Perspectives and advances in probiotics and the gut microbiome in companion animals

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Abstract
As the number of households that raise dogs and cats is increasing, there is growing interest in animal health. The gut plays an important role in animal health. In particular, the microbiome in the gut is known to affect both the absorption and metabolism of nutrients and the protective functions of the host. Using probiotics on pets has beneficial effects, such as modulating the immune system, helping to reduce stress, protecting against pathogenic bacteria and developing growth performance. The goals of this review are to summarize the relationship between probiotics/the gut microbiome and animal health, to feature technology used for identifying the diversity of microbiota composition of canine and feline microbiota, and to discuss recent reports on probiotics in canines and felines and the safety issues associated with probiotics and the gut microbiome in companion animals.

Keywords: Probiotics, Gut microbiome, Companion animal, Canine, Feline

INTRODUCTION
Terms such as ‘companion animals’ apply to households with pets, companion dogs and companion cats that are frequently encountered in the surroundings. The word ‘companion’, with which we are already familiar, refers to an animal that lives with humans and was first proposed by zoologist and Nobel Prize winner Konrad Lorenz at an international symposium held in Vienna, Austria in 1983 [1]. Households that raise these pets accounted for 29.7% of the total households in Korea, with 6.04 million households at the end of 2020 [2]. As the number of people raising companion animals is increasing, the relationship between humans and companion animals is further developing.

Most pet owners currently treat their pets as family, colleagues, and friends [3]. Pet humanization, a phenomenon that recognizes companion animals as family members and treats them as individuals with emotions, has been established as a global trend [4]. In Korea, the trend of pet humanization is...
Probiotics and the gut microbiome in companion animals

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also spreading, with 88.9% of companion households and 64.3% of general households agreeing to the phrase ‘pets are part of the family’ [2]. A typical example is that the pets do the same things as humans do, such as having birthday parties for dogs and cats, sleeping with the owner in the bed, and others. Companion animals have become an increasingly important part of human life, and therefore, the health and well-being of pets have increasingly attracted interest in recent decades [5].

Dogs and cats have evolved into carnivores with high-protein diets and have relatively simple gastrointestinal tracts (GITs) [5,6]. Cats are carnivores that rely on high-protein animal tissues to meet their unique nutritional requirements in the wild and consume protein-containing feed to meet their nutrients in the case of household felines. They are metabolically adapted to low glucose utilization and high protein metabolism [5,7]. Although dogs share many anatomical and metabolic characteristics with cats, they are metabolically omnivorous and can digest, absorb and metabolize significant amounts of carbohydrates [8].

The gut plays an important role in animal health, and the GIT contains a complex microbial community. A healthy gut is known to affect host physiology and well-being. This microbial ecosystem acts in several ways, affecting both the absorption and metabolism of nutrients and the protective functions of the host. Probiotics are defined as ‘living microorganisms that provide health benefits to the host when administered in appropriate amounts’ [9]. Recently, gut-related probiotic products aimed at pets, particularly dogs and cats, have also gained in popularity among owners [10]. The benefits of using probiotics for pets include their modulation of the immune system, help in reducing stress, protection against infections caused by intestinal pathogens and growth performance development [11]. As dogs and cats become family members, the number of studies about dogs and cats has been increasing. Among their topics, knowledge about the gut microbiome and probiotics in dogs and cats is still expanding. However, published papers on the application of probiotics in companion animals are significantly limited compared to those in humans. The purpose of this review is to describe the current knowledge about the gut microbial communities in dogs and cats in relation to probiotics.

PROBIOTICS FOR COMPANION ANIMALS

Probiotics that are living and beneficial microbiota have been used for companion animal’s health [9]. As people’s desire to have their pets for a long time has increased, interest in probiotics has also attracted more attention [10]. Probiotics provide beneficial health effects to the host animal by altering the gastrointestinal (GI) flora. The GI benefits for dogs and cats include maintaining a balanced and healthy gut microbiome, preventing diarrhea, and managing small intestinal bacterial overgrowth and inflammatory bowel disorders [12]. Since dogs and cats have different dietary needs and digestive systems, their needs for and effects from probiotics differ.

Probiotics for canines

Canines are considered animal models for the study of the human microbiome because of the high structural and functional similarity between the canine and human microbiomes [13]. The study of the dog microbiome can be predictive of the human microbiome. Thus, the study of dogs offers two advantages not only directly for dogs but also for its potentially benefits for humans [14]. Although the beneficial effects of probiotics have been extensively studied in humans and animals, the exact mechanisms of probiotic-based immune modulation are not entirely clear, and the efficacy of probiotic applications varies depending on many different factors [15]. Recent reports of using probiotics in canines are shown in Tables 1 and 2.

GI disorders are one of the most common health problems in dogs [16,17]. Regardless of the
cause, most GI disorders present with acute or chronic diarrhea, or, in some cases, vomiting or anorexia [5,18,19]. Many previous studies have shown positive results regarding the treatment of dogs with different types of probiotics [20,21]. Dogs on diets supplemented with $2 \times 10^{10}$ CFU/day canine-derived probiotic Bifidobacterium animalis AHC7 had a significantly more rapid resolution of acute diarrhea than dogs that received placebo [22]. The administration of Lactobacillus rhamnosus MP01 and L. plantarum MP02, two strains isolated from canine milk, decreased the Faecalibacterium in feces [23]. Supplementing $5 \times 10^5$ CFU/day of L. murinus LbP2 in dogs improved their stool output, fecal consistency, mental status, and appetite compared to the control [24]. A total of 15 adult female dogs who were given $2.3 \times 10^8$ CFU/day canine-origin probiotic L. johnsonii CPN23 exhibited increased fiber digestibility and concentrations of short-chain fatty acids in their feces and reduced fecal ammonia concentrations compared to the control [25]. Dogs that consumed $10^7$–$10^9$ CFU/day canine-derived probiotic L. fermentum CCM 7421 displayed an increased lactic acid bacteria population, reduced Clostridia population and some gram-negative bacterial genera. Additionally, dogs that consumed probiotics showed improved total protein, cholesterol and alanine transaminase in blood samples [26]. Three milliliters of $10^8$ CFU/mL of the new potential probiotic L. fermentum AD1 significantly increased total lipids and total protein and significantly decreased the glucose concentration in the bloodstream [27]. Dogs fed $1.04 \times 10^8$ CFU/mL B. animalis B/12 showed a significantly decreased concentration of triglycerides and albumin and increased acetic, acetocetic, and valeric acid in feces [28]. Supplementing $10^8$ CFU/mL canine-origin probiotic L. johnsonii CPN23 in adult female dogs decreased their plasma glucose and cholesterol levels and increased the high-density lipoprotein and low-density lipoprotein ratio [29]. Dogs receiving Enterococcus faecium DSM 32820 had optimal fecal consistency throughout the experiment, significantly stimulated phagocytic activity and a metabolic burst activity of leukocytes and lower serum glucose concentrations [30]. Healthy dogs receiving $5 \times 10^5$ CFU/kg L. acidophilus D2/CSL showed higher body condition scores than the control dogs and there was a positive effect on their fecal consistency [31]. The probiotic feed additive contained three different bacterial strains, namely, L. casei Zhang, L. plantarum P-8, and B. animalis subsp. lactis V9 promoted the average daily feed intake, improved average daily weight gain, increased beneficial bacteria and decreased potentially harmful bacteria [14]. The probiotic E. faecium SF68 improved diarrhea symptoms compared to the control, and Giardia cysts were eliminated [32]. Adding $5 \times 10^8$ CFU/day E. faecium SF68 significantly increased the triglyceride concentration and decreased the cholesterol concentration [33].

The gut microbiome greatly affects the health and disease of the host so maintaining it in good condition is important for the health of the host [34]. Many factors influence the composition of the gut microbiome and aging is one of the greatest impacts [35]. After all, this aging which is defined as the gradual changes that occur after maturation in various organs, resulting in decreased functional capacity in the gut microbiome is thought to be somehow related to the health of the host [36]. Masuoka et al. [34] conducted the experiment with dogs of 5 different age groups (pre-weanling, weanling, young, aged and senile) and analyzed the composition of their intestinal microbiota of dogs in different age groups. As a result, the composition of the dog’s intestinal microbiota changed with age. Lactobacillus and Bifidobacterium were found to decrease as the dog aged. This experiment showed that the gut microbiome of dogs can be changed regarding the age at the level of bacterial groups and species. Further studies are needed to be done to identify whether different probiotics are needed for different phases of life.
Probiotics for felines

Cats have trillions of live bacteria in their bodies, which are mostly in their intestines [21]. Each cat’s bacterial population is different for individuals and can be changed based on diet, health status, and lifestyle choices [37,38]. During times of stress and infection, the microbiome balance can increase the number of bad bacteria, disrupting the system’s balance and potentially causing digestive problems such as decreased appetite, vomiting, diarrhea or stool changes [39,40]. Supplementing probiotics for felines can be one of the best ways to add good bacteria to the cat body [21]. Recent reports of using probiotics in felines are listed in Tables 1 and 2.

