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Nutrition and Immune Function

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Illness is commonly associated with anorexia and altered nutrient requirements as a consequence of biochemical, metabolic, and pathologic abnormalities that influence nutrient use. This complicated cascade of events directly and indirectly involves immunocompetence. Several questions of importance to patient management should be considered. First, should the immune system be enhanced during disease and/or illness? If yes, how can nutrients enhance immunocompetence during disease and/or illness and which nutrients are efficacious immunomodulators? The complex workings of the immune system can be simplified to the fact that disease initiation and progression are correlated with a break in immunocompetence. Something in the system went awry, and the assumption is that nutritional intervention can potentially get the system back on track to help manage, resolve, and, in the future, prevent the disease process.

OVERVIEW OF THE IMMUNE SYSTEM

The immune system is part of the host's defense against destructive forces from outside the body, such as bacteria, viruses, and parasites, or from within, such as malignant cells or those that produce autoantibodies [1]. This system is composed of two components: the innate or nonspecific immune system and the adaptive or specific immune system.

The system of nonspecific immunity consists of anatomic barriers and a cellular component. The various barriers, including skin and mucous and gastrointestinal (GI) mucosa, are the true "first line of defense." Once compromised by microorganisms, endotoxins, or any substance considered to be foreign, the complement system may be activated. The complement system is a complex cascade of proteins that promote such functions as phagocytosis, viral neutralization, and destruction of virus-infected cells. Complement system defects are associated with increased susceptibility to bacterial infections. Inflammatory mediators, in addition to being products of cell membrane destruction, increase vascular permeability, causing the accumulation of acute-phase proteins and immune complexes that promote the cellular phase of acute inflammation [1].

The cellular phase of the nonspecific immune system includes circulating and "fixed" phagocytes. Initially, neutrophils bind to pathogenic microorganisms,

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phagocytose them, and kill them. Phagocytosis is facilitated by opsonization. Opsonins activate neutrophils, resulting in an oxidative burst that includes production of H_2O_2 and O_2^- free radicals. These substances kill the bacteria and the neutrophil with release of toxic waste products. Although this response is beneficial in moderation, prolongation of the inflammatory phase can be detrimental to the host. Monocytes and macrophages are also components of nonspecific immunity. They phagocytize antigens, process them through an oxidative burst reaction, and present antigen particles to T cells via major histocompatibility complex (MHC) class I or II receptors [2].

The specific immune system is composed of B and T cells, which are associated with humoral immunity and cell-mediated immunity. B lymphocytes mature in bone marrow and react to stimulation by certain antigens to differentiate into plasma cells, which synthesize and secrete antibodies commonly termed *immunoglobulins* (Table 1). Cell-mediated immunity, however, relies primarily on T lymphocytes derived from the thymus. Antigen-presenting cells, such as macrophages, are responsible for triggering the specific immune response. Interaction of an antigen and macrophage leads to production of interleukin (IL)-1 by means of arachidonic acid (AA) metabolism. The IL-1 produced by macrophages causes T cells to produce IL-2 and other lymphokines. Production of IL-2 helps to stimulate T and B cells to form clones that carry receptors specific to the sensitizing antigen. These clones form the long-lived memory cells, which proliferate and release lymphokines on re-exposure to the same antigen. These clones, in conjunction with macrophages, can destroy the antigen. Defects in cell-mediated immunity are associated with infections of bacteria, mycobacteria, viruses, fungi, and parasites [1,2].

T cells are not only responsible for mediating delayed hypersensitivity, graft rejection, destruction of pathogenic microorganisms, and destruction of malignant cells but also regulate responses of other immune cells. The subsets of T cells

Table 1
Immunoglobulins

Immunoglobulin	Type	Role and location
IgG	4	Coats microorganisms for uptake by other cells (opsonization); crosses placenta to affect passive immunity; enhances complement function; primarily present in serum
IgM	2	First to respond to antigens via agglutination and bacteriolysis; present in blood
IgA	1	Protects mucous membranes by preventing bacteria from attaching to mucosal surface; present in body fluids
IgE	1	Involved in hypersensitivity reactions and allergic responses; phagocytosis and other immunoglobulin activity; present in plasma and tissue and on surface membranes of basophils and mast cells
IgD	1	Involved in differentiation of B lymphocytes; present in serum and in plasma membrane of B lymphocytes

include helper-inducer T cells (CD4), which help plasma cells to produce antibodies and release lymphokines, which modulate the interaction between lymphocytes and other cells. Cytotoxic-suppressor T cells (CD8) may destroy target cells, inhibit antibody responses, or inhibit the inflammatory response [1,2].

