

Getting Dirty: Does Soil Affect a Dog's Microbiome?

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Abstract

For my project, I tested a dog's microbiome and compared it to the health of the dog's soil. I also looked at the time the dog spends in the soil. I ran two samples. The first sample was a stool sample. The second sample was a soil sample. Both were taken at the same time. I tested dogs that are in the city vs. country dogs. I found out that farm dogs have a healthier microbiome, but city dogs have healthier soil. The farm dogs' average microbial biomass in the soil is 221.0 ug C/g. The city dogs' average microbial biomass in the soil is 273.4 ug C/g. Farm dogs average for F% is 20%. City dogs average for F% is 32%. Farm dogs average for B% is 80%. City dogs average for B% is 68%. The farm dogs were outside in the soil for longer periods of time. The farm dogs might be healthier due to spending more time in the soil.

Introduction

A microbiome is a collection of living organisms in your body. You can find microbiomes all over the body, but it mostly appears in your intestinal tract. A dog's intestinal microbiome has a lot of organisms that help the dog live and fight off bacteria. Organisms include bacteria, fungus, viruses, protozoa, and archaea (Morris Animal Foundation, 2022). In order to stay healthy a dog needs a balanced meal and a balanced microbiome. If a dog does not have a balanced microbiome they could get allergies, asthma, skin conditions, or inflammation. A dog gets a balanced microbiome through their internal body functions and their environment. Good food that is healthy and better or more soil exposure are environmental factors that change their microbiome.

Soil can affect a dog's microbiome because dog's spend time in soil. A dog's microbiome could get improved or worsened by the soil the dog is exposed to. If there is healthy soil, then the dog's microbiome should be healthy. If the soil is unhealthy, then the dog will have a worsened microbiome. The dog also needs exposure time in the soil. My research is going to measure the soil health where a dog lives and it is going to measure the dog's microbiome. The research will help us understand the need for healthy soil and can help keep dogs healthier.

Research

The dog gut microbiome is a new area of research that is helping to save the lives of dogs and improving their health. A dog's microbiome is developing at birth and it changes throughout the lifespan of the dog. A dog's microbiome has five major groups of bacteria. Those groups are 1). Firmicutes, 2). Fusobacteria, 3). Bacteroidetes, 4). Proteobacteria, and 5). Actinobacteria (Garrigues, Apper, Chastant, & Mila, 2022). Firmicutes help to break down carbohydrates in the digestive tract. A dog with low firmicutes could get gut disease (Stay Labs, 2022). Firmicutes are

a big group of bacteria. The bacteria in the group include: lactobacillus, megamonas, ruminococcus, dorea, blautia, and clostridium. Fusobacteria mostly appear in an adult dog's microbiome. When it does appear, it helps the dog break down amino acids. Fun fact, in humans fusobacteria is connected to disease. Fusobacteria is an important bacteria for animals that are carnivores. Bacteroidetes also break down carbohydrates. Higher numbers of bacteroidetes are found in leaner dogs. Proteobacteria metabolize protein, carbohydrates, and vitamins. When they are at higher levels, the dog's microbiome might be out of balance. A type of proteobacteria is escherichia. Finally, actinobacteria is one of the first bacteria found in puppies. These bacteria are found in soil environments and in a dog's microbiome. They are helpful in carbohydrate digestion and can protect against diarrhea, some cancer, and inflammatory bowel disease (NomNom, 2021). A type of actinobacteria is bifidobacterium.

Dog's need balance between the five major groups of bacteria. Research on allergies and asthma in humans found that kids living on farms have less allergies and asthma because they are by more natural microbes in the soil. Dogs could have the same results and have less allergies and asthma when they are out in the soil more (Camp Run a Mutt, 2021). The dog needs soil because of its great bacteria. In one teaspoon of soil it can contain between 100 million and 1 billion bacteria (Hoorman, 2016). When dogs go outside, they get exposed to the soil and bacteria and this can improve their microbiome. Research shows that animals that are exposed to soil have more bacteroidetes and better overall health (Choi, 2020).

If your dog's microbiome is out of balance, then you might want to take it to the vet and get medicine. The veterinarian can give probiotics. Probiotics are good bacteria and they can make your dog healthier. They also can affect the dog's mood through the gut-brain axis. Probiotics have effects on chemicals in the brain that can make you happier.