Although many studies have investigated the use of probiotics in dogs, studies in cats are relatively scarce. Few studies on probiotic usage in cats have been reported to date, and because of differences in host physiologic characteristics and the diet, the probiotic efficacy in cats cannot be extrapolated from studies in dogs [41]. The purpose of this review paper is to discuss various results about treating cats with different types of probiotics. Kittens receiving $2.85 \times 10^8$ CFU/day *E. hirae* showed high intestinal colonization and fecal shedding of live *E. hirae* during administration [42]. Supplementing $2 \times 10^8$ CFU/day *L. acidophilus* DSM13241 as a probiotic in healthy adult cats increased the numbers of beneficial *L.* and *L. acidophilus* groups in feces and decreased the numbers of *Clostridium* spp. and *E. faecalis*. It also decreased the fecal pH and plasma endotoxin concentrations and resulted in systemic and immunomodulatory changes in treated cats [41]. Kittens fed $5 \times 10^8$ CFU/day *E. faecium* SF68 showed a significantly higher percentage of CD4+ lymphocytes than controls [43]. Healthy adult cats fed $5 \times 10^7$ CFU/kg *L. acidophilus* D2/CSL had better results in terms of their fecal quality parameters and had increased *Lactobacillus* counts and decreased total coliform bacteria counts [44]. The percentage of cats with diarrhea was significantly lower in the $2.1 \times 10^9$ CFU/day *E. faecium* SF68 group than in the control group [45]. Young adult cats receiving *E. faecium* SF68 had significantly lower total diarrhea scores for days 1–11 compared to the control. Additionally, feeding *E. faecium* SF68 could lessen some associated clinical abnormalities [46]. Feeding Enterococcus faecium SF68 in cats with chronic feline herpesvirus-1 (FHV-1) infection showed fecal microbial diversity throughout the study which indicates a more stable microbiome. It also lessened the morbidity associated with chronic FHV-1 infections [47]. Healthy cats with $5 \times 10^9$ CFU from a mixture of seven bacterial species per day (Proviable®-DC) showed an increased abundance of probiotic bacteria in the feces. Probiotics also improved diarrhea after 21 days of feeding [48]. Cats with chronic gingivostomatitis that were fed $1 \times 10^6$ CFU/mL *L. plantarum* showed many positive results in gingivostomatitis symptoms. There was an improvement in the time of recurrence, and the symptoms of chronic feline gingivostomatitis disappeared after two weeks of administration. Additionally, ulceration, inflammation and oral cavity pain decreased, and halitosis disappeared [49]. Giving multistrain probiotic products to 8-month-old male cats with feline idiopathic cystitis effectively managed this disease due to the effects of bactericidal, anti-inflammatory, and immunomodulatory actions [50].

The health and disease of the host are affected by gut microbiota, maintaining the gut microbiota is getting more important as cats get aged. Masuoka et al. [51] conducted the experiment with cats of 5 different age groups (pre-weanling, weanling, young, aged and senile) and analyzed the composition of their intestinal microbiota of cats in different age groups. The results suggested that the composition of the cat’s gut microbiome changed with age, whereas the change was different from that of dogs. *Bifidobacterium* which predominated in the gut of dogs did not appear to be important in the gut of cats. Instead, *enterococi* appeared to be the main lactic acid-producing bacteria in cats. Ultimately, the results of this study indicated that the compositions of the gut microbiome between dogs and cats are different and those compositions are changing with aging. Not only are different probiotics might need for dogs and cats but also for regarding aging. Further
| Bacterial strains                          | Amount          | Source                        | Group                                 | Tested for                                      | Result                                                                 |
|------------------------------------------|-----------------|-------------------------------|---------------------------------------|------------------------------------------------|----------------------------------------------------------------------|
| *Bifidobacterium animalis* AHC7          | 2 × 10^10 CFU/day| Canine                        | Young adult dogs with acute diarrhea  | Assessment for managing acute diarrhea          | Reduced diarrhea compared to placebo group                             |
| *Lactobacillus rhamnosus* MP01           | 5 × 10^9 CFU/day | Canine                        | 1 month old puppies                  | Assessments for preventing gastrointestinal infection in puppies | Increased colonic bacterial population with some of the gram-negative bacterial genera in feces. |
| *Bifidobacterium animalis* 7421          | 2 × 10^9 CFU/day | Canine                        | Dogs with canine distemper virus (CDV)-associated diarrhea | Assessment of fecal and mental status          | Fecal consistency, mental status and appetite were significantly improved |
| *Lactobacillus johnsonii* CPN23          | 2.3 × 10^8 CFU/day| Canine                        | Adult female Labrador dogs            | Assessment of nutrient digestibility and fermentative metabolites | Increased crude fiber digestibility. Increased concentrations of SCFAs in feces. Reducing fecal ammonia concentration. |
| *Lactobacillus acidophilus* DSM13241      | 2 × 10^8 CFU/day | Feline                        | Healthy adult cats                   | Assessment for improving intestinal health in cats | Increased numbers of beneficial *Lactobacillus* and *Lactobacillus plantarum* in feces. Increased SCFAs in feces. Modulate liquid stools to normal consistency. |
| *Lactobacillus johnsonii* CPN23          | 10^8 CFU/mL (1 mL/kg BW) | Canine                        | Adult female dogs                    | Assessment of blood sample profile             | Decreased plasma glucose and cholesterol level. Decreased ALT and ALP. |
| *Enterococcus faecium* DSM                | 2 × 10^9 CFU/day | Feline                        | Kittens                              | Assessment by improving intestinal health in cats | Decreased fecal pH and plasma endotoxin concentrations resulting in systemic and immunomodulatory changes. Ameliorated the effects of atypical EPEC experimental infection. |
| *Enterococcus hirae*                     | 2.85–4.28 × 10^8 CFU/day | Feline                        | Kittens                              | Assessment by providing protective effects of *E. hirae* (EPEC) in kittens | Highly effective at promoting intestinal colonization and fecal shedding of live *E. hirae* during administration. |

SCFAs, short-chain fatty acids; ALT, alanine aminotransferase; ALP, alkaline phosphatase; BW, body weight; HDL, high-density lipoprotein; LDL, low-density lipoprotein.
Table 2. List of bacterial strains of non-canine and non-feline origin when used for their probiotic properties in canines and felines

| Bacterial strains | Amount | Source | Group | Tested for | Result | Reference |
|------------------|--------|--------|-------|-----------|--------|-----------|
| Lactobacillus casei Zhang | 2 × 10^8 CFU (2 g for young, 4 g for training, 10 g for elderly dogs) | Lactobacillus casei Zhang (koumiss) | Young, training and elderly dogs | Assessment of nutrition, immunity and composition of fecal microbiome | Promoted the average daily feed intake of elderly dogs Improved average daily weight gain on all dogs | [14] |
| Lactobacillus plantarum P-8 | 5.0 × 10^9 CFU/dg of diet | BIldobacterium animalis subsp. Lacidi V9 | Healthy dogs | Assessment of nutritional and faecal status | Higher body condition score than control group Positive effect on faecal consistency | [31] |
| Enterococcus faecium SF68 | 5 × 10^7 CFU/day | Feces of a healthy breast-fed baby | Dogs with diarrhoea | Assessment of the effect of administering metronidazole in Enterococcus faecium SF68 to treat diarrhoea | Dual therapy that administers metronidazole with Enterococcus faecium SF68 improved diarrhoea more than administering metronidazole alone | [32] |
| Enterococcus faecium SF68 | 5 × 10^7 CFU/day | Feces of a healthy breast-fed baby | Healthy dogs | Assessment of blood sample profile | Mean cholesterol concentration significantly decreased Mean triglyceride concentration significantly increased | [33] |
| Lactobacillus acidophilus D2/ CSL (CECT 4529) | 5 × 10^9 CFU/dg of food | Conventional foods such as milk, yogurt and dietary supplements | Healthy adult cats | Assessment of the effects on nutritional condition and faecal quality | Improved faecal quality parameters Increased Lactobacillus count and decreased total coliform bacteria counts | [44] |
| Enterococcus faecium SF68 | 2.1 × 10^7 CFU/day | A healthy breast-fed newborn baby | Cats | Effects on Enterococcus faecium SF68 in diarrhoea | The percentage of cats with diarrhoea was significantly lower in the probiotic group when compared with the placebo group | [45] |
| Enterococcus faecium SF68 | 1/4 can of canned food mixed with Enterococcus faecium | A healthy breast-fed newborn baby | Young adult cats | Description of the G1 abnormalities associated with the administration of amoxicillin-clavulanate to cats and an assessment of whether feeding Enterococcus faecium SF68 could ameliorate those abnormalities | The total diarrhoea scores for days 1–11 were significantly lower in the cats fed Enterococcus faecium SF68 compared to the cats fed the placebo Feeding Enterococcus faecium SF68 can lessen some associated clinical abnormalities | [46] |
| Enterococcus faecium SF68 | 5 × 10^5 CFU/day | A healthy breast-fed newborn baby | Cats with chronic Feline Herpes virus-1 (FHV-1) infection | Assessment of the effect of feeding Enterococcus faecium SF68 in clinical signs of FHV-1 infection | Fecal microbial diversity was maintained throughout the study in cats supplemented with Enterococcus faecium SF68, indicating a more stable microbiome in cats receiving Enterococcus faecium SF68 Lessened morbidity associated with chronic FHV-1 infection in some cats | [47] |
| Proviable®-DC (7 bacterial species) | 5 × 10^1 CFU of a mixture of seven bacterial species per day | Multistrain probiotic product | Adult cat | Improvement in stool character | Improved diarrhoea symptoms after 21-day feeding | [48] |
| Lactobacillus plantarum | 1 × 10^8 CFU/mL | Mare’s milk | Cats with chronic gingivostomatitis | Assessment of preventive and therapeutic oral pathology | The administration of the probiotic to the two immunosuppressed cats affected by gingivostomatitis led to an improvement in the time of recurrence The symptoms of chronic feline gingivostomatitis disappeared after two weeks of administration The ulceration, inflammation and pain of the oral cavity decreased, thrall and haemorrhage disappeared | [49] |
| Probiotic combination | Lactobacillus casei 4 × 10^7 CFU, Lactobacillus rhamnosus 3 × 10^7 CFU, Lactobacillus acidophilus 5 × 10^7 CFU, Lactobacillus bulgaricus 1 × 10^7 CFU, BIldobacterium infantis 4 × 10^7 CFU, BIldobacterium breve 5 × 10^7 CFU, Streptococcus thermolphlus 1 × 10^7 CFU | Multistrain probiotic product | 8-month-old male cats | Management of feline idiopathic cystitis (FIC) using probiotics | Probiotic combination treatment effectively managed this disease due to the effect of bactericidal, anti-inflammatory, and immunomodulatory actions | [50] |
| Proviable®-DC (7 bacterial species) | 5 × 10^5 CFU of a mixture of seven bacterial species per day | Multistrain probiotic product | Healthy cats | Assessment of a multiplicity of strains in the faecal microbiome of healthy cats | Increased abundance of probiotic bacteria in the feces of healthy cats | [55] |

IgG, Immunoglobulin G; IFN, Interferon; TNF, tumor necrosis factor.
studies are needed to use different probiotics for different phases of life.

