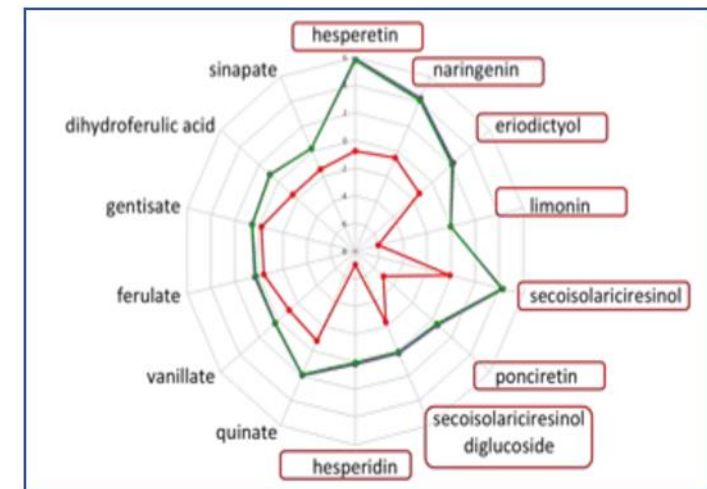
TF2: Powdered cellulose, Rice hulls, Dried beet pulp, Psyllium seed husk and Fructooligosaccharides



Plot shows log-transformed data. Red: Control food; Green: Test food



Fritsch et al, 2023

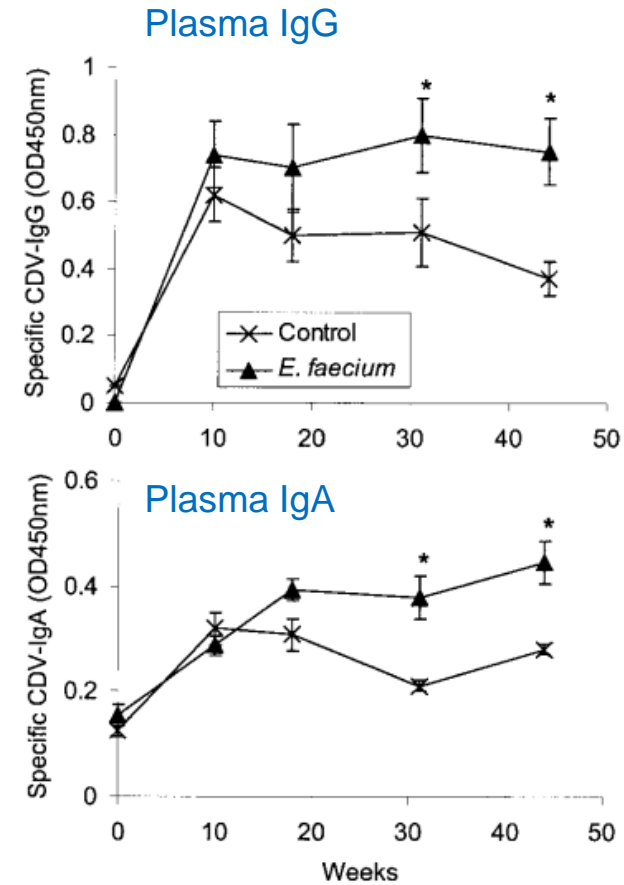
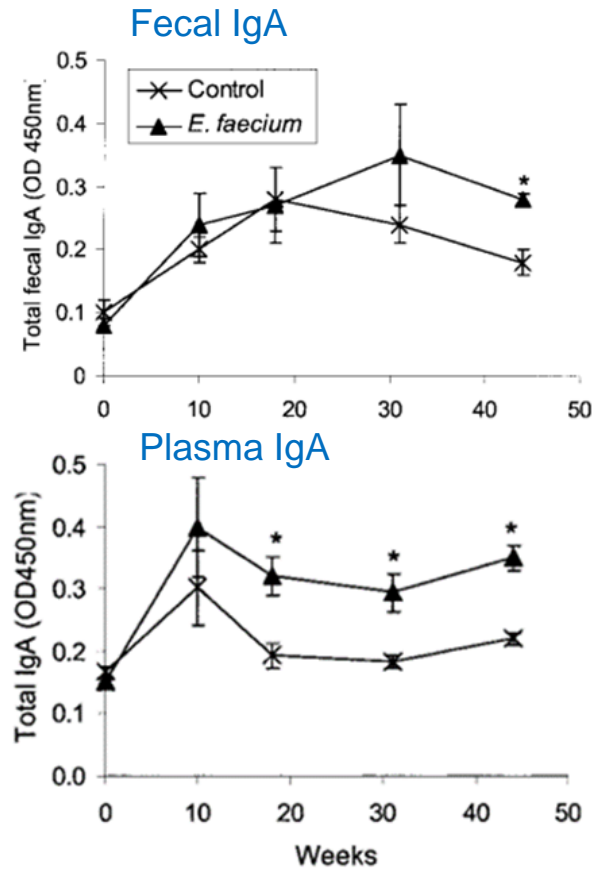