In the dog microbiome there are five major groups of bacteria, those groups help with different things. They all help with digesting the food the dog eats and keeping the dog healthy. The soil that a dog's on can affect its microbiome. The microbiome is important because it is what keeps the dog healthy and it can affect the dog's mood.

Research Question

Does where a dog lives make a healthier microbiome?

Hypothesis

I think that farm dogs will have healthier microbiomes and healthier soil. I also think that farm dogs will spend more time in the soil.

Variables

Variable: Dog's microbiome

Variable: Soil health

Variable : Time in soil

Materials and Methods

I used NomNom microbiome kits to test the dog's microbiome. To test the health of the soil, I used the Microbiometer. My study followed the steps below.

Step One: I got permission from 20 dog owners. I sent them a short questionnaire to consent to participate and on their dog's health (see Appendix A). Then, I scheduled a date to collect the stool and soil samples.

Step Two: I collected a stool sample from the dog using the instructions from the NomNom kit. To collect the sample, I inserted the entire tip of a swab into the recent stool sample and rotated the swab. I made sure that the swab was coated and there was at least a pea sized amount on the swab. Then, I placed the swab into a vial and discarded the upper part of the swab. The vial was

capped and I shook the vial until the sample dissolved in the liquid that is in the vial. The liquid in the vial came with the kit and was not hazardous. I repeated the process with a second swab and vial for each stool sample as per the NomNom instructions. Then, I mailed the vials to Ashley Snipe in the research department at NomNom Now. I collected 20 stool samples - 10 samples from city dogs and 10 samples from country dogs.

Step Three: Next, I collected a soil sample using the Microbiometer instructions. I collected a moist soil sample from the top 2–5 inches of soil. I used a sifter to remove any debris from the soil sample. I compressed the soil sample to .5 ml using the syringe provided by the Microbiometer kit. Then I mixed the soil with water and the extraction powder in the vial provided. The extraction powder contains sodium chloride and calcium chloride which are non-hazardous in the small amounts that are present. After mixing, the liquid rested for 5 minutes. Then, I tapped the bottom of the vial and then let it rest for 15 minutes. After 15 minutes, I used a small pipette to draw up liquid from the sample. Then I placed 3 drops of liquid on the test card. Then I used the app to analyze the results. I collected a total of 11 soil samples from the dogs' houses; one sample per household (Microbiometer, 2020).

Data and Result

The survey showed that none of the dogs had any skin conditions or allergies.

After I got the microbiome reports back from NomNom, I put all the data into a spreadsheet and found the averages for the detailed overall microbiome, the overall microbiome, the microbiome richness, the microbiome evenness, and the types of bacteria in the microbiome. The reports labeled each dog as improvable, healthy, or very healthy. To input this, I coded improvable as a 1, healthy as a 2, and very healthy as a 3. Look at Appendix B for a sample microbiome report.

To get more detail I used the microbiome graph on the NomNom report. I got the detailed number by counting the bars in the sections for improvable, healthy, and very healthy. Each section had 10 numbers in it. For example, if one of the dogs got a 17 they would be healthy. See Figure 1 below for more information.

Figure 1

This is a picture of the NomNom graph describing the overall report of the microbiome. For the overall report this dog would be a 1. For the detailed overall report it would be an 8.