GUT MICROBIOME FOR COMPANION ANIMALS

Gut microbiome and nutrient metabolism
Microorganisms affect the absorption of nutrients in the host and provide beneficial metabolites in return for using host nutrients [52]. Each intestine harbors a different unique microbial ecosystem due to anatomical and physiological differences [53]. Additionally, each animal harbors a different and unique microbial profile. For example, at the species and strain levels, only a few overlap between individual animals. However, the bacterial phyla, order and genera are shared by most mammals [54].

The most predominant bacterial gene category in the canine gut is carbohydrate metabolism, such as that related to mannose, oligosaccharide and raffinose metabolism. The fermentation of carbohydrates by colonic organisms such as Bacteroides, Roseburia, Ruminococcus and Lachnospiraceae results in the synthesis of short-chain fatty acids (SCFAs), such as acetate, propionate and butyrate which are sources of energy for the host [20]. SCFAs have beneficial effects on host health, including immunomodulatory effects, anti-diarrheic effects, and a regulatory effect on GI motility. In the case of felines, which are obligate carnivores, consuming raw meat increased Clostridium and Eubacterium, which are known to produce SCFAs [55].

The synthesis of vitamin K and several components of vitamin B are important functions of the intestinal microbiota [56,57]. Vitamin K, which is included in fat-soluble vitamins, plays an important role in prothrombin coagulation factor activity. Therefore, there is a risk of intestinal bleeding in cases of vitamin K deficiency [58]. Vitamin B12 (also known as cobalamin) is important for many aspects of a dog's health [59]. It is crucial for a healthy nervous system and brain function as well as for the formation and growth of blood cells [60]. Additionally, it is needed to maintain healthy digestion [61]. As a result of a metagenome analysis using dog feces, genes affecting lipoprotein lipase activity in adipocytes were identified in intestinal microbial genes, confirming that microorganisms are also related to lipid metabolism [62].

Gut microbiome and the immune system
The microbiome plays an important role in the immune system of the intestinal tract. In particular, early microbial exposure significantly affects gut microbiome formation and immune modulation, which affects susceptibility to intestinal diseases [63]. When comparing animals born through vaginal delivery with germ-free animals through cesarean section, the germ-free animals have fewer and smaller peyer’s patches, mesenteric lymph nodes and CD4+ T cells in the lamina propria of the gut wall [64]. In germ-free animals, a reduction in B cells, macrophages and neutrophils was confirmed [65]. Additionally, in germ-free animals, immunoglobulin was found at a level of 2%, which was significantly lower than that in normal healthy animals [66]. The microbiome also plays a role as a signal indicating health [65]. This characteristic is expected because animals evolved in coexistence with symbiotic microorganisms for a very long time [67]. Microorganisms that coexist with animals communicate directly and effectively with their host’s immune system through metabolites and nutrients [64,65].

Identifying diversity in the canine and feline microbiomes
The intestine is a major part of the body that influences host health. Numerous microbes form the complex microbial community in the GIT. Disrupting the gut microbiome may cause dysbiosis and lead to several diseases and disorders, such as diarrhea, allergies and obesity [21]. GI disease caused
by the dysbiosis of the gut microbial community is also generally observed in dogs and cats [68,69]. Knowing the diversity and taxonomic bacterial distribution of the gut microbiota of healthy dogs and cats is important as a baseline in future studies evaluating GI diseases in dogs and cats [70,71].

Previous studies have focused on the cultivation of intestinal content to characterize and identify the microbiota [72–74]. Most of the bacterial groups cultivated from the canine intestine belonged to *Enterobacteriaceae*, *Bacteroides*, *Clostridium*, *Lactobacillus*, and *Bifidobacterium* spp. [75,76]. However, culturing the bacteria to evaluate the complex diversity of the microbiota had limitations. Bacterial species that can be cultivated by using bacterial culture techniques are only a small portion of microbiota composition [77,78], anaerobic bacteria can be easily damaged during sample handling [79,80], the cost used for culture techniques is expensive [80], a great amount of time is used for isolation and cultivation [80,81]. A novel molecular method that uses the 16S ribosomal RNA (rRNA)-enabled the evaluation of the diversity and abundance of bacteria in the sample without culturing [82,83].

The development of next-generation sequencing technologies has helped to characterize bacterial communities and to understand interactions between hosts and bacteria. Using next-generation sequencing, dog and cat organ microbiotas have been described. These microbiota include those of the GIT [70, 84–86], skin [87], oral cavity [88,89], nasal cavity [90], and vagina [91].

### Composition of canine and feline microbiome

All animals, including dogs and cats, harbor numerous microorganisms in the GIT [8]. Dogs and cats have different microbiota compositions and they also differ in the same species [21]. There are lots of factors that can affect microbiota compositions such as age [92–94], breed [8,95,96], diet composition [39,92,94], disease [92,93], environment [92,96,97], food type [93,98] and sex [99,100].

The gut microbiome which is highly related to a healthy life could be affected by dogs’ breed. There was a relationship between GI conditions and dog breeds [101,102]. According to You and Kim’s experiment [103], there was a difference in microbial composition in Poodle and Maltese groups. Also, phylum Fusobacterium was differed by breeds (Maltese, Poodle, and Miniature Schnauzer). From these data, they suggested that there might be differences in the gut microbiome composition depending on the dog breeds. According to Lehtimäki et al.’s experiment [104], living conditions have a significant impact on the skin microbiome in humans and dogs, but not the gut microbiome. Dogs living in rural and urban environments participated in the study. The skin microbiome was more diverse among individuals in rural areas compared to urban areas. This study showed that the living environment had a much greater effect on the skin microbiome than the guts of dogs. Experiments on changes in the gut microbiome of cats according to breeds and living environments have been limited. Further research is needed as with dogs. According to Older et al.’s [96] experiment, breed and living environment played an important role in shaping the cat skin microbiome. In particular, it seems that the hair coat and grooming according to the cat breeds have a great influence on the microbiome of the cat’s skin microbiome.

The bacterial count in the stomach is between $10^4$ and $10^5$ CFU/g [105]. In the duodenum and jejunum, the bacterial counts are generally low ($10^5$ CFU/g) but can reach $10^9$ CFU/mL in some dogs and cats [106]. The ileum contains an increasing number of diverse microbiota, mostly at $10^7$ CFU/mL. The bacterial counts in the colon are between $10^9$ and $10^{11}$ CFU/g [38,73].

The healthy canine stomach has a comparably low number of total bacteria. Most belonged to Proteobacteria (99.6%), and few belonged to Firmicutes (0.3%). The dominant species are *Helicobacter* and *Lactobacillus* spp. [85]. Using 16S rRNA sequences, four phyla (Firmicutes, Fusobacteria, Bacteroidetes and Proteobacteria) predominated in the small intestine [70]. The
duodenum of healthy canines consisted of six phyla. Firmicutes predominated followed by Proteobacteria, Bacteroidetes, Spirochaetes, Fusobacteria and Actinobacteria [37,107]. Healthy dog microbiota in the jejunum were evaluated, and the most predominant phylum was Proteobacteria (46%), followed by Firmicutes (15%), Actinobacteria (11.2%), Spirochaetes (14.2%), Bacteroidetes (6.2%) and Fusobacteria (5.4%) [84]. The ileum microbiota of healthy dogs predominantly consists of Fusobacteria, Firmicutes and Bacteroidetes [70].

Lactobacillales was present in all parts of the intestines (22% in the duodenum and 10% in the jejunum). Enterobacteriales were more frequently detected in the small intestine than in the colon. Clostridiales were highly abundant in the duodenum (40%), jejunum (39%), ileum (25%) and colon (26%) [70]. Facultative anaerobic Lactobacillus strains predominated in the jejunal microbiota, and L. acidophilus was the most abundant among them [108]. In the jejunal samples, facultative anaerobic and anaerobic bacteria were similarly detected, while anaerobic bacteria predominated in the fecal samples. The number of bacteria in the jejunal microbiota was 10^8 to 10^11 CFU/g, while the number in the feces was 10^8 to 10^11 CFU/g. Despite the lower number in the small intestine, some microbial groups were more prevalent in the small intestine than in feces: staphylococci, 64% versus 36%; non fermentative gram-negative rods, 27% versus 9%; and yeasts, 27% versus 5% [73].

