PURINA Institute

Advancing Science for Pet Health

UNDERSTANDING PROBIOTICS

And Their Impact On Pet Health

PURINA Institute

Advancing Science for Pet Health

A probiotic supplement for dogs or cats or a probiotic in a pet food can be defined as a viable microorganism (or multiple viable microorganisms) that provides health benefits for the host when fed at a specific amount. Probiotics have the potential to provide many benefits to pets. These benefits include promoting digestive health, supporting immunity, and other benefits outside the gastrointestinal tract in systems affected by the gut microbiome, such as the brain. The specific bacterial strain in the probiotic should have demonstrated efficacy in the target species.

Bifobacterium

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MICROBIOME AND PROBIOTIC BASICS

What Is the Gut Microbiome?

The gastrointestinal tract contains thousands of species of bacteria, some of which are beneficial and some of which are potentially pathogenic. The gut microbiome contains an entire ecosystem of microorganisms – called microbiota – that includes these bacteria, as well as viruses and fungi. The microbiome is the full collection of genetic material of all the gut microbiota. The majority of microbiota are in the large intestine, with bacteria from genera including *Bifidobacterium*, *Lactobacillus*, and *Enterococcus*, as well as some potentially pathogenic bacteria from genera such as *Clostridium* and *Staphylococcus*.[1-4] The predominant phyla present in the gut microbiome of healthy dogs and cats include Firmicutes, Bacteroidetes, Actinobacteria, and Fusobacteria.[5-7] Despite phyla similarities, the microbiome is unique for each dog. In one study analyzing the fecal microbiota in 76 healthy dogs by qPCR and shotgun sequencing, only a small number of bacterial species (17/1190) were consistently present in the microbiomes of these dogs.[8]

In the human body, the number of bacterial cells present in the gut microbiome is estimated to be in the trillions and is approximately equivalent to the total number of human cells in the body.[9] Previous resources considered the bacterial count to be about ten times higher than the number of host cells.[1]

To understand the gut microbiome, it is important to understand the function of the GI tract. Each section of the GI tract has unique characteristics and different functions:

- The role of the stomach is to begin the digestive process with the mechanical and chemical breakdown of food.
- The small intestine's role is to complete digestion of macronutrients and allow for nutrient absorption.
- The large intestine (colon) primarily is responsible for handling waste from the digestive process, which involves stool production and further processing of the waste by the microbiome, allowing for fermentation, production of short chain fatty acids, and other processes. The large intestine is also responsible for water absorption and balance.

The stomach contains low levels of bacteria. In the small intestine, there is a small population of bacteria that gradually increases in size closer to the large intestine. The colon is inhabited by many bacterial species, and the bacterial population can vary among individuals as well as species. However, there is a core population of similar bacteria that are present in every animal of a certain species.

There are numerous known functions of the gut microbiome and additional functions that have yet to be discovered. Some of the known functions of the gut microbiome include:[10,11]

- Providing nutrients for the host via fermentative and metabolic activities
- Breaking down dietary fibers
- Helping develop the immune system and maintaining homeostasis
- Defending against intestinal pathogens
- Aiding nutrient digestion and absorption in the gastrointestinal tract
- Supporting gastrointestinal health

Many of these functions result in improved stool quality, enhanced immune function, decreased stress-related GI signs, and reduced anxious behaviors. In addition to directly impacting GI health, the gut microbiome may affect other organs and systems, such as the brain. Maintaining microbial balance in the gastrointestinal tract can help ensure optimal gastrointestinal and immune function and influence the overall health of the animal. There are connections between the gut microbiome and

other systems including the brain and heart.[12,13] Nutritional modifications, including probiotics, can help maintain microbiome health.