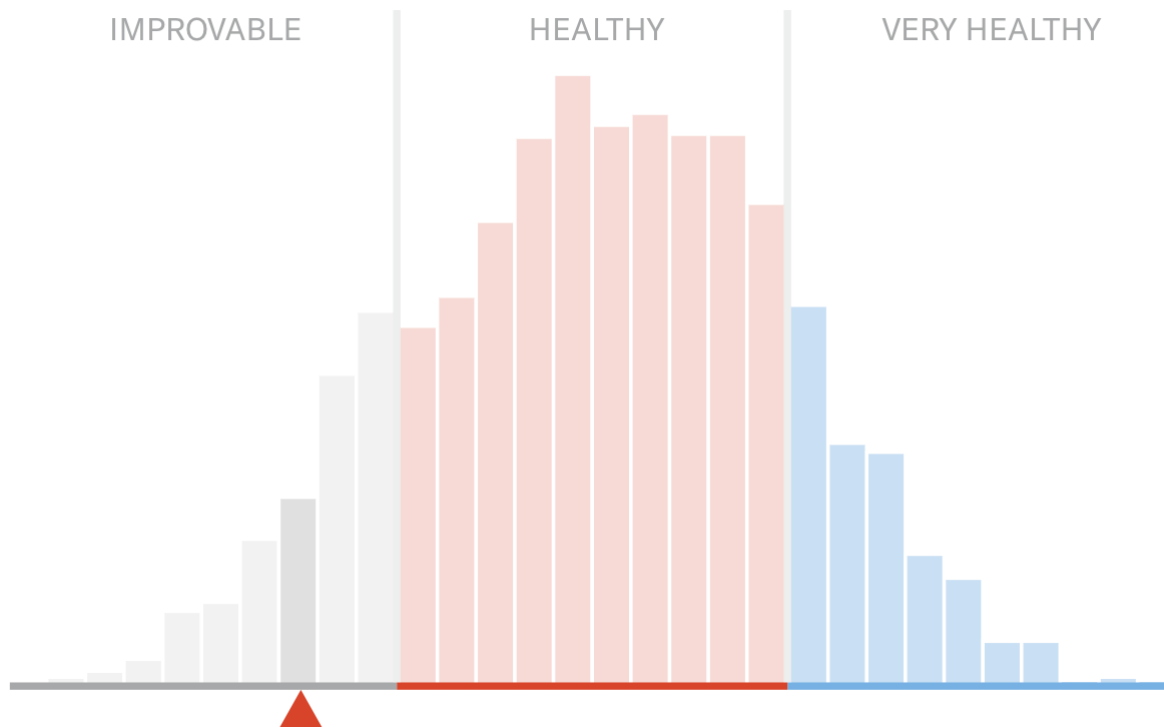


Image from NomNom Report for farm dog number 5.

I used the detailed overall report to see more differences between the dogs. The average for the farm dogs detailed overall report was a 19.4. The average for the city dogs detailed overall report was a 13.4. This gave me an average for the overall report. I used an overall score

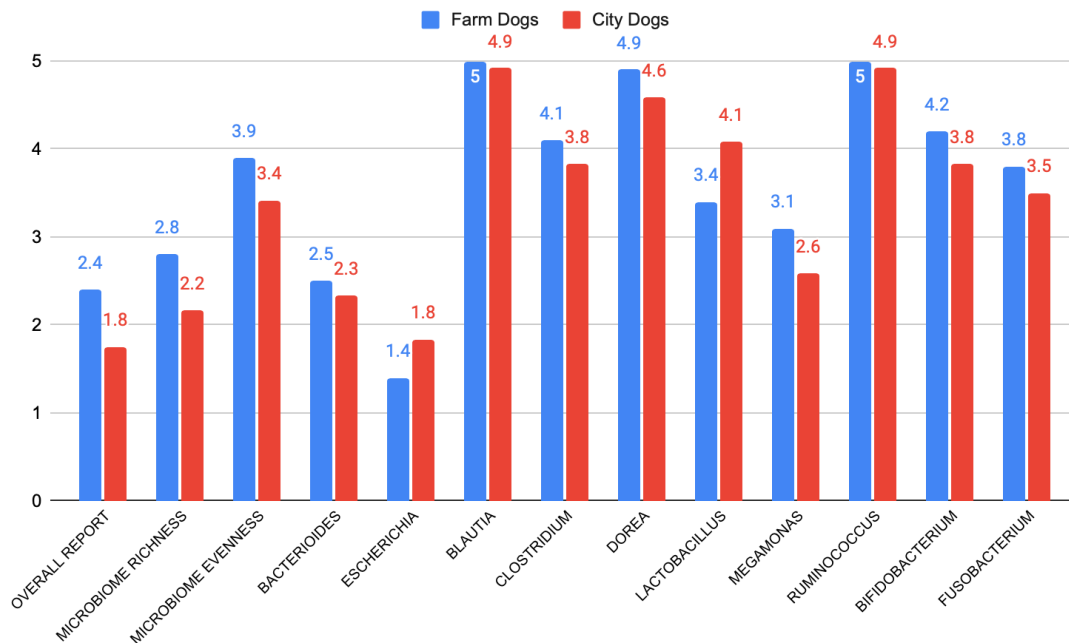
to simplify the detailed overall report. The farm dogs overall report was a 2.4. The city dogs overall report was a 1.8.

The microbiome richness was a group of the NomNom report. The average for the farm dogs microbiome richness was a 2.8. The average for the city dogs microbiome richness was a 2.2. The microbiome evenness was also a group in the NomNom report. The average for the farm dogs microbiome evenness was a 3.9. For the city dogs microbiome evenness was a 2.1.