Firmicutes, Bacteroidetes, Fusobacteria, Proteobacteria and Actinobacteria were the most abundant phyla in the fecal microbiota of healthy dogs [70, 86, 109]. However, Fusobacteria (39.17%) were dominant, followed by Bacteroidetes (33.36%) and Firmicutes (15.81%) in healthy adult Miniature Schnauzer dogs [86], while the abundances of Fusobacteria, Bacteroidetes and Firmicutes were similar (approximately 30% each) in six Hound dogs [109].

Clostridia was the most predominant bacterial class in the dog fecal microbiota [109]. At the genus level, Lactobacillus was the most predominant, followed by Bifidobacterium, Enterococcus, Streptococcus and Pediococcus, in the dog fecal microbiota [110]. Many Lactobacillus spp. including L. casei, L. salivarius, L. rhamnosus, L. mucosae, L. fermentum, L. reuteri, L. animalis, L. acidophilus and L. johnsonii were the most frequently isolated ones from the feces. L. reuteri, L. animalis, and L. johnsonii were the most predominant species in dogs [110–112]. Weissella confusa, Pediococcus acidilactici, Enterococcus spp. and B. animalis ssp. lactis were also frequently isolated from dog feces [111–113]. At the fungal kingdom-phylum level, Ascomycota, Basidiomycota, Glomeromycota, and Zygomycota were detected in dog feces [109].

In the skin microbiota, the most predominant phyla and families were Proteobacteria and Oxalobacteriaceae [87]. At the oral microbiota phylum level, Bacteroidetes (60%) was the most predominant, followed by Proteobacteria (20.8%), Firmicutes (11.4%), Fusobacteria (4.7%) and Spirochaetes (1.7%). At the genus level, the oral microbiota consisted of Porphyromonas (39.2%), Fusobacterium (4.5%), Capnoclostridium (3.8%), Derxia (3.7%), Moraxella (3.3%) and Bacteroides (2.7%) [88]. In the nasal microbiota of healthy dogs, Moraxella spp. was the most abundant species, followed by Phyllobacterium spp., Staphylococcus spp., and Cardiobacteriaceae [90]. The most frequently isolated bacteria from the dog's vaginal tract were Lactobacillus, Escherichia coli and Staphylococcus pseudointermedius [21, 91].

The feline GIT has different bacterial species than other animals. Helicobacter is known to reside in the stomachs of cats [114]. For the microbiota composition of the GI (stomach, duodenum, jejunum, ileum, and colon) contents, which were collected from 5 healthy felines, Firmicutes (68%) predominated, followed by Proteobacteria (14%), Bacteroidetes (10%), Fusobacteria (5%) and Actinobacteria (4%). At the order level, Clostridiales (54%) prevailed, followed by Lactobacillales, Bacteroidales, Campylobacteriales, and Fusobacteriales [37]. Based on several studies, it is known that Bacteroides spp., Clostridium spp., Enterococcus spp., Streptococcus spp., Fusobacteria spp., and Eubacteria spp. are present in the small intestines of felines [69,115]. Representative lactic acid
Probiotics and the gut microbiome in companion animals

Bacteria present in the GIT of felines include *L. acidophilus*, *L. salivarius*, *L. johnsonii*, *L. reuteri* and *L. sakei*, which are typical intestinal lactic acid bacteria found in animals, including humans, although the amount varies by individual [21,37]. The major phyla in the feline fecal microbiota were Firmicutes, Bacteroidetes, Actinobacteria and Proteobacteria [69,109,116]. These four phyla make up more than 99% of the fecal microbiota [69]. Handl et al. [109] reported that Firmicutes was the most prevalent phylum in the fecal microbiota, followed by Bacteroidetes and Actinobacteria; however, Tun et al. [116] reported that Bacteroidetes was the most predominant phylum, followed by Firmicutes and Proteobacteria. *Bacteroides, Fusobacterium*, and *Prevotella* were the most predominant genera in the feline fecal microbiota, which indicated that these genera play a major role in the feline intestine [117]. Fungi, archaea and viruses compose a minor part of intestinal microbial communities. Ascomycota was the only phylum of fungi detected in cats [109,116].

*Malassezia* spp. were the most prevalent fungi in feline skin mycobiota. *M. restricta* and *M. globosa* were the most predominant fungal species in all cat breeds [96]. *M. pachydermatis* is known as a yeast that is present in the skin microbiome, yet it can also act as a pathogen that can cause dermatitis [118]. The phylum level of the oral microbiota is generally conserved between cats. These phyla are predominated by Proteobacteria (75.2%), followed by Bacteroidetes (9.3%), Firmicutes (6.7%), SR1 (2.7%), Spirochaetes (1.8%), Fusobacteria (1.3%), and Actinobacteria (0.6%) [89]. The composition of the canine and feline microbiota is shown in Figs. 1 and 2.

**Fig. 1.** The dynamic community of nasal, oral and gut microbiota in canines ([21], [37], [38], [70], [73], [86]–[88], [90], [91], [107]–[113]).
SAFETY ISSUES OF PROBIOTICS AND THE GUT MICROBIOME IN COMPANION ANIMALS

The Food and Agriculture Organization of the United Nations and World Health Organization (FAO/WHO) defined probiotics as “live microorganisms, which when administered in adequate amounts, confer a health benefit on the host” [9]. Tremendous scientific evidence for the efficacy of probiotic candidates has been available for decades, but insufficient information about their safety is available. While known to be safe in general, a few adverse effects associated with probiotics use have been documented in patients [119,120]. Moreover, there is a lack of information on the inherent characteristics of each probiotic strain that may be associated with health risks [121].

The European Food Safety Authority (EFSA) recommended the qualified presumption of safety (QPS) status for microorganisms used in feed and food production in 2003. Based on the QPS guidelines, microbes that produce toxins or possess virulence factors that may contribute to their pathogenicity cannot be used as probiotics. In addition, it must be ensured that there are no acquired genes encoding antimicrobial resistance (AMR). The existence of knowledge, including a history of use, ecology, industrial application, clinical reports, and a public database, is considered important evidence for evaluating the safety of microbial species [122]. QPS list includes several taxonomic units for bacteria, yeasts, and viruses [123], of which *Lactobacillus* and *Bifidobacterium* are representative because their reasonable certainty of no harm has been supported by an extensive record of safe use [124].

With the recent focus on the beneficial effects of probiotics in companion animals and their relationship to gut microflora and health, probiotic products are being increasingly marketed in the form of feed additives, dietary supplements, and probiotic-containing foods [125]. Although there have been no reports of adverse events when probiotics are administered to small animals, safety concerns remain to be addressed [126]. The microorganisms used in feed additives require safety verification for target animals, manufacturers, and owners/consumers. In particular, *Enterococcus*, known as canine and feline intestinal commensal bacteria, have been used as probiotics for small animals, but their use is restricted in some countries because of the risk of host infection by AMR gene transfer [125,127]. Rinkinen [128] demonstrated that some *Enterococcus faecium* strains...
promoted the adherence of the zoonotic pathogen *Campylobacter jejuni* in the intestines of canines. Notably, the administration of *E. faecium* strain SF68 (deposited as strain NCIMB 10415), which originated from infant feces in 1968, reduced the occurrence of diarrhea in dogs and cats housed in animal shelters [45], and it has been verified that the strain may not cause any safety concerns for companion animals and their owners [129–131]. Due to these conflicting outcomes, stringent safety evaluations are required in a strain-specific manner with regard to probiotic use.

Host specificity is considered an important criterion for selecting probiotic candidates, primarily due to differences in physiological structure, immune systems, and microbial composition [132,133]. However, most commercial probiotics used as feed additives originate from humans and are verified by human-based methods and criteria [134]. The clinical results from Weese and Anderson [135] showed that *Lactobacillus rhamnosus* GG, a commercial probiotic strain isolated from a healthy human GIT, may not be suitable for use in canines because of its short persistence. In addition, in an in vitro test, probiotic strains of canine origin inhibited the adhesion of enterotoxigenic *Clostridium perfringens* to canine jejunal chyme more efficiently than non-canine strains [128]. Recent studies have focused on strains isolated from the intestines of healthy dogs and cats to demonstrate their impact on pathogen inhibition, attenuation of inflammatory status, and modulation of the gut microbiome [136,137]. Host specificity has been discussed with respect to probiotic efficacy in most publications but must also be addressed from a safety perspective. Furthermore, clinical outcomes for the safe use of probiotics in target animal species should be documented. Given the host specificity and broad diversity of potential probiotic candidates for small animals, the availability of novel species with no record of use requires attention [133]. For a complete characterization, documentation of genetic and biochemical properties and long-term clinical trials are needed. Additional efforts are required to standardize safety assessment methods for novel species to be considered QPS or as having a generally recognized as safe status based on the opinions of regulatory bodies and expert panels [138].

Quality control issues associated with probiotics relate to products intended for animals as well as humans [21]. The global market for probiotics for companion animals is growing, but insufficient quality control regulations create serious problems for the safety of consumers and target animals. Some investigators have disclosed that many commercial animal probiotics or pet foods that claim to contain probiotics did not contain microbial species listed on the label or even contain other species. Moreover, bacterial viability, a key concept to stipulate probiotics, was inconsistent with labeled values expressed in colony-forming units [139–141]. The current low level of quality control may lead to exposure to unknown health risks not only for the animals but also for the owners and the environment. Recent advances in meta-omics technologies (metagenomics, metatranscriptomics, metaproteomics, and metabolomics) have promoted a correct evaluation of the quality of probiotic products. For the multistrain product VSL#3, Mora and colleagues [142] successfully identified microbial taxa with metagenomics and viability by flow cytometry and confirmed reproducibility using metaproteomics. Metagenomic approaches have also enabled the analysis of genes related to safety concerns in a culture-independent manner. Stringent oversight by regulatory bodies, high manufacturer awareness, and the development of rigorous evaluation methods by researchers are needed for the safe selection and production of probiotic candidates intended for both companion animal and human use.

