Effects of Nutrient Supplementation on Immune Function in Cats and Dogs

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Abstract
Immune function is known to decline with age, and chronic stress can impair immune function and exacerbate the effects of aging. Dietary supplementation, with immunomodulatory ingredients, has potential in preventing or slowing such changes. Any modulation of the immune system should be balanced so that the immune system is primed to be optimally and appropriately active; however, not overstimulated, which could lead to negative effects such as chronic inflammation, allergy and autoimmune disorders. This article will explore the advantages and disadvantages of nutritional immunomodulation and examine the evidence behind the immunomodulating effects of certain nutrients.

Glossary of Abbreviations
ConA: Concanavalin A
CRP: C-Reactive Protein
DM: Dry Matter
IL-1: Interleukin 1
IL-2: Interleukin 2
IL-6: Interleukin 6
IL-10/STAT3: Interleukin 10/Signal Transducer and Activator of Transcription 3
MHC: Major Histocompatibility Complex
NK: Natural Killer
ω-3: Omega 3
ω-6: Omega 6
PHA: Phytohaemagglutinin
Se: Selenium
TNF-α: Tumor Necrosis Factor α
Vit: Vitamin

Introduction
Dietary components are able to impact the health of an animal in two ways; first, in a purely nutritive sense, supplying the necessary energy and amino acids to the animal, and second, by acting as bioactive molecules and influencing a number of functions within the animal, including immune health. The field of immunonutrition encompasses the latter by studying the effects of dietary supplementation with nutrients (including macronutrients, vitamins, minerals, and trace elements) on aspects of the immune system including inflammation, leukocyte function, antibody production, as well as resistance to disease.

Factors Causing Changes in Immune Function
It is widely recognized that malnutrition results in an impairment of the immune system, with a reduction of antibody responses and defective T-helper cell subpopulations[1] that can be reversed by feeding a high-protein diet.[2] However, research in several species, including dogs, indicates that overfeeding also has a detrimental effect on the immune system, resulting in a decreased resistance to the canine distemper virus[3] and salmonella infection, increased severity and frequency of disease,[4] reduced wound healing, and a greater mortality rate. With more and more cats and dogs being categorized as overweight or obese, weight control represents a simple mechanism to maintain or improve immune health in companion animals.

Excesses of individual amino acids also can be detrimental to the function of the immune system. It is well known that excessive intake of leucine has a detrimental effect on both cellular and humoral immunity in a number of species leading to decreased numbers of T lymphocytes and decreased antibody-forming cells or antibody titres.[5,6] These findings emphasize the importance of a balanced diet.

Aging and natural stress are two of the key causes of impaired function of the immune system and cause many of the same specific changes in immune function. The effects of age and psychological stress are viewed as being interactive, with chronic stress thought to exacerbate the effects of aging on the immune system.[7,8] Increased stress is associated with lower telomerase activity and shorter telomere length, which are both markers of aging.

Changes in Immune Function with Aging
In all species, age-related changes in the immune system

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are considered to impact risk of infection and diseases such as cancer, diabetes and autoimmune diseases,[9-11] making these changes one of the key factors that influence life expectancy, mortality and survival.[9,12-15] This age-related deterioration of the immune system is known as immunosenescence.[16]

A fully functioning immune system is comprised of both innate and adaptive arms, which are interdependent, relying on interaction between the two for activation and therefore generation of an efficient immune response. However, since individuals are exposed to an unique set of antigens and the nature and duration of the accompanying immune response differs considerably, the status of the immune system should be considered the result of both chronological age and immunological history of the individual.[15] The status of the immune system will also reflect differences as a result of genetics, nutrition and environment.[10]

Immunosenescence is generally considered to be due to a decline in adaptive responses rather than innate immunity and can be attributed at least in part to the involution of tissues such as the lymph nodes, Peyer's patches, the thymus, and tonsils and to physiological changes in the spleen and bone marrow that occur with age.[17] However, in the absence of disease, innate immunity remains relatively intact or even increased in older individuals,[15] and compensatory mechanisms appear to help maintain it. For example, the function of natural killer (NK) cells appears to decline with age; however, NK cell numbers increase thus compensating for the lower efficacy. The importance of this elevation in NK cell numbers was shown in a study in a group of over 85-year-old humans, in which individuals with NK cell numbers in the lowest quartile were three times more likely to die over the following two years.[9]