NUTRIENT INFLUENCE ON SPECIFIC COMPONENTS OF THE IMMUNE SYSTEM

The small bowel contains an abundant amount of lymphoid tissue and is a primary component of innate immunity through its cellular component and protective barrier function. It is often considered to be the first line of defense to invasion by microbes into the systemic circulation. The physical barrier is created by the tight junctions between intact epithelial cells, gastric acid, digestive enzymes, mucus production, intestinal motility, and normal bacterial flora [3]. The gut-associated lymphoid tissue (GALT) or intestinal immune system consists of lymphocytes and macrophages situated throughout the intestinal wall. IgA is also secreted into the GI lumen to prevent adherence of microbes to the mucosa. By design, these two components prevent or minimize the spread of pathogens, or their products, across the intestinal wall into the systemic circulation, a process termed *translocation*. Multiple factors promote translocation, including luminal bacterial overgrowth, impaired host defense mechanisms, protein-calorie malnutrition (PCM), trauma, critical illness, interruption of the luminal nutrient stream, or any other process that leads to mucosal atrophy [4]. Fig. 1 depicts the potential life-threatening sequelae to bacterial translocation. Maintenance of an intact barrier and functioning GALT are imperative to preventing translocation. Certain nutrients, specifically glutamine (GLN), arginine, nucleotides, ω-3 fatty acids, and dietary fiber (a source of short-chain fatty acids), are necessary for growth and normal function of the mucosal epithelial cells and the lymphoid cells of the GALT intestinal barrier [3,5].

MOLECULAR ASPECTS OF NUTRITION AND IMMUNE FUNCTION

Basic Concepts

The structural complexity of mammalian cells has its basis in regulated expression of thousands of different proteins. Most of the information that defines these molecules and structures is contained in DNA sequences within the cell nucleus. The primary focus for the technology of molecular biology is DNA and the processes that translate the informational content of DNA into cellular structures and functions. Much of the informational content of DNA consists of regions of nucleotide or base sequences that define or code for the amino acid sequences of proteins. Steps involved in the information transfer from DNA to proteins are illustrated in Fig. 2. The DNA nucleotide sequence is transcribed into an mRNA nucleotide. The mRNA sequence is then translated into a protein. The proteins generated in this manner form cellular structures or function as enzymes or membrane transporters that dictate much of the overall structure and function of the cell [6].

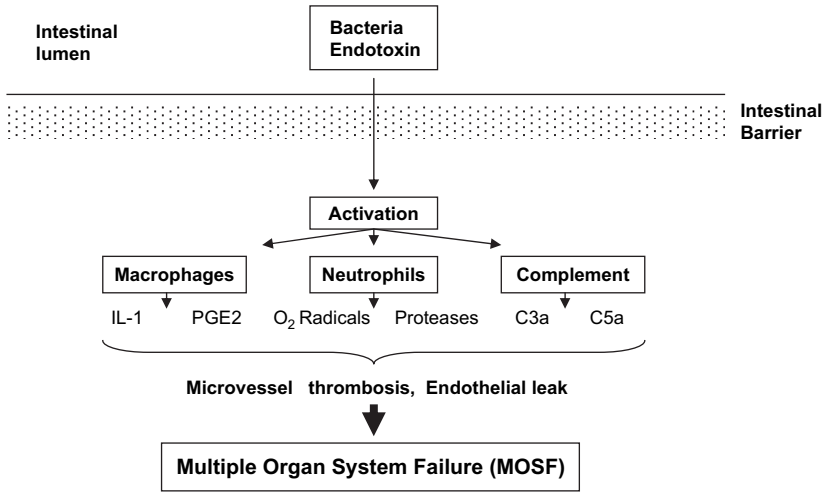

Fig. 1. A summary of the stepwise progression of bacterial translocation through the mucosal barrier resulting in multiple organ failure. PGE_2, prostaglandin E_2. (*Modified from* Rombeau JL. Enteral nutrition and critical illness. In: Borlase BC, Bell SJ, Blackburn GL, et al, editors. Enteral nutrition. New York: Chapman & Hall; 1994. p. 30; with permission.)