**CONCLUSION**

Although not enough research has been conducted on the probiotics and gut microbiome thus far, because the number of companion animal people and the companion animal market are
growing, research related to the companion animal microbiome is also growing. Recently, in-depth research has been performed to identify the functionality of probiotics and the gut microbiome of companion animals and to make them with various materials, ranging from feed, snacks, supplies, and treatments for the diseases of companion animals. The current evidence suggests that specific probiotic strains and/or their defined combinations may be useful in canine and feline nutrition, therapy and care. However, probiotics and the gut microbiota used in the present study are of human origin; thus, the companion animal-specific health benefits are not unclear. Therefore, the most important step is to secure pet-originated microorganisms for their health claims. Moreover, detailed in vivo designs and trials using companion animals are needed to identify and characterize newly isolated pet-originated microbiomes with an impact on health maintenance in both dogs and cats. Corroboration of these health-promoting effects and microbiological safety issues should be assessed regarding potential probiotics and the gut microbiome for animal health and welfare.

REFERENCES

1. Krebs JR, Sjolander S. Konrad zacharias lorenz, 7 November 1903 - 27 February 1989. Biogr Mem Fell R Soc 1992; 38:209-28.
2. Hwang EK, Sohn KP. Companion animal in Korea report. Seoul: KB financial group; 2021.
3. Mosteller J. Animal-companion extremes and underlying consumer themes. J Bus Res. 2008;61:512-21. https://doi.org/10.1016/j.jbusres.2007.07.004
4. Do S, Phungviwatnikul T, de Godoy MRC, Swanson KS. Nutrient digestibility and fecal characteristics, microbiota, and metabolites in dogs fed human-grade foods. J Anim Sci. 2021;99:skab028. https://doi.org/10.1093/jas/skab028
5. Redfern A, Suchodolski J, Jergens A. Role of the gastrointestinal microbiota in small animal health and disease. Vet Rec. 2017;181:370. https://doi.org/10.1136/vr.103826
6. MacDonald ML, Rogers QR, Morris JG. Nutrition of the domestic cat, a mammalian carnivore. Annu Rev Nutr. 1984;4:521-62. https://doi.org/10.1146/annurev.nu.04.070184.002513
7. Clauss M, Kleffner H, Kienzle E. Carnivorous mammals: nutrient digestibility and energy evaluation. Zoo Biol. 2010;29:687-704. https://doi.org/10.1002/zoo.20302
8. Deng P, Swanson KS. Gut microbiota of humans, dogs and cats: current knowledge and future opportunities and challenges. Br J Nutr. 2015;113:S6-17. https://doi.org/10.1017/S0007114514002943
9. Morelli L, Capurso L. FAO/WHO guidelines on probiotics: 10 years later. J Clin Gastroenterol. 2012;46:S1-2. https://doi.org/10.1097/MCG.0b013e318269fdD5
10. Hill C, Guarner F, Reid G, Gibson GR, Merenstein DJ, Pot B, et al. The International Scientific Association for Probiotics and Prebiotics consensus statement on the scope and appropriate use of the term probiotic. Nat Rev Gastroenterol Hepatol. 2014;11:506-14. https://doi.org/10.1038/nrgastro.2014.66
11. Sarowska J, Choroszy-Król I, Regulska-Ilow B, Frej-Mađrak M, Jama-Kmieciec A. The therapeutic effect of probiotic bacteria on gastrointestinal diseases. Adv Clin Exp Med. 2013;22:759-66.
12. Case LP, Daristotle L, Hayek MG, Raasch MF. Canine and feline nutrition: a resource for companion animal professionals. 3rd ed. London: Elsevier Health Sciences; 2010.
13. Coelho LP, Kultima JR, Costea PI, Fournier C, Pan Y, Czarnecki-Maulden G, et al. Similarity of the dog and human gut microbiomes in gene content and response to diet. Microbiome. 2018;6:72. https://doi.org/10.1186/s40168-018-0450-3
14. Xu H, Huang W, Hou Q, Kwok LY, Laga W, Wang Y, et al. Oral administration of
compound probiotics improved canine feed intake, weight gain, immunity and intestinal microbiota. Front Immunol. 2019;10:666. https://doi.org/10.3389/fimmu.2019.00666

15. Culligan EP, Hill C, Sleator RD. Probiotics and gastrointestinal disease: successes, problems and future prospects. Gut Pathog. 2009;1:19. https://doi.org/10.1186/1757-4749-1-19

16. Suchodolski JS. Companion animals symposium: microbes and gastrointestinal health of dogs and cats. J Anim Sci. 2011;89:1520-30. https://doi.org/10.2527/jas.2010-3377

17. Mondo E, Marliani G, Accorsi PA, Cocchi M, Di Leone A. Role of gut microbiota in dog and cat's health and diseases. Open Vet J. 2019;9:253-8. https://doi.org/10.4314/ovj.v9i3.10

18. Unterer S, Busch K. Acute hemorrhagic diarrhea syndrome in dogs. Vet Clin Small Anim Pract. 2021;51:79-92. https://doi.org/10.1016/j.cvsm.2020.09.007

19. Jergens AE, Simpson KW. Inflammatory bowel disease in veterinary medicine. Front Biosci (Elite Ed). 2012;4:1404-19. https://doi.org/10.2741/e470

20. Huang Z, Pan Z, Yang R, Bi Y, Xiong X. The canine gastrointestinal microbiota: early studies and research frontiers. Gut Microbes. 2020;11:635-54. https://doi.org/10.1080/19490976.2019.1704142

21. Grześkowiak Ł, Endo A, Beasley S, Salminen S. Microbiota and probiotics in canine and feline welfare. Anaerobe. 2015;34:14-23. https://doi.org/10.1016/j.anabio.2015.04.002

22. Kelley RL, Minikhiem D, Kiely B, O'Mahony L, O'Sullivan D, Boileau T, et al. Clinical benefits of probiotic canine-derived Bifidobacterium animalis strain AHC7 in dogs with acute idiopathic diarrhea. Vet Ther. 2009;10:121-30.

23. Fernández L, Martínez R, Pérez M, Arroyo R, Rodríguez JM. Characterization of Lactobacillus rhamnosus MP01 and Lactobacillus plantarum MP02 and assessment of their potential for the prevention of gastrointestinal infections in an experimental canine model. Front Microbiol. 2019;10:1117. https://doi.org/10.3389/fmicb.2019.01117

24. Delucchi L, Fraga M, Zunino P. Effect of the probiotic Lactobacillus murinus LbP2 on clinical parameters of dogs with distemper-associated diarrhea. Can J Vet Res. 2017;81:118-21.

25. Kumar S, Pattanaik AK, Sharma S, Gupta R, Jadhav SE, Dutta N. Comparative assessment of canine-origin Lactobacillus johnsonii CPN23 and dairy-origin Lactobacillus acidophilus NCDC 15 for nutrient digestibility, faecal fermentative metabolites and selected gut health indices in dogs. J Nutr Sci. 2017;6:e38. https://doi.org/10.1017/jns.2017.35

26. Strompfová V, Kubašová I, Lauková A. Health benefits observed after probiotic Lactobacillus fermentum CCM 7421 application in dogs. Appl Microbiol Biotechnol. 2017;101:6309-19. https://doi.org/10.1007/s00253-017-8425-z

27. Strompfová V, Marcínáková M, Simonová M, Bogovič-Matijašić B, Lauková A. Application of potential probiotic Lactobacillus fermentum AD1 strain in healthy dogs. Anaerobe. 2006;12:75-9. https://doi.org/10.1016/j.anabio.2005.12.001

28. Strompfová V, Pogány Simonová M, Gancarčíková S, Mudroňová D, Farbáková J, Maďari A, et al. Effect of Bifidobacterium animalis B/12 administration in healthy dogs. Anaerobe. 2014;28:37-43. https://doi.org/10.1016/j.anabio.2014.05.001

29. Kumar S, Pattanaik AK, Sharma S, Jadhav SE. Species-specific probiotic Lactobacillus johnsonii CPN23 supplementation modulates blood biochemical profile and erythrocytic antioxidant indices in Labrador dogs. Indian J Anim Sci. 2016;86:918-24.