The decline in the adaptive immune system, primarily the T-cell system that characterises immunosenescence, begins following puberty and is manifested by decreasing CD8+ naive T cells numbers, decreased proliferative capacity, increased numbers of memory T and B cells, and a reduction in the diversity of antigen recognition.[14]

Since a successful humoral response is highly dependent on a fully functioning cell-mediated response, antibody responses are affected, and decreased responses to vaccination have been reported in both older individual humans (17-53% cf, 70-90% in young adults[9]) and laboratory animal models.[15] Even when older individuals show adequate antibody responses, the level of protection from infection is less than in younger adults. This is thought to be due to the antibodies produced having a lower affinity[10] and therefore, being less effective at neutralizing the viral pathogen.[9] Despite this, the immune system can function successfully for the majority of adult life without the deleterious effects that normally are seen only in the very late stages of life.

There also appears to be age-associated defects in non-T cells, such as antigen-presenting cells and dendritic cells, which fail to activate T cells effectively[15] and contribute to a loss of immune function by affecting regulatory control, primarily via indirect changes in cytokine expression.[9]

Inflammatory processes are also closely linked with immune function, and in older individuals conditions that would normally cause an acute inflammatory response such as acute infection or tissue damage tend to take longer to heal due to a reduction in the production of proinflammatory cytokines.[18] In contrast, chronic inflammation, which is prevalent in older individuals, confers an increased risk of both the development and the severity of diseases such as rheumatoid arthritis, cancer, cardiovascular disease, atherosclerosis, and osteoporosis.[8] Such chronic inflammation is manifested by high levels of proinflammatory cytokines such as IL-6 and TNF-α, as well as increased levels of C-reactive protein (CRP), which is produced in response to elevated IL-6.[8]

Changes in Immune Function with Stress

Physical and mental stress both impact the immune system, with acute stress tending to enhance immune responses, potentially as an adaptive response to help ensure survival by preparing the immune system to respond to challenges resulting from the stress, such as wounding or infection. In contrast, chronic stress results in immunosuppression, with decreased responses to vaccines,[19] a reduction in lymphocyte numbers due to apoptosis following activation of the Il-10/STAT3 axis,[20] decreased mitogen-induced lymphocyte proliferation, decreased NK cell function, altered cytokine production, and decreased skin cell-mediated immunity, all leading to slower wound healing and an increased risk of infectious disease, with the severity of the disease increasing as the duration of the stress increases.[21] In addition, dysregulation of the immune system is often still apparent several years after the cessation of the chronic stress;[8] hence, the individual remains at greater risk of infection and disease though the stress episode has been resolved.

The risks of maladaptive immune responses to stress in later life are also increased following prenatal or early life stressors.[8] Aside from maternal stress resulting in decreased maternal antibody transfer, early life stress results in decreased NK cell activity and lower antibody responses against vaccines and hence decreased resistance to infection and disease. These effects have also been shown to persist for at least several years after the stress has been resolved and are reflected in faster pathogenesis of viruses in adulthood, indicating that the early life stress can result in a significant and perhaps permanent compromise in immune cell populations.

It is not only the immune system that exhibits long-term effects of early life stress. Stress hormone (cortisol) responses to acute stress in adulthood are greater in those who experienced stress effects during early life. Due to the connections among the nervous, endocrine and immune systems, early life stress events could lead to significant lifelong health implications.

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Potential and Issues of Immunomodulation

There is increasing evidence that supplementing a complete and balanced diet with certain dietary compounds is frequently able to enhance immune function, thus indicating that ingestion of a complete and balanced diet is not always sufficient to achieve an optimally performing immune system and compensate for factors that impair immune function.

Given that both age and stress can negatively impact the immune system, on the surface it would appear that dietary enhancement of the immune system could be beneficial. There is also the question whether it is possible to use targeted dietary supplementation to prime the immune system for upcoming events, such as an upregulation of the immune system in preparation for a potential disease challenge (e.g., before placing pets in catteries/boarding kennels), or downregulation of the immune system with a dampening of excess inflammatory/allergic responses before the challenge is apparent (e.g., spring pollen levels, etc.).