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Gut Microbiome Composition and Imbalance

Individual dogs and cats have a unique gut microbiome, although there is a group of "core" microbial species found in every pet. *Bacteroides, Lactobacillus, Bifidobacterium*, and *Enterococcus* are the predominant bacterial genera cultured from canine and feline GI tract.[1] In addition to beneficial bacteria, there are potentially harmful microorganisms in the gut microbiome. The composition of the microbiota can be affected by the pet's environment, genetics, diet, stress, medications, host health, and other factors. Microbiota imbalance, or changes in the composition of the gut microbiome that impact microbiome function, is called dysbiosis and can significantly impact the health of the GI tract and other systems.[14,15] For example, dysbiosis can result in decreased production of short-chain fatty acid (SCFA production).[14] With dysbiosis, restoring the gut microbiome is crucial to maintaining health and to helping manage GI disease. Benefits of preserving and restoring microbiome health include:

- Supporting immune function
- Aiding digestion
- Suppressing colonization and growth of potentially pathogenic organisms
- Production of certain vitamins

Animals with GI diseases such as chronic enteropathies may have a greater degree of dysbiosis versus healthy dogs and cats.[14] Nutritional interventions, such as a change in diet to include more prebiotic fibers and/or the addition of probiotics, may be beneficial in cases where intestinal dysbiosis is present.

What Is a Probiotic?

A probiotic is defined by the International Scientific Association for Probiotics and Prebiotics (ISAPP) as "live microorganisms that, when administered in adequate amounts, confer a health benefit on the host."[16,17] Probiotics have the potential to provide many benefits to pets. These benefits include promoting digestive health, supporting immunity, and many other benefits outside the gastrointestinal tract in systems affected by the gut microbiome, such as the brain.

Probiotics are referred to by their genus, species, and strain. Probiotics are strain-specific and dose-dependent, meaning different strains have different effects, and a specific amount must be provided to deliver benefits to the pet.

Probiotics can be provided as a supplement or can be found in pet food. Regardless of the delivery method, the probiotic should be viable when consumed. Probiotic supplements are becoming increasingly popular with pet owners and have the potential to support pet health. When recommending probiotic supplements, veterinarians should research the probiotic in the supplement to ensure it has demonstrated efficacy in the target species.

INFLUENCING HEALTH WITH PROBIOTICS

Probiotics are one way to influence gut health and other aspects of pet health. Probiotic supplementation or providing a probiotic in a pet food may aid in maintaining or improving digestive health by:

- Competition with potentially pathogenic bacteria in the GI tract
- Production of antibacterial substances
- Creation of an environment that is unfavorable for pathogens
- Production of metabolites that can be beneficial to the host and other microbiota (such as SCFAs)

Probiotics may be administered for multiple clinical purposes. However, to achieve an effect, the probiotic bacteria must be alive and in sufficient numbers in the supplement or diet. It is important to consider that the desired effects associated with probiotic usage often require continuous administration of the probiotic.

Intestinal dysbiosis can be associated with many health concerns including gastrointestinal conditions, changes in immune function, and anxiety/stress. As a result, probiotics can help veterinarians manage several different health issues in dogs and cats. These include reducing GI signs such as diarrhea associated with stress, dietary change, dietary indiscretion, or antibiotic usage.[18-20] Some probiotics are also safe for these uses in puppies and kittens over eight weeks of age. Some probiotics can also reduce anxious behavior and increase social interactions by impacting the gut – brain axis.[13,21,22] Probiotics can also be immunomodulatory; supplementation or feeding of certain probiotics has been shown to support immune function.[23]

To influence health, a probiotic provided as a supplement or in food should meet some or all of the characteristics of an effective probiotic for pets, which include the following:

1. **Be resistant to digestion by gastric acid or intestinal enzymes**
2. **Reduce or prevent the adherence of pathogenic bacteria in the gut**
3. **Produce products unfavorable to pathogen growth (example: short-chain fatty acids that lower intestinal pH)**
4. **Remain viable until consumption by the pet**
5. **Promote a normal and balanced microbiome**
6. **Provide a health benefit to the dog or cat**
7. **Be safe for the pet, noninvasive, and nonpathogenic**

There are multiple probiotics that meet these characteristics and requirements, such as *Enterococcus lactis* (formerly *Enterococcus faecium*) SF68 (NCIMB 10415 4b1705), *Bifidobacterium longum* BL999 NCC 3001 (BL999), and *Bacillus coagulans* GBI-30, 6086, commonly called BC30, and others. Results of clinical studies using these probiotics are applicable only to the specific strains in the probiotic supplement or pet food and cannot be assumed to be effective for all strains of the bacterial species. Extensive research has led to the development of probiotics that are both effective and safe for dogs and cats.