30. Strompfová V, Kubašová I, Ščerbová J, Maďari A, Gancarčíková S, Mudroňová D, et al. Oral administration of bacteriocin-producing and non-producing strains of Enterococcus faecium in dogs. Appl Microbiol Biotechnol. 2019;103:4953-65. https://doi.org/10.1007/s00253-019-09847-3
31. Marelli SP, Fusi E, Giardini A, Martino PA, Polli M, Bruni N, et al. Effects of probiotic Lactobacillus acidophilus D2/CSL (CECT 4529) on the nutritional and health status of boxer dogs. Vet Rec. 2020;187:e28. https://doi.org/10.1136/vr.105434

32. Fenimore A, Martin L, Lappin MR. Evaluation of metronidazole with and without Enterococcus faecium SF68 in shelter dogs with diarrhea. Top Companion Anim Med. 2017;32:100-3. https://doi.org/10.1053/j.tcam.2017.11.001

33. Lucena R, Novales M, Blanco B, Hernández E, Ginel PJ. Effect of probiotic Enterococcus faecium SF68 on liver function in healthy dogs. J Vet Intern Med. 2019;33:2628-34. https://doi.org/10.1111/jvim.15625

34. Masuoka H, Shimada K, Kiyosue-Yasuda T, Kiyosue M, Oishi Y, Kimura S, et al. Transition of the intestinal microbiota of dogs with age. Biosci Microbiota Food Health. 2017;36:27-31. https://doi.org/10.12938/bmfh.BMFH-2016-021

35. Mitsuoka T. Establishment of intestinal bacteriology. Biosci Microbiota Food Health. 2014;33:99-116. https://doi.org/10.12938/bmfh.33.99

36. Armstrong PJ. Changes in body composition and energy balance with aging. Vet Clin Nutr. 1996;3:83-7.

37. Ritchie LE, Steiner JM, Suchodolski JS. Assessment of microbial diversity along the feline intestinal tract using 16S rRNA gene analysis. FEMS Microbiol Ecol. 2008;66:590-8. https://doi.org/10.1111/j.1574-6941.2008.00609.x

38. Suchodolski JS. Intestinal microbiota of dogs and cats: a bigger world than we thought. Vet Clin North Am Small Anim Pract. 2011;41:261-72. https://doi.org/10.1016/j.cvsm.2010.12.006

39. Honneffer JB, Minamoto Y, Suchodolski JS. Microbiota alterations in acute and chronic gastrointestinal inflammation of cats and dogs. World J Gastroenterol. 2014;20:16489-97. https://doi.org/10.3748/wjg.v20.i44.16489

40. Willard MD. Feline inflammatory bowel disease: a review. J Feline Med Surg. 1999;1:155-64. https://doi.org/10.1016/S1098-612X(99)90204-8

41. Marshall-Jones ZV, Baillon MLA, Croft JM, Butterwick RF. Effects of Lactobacillus acidophilus DSM13241 as a probiotic in healthy adult cats. Am J Vet Res. 2006;67:1005-12. https://doi.org/10.2460/ajvr.67.6.1005

42. Watson VE, Jacob ME, Bruno-Bárcaena JM, Amirsultan S, Stauffer SH, Piquéras VO, et al. Influence of the intestinal microbiota on disease susceptibility in kittens with experimentally-induced carriage of atypical enteropathogenic Escherichia coli. Vet Microbiol. 2019;231:197-206. https://doi.org/10.1016/j.vetmic.2019.03.020

43. Veir JK, Knorr R, Cavadini C, Sherrill SJ, Benyacoub J, Satyaraj E, et al. Effect of supplementation with Enterococcus faecium (SF68) on immune functions in cats. Vet Ther Res Appl Vet Med. 2007;8:229-38.

44. Fusi E, Rizzi R, Polli M, Cannas S, Giardini A, Bruni N, et al. Effects of Lactobacillus acidophilus D2/CSL (CECT 4529) supplementation on healthy cat performance. Vet Rec Open. 2019;5:e000368. https://doi.org/10.1136/vetreco-2019-000368

45. Bybee SN, Scorza AV, Lappin MR. Effect of the probiotic Enterococcus faecium SF68 on presence of diarrhea in cats and dogs housed in an animal shelter. J Vet Intern Med. 2011;25:856-60. https://doi.org/10.1111/j.1939-1676.2011.0738.x

46. Torres-Henderson C, Summers S, Suchodolski J, Lappin MR. Effect of Enterococcus faecium strain SF68 on gastrointestinal signs and fecal microbiome in cats administered amoxicillin-clavulanate. Top Companion Anim Med. 2017;32:104-8. https://doi.org/10.1053/j.tcam.2017.11.002
47. Lappin MR, Veir JK, Satyaraj E, Czarnecki-Maulden G. Pilot study to evaluate the effect of oral supplementation of Enterococcus faecium SF68 on cats with latent feline herpesvirus 1. J Feline Med Surg. 2009;11:650-4. https://doi.org/10.1016/j.jfms.2008.12.006

48. Hart ML, Suchodolski JS, Steiner JM, Webb CB. Open-label trial of a multi-strain synbiotic in cats with chronic diarrhea. J Feline Med Surg. 2012;14:240-5. https://doi.org/10.1177/1098612X11434386

49. Segovia BM, Torras M, Díaz JM. Communication of the results of the treatment with probiotics in two cats with chronic gingivostomatitis. Open J Vet Med. 2018;8:9-14. https://doi.org/10.4236/ojvm.2018.82002

50. Sofyan MS, Rosman N, K시스n B, Kamaludeen JB, Dadi TB, Pertwii H. Management of feline idiopathic cystitis (FIC) using probiotic combination treatment. Indian Vet J. 2019;96:20-2.

51. Masuoka H, Shimada K, Kiyosue-Yasuda T, Kiyosue M, Oishi Y, Kimura S, et al. Transition of the intestinal microbiota of cats with age. PLOS ONE. 2017;12:e0181739. https://doi.org/10.1371/journal.pone.0181739

52. Suchodolski JS, Ruaux CG, Steiner JM, Fetz K, Williams DA. Application of molecular fingerprinting for qualitative assessment of small-intestinal bacterial diversity in dogs. J Clin Microbiol. 2004;42:4702-8. https://doi.org/10.1128/JCM.42.10.4702-4.708.2004

53. Suchodolski JS, Ruaux CG, Steiner JM, Fetz K, Williams DA. Assessment of the qualitative variation in bacterial microflora among compartments of the intestinal tract of dogs by use of a molecular fingerprinting technique. Am J Vet Res. 2005;66:1556-62. https://doi.org/10.2460/ajvr.2005.66.1556

54. Ritchie LE, Burke KE, Garcia-Mazcorro JF, Steiner JM, Suchodolski JS. Characterization of fecal microbiota in cats using universal 16S rRNA gene and group-specific primers for Lactobacillus and Bifidobacterium spp. Vet Microbiol. 2010;144:140-6. https://doi.org/10.1016/j.vetmic.2009.12.045

55. Pilla R, Suchodolski JS. The gut microbiome of dogs and cats, and the influence of diet. Vet Clin North Am Small Anim Pract. 2021;51:605-21. https://doi.org/10.1016/j.cvsm.2021.01.002

56. Altveş S, Yildiz HK, Vural HC. Interaction of the microbiota with the human body in health and diseases. Biosci Microbiota Food Health. 2020;39:23-32.

57. Peterson CT, Rodionov DA, Osterman AL, Peterson SN. B vitamins and their role in immune regulation and cancer. Nutrients. 2020;12:3380. https://doi.org/10.3390/nu12113380

58. Vermeer CV. Vitamin K: the effect on health beyond coagulation – an overview. Food Nutr Res. 2012;56:5329. https://doi.org/10.3402/fnr.v56i0.5329

59. Kather S, Grützner N, Kook PH, Dengler F, Heilmann RM. Review of cobalamin status and disorders of cobalamin metabolism in dogs. J Vet Intern Med. 2020;34:13-28. https://doi.org/10.1111/jvim.15638

60. Weiss DJ, Wardrop KJ. Schalm's veterinary hematology. New York, NY: John Wiley & Sons; 2011.

61. Xu H, Zhao F, Hou Q, Huang W, Liu Y, Zhang H, et al. Metagenomic analysis revealed beneficial effects of probiotics in improving the composition and function of the gut microbiota in dogs with diarrhoea. Food Funct. 2019;10:2618-29. https://doi.org/10.1039/C9FO00087A

62. Swanson KS, Dowd SE, Suchodolski JS, Middelbos IS, Vester BM, Barry KA, et al. Phylogenetic and gene-centric metagenomics of the canine intestinal microbiome reveals
similarities with humans and mice. ISME J. 2011;5:639-49. https://doi.org/10.1038/ismej.2010.162

63. Nash MJ, Frank DN, Friedman JE. Early microbes modify immune system development and metabolic homeostasis—the “restaurant” hypothesis revisited. Front Endocrinol. 2017;8:349. https://doi.org/10.3389/fendo.2017.00349

64. Kamada N, Núñez G. Role of the gut microbiota in the development and function of lymphoid cells. J Immunol. 2013;190:1389-95. https://doi.org/10.4049/jimmunol.1203100

65. Tomkovich S, Jobin C. Microbiota and host immune responses: a love–hate relationship. Immunology. 2016;147:1-10. https://doi.org/10.1111/imn.12538

66. Tizard IR, Jones SW. The microbiota regulates immunity and immunologic diseases in dogs and cats. Vet Clin North Am Small Anim Pract. 2018;48:307-22. https://doi.org/10.1016/j.cvsm.2017.10.008

67. Zilber-Rosenberg I, Rosenberg E. Role of microorganisms in the evolution of animals and plants: the hologenome theory of evolution. FEMS Microbiol Rev. 2008;32:723-35. https://doi.org/10.1111/j.1574-6976.2008.00123.x

68. Qin X. What is human inflammatory bowel disease (IBD) more like: Johne’s disease in cattle or IBD in dogs and cats? Inflamm Bowel Dis. 2008;14:138. https://doi.org/10.1002/ibd.20240

69. Minamoto Y, Hooda S, Swanson KS, Suchodolski JS. Feline gastrointestinal microbiota. Anim Health Res Rev. 2012;13:64-77. https://doi.org/10.1017/S1466252312000060

70. Suchodolski JS, Camacho J, Steiner JM. Analysis of bacterial diversity in the canine duodenum, jejunum, ileum, and colon by comparative 16S rRNA gene analysis. FEMS Microbiol Ecol. 2008;66:567-78. https://doi.org/10.1111/j.1574-6941.2008.00521.x