In any modulation of the immune system there needs to be a balance such that the immune system is primed to be optimally and appropriately active when faced with an immunological challenge, and not overstimulated, which could lead to negative effects such as chronic inflammation, allergy and autoimmune disorders. It is, therefore, necessary not only to explore the issues surrounding nutritional immunomodulation but also to consider the criteria by which you would determine optimum immune function and how to assess it. This is particularly important given the many factors that can occur over the life span of the animal (including *in utero*) to influence immune function and hence affect the potential diversity of immune status of our companion animals.

There is considerable literature describing the immunomodulatory effects of various feed ingredients and pronutrients in a wide range of species.[22] For most immunomodulatory nutrients, a concentration range will exist within which consumption will result in optimal immune function; however, consumption at levels below or above this range can result in immunosuppression. For many immunomodulatory nutrients, excess consumption is not an issue as the levels required to elicit immunosuppression would be difficult to achieve. For other nutrients, the excess level at which immunosuppression occurs may be relatively low and easily achievable even with moderate supplementation. It is, therefore, important to clearly define the optimal immunomodulatory concentration levels for each species and also to educate the consumer of potential dangers of overconsumption.

The question as to what constitutes optimal immune function and how it can be measured is an important one. On the surface, optimal immune function would seem to be the ability to fight and/or prevent infection and disease; the difficulty arises in how it can be assessed. Frequency of illness and long-term survival would seem to be one answer, but neither is particularly informative, especially in the short term, and large numbers of subjects are needed to investigate this. There is considerable debate as to whether changes in immune parameters such as lymphocyte proliferation, phagocytosis and natural killer cell activity observed with *in vitro* and *ex vivo* studies will, in fact, result in a health benefit such as resistance to disease or infection *in vivo*. Although these methods can give indications of changes in immune function parameters, the clinical outcome remains unclear. Information on the frequency, severity and mortality rates from infection and disease challenge studies could potentially give a much clearer answer as to immune status, but this would come at a potentially very high ethical cost. Studies investigating the ability of ingredients to enhance antibody production against vaccines do provide relevant information on the immune function system; however, such studies are only providing information on one arm of the immune system, the adaptive arm. Taken together, most of the currently available methods for assessing optimal immune function are imperfect and therefore the results from work assessing the impact of dietary supplementation on immune function should be interpreted with this caveat in mind.

Effect of Nutritional Components on the Immune System

Omega 3 (ω-3) and Omega 6 (ω-6) Fatty Acids

The potential therapeutic benefits of dietary supplementation with ω-3 fatty acids (including eicosapentaenoic acid), which are found primarily in fish oils including salmon, and the optimal ratio of ω-6 (including arachidonic acid) to ω-3 fatty acids, has led to great interest in the functional foods arena. Interest in these fatty acids was triggered in humans in 1976 following the observation that Inuit people, who normally have a diet high in fish, also have a low incidence of heart disease.[23] Since then, investigations into the effects of ω-6 and ω-3 fatty acids on immune function, eicosanoid production, inflammatory responses, lipid peroxidation cancer, rheumatoid arthritis, ulcerative colitis, psoriasis, organ transplantation, and heart disease have been carried out in a number of species including dogs,[24-28] rats,[29] monkeys,[30] and humans.[31,32]

Results from these studies have been somewhat contradictory in nature, particularly among different species.[22] In addition, since the effects on the immune system, either immunoenhancing or immunosuppressive, appear to be ω-6:ω-3 fatty acid ratio dependent, this highlights the importance of determining the dose response effects of immunomodulatory components in the relevant species. It also shows the importance of raising consumer awareness that consumption of more of an immunomodulatory ingredient isn't necessarily advantageous and in some circumstances could even be deleterious for health.

Wound inflammation has been shown to be reduced in dogs following dietary supplementation with ω-3 fatty acids.[25]

Since ω-3 fatty acids act as substrates for eicosanoid metabolism, and changes in the feeding levels of ω-3:ω-6 fatty acids in dogs result in altered eicosanoid metabolism,[26,28] the likely mechanism of action is through the production of eicosanoids with lower inflammatory potential than those produced from ω-6 fatty acids.[33]