The farm dogs outscored the city dogs in every category of bacteria but two categories. The first was escherichia. Escherichia is a bacteria that dogs do not want to have a high number in. Escherichia is associated with diseases like inflammatory bowel disease and diarrhea. The second was lactobacillus. This bacteria helps dogs recover from diarrhea and to stay healthy. See Figure 2 below to see the averages of the different types of bacteria.

Figure 2

Averages of the NomNom microbiome report for farm and city dogs.

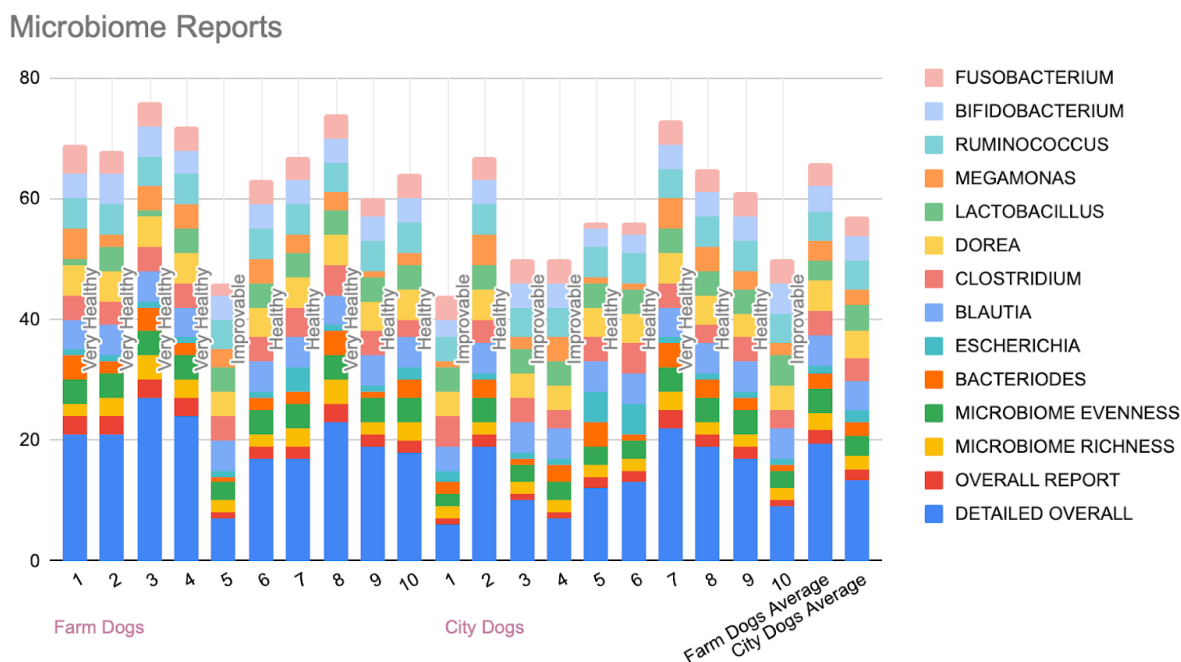


Graph made by student with help from parent.

Individual microbiome reports also show increased health in the farm dogs. See Figure 3 below.