71. Inness VL, McCartney AL, Khoo C, Gross KL, Gibson GR. Molecular characterisation of the gut microflora of healthy and inflammatory bowel disease cats using fluorescence in situ hybridisation with special reference to Desulfovibrio spp. J Anim Physiol Anim Nutr. 2007;91:48-53. https://doi.org/10.1111/j.1439-0396.2006.00640.x

72. Werner M, Suchodolski JS, Lidbury JA, Steiner JM, Hartmann K, Unterer S. Diagnostic value of fecal cultures in dogs with chronic diarrhea. J Vet Intern Med. 2021;35:199-208. https://doi.org/10.1111/jvim.15982

73. Mentula S, Harnoinen J, Heikkilä M, Westermarck E, Rautio M, Huovinen P, et al. Comparison between cultured small-intestinal and fecal microfloras in beagle dogs. Appl Environ Microbiol. 2005;71:4169-75. https://doi.org/10.1128/AEM.71.8.4169-4175.2005

74. German AJ, Day MJ, Ruaux CG, Steiner JM, Williams DA, Hall EJ. Comparison of direct and indirect tests for small intestinal bacterial overgrowth and antibiotic-responsive diarrhea in dogs. J Vet Intern Med. 2003;17:33-43. https://doi.org/10.1111/j.1939-1676.2003.tb01321.x

75. Benno Y, Nakao H, Uchida K, Mitsuoka T. Impact of the advances in age on the gastrointestinal microflora of beagle dogs. J Vet Med Sci. 1992;54:703-6. https://doi.org/10.1292/jvms.54.703

76. Buddington RK. Postnatal changes in bacterial populations in the gastrointestinal tract of dogs. Am J Vet Res. 2003;64:646-51. https://doi.org/10.2460/ajvr.2003.64.646

77. Greetham HL, Giffard C, Hutson RA, Collins MD, Gibson GR. Bacteriology of the Labrador dog gut: a cultural and genotypic approach. J Appl Microbiol. 2002;93:640-6. https://doi.org/10.1046/j.1365-2672.2002.01724.x

78. Leser TD, Amenuvor JZ, Jensen TK, Lindecrora RH, Boye M, Møller K. Culture-independent analysis of gut bacteria: the pig gastrointestinal tract microbiota revisited. Appl
Probiotics and the gut microbiome in companion animals

Environ Microbiol. 2002;68:673-90. https://doi.org/10.1128/AEM.68.2.673-690.2002

79. Holland KT. Anaerobic bacteria. Cham: Springer Science & Business Media; 2013.

80. Lagier JC, Dubourg G, Million M, Cadoret F, Bilen M, Fenollar F, et al. Culturing the human microbiota and culturomics. Nat Rev Microbiol. 2018;16:540-50. https://doi.org/10.1038/s41579-018-0041-0

81. Pereira AC, Cunha MV. An effective culturomics approach to study the gut microbiota of mammals. Res Microbiol. 2020;171:290-300. https://doi.org/10.1016/j.resmic.2020.09.001

82. Hayashi H, Sakamoto M, Benno Y. Phylogenetic analysis of the human gut microbiota using 16S rDNA clone libraries and strictly anaerobic culture-based methods. Microbiol Immunol. 2002;46:535-48. https://doi.org/10.1111/j.1348-0421.2002.tb02731.x

83. Lan PTN, Hayashi H, Sakamoto M, Benno Y. Phylogenetic analysis of cecal microbiota in chicken by the use of 16S rDNA clone libraries. Microbiol Immunol. 2002;46:371-82. https://doi.org/10.1111/j.1348-0421.2002.tb02709.x

84. Suchodolski JS, Dowd SE, Westermark E, Steiner JM, Wolcott RD, Spillmann T, et al. The effect of the macrolide antibiotic tylosin on microbial diversity in the canine small intestine as demonstrated by massive parallel 16S rRNA gene sequencing. BMC Microbiol. 2009;9:210. https://doi.org/10.1186/1471-2180-9-210

85. Garcia-Mazcorro JF, Lanerie DJ, Dowd SE, Paddock CG, Grützner N, Steiner JM, et al. Effect of a multi-species symbiotic formulation on fecal bacterial microbiota of healthy cats and dogs as evaluated by pyrosequencing. FEMS Microbiol Ecol. 2011;78:542-54. https://doi.org/10.1111/j.1574-6941.2011.01185.x

86. Hand D, Wallis C, Colyer A, Penn CW. Pyrosequencing the canine faecal microbiota: breadth and depth of biodiversity. PLOS ONE. 2013;8:e53115. https://doi.org/10.1371/journal.pone.0053115

87. Rodrigues Hoffmann A, Patterson AP, Diesel A, Lawhon SD, Ly HJ, Elkins Stephenson C, et al. The skin microbiome in healthy and allergic dogs. PLOS ONE. 2014;9:e83197. https://doi.org/10.1371/journal.pone.0083197

88. Sturgeon A, Stull JW, Costa MC, Weese JS. Metagenomic analysis of the canine oral cavity as revealed by high-throughput pyrosequencing of the 16S rRNA gene. Vet Microbiol. 2013;162:891-8. https://doi.org/10.1016/j.jvetmic.2012.11.018

89. Sturgeon A, Pinder SL, Costa MC, Weese JS. Characterization of the oral microbiota of healthy cats using next-generation sequencing. Vet J. 2014;201:223-9. https://doi.org/10.1016/j.tvjl.2014.01.024

90. Dorn ES, Tress B, Suchodolski JS, Nisar T, Ravindran P, Weber K, et al. Bacterial microbiome in the nose of healthy cats and in cats with nasal disease. PLOS ONE. 2017;12:e0180299. https://doi.org/10.1371/journal.pone.0180299

91. Hutchins RG, Vaden SL, Jacob ME, Harris TL, Bowles KD, Wood MW, et al. Vaginal microbiota of spayed dogs with or without recurrent urinary tract infections. J Vet Intern Med. 2014;28:300-4. https://doi.org/10.1111/jvim.12299

92. Pilla R, Suchodolski JS. The role of the canine gut microbiome and metabolome in health and gastrointestinal disease. Front Vet Sci. 2020;6:498. https://doi.org/10.3389/fvets.2019.00498

93. Bermingham EN, Young W, Butowski CF, Moon CD, Maclean PH, Rosendale D, et al. The fecal microbiota in the domestic cat (Felis catus) is influenced by interactions between age and diet; a five year longitudinal study. Front Microbiol. 2018;9:1231. https://doi.org/10.3389/fmicb.2018.01231

94. Hasan N, Yang H. Factors affecting the composition of the gut microbiota, and its
modulation. PeerJ. 2019;7:e7502. https://doi.org/10.7717/peerj.7502
95. Reddy KE, Kim HR, Jeong JY, So KM, Lee S, Ji SY, et al. Impact of breed on the fecal microbiome of dogs under the same dietary condition. J Microbiol Biotechnol. 2019;29:1947-56. https://doi.org/10.4014/jmb.1906.06048
96. Older CE, Diesel AB, Lawhon SD, Queiroz CRR, Henker LC, Rodrigues Hoffmann A. The feline cutaneous and oral microbiota are influenced by breed and environment. PLOS ONE. 2019;14:e0220463. https://doi.org/10.1371/journal.pone.0220463
97. Du G, Huang H, Zhu Q, Ying L. Effects of cat ownership on the gut microbiota of owners. PLOS ONE. 2021;16:e0253133. https://doi.org/10.1371/journal.pone.0253133
98. Bermingham EN, Young W, Kettelmann S, Kerr KR, Swanson KS, Roy NC, et al. Dietary format alters fecal bacterial populations in the domestic cat (Felis catus). MicrobiologyOpen. 2013;2:173-81. https://doi.org/10.1002/mbo3.60
99. Scarsella E, Stefanon B, Cintio M, Licastro D, Sgorlon S, Dal Monego S, et al. Learning machine approach reveals microbial signatures of diet and sex in dog. PLOS ONE. 2020;15:e0237874. https://doi.org/10.1371/journal.pone.0237874
100. Kim YS, Unno T, Kim BY, Park MS. Sex differences in gut microbiota. World J Men's Health. 2020;38:48-60. https://doi.org/10.5534/wjmh.190009
101. Serpell JA, Duffy DL. Dog breeds and their behavior. In: Horowitz A, editor. Domestic dog cognition and behavior: the scientific study of Canis familiaris. Berlin: Springer; 2014. p. 31-57.
102. Kahrnani A, Werling D, Allenspach K. Canine breeds at high risk of developing inflammatory bowel disease in the south-eastern UK. Vet Rec. 2011;169:635. https://doi.org/10.1136/vr.d5380
103. Kim SY, Adachi Y. Biological and genetic classification of canine intestinal lactic acid bacteria and bifidobacteria. Microbiol Immunol. 2007;51:919-28. https://doi.org/10.1111/j.1348-0421.2007.tb03983.x
104. Beasley SS, Manninen TJK, Saris PEJ. Lactic acid bacteria isolated from canine faeces. J ANIM SCI. 2011;89:1498-505. https://doi.org/10.2527/jas.2010-3498
105. Karen LJ. Small intestinal bacterial overgrowth. Vet Clin North Am Small Anim Pract. 1999;29:523-50. https://doi.org/10.1016/S0195-5616(99)50033-8
106. Xenoulis PG, Palculict B, Allenspach K, Steiner JM, Van House AM, Suchodolski JS. Molecular-phylogenetic characterization of microbial communities imbalances in the small intestine of dogs with inflammatory bowel disease. FEMS Microbiol Ecol. 2008;66:579-89. https://doi.org/10.1111/j.1574-6941.2008.00555.x
107. Tang Y, Manninen TJ, Saris PE. Dominance of Lactobacillus acidophilus in the facultative jejunal Lactobacillus microbiota of fistulated beagles. Appl Environ Microbiol. 2012;78:7156-9. https://doi.org/10.1128/AEM.01975-12
108. Handl S, Dowd SE, Garcia-Mazcorro JF, Steiner JM, Suchodolski JS. Massive parallel 16S rRNA gene pyrosequencing reveals highly diverse fecal bacterial and fungal communities in healthy dogs and cats. FEMS Microbiol Ecol. 2011;76:301-10. https://doi.org/10.1111/j.1574-6941.2011.01058.x
109. Kim SY, Adachi Y. Biological and genetic classification of canine intestinal lactic acid bacteria and bifidobacteria. Microbiol Immunol. 2007;51:919-28. https://doi.org/10.1111/j.1348-0421.2007.tb03983.x
110. Beasley SS, Manninen TJK, Saris PEJ. Lactic acid bacteria isolated from canine faeces. J ANIM SCI. 2011;89:1498-505. https://doi.org/10.2527/jas.2010-3498
Probiotics and the gut microbiome in companion animals