Enterococcus lactis SF68, *or E. lactis* SF68, has a long history of safe use in both animals and humans. It is nonpathogenic and nontoxic and does not have abnormal resistance to antibiotics. Importantly, it survives the conditions of the gastrointestinal tract and reaches the intestines but does not colonize the GI tract permanently.[24] An extensive number of publications show *E. lactis* SF68 supplementation results in an improvement of diarrhea associated with multiple causes, and that it supports a strong immune system.[18-21,23,25-29] One example is *E. lactis* SF68 improving diarrhea resulting from stress and antibiotic usage.[18,20,27] *E. lactis* SF68 can be safely fed to puppies and kittens 8 weeks and older, in addition to adult and senior dogs and cats.

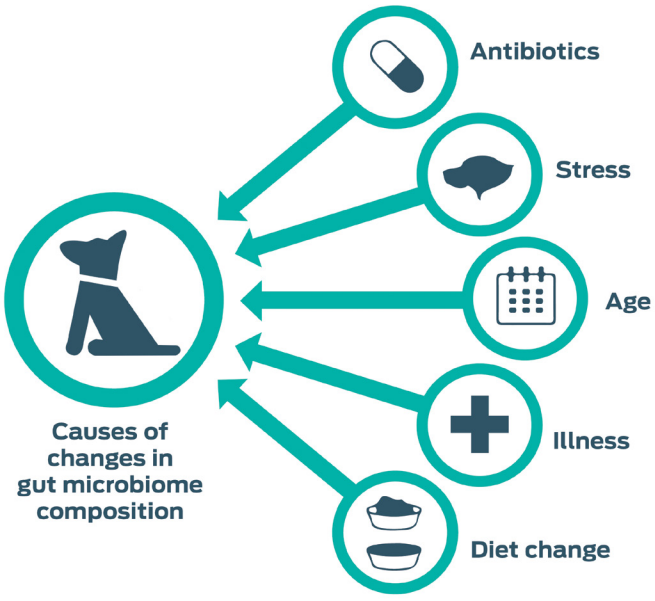
Bifidobacterium is one of the most significant bacterial genera of the large intestine in mammals. *Bifidobacterium longum* BL999, or *B. longum* BL999, lacks pathogenicity in all age groups and in immunocompromised individuals.[30,31] *Bifidobacterium* species have a wide variety of effects on

host health including inhibition of adhesion of pathogenic bacteria, stimulation of immune function, and reducing anxious behaviors.[13,22,32]

BC30 is another probiotic used to support pet health. In most situations, probiotics added to pet food are sensitive to the heat necessary for manufacturing. BC30 is more resistant to stresses associated with dry pet food production and can be added to dry pet food as a topical coating.[33] It tends to be more resilient because it is a spore-forming bacteria.[34] BC30 is nonpathogenic, and previous studies show it may help enhance the immunologic response,[34] and has no known adverse effects on health in dogs or cats.

Gastrointestinal Health and Probiotics

For gastrointestinal disease, effective probiotics may compete with pathogens for adhesion sites on the gut wall. Some researchers suggest that toxin receptors on the intestinal mucosa may be affected by probiotics.[35] Probiotics can help inhibit colonization by other strains and encourage growth of other beneficial bacteria.[35] Probiotics may also be useful in times when the microbiome is challenged and at risk of developing dysbiosis. Potential challenges to the gut microbiome include environmental changes, dietary modification, antibiotics, stress, or dietary indiscretion.



Immunity and Probiotics

The gastrointestinal tract has an extensive network of immune cells, comprising over 70% of the body's immune system.[36] The immune system can be affected by the microbiome at both a systemic and mucosal level. Probiotics have the potential to beneficially influence both the innate and acquired immunity by a variety of proposed mechanisms. Some of the proposed mechanisms include altered cytokine production, natural killer cell activity, and specific and non-specific immunoglobulin production.[37] However, the effect will vary depending on the probiotic.