Practical Applications to Clinical Nutrition

Polysomes, the "preprotein" molecule sequence that builds on the mRNA backbone, can be isolated and quantified as a marker of nutrient intervention. GLN-supplemented total parenteral nutrition (TPN) versus a GLN-free control formula was given to patients for the first 3 days after abdominal surgery. Polysome content of the quadriceps femoris muscle obtained before and after day 3 after surgery showed a significant improvement in nitrogen balance and a sparing of free muscle GLN in the GLN-supplemented patients [7].

A second application involves DNA cloning (cDNA). A segment of DNA, or cDNA, that encodes for a specific protein of interest can be developed and used as a probe to assess expression of its corresponding mRNA in cells or tissues. Nutritionally relevant sequences derived from cDNA include proteins that function in carbohydrate (CHO; pyruvate kinase) and lipid (apolipoproteins) metabolism, growth control (growth hormone), and specific micronutrient actions (retinol-binding protein [RPB]). cDNA can be replicated multiple times over through host bacteria cultures. These recombinant proteins can be used to study normal and abnormal nutritionally important mechanisms of disease, including inflammation, cachexia, and oxidative stress, through such proteins as tumor necrosis factor and ILs [6].

Fig. 2 illustrates the process of creating and transferring a protein molecule from within the nucleus out to the cell cytoplasm. Examples of how the technologies associated with molecular biology can be used to monitor expression of nutrition-related molecules have been summarized. Interestingly, there seems to be a flip side of molecular nutrition, which involves the aspect of

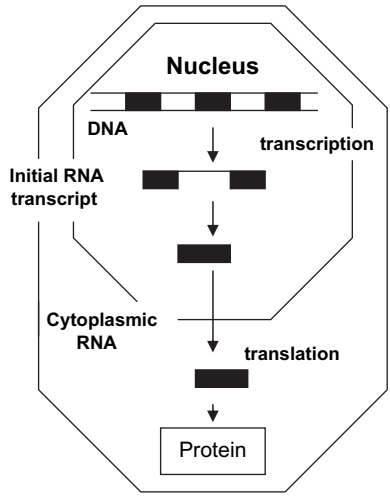

Fig. 2. Steps involved in the information transfer from DNA to protein. (*Modified from* Smith RJ. Molecular biology in nutrition. Nutr Clin Pract 1992;7:34; with permission from the American Society for Parenteral and Enteral Nutrition [ASPEN].)

uncovering the cellular mechanisms by which nutrients can influence gene expression. The transmission of signals from outside the cell into the nucleus can be profoundly influenced by nutrients. This area of technology is helping to identify how and which specific nutrients influence cell signal transduction and, ultimately, gene expression associated with immune function. Fig. 3 summarizes the complex relation between fatty acids, cell signal transduction cascades, apoptosis, and proliferation. This is one of numerous relations that are being clarified through nutrition research. The topic of nutrients as cell signals, although intriguing, is not meant to be a primary focus of this article. Several excellent reviews are available to develop this topic further. This level of nutritional science is likely to be extremely influential in optimizing patient management in the near future.

PATHOGENESIS OF ALTERED IMMUNOCOMPETENCE

Immunoimbalance is a systemic stress response to illness-induced anorexia or hypermetabolic food deprivation. It is associated with an increased metabolic rate, protein catabolism, and a release of multiple cell mediators, including cytokines, prostaglandins, and leukotrienes [8]. Oxidative (immune) cell function is escalated with the production of highly reactive radicals that damage cell constituents (membranes, proteins, lipids, and DNA), leading to cell death and multiple organ dysfunction. From a biologic perspective, stress is more accurately defined as oxidative stress, or the imbalance between production of damaging free radicals and antioxidant protection [9]. Oxidative stress plays a major role in many degenerative pathologic conditions, and free radical formation is considered to be a pathologic biochemical mechanism involved in the