Plot shows log-transformed data. Red: Control food; Purple: Test food 4 weeks; Green: Test food 8 weeks.



Wernimont et al, 2019

# Gut microbiome modulation: Probiotics



Benyacoub et al, 2003

# Gut microbiome modulation: Probiotics



Medians (1st;3rd quartiles) of fecal score, ammonia, and fecal odor of dogs fed the control or *B. subtilis* C-3102 diet.

Item	Control	<i>B. subtilis</i> C-3102	P value
Score	3 (3;4)	4 (3;4)	<0.001
Ammonia (g/kg)	0.705 (0.601;0.776)	0.602 (0.589;0.625)	0.037
Odor	2 (2;2)	1 (1;2)	<0.001

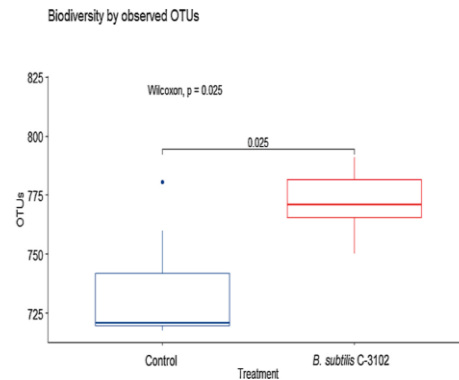


Fig. 2. Richness of bacterial communities (number of OTUs per 12,804 reads) of feces of dogs fed the control or *Bacillus subtilis* C-3102 diet ( $P = 0.025$ ).