The Gut-Brain Axis

The gut microbiome impacts body systems in addition to the gastrointestinal tract and has a significant effect on health. There is evidence showing that manipulation of the gut microbiome can reduce some anxious behaviors.[13,22] The bidirectional communication between the microbiome, the gastrointestinal tract, and the brain is called the gut-brain axis. The gut microbiota plays a critical role in the gut-brain axis to influence behavior, brain biochemistry, and neural development, with both the composition and diversity of the microbiota affecting this communication.[38,39] This communication occurs to some degree through the enteric nervous system, which is a network of neurons along the length of the digestive tract that communicates with the brain via the vagus nerve.[40] The enteric nervous system is part of the autonomic nervous system. The gastrointestinal immune system and hormonal changes may also impact communication between the gut microbiome and the brain through chemical messengers or metabolites of the gut microbiota.[41]

The gut-brain communication via the microbiota can be further impacted by stress hormone production, including cortisol and epinephrine, which have an effect on the gut.[39] Production of hormones and neurotransmitters can be affected by the microbiome, such as the regulation of tryptophan availability for serotonin production.[42] For these reasons, intestinal dysbiosis and altering the microbiome

can affect health conditions including cognitive changes and anxiety.[13,22,43-45] The display of anxious behaviors can be accompanied by increased susceptibility to illness and gastrointestinal signs in addition to behavioral signs[46,47] Some of the signs of anxiety may be difficult for pet owners to recognize.

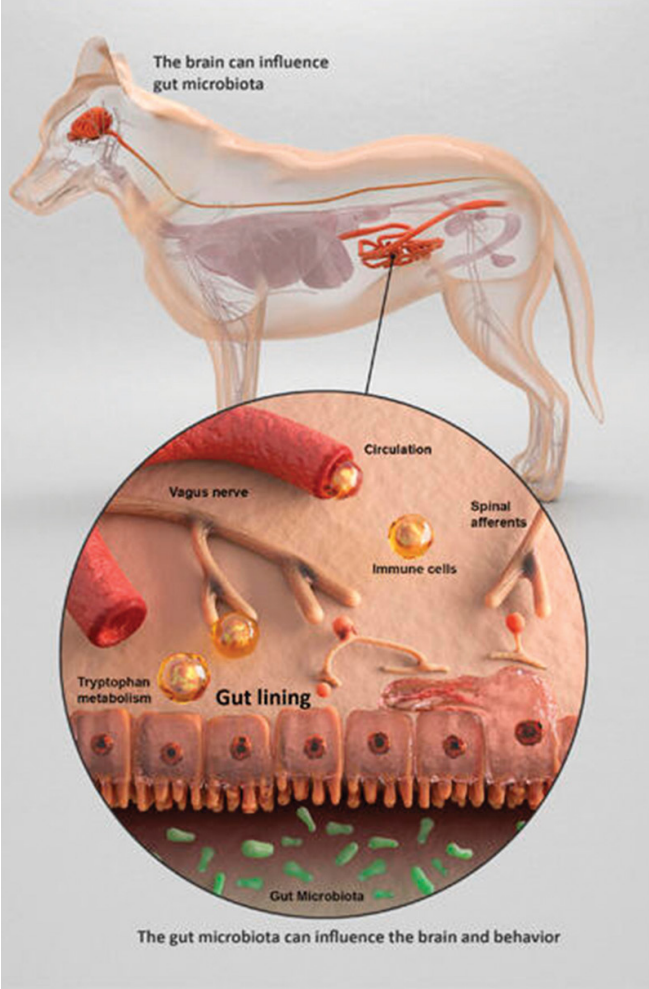


Figure:
The gut-brain axis involves bidirectional communication

RESEARCH ON PROBIOTICS

Because effects vary by probiotic strain and by host species, probiotics are currently a vast area of research in veterinary medicine. In addition, there is still much more to be learned about the microbiome, and the interactions between the microbiome, the host, and probiotics.