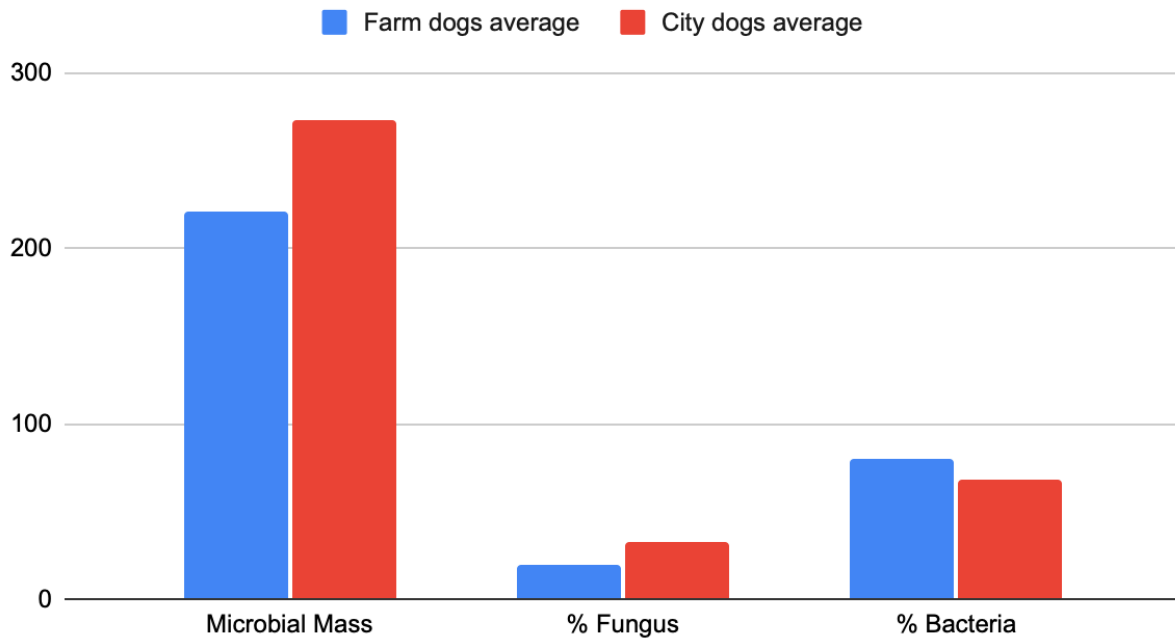
Figure 3

Individual microbiome reports.



Graph made by student with help from parent.

I also did the same thing to the soil data. I found out that the city dogs have healthier soil than the farm dogs. The average microbial mass for the city dogs was 273.4 ug C/g. For the farm dogs, the average microbial mass was 221.0 ug C/g. Microbial mass is a measure of overall soil health. The microbiometer also measured the fungus and the bacteria in the soil. The city dogs' average for percent fungus in the soil was 32%. The farm dogs' average for percent fungus was 20%. For bacteria, the city dogs' average was 68%. For farm dogs' the average percent bacteria was 80%. See Figure 4 below.

Figure 4*Soil**averages***Soil Averages**

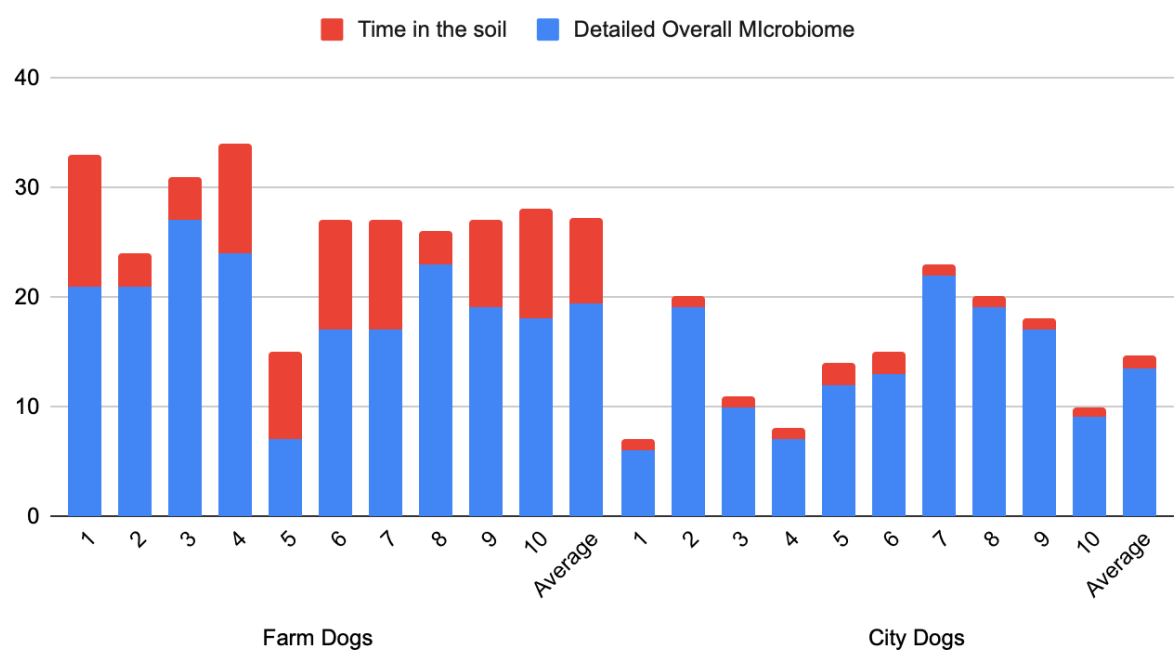
Graph made by student with help from parent.