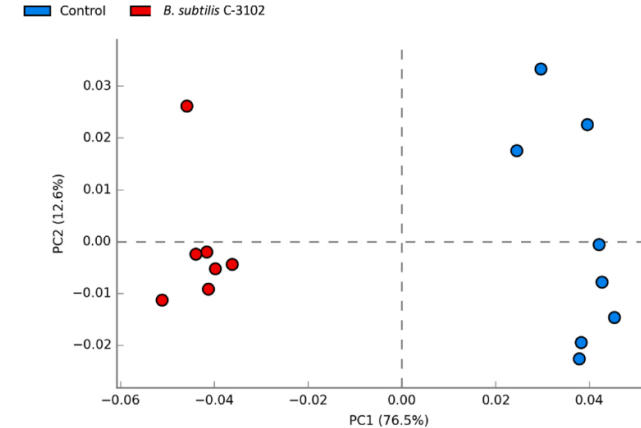


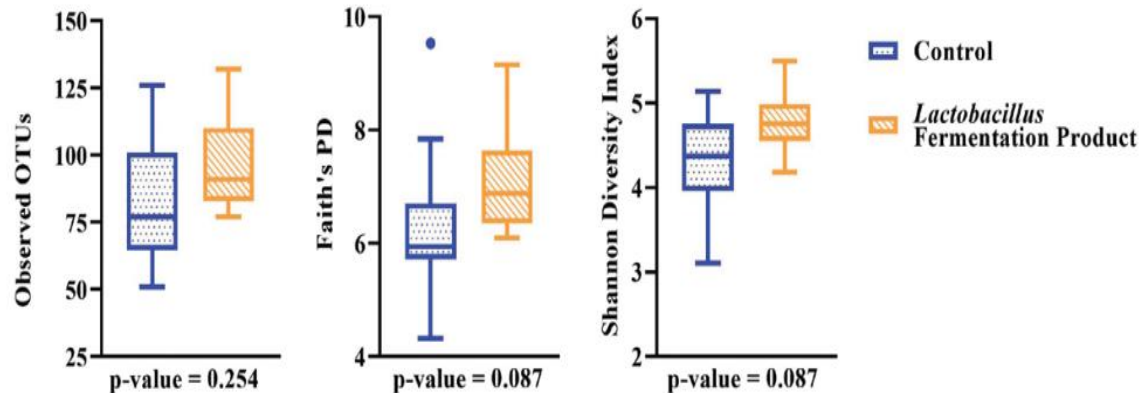
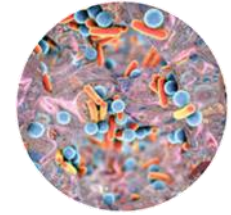
Fig. 1. Principal component analysis (PCA) of the bacterial community in feces of dogs fed the control or *Bacillus subtilis* C-3102 diet.

De Lima et al., 2020

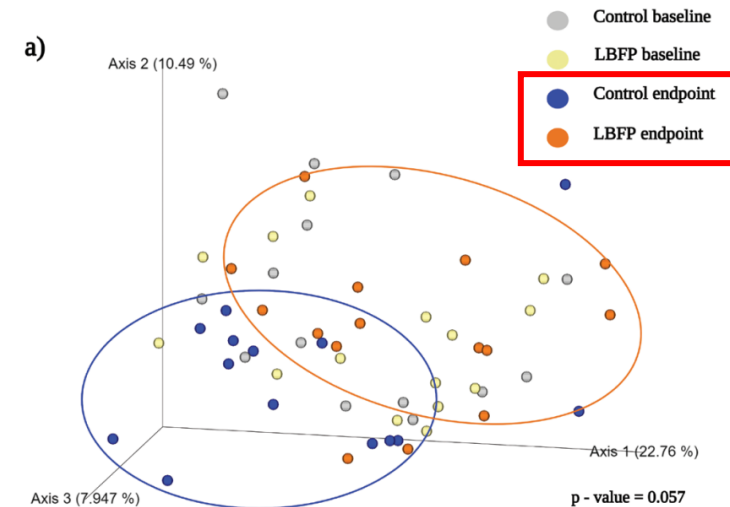
# Gut microbiome modulation: Postbiotics



Heat killed *Lactobacillus delbrueckii* & *Lactobacillus fermentum*



**Figure 2.** Alpha diversity measures of fecal samples collected from dogs supplemented with *Lactobacillus* fermentation product or placebo (dextrose) control. Alpha-diversity is represented by observed OTU, Faith's phylogenetic diversity (PD), and Shannon diversity index. Faith's PD and Shannon diversity index suggest that species richness tended to be higher in dogs supplemented with *Lactobacillus* fermentation product than those fed the control. The observed OTU comparison, however, did not reach statistical significance.