It is known that the benefit of probiotics is not solely gut-related. Systems or areas that have been shown to benefit most from probiotics to date include the gastrointestinal tract, the immune system, and behavior and stress. There has been research on the effects of and relationships between the gut microbiome and the kidneys,[48] heart,[12] and other systems. Many of these other areas require ongoing research to determine if these body systems can benefit from probiotic supplements. For parts of the body with their own microbiome—such as the skin and oral cavity —additional research could determine the influence of probiotics on those microbiomes, what those benefits could be, how the probiotics could be given, and what strain or strains of microorganisms would be most beneficial.

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Enterococcus lactis SF68: Research on GI Signs and Immune System Support

Enterococcus lactis SF68 is a nontoxic, nonpathogenic strain of *E. lactis* that influences gastrointestinal and immune health in pets. It is very unlikely to contribute to the antimicrobial resistance challenges facing public health.[24] *E. lactis* SF68 has been shown to be safe and has a long history of safe use in both animals and humans. It can encourage beneficial bacteria such as *Lactobacilli* to thrive and discourages the growth of potentially pathogenic bacteria such as *E. coli* and *Clostridium perfringens*.[49,50] Many published studies demonstrate *E. lactis* SF68 has potential to promote digestive health and support a strong immune system in dogs and cats and are shown in the table. *E. lactis* SF68 can be microencapsulated to increase stability and viability.

System Studied	Summary	Reference Number
Gastrointestinal	Kittens fed *E. lactis* SF68 for one year had significantly reduced incidence of diarrhea and a significantly lower number of days with diarrhea during the year versus a placebo.	19
Gastrointestinal	Alaskan sled dogs with acute diarrhea had a significantly fewer number of days with severe watery diarrhea and improved sooner when they received *E. lactis* SF68, versus dogs that received a placebo.	20
Gastrointestinal	Shelter dogs treated with *E. lactis* SF68 and metronidazole experienced faster resolution of diarrhea than dogs treated with metronidazole alone.	18
Gastrointestinal	Oral administration of amoxicillin-clavulanate commonly induces diarrhea and alters the gastrointestinal microbiome. Feeding *E. lactis* SF68 can lessen some of the associated clinical abnormalities.	27
Gastrointestinal	Adult dogs fed a complete and balanced commercial diet and a supplement with *E. lactis* SF68 for two weeks had reduced flatulence, with a reduction in the number of total flatulence events and a lower amount of hydrogen sulfide released.	29
Gastrointestinal	Cats fed *E. lactis* SF68 in a shelter environment had fewer episodes of diarrhea lasting more than two days compared to cats receiving a placebo.	25
Immunity	Puppies supplemented with *E. lactis* SF68 from weaning to one year showed a greater antibody response to the canine distemper virus vaccine versus the control group, demonstrating SF68 promoted a strong immune system.	23
Immunity	Some cats with Feline Herpesvirus-1 fed *E. lactis* SF68 for 20 weeks had reduced morbidity associated with chronic FHV-1 infection.	21
Immunity	Supplementing adult beagles with *E. lactis* SF68 can induce immunomodulation as early as 4 weeks.	26
Immunity	Cats supplemented with *E. lactis* SF68 had a higher percentage of CD4+ lymphocytes versus cats receiving a placebo.	28

Bifidobacterium longum BL999 NCC 3001 and the Gut-Brain Axis

Certain probiotics offer an opportunity to address anxious behaviors in dogs and cats. Studies evaluating the effects of *Bifidobacterium longum* BL999 NCC 3001 have shown benefits on psychological stress in humans.[51] *B. longum* BL999 NCC 3001 has also been shown to reduce the expression of anxious behaviors in dogs and in cats. Dogs fed *B. longum* BL999 for 6 weeks had improved behavioral evaluations versus dogs receiving a placebo. Improvements such as reduced barking, reduced pacing, and increased exploratory behavior were noted.[13] Dogs supplemented with *B. longum* BL999 had a decreased heart rate and increased heart rate variability, suggesting improved adaptation to stimuli of potential anxious behaviors. The researchers also noted decreased salivary cortisol in response to stimuli.[13]