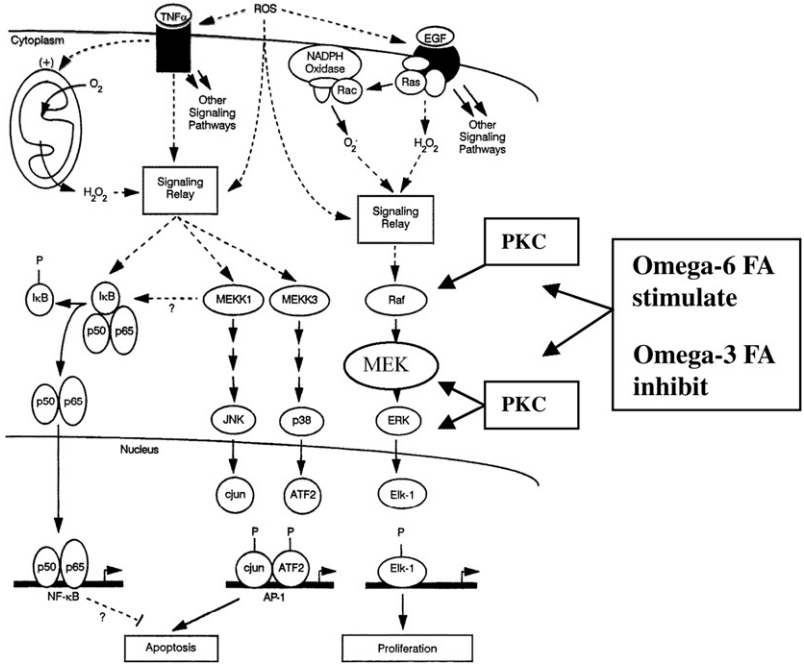

Fig. 3. Overview of cell signal transduction pathways suggesting points of signal enhancement or interruption via fatty acids (FA). AP-1, activator protein-1; EGF, epidermal growth factor; NF-κB, nuclear factor-κB; PKC, protein kinase C; ROS, reactive oxygen species; TNFα, tumor necrosis factor-α. (*Adapted from* Cowing BE, Saker KE. Polyunsaturated fatty acids and epidermal growth factor receptor/mitogen-activated protein kinase signaling in mammary cancer. J Nutr 2001;131:1127; with permission.)

initiation or progression phase of various diseases. These highly reactive radicals that can be so damaging if left unchecked are oxygen (reactive oxygen species [ROS]) and nitrogen (reactive nitrogen species [RNS]) based and are continuously and inescapably produced from the energy production cycle (mitochondrial electron transport chain), the detoxifying process via cytochrome P450 enzymes, reactions with transition metals (ie, copper [Cu], iron [Fe]) released during high-oxidative events (ie, injury, drug therapy, during the synthesis of fatty acid metabolites [eg, prostaglandins, leukotrienes]), and the immune system as a deliberate function of innate immune cells [10]. Based on the diversity of radical producers, it is logical that numerous body systems, including cardiopulmonary, endocrine, hematologic, integument, neurologic, and GI systems, are affected by oxidative stress.

The relation of nutrition, disease, and oxidative status is complex. Again, the aspect of molecular immunology surfaces as a technology that is being used to identify mechanisms involved in these relations. Fig. 4 illustrates one example of the complexity of nutrient manipulation focused on immunity (inflammation) and cell growth. In this case, ω-fatty acids are the potential influence on

the cell signal cascade and nuclear transcription through inflammatory and tumor cell mediators.

MALNUTRITION, NUTRIENTS, AND IMMUNITY

Malnutrition, associated with a single nutrient or multiple nutrient inadequacies, is consistently associated with metabolic and clinical alterations of immunity. The association of malnutrition with reduced resistance to infection has been observed for centuries. Early work with children in developing countries suggested that the degree of immunocompromise depended on the degree of protein-energy malnutrition, presence of infection, and age at the onset of malnutrition [11]. In industrialized societies, PCM has been described more frequently in the elderly and in hospitalized patients [1]. Malnutrition should not only be considered to be of protein-calorie origin, however, because vitamin and mineral deprivation can also adversely influence the aspects of immunity. Immunocompromise and malnutrition in hospitalized patients contribute to the development of infection, sepsis, organ failure, poor wound healing, and a general increase in morbidity and mortality. Acutely ill patients with sepsis or a sepsis syndrome may exhibit immunosuppression without prior starvation [12].