A study in geriatric Beagles investigating the effects of feeding three different ratios of ω-6:ω-3 fatty acids (31:1, 5.4:1 and 1.4:1) for 12 weeks reported that PGE_2 production and DTH responses tended to decrease with increasing ω-3 fatty acids in the diet, with a significant decrease observed at an ω-6:ω-3 ratio of 1.4:1.[28] Humoral responses were not affected, however, increased lipid peroxidation and decreased plasma vitamin E levels were also observed in the dogs as the level of enrichment of ω-3 fatty acids in the diet increased. Old and young Fox Terriers and Labrador Retrievers fed a diet containing an ω-6:ω-3 ratio of 5:1 for 60 days showed increases in PGE_3 production, but no effect on PGE_2 production (indicating no effect on eicosanoid production), and no effects on production of proinflammatory cytokines (IL-1, IL-6, TNF-α) or antibody production.[27] However, enhancement of T and B cell mitogenic responses were observed in the young dogs consuming the ω-3 enriched diets. The authors concluded that feeding Fox Terriers and Labrador Retrievers a diet containing an ω-6:ω-3 ratio of 5:1 had no negative effects on immune function. However, perhaps most important, they reported that the regulation of metabolism of ω-3 fatty acids appeared to differ between the two breeds.[27] These results emphasis the fact that feeding different ratios of ω-6:ω-3 can elicit different effects on the immune system and also may have different effects in different dog breeds.

In cats, some studies investigating the effects of dietary supplementation with ω-3 fatty acids on the immune system have shown that supplementation results in immunosuppression.[34-36] Cats consuming fatty acids derived from fish oil at an ω-6:ω-3 ratio of 5:1 for 12 weeks exhibited decreased lymphocyte proliferation to pokeweed mitogen, but no changes in responses to ConA or PHA[36] and a reduction in the numbers of B, T and Th cells.[34,36] Supplementation at the ω-6:ω-3 ratio of 5:1 also resulted in decreased skin inflammatory responses in cats.[36] In contrast, results from our study[37] that showed cats fed a diet supplemented with fish oil to an ω-6:ω-3 ratio of 1.77:1 for 35 days exhibited enhancement of lymphocyte proliferative responses to PHA, increased peripheral blood phagocytic activity, and no changes in the expression of B or Th cell subsets. These differences in the results may be due to the different lengths of the trials, the different ω-6:ω-3 ratio fed, or perhaps as suggested in previous canine studies, the breed of cat, and thus indicate that more work is required to determine the optimum ratio of ω-6:ω-3 fatty acids for overall health in the cat. Trials investigating the effects of feeding different ω-6:ω-3 ratios over varying periods of times and in different breeds of cats are also needed.

Potential Issues with Omega-3 Supplementation

Studies in cats,[38] dogs[39] and humans[40] indicate that consumption of diets enriched with ω-3 fatty acids (e.g., 1.31:1 ω-6:ω-3 ratio) led to prolonged bleeding times. In addition, some studies suggest that diets enriched with ω-3 fatty acids may lead to impaired immune responses and have other deleterious effects such as increased lipid peroxidation and adverse effects on the gut and wound healing (reviewed in[41]). Therefore, due to the apparent broad spectrum of effects that ω-3 fatty acids seem able to elicit, caution is needed when supplementing diets with ω-3 fatty acids. In stimulating one function, such as controlling inflammatory responses, other deleterious effects may result.

Trace Elements

Minerals have many roles within the body such as acting as cofactors for enzymes, maintenance of the 3D structure of proteins, forming a vital part of metalloenzymes, and relaying intercellular and intracellular messages. Minerals such as selenium and zinc are important for the endogenous synthesis of other compounds with antioxidant capacity including glutathione, glutathione peroxidase and superoxide dismutase. It is not surprising, therefore, that minerals play a role in maintaining a healthy immune system, with at least 20 minerals reported to have an impact on immune function in some way.[42] Even moderate deficiencies of trace elements such as zinc can prove detrimental to health by decreasing cell-mediated, humoral and nonspecific immunity in animals and leading to greater susceptibility to disease.[42-47] Conversely, consumed in excess, zinc has also been shown to impair immune function, reducing the phagocytic and bactericidal activity of neutrophils and macrophages.[48-50] Again, these results emphasize the importance of characterizing the levels required for optimal functioning of the immune system in each species.