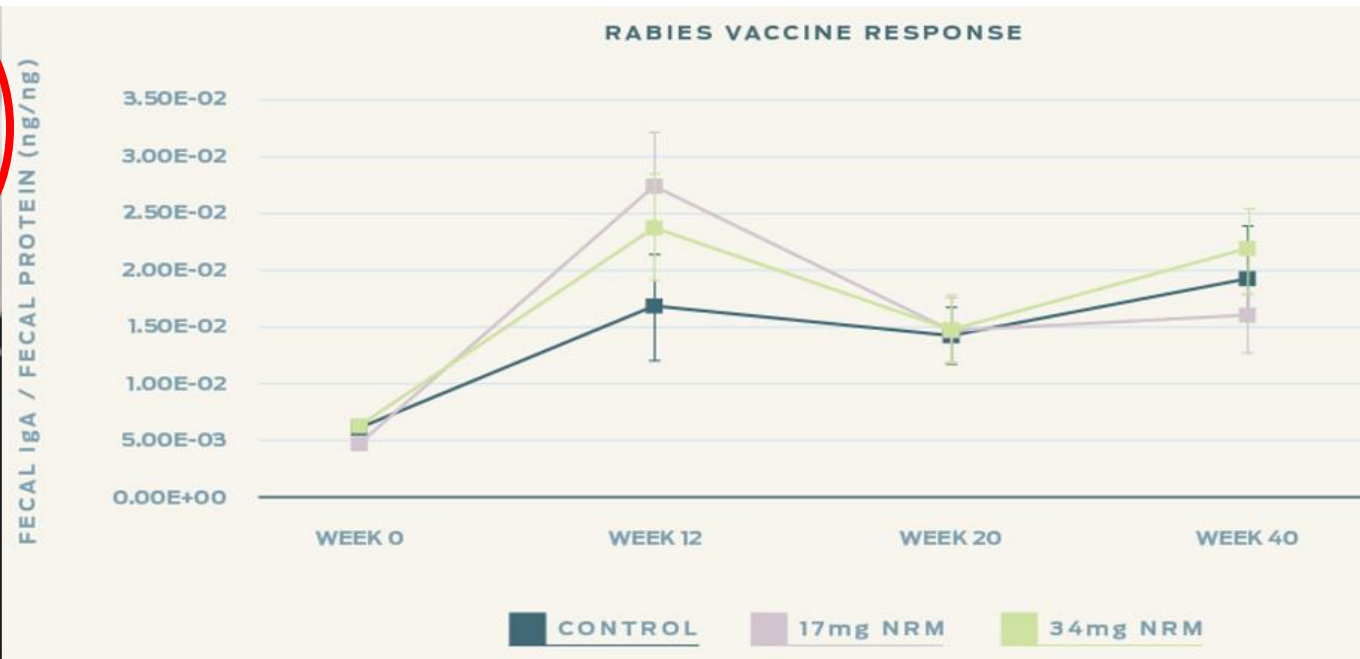
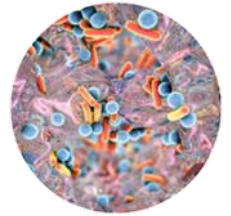
**Figure 3.** Beta diversity measures of fecal samples collected from dogs supplemented with *Lactobacillus* fermentation product or placebo (dextrose) control. Principal coordinates analysis (PCoA) plots of unweighted (a) and weighted (b) UniFrac distances of fecal microbial communities were performed on the 97% OTU abundance matrix using QIIME2. Unweighted UniFrac distances suggest a trend of bacterial population separation between treatment groups.

Kozioł et al, 2023

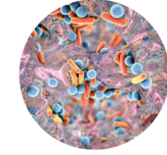
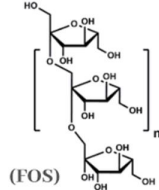


# Gut microbiome modulation: Postbiotics

Heat killed *Lactobacillus delbrueckii* & *Lactobacillus fermentum*



# Comparison of Pre-, Pro- & Postbiotics



	Prebiotics	Probiotics	Postbiotics
Characteristics	Mostly polysaccharides	Live microorganisms	Dead microorganisms
Health benefits	Yes	Yes	Yes
Heat resistance	Yes	No	Yes/No
Applications	Dry, Wet, Treat, Supp.	Dry, Treat, Supp.	Dry, Wet, Treat, Supp.
Availability	Many	Limited (EU)	Limited/Increasing
Storage stability	Stable	Unstable	Stable/Unstable
Awareness of pet owners	Good	Good	Poor/Improving
Market penetration	Excellent	Good (US)	Poor/Increasing



# Thank You!



<https://www.istockphoto.com/photo/cats-and-dogs-over-thanksgiving-pumpkins-gm877205556-244802876>