Marasmus or semistarvation malnutrition is a syndrome that develops gradually over months to years with insufficient energy intake. The body responds by decreasing basal energy expenditure through a decrease in thyroid and sympathetic nervous system activity. Additionally, there is a shift of fuel sources in

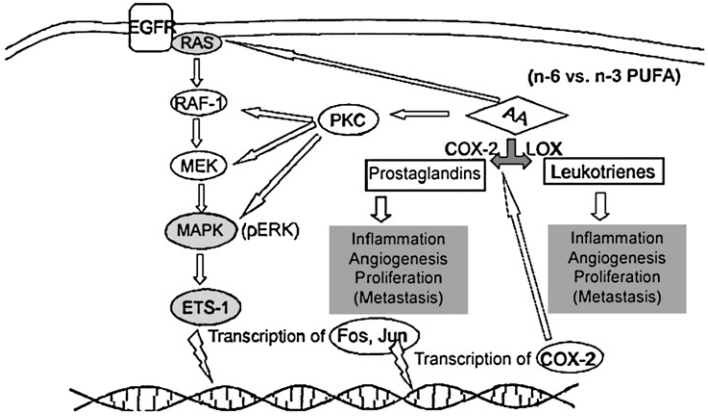

Fig. 4. Overview of the proposed relationship between inflammation and cell growth. Omega-6 and omega-3 fatty acids may influence the cell signaling cascade and nuclear transcription. COX-2, cyclooxygenase-2; EGFR, epidermal growth factor receptor; LOX, lipoxygenase; MAPK, mitogen-activated protein kinase; PKC, protein kinase C; PUFA, polyunsaturated fatty acid; RAS, p21 ras; RAF-1, p74 raf-1; MEK, MAPK (Erk) kinase. (*Adapted with modification from* Cowing BE, Saker KE. Polyunsaturated fatty acids and epidermal growth factor receptor/mitogen-activated protein kinase signaling in mammary cancer. J Nutr 2001;131: 1127; with permission.)

response to the depletion rate of stored nutrients. As glucose and glycogen reserves are depleted, protein and fat are sequestered for energy. To sustain primary protein needs for as long as possible, fat becomes the predominant fuel source. This type of malnutrition can be observed in patients with chronic disease processes that adversely affect energy intake, such as cachexia (cardiac or cancer) or malassimilation disorders [13].

In contrast, hypoalbuminemic malnutrition is a manifestation of the body's response to infection or inflammation with or without nutrient deprivation [14]. The association of malnutrition with metabolic and clinical alterations of immunity is more clearly evident in this situation. Hypoalbuminemic malnutrition is modulated by hormones and cytokines (ie, IL-1, tumor necrosis factor-α [TNFα]) secreted during the acute response to major stressors, such as sepsis, head injury, burns, or trauma [14]. It occurs quickly to deplete visceral protein (albumin) stores, and the multiplicity of sequelae to these events adversely affects metabolism and immune function.

The effects of PCM, regardless of the category, can be quite complex. Humoral immunity can be affected by PCM as a decline in the production of immunoglobulins, secretory antibodies, and complement. Cell-mediated immunity is commonly affected in hypoalbuminemic or severely marasmic patients. The thymus and lymphoid tissues atrophy, peripheral T lymphocytes decrease in number, alterations in cell-mediated delayed cutaneous hypersensitivity and graft-versus-host reactions are apparent, there is an impaired response of lymphocytes to mitogens, and patients exhibit a poor response to contact sensitization or inflammatory reactions as well as a depressed response to vaccines [15]. Neutropenia may occur to varying degrees in patients with PCM. Although neutrophils seem to be morphologically normal, cell function is decreased, specifically the capacity of neutrophils to kill phagocytosed bacteria or molds and to secrete chemokines [16]. Complement components of innate (nonspecific) immunity are depressed. Interferon production, opsonization, plasma lysosome production, and acute-phase reactants (ie, C-reactive protein) are adversely affected. Likewise, alterations in the anatomic barriers to infection included in the nonspecific immune system, such as atrophy of the skin and GI mucosa, may increase the risk of infection [13].

Whether correction of malnutrition improves patient outcome in all cases has yet to be proven, although, intuitively, it is assumed to be associated with a beneficial effect. Several studies [17,18] that clearly demonstrate the acute impact of nutrient deprivation on immune function and the reversal of innate and cell-mediated immune system compromise by appropriate nutrition support are summarized in Table 2. Current research in nutrition support is beginning to focus on exerting organ-specific effects by modulating metabolic processes rather than by simply improving nutrition. The effect of specific nutrients on the immune system is showing great promise in this regard. Many nutrients have a role in immune function. For some, mechanisms of immunomodulation have been clearly delineated, whereas much is yet to be learned in others.