I asked the owners how long is your dog in soil to get a better understanding of soil exposure. I found that although the city dogs have healthier soil, the dogs are not in the soil as much as the farm dogs. The average time in soil for the city dogs was 1.2 hours per day. The average time in soil for the farm dogs was 7.8 hours per day. See Figure 5 below.

Figure 5

Dog's microbiome health and time in the soil.

Microbiome Health and Time in the Soil



Graph made by student with help from parent.

Conclusion

My hypothesis was partially supported. Farm dogs do have a healthier microbiome and farm dogs did spend more time in the soil. What was not supported was that farm dogs will have healthier soil. Instead, city dogs had the healthier soil. The farm dogs could have had decreased soil health because of where I took the soil sample from. Also, farm dogs had more bacteria but not fungus. More studies need to be conducted on fungus to bacteria level.

The breed of the dog could also explain my results. The farm dogs were all large dog breeds. The city dogs were all small dog breeds.

Application and Future Research

This research might be helpful to tell the dog's owner how healthy the dog's microbiome is. If the dog was sick, the report could help the owner understand how to take care of the dog. I could also expand on this project next year. I would try to get more exact measurements of the microbiome bacteria numbers and the soil bacteria and fungus types. I could also look at the dog's activity level, the dog's behavior, and their age. It would be helpful for other people if they would want to do a similar project as me. Another study could look at the breed or size of a dog.

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Appendix A

Dog Permission Slip and Questionnaire

* Required

1. Email Address *

2. Dog's name

3. I give permission for my dog to participate in Kenley Mitchell's study. I give permission for Kenley to be on my property and to collect a stool sample from my dog and to collect a soil sample from my property.*

I understand that I will get a free report of my dog's microbiome for participating in this study.

To consent to the above statements, please type your name and the date in the space below.

4. Dog's Address*

5. About how many hours per day is your dog exposed to dirt?

6. What brand of dog food does your dog eat?

7. Do you use Roundup at your house? Mark only one oval.

Yes No

8. Does your dog have any allergies? Mark only one oval.

Yes No

9. Please explain any allergies below.

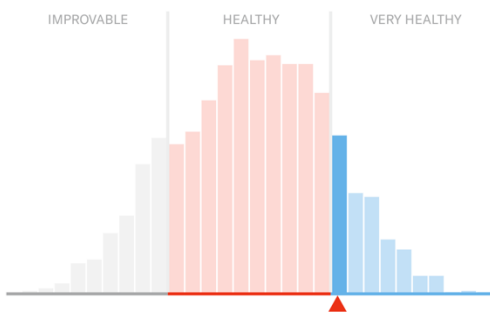
10. Does your dog have skin conditions?

Yes No

Please explain any skin conditions.

Appendix B

ELLIE'S MICROBIOME REPORT



Your dog's intestinal tract contains an incredible variety of microbes whose genetic material is referred to as the gut microbiome.

The specific types and amounts of microbes that colonize your dog's gut are affected by both your dog genetics as well as modifiable factors such as diet, physical activity, and antibiotic use. This means every pet has a unique gut microbiome that can change over time!

ELLIE'S MICROBIOME DIVERSITY

A diverse microbiome helps keep your pet healthy because it has microbes to fulfill a wide range of functions that are present in the right amounts.

- RICHNESS** Richness is simply the number of different bacteria found in your sample. 👍👍👍👍
- EVENNESS** Evenness specifies how evenly represented each type of bacteria is within the sample. 👍👍👍👍

BACTERIA LIVING IN ELLIE'S GUT

The bacteria represented below are regularly found in dog microbiomes. If you'd like to learn a little more about them, keep reading.

OUR BASELINE



ELLIE



< **DOREA** >

ABUNDANCE 👍👍👍👍

Prevalent in the guts of healthy dogs

Breaks down carbohydrates to produce beneficial nutrients for its host

Abundance decreased when dogs are switched to a low fat high fiber diet



BACTERIA LIVING IN ELLIE'S GUT

The bacteria represented below are regularly found in dog microbiomes. If you'd like to learn a little more about them, keep reading.