Selenium (Se) is a component of several enzymes that are involved in protecting cellular components from oxidative damage[51-53] and provide some of the mechanisms through which Se is proposed to modulate immune function and reduce inflammation.[54] The role of glutathione peroxidase, which contains Se, is to remove immunosuppressive radical peroxides and regulate the lipoxygenase and cyclo-oxygenase pathways of the arachidonic acid cascade, which, in turn, control the synthesis of leukotrienes, thromboxanes, prostaglandins, and lipoxins, and modulate the products of the respiratory burst of phagocytes.[55,56] In addition, Se-containing enzymes (the glutathione peroxidase family of enzymes) control the production of proinflammatory cytokines (e.g., IL-1, IL-6, TNF-α), and prevent the activation of the proin-

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flammatory nuclear factor κB cascade by the removal of free radicals from the cell.[56] Se also is thought to increase T-cell activity by increasing expression of the high-affinity interleukin-2 receptor on T cells.[57]

Se deficiency has been reported to impair humoral and nonspecific immune responses in a variety of animal species, resulting in reduced neutrophil bactericidal activity,[51] lower levels of antibody producing cells, and hence lower levels of antibodies,[58,59] and the animal being more susceptible to infection than normal controls.[60] These effects are easily reversed by providing an adequate Se-containing diet.[51] Se toxicity also suppresses humoral and nonspecific immune function, while adequate and supplementary Se can improve immune function parameters, including lymphocyte proliferation, NK cell activity, expression of IL-2R, and cytotoxic T cell activity.[55,56,61] These changes are due at least in part to the ability of Se to enhance the clonal expansion of a range of immunocompetent cells.[22]

We carried out a trial[62] to determine if cats responded to dietary supplementation of Se with enhancement of certain immune parameters as has been seen in other species. Cats were fed a complete and balanced diet with and without Se supplementation (Se content DM basis: control, 0.38mg/kg (0.3mg/kg DM minimum SE requirement;[63] moderate Se, 2mg/kg; and high Se 10mg/kg)) for a period of four weeks. The results showed that in contrast to work in other species, dietary supplementation with Se had no effect on any immune function parameters measured (lymphocyte proliferative responses, peripheral blood phagocytosis and expression of lymphocyte subsets).[62] Outcomes such as this support the fact that results from trials assessing the effects of immunomodulatory
nutrients on immune enhancement cannot be extrapolated across species.

Antioxidants

Immune cells are particularly susceptible to oxidative damage for two reasons: First, their cell membranes have high levels of unsaturated fatty acids, and second, they are capable of producing large concentrations of free radicals during periods of intense activity such as inflammation.[22] Such oxidative damage can lead to a decline in immune function. There is growing interest in the utilization of antioxidants for improving health status by slowing the aging process[64] and preventing the generation of free radicals that can damage healthy cells and hence the immune system.

The antioxidants most studied in cats and dogs include vitamin E, ascorbic acid, β-carotene, lutein, and isoflavonoids.[62,65-71] The effects of some of these compounds on the immune system will be described here.

Vitamin C (Vit C) deficiency has been shown to result in impairment of the immune system in a number of species including guinea pigs and primates. Under normal conditions Vit C levels are high in leukocytes, and during infection are rapidly reduced in order to prevent free radicals from causing DNA damage to immune cells.[22] Vit C supplementation has been shown to be beneficial in humans by enhancing mitogen-induced lymphocyte proliferation, phagocytic function of neutrophils,[72] and in the treatment of several autoimmune diseases including HIV (reviewed in[73]). In addition, Vit C has been shown to reduce the toxic effects of chemicals on the immune system in both humans and animals.[22]

Lutein has been shown to have immunomodulatory effects in several species including cats and dogs. Kim, et al.[69] showed that a 12-week supplementation with dietary lutein in cats increased the levels of CD4+ and CD21+ lymphocytes and lymphocyte proliferative responses to ConA and pokeweed mitogen but did not affect pan T, CD8 or MHC class II markers. Lutein supplementation also resulted in a significant increase in plasma IgG levels in cats[69] and dogs,[74] possibly due to alterations in the cell membrane, which influence antigen presentation.[75] Significant increases in the levels of CD5+, CD4+, T and MHC II+ lymphocytes following dietary supplementation with lutein have been observed in dogs.[74]

The protective effects of Vit E on immune function can largely be explained by its antioxidant activity,[76,77] with Vit E reducing the immunosuppressive effects of free radicals and lipid peroxidation.[78] However, Vit E supplementation has also been shown to influence both the innate and acquired immune systems in various production animals, laboratory animals and humans,[77,79] though results are mixed. Enhancement of phagocytic activity by Vit E supplementation is likely due to its effects reducing the production of immunosuppressive compounds such as free radicals and PGE_2 [80-82] and the cell signaling molecule, nuclear factor κB.[56] Vit E also stimulates IL-2 production by T helper 1 cells, and since IL-2 is an important promoter of T and B cell proliferation and differentiation,[77,80] this mechanism may also explain the immune-enhancing effects of Vit E.