Table 2
Differences in the monocyte phagocytic activity and CD4+/CD8+ lymphocyte expression in response to nutrient deprivation and refeeding in adult cats[a]

	Day[b]			
Item	4	7	11	14
Phagocytic activity[c]	−4.1*	−4.2*	−5.8**	−4.7**
CD4+[d]	−7.0	−9.0	−10.0	+5.0**
CD8+[d]	+15.0*	+9.0	+5.0	+25.0*
Lymphocyte proliferation[e]	−0.04	−0.09	−0.02*	−0.01*

Abbreviations: Con-A, conanavalin-A; DER, daily energy requirement; FITC; fluorescein isothiocyanate.

[a]N = 23 cats.

[b]Cats underwent nutrient deprivation (excluding water) for days 1 through 7 and were then refed to meet DER for days 8 through 14.

[c]Values represent the change from baseline (day 0) in percentage of cells that phagocytosed fluorescent polystyrene beads. Fluorescence detected by flow cytometry.

[d]Values represent the change from baseline in expression of CD4/CD8 on lymphocyte cell membranes after stimulation with con-A and staining with FITC and specific monoclonal antibody. Fluorescence detected by flow cytometry.

[e]Values represent change from baseline in proliferative capacity of 3.0×10^9 cells/mL stimulated with con-A. Proliferation determined by Alamar blue staining.

*Values differ from Day 0 ($P<.01$).

**Values differ from Day 7 ($P<.01$).

KEY NUTRIENTS AS MODULATORS OF IMMUNE FUNCTION

The list of key nutrients that may influence immunity seems to expand almost daily. Several decades ago, the list was short. Protein was the key nutrient, and micronutrient nutrition was a radical concept. Currently, a list of key nutrients includes specific amino acids, fatty acids, vitamins, microminerals, and nucleic acids in addition to the less well-defined micronutrients, such as flavonoids.

PROTEIN AND AMINO ACIDS

Along with total dietary protein content, form of protein delivery as an amino acid or an intact molecule and the individual amino acid concentration in the diet have been shown to influence the immune response. Early human studies evaluated increasing the percentage of protein in diets of children with extensive burn injuries. An increase from 15% to 23% resulted in a twofold increase in survival [19]. The higher dietary protein likewise resulted in significantly higher levels of serum total protein, transferrin (an acute-phase protein), complement C_3, and IgG in these patients. Investigators found no significant enhancement in serum protein levels, nitrogen balance, and complement C_3 in animals fed free amino acids compared with an intact whey protein source [20]. Conversely, there have been numerous reports of immune enhancement resulting from single amino acid enrichment to patient diets, particularly with arginine or GLN.

Arginine

Arginine is an essential amino acid in the cat for growth and maintenance of the urea cycle. It falls into the "semiessential" or "conditionally essential" category

under a variety of stress situations, including burns, trauma, sepsis, and rapid growth in other species [21]. Arginine has also been shown to play a necessary role in collagen synthesis for wound healing and is required for nucleotide synthesis. Multiple secretagogue activities have been associated with arginine because it enhances secretion of prolactin, growth hormone, and insulin-like growth factor-1 (IGF-1) [22]. Prolactin induces maturation of dendritic cells by increasing the expression of antigen-presenting MHC class II and costimulatory molecules and stimulates release of T helper (Th) 1 cytokines by T lymphocytes. Growth hormone and IGF-1 can potentiate cytokine responses of T cells, increase progenitor cells in the bone marrow, and increase lymphocyte number [23].

Arginine also has documented immunoregulatory function in the stressed animal. Overall, it augments cellular immunity, and the specific effects can be summarized as increased thymic lymphocyte blastogenesis, responsiveness to mitogens, IL-2 production and IL-2 receptors, natural killer (NK) cells, and macrophage cytotoxicity to tumor cells and bacteria [24]. Arginine also seems to affect induction and development of malignant tumors through its effects on the immune system. These actions seem to be linked to arginine-derived nitric oxide (NO) and, depending on the surrounding microenvironment, the net biologic effect of arginine-derived NO can inhibit or promote tumor growth [25]. Bower [1] summarized numerous studies evaluating the role of arginine in animal tumor models and clinical studies. Arginine was reported to decrease the incidence of tumors after exposure to carcinogens, increase the latency period, shorten the interval required for tumor regression, and increase host survival in animals with malignant lesions [26]. It is thought that retardation of tumor growth and metastatic spread may be caused by the arginine-enhanced phagocytic function of macrophages, increased T-cell blastogenesis, and increased IL-2 production [1]. Adults with a GI malignancy demonstrated a quicker and more advantageous lymphocyte proliferative response to arginine (25 g) versus glycine (43 g) in the postoperative period [27]. These and numerous other studies are suggesting that the antitumorigenic effects of arginine can be via the specific and nonspecific immune systems.