112. Silva BC, Jung LRC, Sandes SHC, Alvim LB, Bomfim MRQ, Nicoli JR, et al. In vitro assessment of functional properties of lactic acid bacteria isolated from faecal microbiota of healthy dogs for potential use as probiotics. Benef Microbes. 2013;4:267-75. https://doi.org/10.3920/BM2012.0048

113. Bunešová V, Vlková E, Rada V, Ročková Š, Svobodová I, Jebraříček L, et al. Bifidobacterium animalis subsp. lactis strains isolated from dog faeces. Vet Microbiol. 2012;160:501-5. https://doi.org/10.1016/j.vetmic.2012.06.005

114. Araujo IC, Mota SB, de Aquino MHC, Ferreira AMR. Helicobacter species detection and histopathological changes in stray cats from Niterói, Brazil. J Feline Med Surg. 2010;12:509-11. https://doi.org/10.1016/j.jfms.2010.01.008

115. Johnston KL, Swift NC, Forster-van Hijfte M, Rutgers HC, Lamport A, Ballavre O, et al. Comparison of the bacterial flora of the duodenum in healthy cats and cats with signs of gastrointestinal tract disease. J Am Vet Med Assoc. 2001;218:48-51. https://doi.org/10.2460/javma.2001.218.48

116. Tun HM, Brar MS, Khin N, Jun L, Hui RKH, Dow SE, et al. Gene-centric metagenomics analysis of feline intestinal microbiome using 454 junior pyrosequencing. J Microbiol Methods. 2012;88:369-76. https://doi.org/10.1016/j.mimet.2012.01.001

117. Alessandri G, Milani C, Mancabelli L, Longhi G, Anzalone R, Lugli GA, et al. Deciphering the bifidobacterial populations within the canine and feline gut microbiota. Appl Environ Microbiol. 2020;86:e02875-19. https://doi.org/10.1128/AEM.02875-19

118. Buommino E, Nocera FP, Parisi A, Rizzotto A, Donnarumma G, Mallardo K, et al. Correlation between genetic variability and virulence factors in clinical strains of Malassezia pachydermatis of animal origin. New Microbiol. 2016;39:216-23.

119. Jacobi CA, Schulz C, Malfertheiner P. Treating critically ill patients with probiotics: beneficial or dangerous? Gut Pathog. 2011;3:2. https://doi.org/10.1186/1757-4749-3-2

120. Kochan P, Chmielarczyk A, Szymaniak L, Bryczynska M, Galant K, Zych A, et al. Lactobacillus rhamnosus administration causes sepsis in a cardiosurgical patient—is the time right to revise probiotic safety guidelines? Clin Microbiol Infect. 2011;17:1589-92. https://doi.org/10.1111/j.1469-0691.2011.03614.x

121. Sanders ME, Akkermans LMA, Haller D, Hammerman C, Heimbach JT, Hörmannspenger G, et al. Safety assessment of probiotics for human use. Gut Microbes. 2010;1:164-85. https://doi.org/10.4161/gmic.1.3.12127

122. EFSA [European Food Safety Authority]. Opinion of the Scientific Committee on a request from EFSA related to a generic approach to the safety assessment by EFSA of microorganisms used in food/feed and the production of food/feed additives. EFSA J. 2005;226:1-12. https://doi.org/10.2903/j.efsa.2005.226

123. European Commission. On a generic approach to the safety assessment of microorganisms used in food/feed and the production of food/feed additives. A working paper open for comment [Internet]. 2003 [cited 2021 Nov 4]. Available at: https://ec.europa.eu/food/sites/food/files/safety/docs/animal-feed_additives_rules_scan-old_report_out178.pdf

124. Hempel S, Newberry S, Ruelaz A, Wang Z, Miles JN, Suttorp MJ, et al. Safety of probiotics used to reduce risk and prevent or treat disease. Evid Rep Technol Assess. 2011;Apr:1-645.

125. Baffoni L. Probiotics and prebiotics for the health of companion animals. In: Di Gioia D, Biavati B, editors. Probiotics and prebiotics in animal health and food safety. Cham: Springer; 2018. p. 175-95.

126. Schmitz SS. Value of probiotics in canine and feline gastroenterology. Vet Clin North Am
127. FEEDAP [EFSA Panel on Additives, Products or Substances used in Animal Feed]. Guidance on the safety assessment of Enterococcus faecium in animal nutrition. EFSA J. 2012;10:2682. https://doi.org/10.2903/j.efsa.2012.2682

128. Rinkinen M, Jalava K, Westermark E, Salminen S, Ouwehand AC. Interaction between probiotic lactic acid bacteria and canine enteric pathogens: a risk factor for intestinal Enterococcus faecium colonization? Vet Microbiol. 2003;92:111-9. https://doi.org/10.1016/S0378-1135(02)00356-5

129. EFSA [European Food Safety Authority. Opinion of the scientific panel on additives and products or substances used in animal feed (FEEDAP) on the efficacy and safety of the coccidiostat Koffogran. EFSA J. 2004;2:16. https://doi.org/10.2903/j.efsa.2004.16

130. FEEDAP [EPSA Panel on Additives], Products or Substances used in Animal Feed]. Scientific opinion on the safety and efficacy of Cylactin® (Enterococcus faecium) as a feed additive for cats and dogs. EFSA J. 2013;11:3098. https://doi.org/10.2903/j.efsa.2013.3098

131. Holzapfel W, Arini A, Aeschbacher M, Coppolecchia R, Pot B. Enterococcus faecium SF68 as a model for efficacy and safety evaluation of pharmaceutical probiotics. Benef Microbes. 2018;9:375-88. https://doi.org/10.3920/BM2017.0148

132. Dogi CA, Perdigón G. Importance of the host specificity in the selection of probiotic bacteria. J Dairy Res. 2006;73:357-66. https://doi.org/10.1017/S0022029906001993

133. Park H, Yeo S, Arellano K, Kim HR, Holzapfel W. Role of the gut microbiota in health and disease. In: Di Gioia D, Biavati B, editors. Probiotics and prebiotics in animal health and food safety. Cham: Springer; 2018. p. 35-62.

134. Yeo S, Lee S, Park H, Shin H, Holzapfel W, Huh CS. Development of putative probiotics as feed additives: validation in a porcine-specific gastrointestinal tract model. Appl Microbiol Biotechnol. 2016;100:10043-54. https://doi.org/10.1007/s00253-016-7812-1

135. Weese JS, Anderson MEC. Preliminary evaluation of Lactobacillus rhamnosus strain GG, a potential probiotic in dogs. Can Vet J. 2002;43:771-4.

136. Strompfová V, Lauková A, Ouwehand AC. Lactobacilli and enterococci — potential probiotics for dogs. Folia Microbiol. 2004;49:203-7. https://doi.org/10.1017/S0042029904000193

137. Kainulainen V, Tang Y, Spillmann T, Kilpinen S, Reunanen J, Saris PEJ, et al. The canine isolate Lactobacillus acidophilus LAB20 adheres to intestinal epithelium and attenuates LPS-induced IL-8 secretion of enterocytes in vitro. BMC Microbiol. 2015;15:4. https://doi.org/10.1186/s12866-014-0337-9

138. Cunningham M, Azcarate-Peril MA, Barnard A, Benoit V, Grimaldi R, Guyonnet D, et al. Shaping the future of probiotics and prebiotics. Trends Microbiol. 2021;29:667-85. https://doi.org/10.1016/j.tim.2021.01.003

139. Weese JS, Arroyo L. Bacteriological evaluation of dog and cat diets that claim to contain probiotics. Can Vet J. 2003;44:212-6.

140. Weese JS, Martin H. Assessment of commercial probiotic bacterial contents and label accuracy. Can Vet J. 2011;52:43-6.

141. Metras BN, Holle MJ, Parker VJ, Miller MJ, Swanson KS. Assessment of commercial companion animal kefir products for label accuracy of microbial composition and quantity. J Anim Sci. 2020;98:skaa301. https://doi.org/10.1093/jas/skaa301

142. Mora D, Filardi R, Arioli S, Boeren S, Aalvink S, de Vos WM. Development of omics-based protocols for the microbiological characterization of multi-strain formulations marketed as probiotics: the case of VSL#3. Microb Biotechnol. 2019;12:1371-86. https://doi.org/10.1111/1751-7915.13476