OUR BASELINE



ELLIE



< **LACTOBACILLUS** >

ABUNDANCE



Several strains are commonly found in healthy dogs
When administered as a probiotic it can shorten recovery time from acute diarrhea and results in measurable improvements in health
Various species frequently used as probiotics



BACTERIA LIVING IN ELLIE'S GUT

The bacteria represented below are regularly found in dog microbiomes. If you'd like to learn a little more about them, keep reading.

OUR BASELINE



ELLIE



< **MEGAMONAS** >

ABUNDANCE



Negatively associated with the rate of weight loss in dogs, i.e., dogs with more Megamonas may lose weight more slowly
Lower abundance in dogs with irritable bowel syndrome



BACTERIA LIVING IN ELLIE'S GUT

The bacteria represented below are regularly found in dog microbiomes. If you'd like to learn a little more about them, keep reading.

OUR BASELINE



ELLIE



< **RUMINOCOCCUS** >

ABUNDANCE



Higher abundance in dogs fed a high protein low carbohydrate diet
Able to break complex carbohydrates down into smaller molecules called short chain fatty acids, (SCFA), which can provide energy for intestinal cells
Dogs with increases in Ruminococcus have decreased rate of weight loss, possibly because SCFAs are used as an additional energy source by a dog's cells within the gut



BACTERIA LIVING IN ELLIE'S GUT

The bacteria represented below are regularly found in dog microbiomes. If you'd like to learn a little more about them, keep reading.

OUR BASELINE



ELLIE



< **BLAUTIA** >

ABUNDANCE [5 icons]

Commonly found in the guts of dogs
Abundance reduced in dogs with gastrointestinal disorders or acute diarrhea
Abundance increases when dogs are fed a diet with supplemental fiber



BACTERIA LIVING IN ELLIE'S GUT

The bacteria represented below are regularly found in dog microbiomes. If you'd like to learn a little more about them, keep reading.

OUR BASELINE



ELLIE



< **FUSOBACTERIUM** >

ABUNDANCE [5 icons]

Common in the guts and oral cavities of healthy dogs
More abundant in dogs fed a diet that includes raw meat
Abundances lower in dogs with gastrointestinal disease



BACTERIA LIVING IN ELLIE'S GUT

The bacteria represented below are regularly found in dog microbiomes. If you'd like to learn a little more about them, keep reading.

OUR BASELINE



ELLIE



< **CLOSTRIDIUM** >

ABUNDANCE [5 icons]

Typically the most abundant group of bacteria in the guts of healthy dogs
Abundance increased in dogs fed a raw red meat diet
Abundance can decrease upon switching to a low fat high fiber diet
Lower abundance associated with inflammatory bowel disease in dogs



BACTERIA LIVING IN ELLIE'S GUT

The bacteria represented below are regularly found in dog microbiomes. If you'd like to learn a little more about them, keep reading.

OUR BASELINE



ELLIE



< **BIFIDOBACTERIUM** >

ABUNDANCE



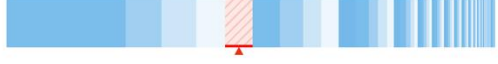
- Several species are regularly found in healthy dogs
- Responds favorably to potato fiber in the diet
- Various species frequently used as probiotics



BACTERIA LIVING IN ELLIE'S GUT

The bacteria represented below are regularly found in dog microbiomes. If you'd like to learn a little more about them, keep reading.

OUR BASELINE



ELLIE



< **BACTEROIDES** >

ABUNDANCE



- Growth is favored when dogs are fed a diet high in carbohydrates and low in protein
- Abundance increases as dogs age
- Abundance can decrease if dogs have been treated with omeprazole, a common treatment for gastrointestinal reflux and ulcers



BACTERIA LIVING IN ELLIE'S GUT

The bacteria represented below are regularly found in dog microbiomes. If you'd like to learn a little more about them, keep reading.

OUR BASELINE



ELLIE



< **ESCHERICHIA** >

ABUNDANCE



- Associated with gastrointestinal diseases such as inflammatory bowel disease and chronic diarrhea
- Abundance increases in dogs fed a diet of raw beef or bones and raw food
- Providing the prebiotics inulin or yeast cell extract may help reduce levels



Photos

Me running the Microbiometer kit.

Photo by Clay Mitchell.



The NomNom kit.

Photo by Kenley Mitchell.



Me running the NomNom kit.

Photo by Gina Mitchell.



The Microbiometer kit.

Photo by Kenley Mitchell.

Acknowledgements

I would like to give a big thanks to my mom and dad for helping me with my project.