# Influence of Probiotic Supplementation on Health of Dogs

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Probiotics are live microorganisms that deliver health benefits to the host when administrated in an adequate amount. The possible mechanism behind probiotics' beneficial effects could be their positive regulation of the host's intestinal microbiota. Probiotics are reported to have therapeutic properties against canine GI and other diseases.

Keywords: canine ; probiotics ; microbiota ; Lactobacillus

## 1. Introduction

Companion dogs need nutritional care to maintain good health [1]. Dogs are fed with foods supplemented with appropriate nutrients such as choline, vitamins (E, B5, B3, B2, B1, K, B6, A, B9, B12, D), and minerals (calcium, potassium, phosphorus, magnesium, etc.) in addition to the dietary components, such as proteins, carbohydrates, fats, and fibers [1]. Malnutrition can occur due to an excess or a deficiency in nutrient intake, which could be harmful to dogs 2. The nutritional requirements of each dog can differ based on the dog's breed, size, developmental stage, and level of activity <sup>[1]</sup>. Even if companion animals are fed with nutritionally balanced diets to maintain good health, any differences in their normal microbiota can facilitate illness upon exposure to harmful environmental influences and pathogens [3][4]. The supplementation of probiotics positively regulates the microbiota of the host and improves their health <sup>[5][6][7]</sup>. The joint Food and Agriculture Organization (FAO) of the United Nations and World Health Organization (WHO) defines the term "probiotics" as "live microorganisms which when administered in adequate amounts confer a health benefit on the host" <sup>[8]</sup>. The term "probiotics" refers to live microbes (isolated from different sources, such as gut commensals, fermented foods, and any other source) that have been characterized and evidenced in adequate controlled studies to have a health beneficial effect, and their safety has been verified. Even though microbial components, dead microbes, and microbial products exhibit health beneficial effects, they should not be considered as probiotics <sup>[9]</sup>. The ideal characteristics of probiotics include being non-pathogenic to the host, the ability to tolerate low pH levels and high concentrations of bile acids in the gastrointestinal environment, and the ability to proliferate in the gut by adhering to the intestinal epithelium. For economically viable production, the probiotics should also preferably have the ability to grow well on inexpensive media and tolerate manufacturing, transportation, and storage processes [10][11]. Probiotic supplementation aids in maintaining gut health by preventing or controlling pathogens in the gastrointestinal tract and promoting the population of favorable gastrointestinal tract microflora. The modes of action of probiotics might be strain-specific [11]. Since the supplementation of probiotics is not universally safe and effective in humans [12], probiotics should also be used with caution in companion animals until the limitations of probiotics usage in companion animals are found. Probiotics have been reported to have several health benefits in humans, such as facilitating dental health [13], aiding in cancer management [14], exerting cholesterol-lowering abilities [15], controlling diabetes mellitus [16], and exhibiting immunestimulating properties <sup>[12]</sup>. Generally, probiotics that are beneficial to humans are considered to be safe, but caninederived probiotics could be used safely for canine animals [18].

The intervention of a probiotic-supplemented diet can protect dogs from various health effects. The supplementation of probiotics has been used for the alleviation or prevention of atopic dermatitis <sup>[19][20]</sup> and treatment of gastrointestinal diseases, such as diarrheal diseases <sup>[21][22][23][24][25]</sup> in companion animals.

## 2. Characterization of the Microbiota in Dogs

The microbiomes of humans and pet animals have similarities  $\frac{[26]}{}$ , and the microbiota play a critical role in health and diseases among pet animals  $\frac{[27][28]}{}$ . Previous bacterial-culture-based studies showed that healthy dogs contain a  $10^2$  to  $10^{11}$  CFU/g bacterial load in their gastrointestinal (GI) tracts. Recently, high-throughput DNA sequencing techniques were implemented to determine the microbiomes of canines  $\frac{[29]}{}$ . Current molecular methods revealed that the microbial loads of

healthy dogs can range from  $10^{12}$  to  $10^{14}$  [30][31][32]. Garcia–Mazcorro et al. <sup>[33]</sup> reported that Firmicutes was the predominant phylum, followed by Actinobacteria, Proteobacteria, and Bacteroidetes, which were identified in the fecal microbiome of healthy dogs. Fusobacteria and Acidobacteria were the least abundant phylum compared to the other identified phyla in the fecal microbiome of healthy dogs <sup>[33]</sup>. Jha et al. <sup>[34]</sup> reported the same set of phyla as the predominant one found in the fecal samples of dogs <sup>[34]</sup>.

The microbial load of each segment in the GI tract varies based on that section's physiological functions <sup>[35][36]</sup>. For instance, the small intestine is rich in aerobic and facultative anaerobic bacteria, while the colon harbors anaerobic bacteria <sup>[35]</sup>. Individual variations in the microbiota of different intestinal compartments (such as higher diversity indices for the colon, rectum than those of duodenum, jejunum, and ileum within individual dogs) may also be present, and these variations must be taken into account to explain the microbiota of healthy dogs <sup>[36]</sup>. The results can fluctuate based on the sequencing method and depth of the study. Some studies showed that Firmicutes (abundant in duodenum, jejunum, ileum, and colon), Bacteroidetes (abundant in ileum, and colon) <sup>[37]</sup>, Proteobacteria (abundant in duodenum <sup>[37][38]</sup>, jejunum, ileum <sup>[38]</sup>, and rectum <sup>[37]</sup>), and Actinobacteria <sup>[37]</sup> are the primary phyla identified in the GI tract of the dog microbiota <sup>[37][38]</sup>.