Anxious behaviors can be reduced in cats supplemented with *B. longum* BL999 as well. Cats given *B. longum* BL999 for 12 weeks showed reduced signs of stress and more social behavior versus cats given a placebo.[22] The cats fed *B. longum* BL999 were less likely to have abnormal serum cortisol concentrations.[22]

BC30: Research on Gastrointestinal Health and Immune System Support

Although published research is limited in dogs and cats, BC30 is a nonpathogenic spore-forming probiotic that has been shown to be resistant to extreme temperatures and to survive the environment in the upper gastrointestinal tract. Studies in humans have demonstrated digestive health and immunity benefits of BC30, and this probiotic seems to especially support immunity during times of stress.[52-56] The spore-forming nature and heat resistance make BC30 a good candidate for a probiotic that is effective when used within pet foods.

Purina internal studies have provided evidence that BC30 can increase gastrointestinal microbial diversity in dogs, support digestive health in dogs and cats, and help reduce the percentage of days with diarrhea in cats.

PRACTICAL ASPECTS OF PROBIOTIC SUPPLEMENTATION

Probiotics can be provided in pet food or as a dietary supplement. A diet or supplement containing a probiotic may provide one strain or multiple strains of bacteria. The probiotic strain(s) should be chosen based on the desired effect and the evidence of the probiotic's efficacy in the target species.

When selecting a probiotic, quality control and safety should also be considered. Quality and safety considerations for probiotics in pet food and probiotic supplements include:

- The manufacturer of the probiotic should be known and trusted
- A quality probiotic survives typical manufacturing, shipping, and storage conditions
- The safety and efficacy of the probiotic should not be altered during typical storage conditions
- The probiotic should remain alive until at least the expiration date and the number of bacterial cells or colony forming units (CFU) should not decline below the package guarantee before the expiration date
- The specific strain(s) and quantity (CFU) should match what is listed on the label
- The specific strain(s) should be tested and show evidence of efficacy in the species in question, without adverse effects

Because the microbiome differs among species, probiotics recommended to clinical patients should have a demonstrated effect in the species in question. However, there is no evidence that probiotics must originate from the same species as the target species. For example, probiotics given to dogs do not have to be derived from a dog. This includes both single-strain and multi-strain

probiotics. Efficacy of a probiotic supplement should be determined regardless of the number of strains present in the supplement. Overall, the most important aspect is high-quality evidence supporting the benefits and safety of the probiotic in the species in question.

To be effective, a probiotic must be stable throughout its shelf life to be present in sufficient numbers or colony-forming units (CFU). This means the probiotic must remain stable during manufacturing, shipping, and typical storage conditions. A study of 25 commercial veterinary probiotic products showed only 2/25 of the products had a label that accurately described the content, including the specific strains and the CFUs.[57] Therefore, it is important to investigate a probiotic's efficacy in the target species and to use products from trusted manufacturers, so an appropriate, safe, and reliable product can be selected.

To be effective, a probiotic must be stable throughout its shelf life to be present in sufficient numbers or colony-forming units (CFUs). A probiotic must contain at least the CFUs on the label guarantee at the time of expiration. Therefore, it is important to investigate a probiotic's efficacy in the target species and to use products from trusted manufacturers.

SUMMARY

Probiotics are live microorganisms that can benefit health when fed in a specific amount to a certain species. Recommending probiotics for a dog or cat have been shown to help promote microbiome health and balance, which can improve overall pet health. The specific bacterial strain in the probiotic supplement having demonstrated efficacy with high-quality scientific evidence in the target species is an important detail.

As more research emerges with respect to the microbiome and the beneficial effects of probiotics, it is hoped that in the future, positive effects can be demonstrated to support more health conditions.

Probiotics are commonly administered to dogs and cats. Before recommending a specific product, veterinarians should ensure there is evidence from the manufacturer indicating the probiotic has benefit in the target species and that there is evidence the microorganism in the probiotic can survive production and storage.

Microbiome Bacteria

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