Trials investigating the immune-enhancing effects of Vit E in cats have shown enhancement of lymphocyte proliferative response to ConA, but not to PHA in aged (9.92 years) though not young cats (2.65 years)[81] when the diet was supplemented at either 250 IU/kg DM (225 mg/kg DM) or 500 IU/kg DM Vit E (450 mg/kg DM). In the same study, young cats exhibited a significant increase in the response to pokeweed mitogen when fed a diet supplemented with 500 IU/kg Vit E (450 mg/kg DM) but not 250 IU/kg DM (225 mg/kg DM).[81] These results demonstrate both age-related and dose response effects when supplementing with Vit E. Results from our own work in a mixed age group of cats (1.5-10 years) showed similar enhancement of lymphocyte proliferative responses to ConA and PHA and enhancement of phagocytic activity when animals were fed a diet containing either 250 IU/kg

DM (225 mg/kg DM) or 500 IU/kg DM Vit E (450 mg/kg DM). This indicated that in this group of cats supplementation at a level above 250 IU/kg DM was of no additional benefit.[62]

Combined supplementation of Vit E and Se has also been found to produce a greater enhancement of immune function in some species; an effect that is thought to be due to the synergistic antioxidant effects of Vit E and Se in the cell membrane and their control of arachidonic acid metabolism.[79] However, our own work in cats showed that diets supplemented with both Vit E and Se had no immune-enhancing effect above the level of Vit E alone.[62]

Trials in dogs have indicated that greater benefits of antioxidant supplementation can be expected during conditions of increased oxidative stress, such as exercise, leading to enhanced endurance[83] and reduction in exercise-induced oxidative damage.[66] Hence, antioxidant supplementation prior to such exercise may be useful in dogs, though the duration of the supplementation and period of administration prior to the event needs to be determined.

Antioxidants impact a range of immune functions, and since each antioxidant benefits the immune system uniquely, it is possible that a blend of several antioxidants may be more effective at enhancing the immune system than a single antioxidant at a high dose rate. In support of this, a study in dogs[84] showed that supplementation with Vit E and Vit C resulted in increased neutrophil bactericidal activity. Again, further trials comparing various dose rates and combinations of antioxidants are needed to provide substantiated evidence of the most effective formulation for each species.

Nucleotides

Studies in animals have shown that nucleotide-free diets lead to impaired cell-mediated and humoral immune responses, including decreased macrophage and NK cell activity, DTH responses, cytokine levels, lower antibody production, and an increased susceptibility to infection.[22,85] These changes can be reversed by dietary nucleotide supplementation. A number of animal studies suggest that the supplementation of diets with nucleotides leads to enhancement of immune function including mitogen-induced lymphocyte proliferation, IL-2 production, improved resistance to infection, enhanced cell-mediated immunity, and enhanced enterocyte and lymphocyte maturation.[86,87] Our own work in cats[37] showed that dietary supplementation with nucleotides for five weeks resulted in enhancement of phagocytic activity and lymphocyte proliferative responses to the T-cell mitogen PHA, which is agreement with work carried out in mice and humans.[87-89]

Conclusion

There is evidence to suggest that a number of dietary ingredients have the potential to modulate the immune system of healthy cats and dogs. However, we are still a considerable way from being able to recommend the targeted consumption of specific immunomodulatory ingredients to overcome upcoming challenges to the immune system. The evidence shows that different immune responses are sometimes seen in different species and possibly even in different breeds; therefore, results cannot be extrapolated across species and may not even be applicable in different breeds. In addition, as excess consumption of some immunomodulatory nutrients can result in deleterious effects, it is important to determine safe but efficacious levels of consumption. While this field is extremely promising, more work is needed to clarify the levels of supplementation required to optimize immune function in individual breeds, the effects of mixtures of immune-enhancing nutrients, and the efficacy and safety of term and even lifelong feeding of immunomodulatory ingredients.

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