## 3. Health Benefits of Probiotic Supplementation in Healthy Dogs

## 3.1. Microbiota Changes and Other Benefits in Healthy Dogs upon Probiotic Supplementation

The supplementation of *Enterococcus faecium* NCIB 10415 (derived from healthy adult canine feces) ( $9.2 \times 10^9$  CFU/dog/day) for 18 days reduced the fecal count of *Clostridium* spp. in healthy dogs. *E. faecium* NCIB 10415 lowered the fecal count of *Campylobacter* spp. and *Salmonella* spp. in a few healthy dogs and insignificantly increased the fecal count of *Campylobacter* spp. and *Salmonella* spp. in the majority of the healthy dogs, indicating that *E. faecium* NCIB 10415 was not effective in inhibiting the growth of *Campylobacter* spp. and *Salmonella* s

Biagi et al. <sup>[39]</sup> studied the effects of *Lactobacillus animalis* LA4 (derived from healthy adult canine feces) on the composition and metabolism of dog intestinal microbiota under both in vitro and in vivo conditions. The in vitro evaluation showed that upon exposure of *L. animalis* LA4 to adult dog fecal cultures, *L. animalis* LA4 increased the count of *Lactobacillus* and reduced the *Clostridium perfringens* count. Moreover, LA4 increased the level of lactic acid in fecal cultures. LA4 reduced the ammonia concentration under in vitro conditions, while fecal concentration of ammonia was not influenced by LA4 under in vivo conditions. When healthy dogs (n = 9 adult) were fed with *L. animalis* LA4 (10<sup>9</sup> CFU/g; 0.5 g per dog per day) for 10 days, the count of *Lactobacillus* was increased, and the enterococci count was reduced. The in vivo evaluations of *Lactobacillus* count also supported the findings of the in vitro experiments. The in vivo results showed that the strain LA4 could withstand the gastrointestinal environment and proliferate in the intestines of the studied adult dogs <sup>[39]</sup>.

## 3.2. Immune Immunomodulatory Properties

*L.johnsonii* CPN23 (canine origin) administration to healthy adult Labrador dogs improved the fecal acetate and butyrate levels. *L. johnsonii* CPN23 also reduced fecal ammonia concentrations. Cell-mediated immune response was also evaluated as type-IV delayed-type hypersensitivity reaction to intradermally injected phytohaemagglutinin-P. CPN23 administration improved the cell-mediated immune response (as delayed-type hypersensitivity response in the form of increase in skin induration) in healthy dogs. There was no change in the antibody response to sheep erythrocytes. The study concluded that *L. johnsonii* CPN23 could be used to achieve improvements in canine health <sup>[40]</sup>.

# 4. Health Benefits of Probiotic Supplementation in Diseased Dogs

## 4.1. Chronic Kidney Disease

VSL#3 supplementation effectively increased the glomerular filtration rate (GFR) in dogs with chronic kidney disease. In more detail, the 60-day administration of VSL#3 (a mixture of probiotics containing *L. casei*, *L. plantarum*, *L. acidophilus*, *Lactobacillus delbrueckii* subsp. *bulgaricus*, *B. longum*, *Bifidobacterium breve*, *Bifidobacterium infantis*, and *Streptococcus salivarius* subsp. *thermophilus*; 112 to  $225 \times 10^9$  CFU/10 kg body weight; every 24 h) to dogs (n = 30) with chronic kidney disease increased the GFR compared to that of the control group (n = 30) and baseline values <sup>[41]</sup>.

#### 4.2. Atopic Dermatitis

The oral administration of probiotic strain *Lactobacillus sakei* probio-65 ( $2 \times 10^9$  CFU per gram per day for two months) to dogs (n = 32) diagnosed with canine AD effectively reduced the disease severity index measured by CADESI scoring system. The double-blind placebo-controlled study demonstrated that *L. sakei* probio-65 could be considered an adjuvant therapeutic agent to manage canine AD <sup>[42]</sup>.

*B.longum* (5 × 10<sup>10</sup> CFU/day) was orally supplemented in dogs with AD for 12 weeks, and the severity of the skin lesions was assessed using the CADESI score. Pruritus severity was also assessed using the pruritus visual analog scale (PVAS). The transepidermal water loss (TEWL) and the medication score was also recorded. The data were collected every four weeks of the study. The results revealed that the administration of *B. longum* progressively reduced the CADESI score compared to the baseline, whereas probiotic supplementation was not effective in reducing the TEWL, PVAS, and medication scores. The study determined that *B. longum* could improve skin lesions in dogs <sup>[43]</sup>.

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