Arginine is an important substrate for the synthesis of NO. The inducible form of nitric oxide (iNOS) is of most relevance to the immune system. iNOS expression, and hence NO production, is induced in monocytes and macrophages in response to stimuli, particularly that of interferon-γ (IFNγ) and lipopolysaccharide (LPS). NO is a regulator of various immune functions, and its inhibition increases host susceptibility to infections, making it essential for host defense [23]. Alternatively, arginine metabolism can involve the enzyme arginase. Arginase is increased in LPS- and cytokine-stimulated macrophages and converts arginine to ornithine. The ornithine produced is involved in the synthesis of polyamines, which are required for maintenance of cell viability. Polyamines act to facilitate DNA, RNA, and protein synthesis; therefore, inhibition of polyamine synthesis leads to a reduction in cell viability and cell differentiation [23]. A profound effect of dietary arginine

supplementation was reported some years ago through a series of animal studies. Injured rats exhibited a reduction in trauma-induced thymic involution, increased T-cell response, sustained body weight, improved wound healing, and prolonged survival [26]. Babineau [21] summarized several studies that highlighted the influence of arginine on immune function. One such study demonstrated that arginine-supplemented diets enhanced cytotoxic T-lymphocyte development and increased NK cell activity and IL-2 receptor expression kinetics on activated T cells. In another study, 2% arginine supplementation resulted in increased survival and an improved delayed hypersensitivity response in animal burn patients [21]. A study involving stressed human subjects indicates that dietary arginine enhanced immune function through an increased peripheral blood lymphocyte and blastogenic response to mitogens, concanavalin A, and phytohemagglutinin [21].

Numerous human clinical studies have used commercially available immune-enhancing diets, which are supplemented with a combination of arginine, omega-3 fatty acids, and nucleotides. Although immune-enhancing effects are clearly demonstrated, because of the complexity of the formula, an arginine-specific effect is often not clearly interpretable [23]. Bansal and coworkers [28] investigated the interactions between fatty acids and arginine metabolism and what implications this may have on immune-enhancing diets. They reported that prostaglandin E_2 (PGE_2) from ω-6 fatty acids upregulates expression of arginase 1, which, subsequently, leads to arginine depletion. Conversely, ω-3 fatty acids protected against arginine depletion by attenuating the upregulation of arginase 1. The duality of arginine metabolism has led to some controversy regarding the immunologic value of increased dietary levels of arginine across all critical illness scenarios [29]. Currently, however, the pendulum swings toward supplementation improving outcomes in patients with sepsis, wounds, ischemia-reperfusion, and thermal injury [29,30].

Glutamine

GLN has traditionally been considered to be a nonessential amino acid in health, only taking on a "conditionally essential" status in states of illness or injury. In catabolic states, amino acids, predominantly alanine and GLN, are released from muscle tissue to provide a fuel source for enterocytes of the small bowel and for rapidly dividing leukocytes and macrophages in the immune system [31]. Particularly during stress, GLN is the preferential fuel source for cells of the gut and can be rapidly depleted despite the significant release from muscle tissue. Plasma GLN levels have been shown to decrease by 58% after critical illness or injury and to remain decreased for up to 3 weeks with increased mortality in seriously ill patients [31]. A GLN deficiency-related impairment of lymphocyte and neutrophil function as well as glutathione (GSH) depletion is a likely mechanism for the increased mortality. Several specific immunomodulatory actions of GLN in vitro and in vivo have been nicely summarized and reported by Calder and Yagoob [23]. Increasing the availability of GLN in culture has been shown to enhance T-lymphocyte proliferation and IL-2 and IFNγ

production by lymphocytes, B-lymphocyte differentiation into antibody-producing cells, phagocytosis, and antigen-presenting activities of macrophages and neutrophils. Animal studies have reported that GLN enrichment of the diet increases T-lymphocyte proliferation of splenic CD4+ cells and cytokine production (eg, TNFα, IL-1, IL-2, IL-6, IFNγ) in injury- or infection-stressed situations. A study of critically ill patients supplemented with enteral GLN reported a significant decrease in the incidence of sepsis, pneumonia, and bacteremia. The mechanism was thought to be associated with enhanced expression of antigen-presenting receptors on the monocyte cell surface [32]. This, of course, is an important aspect of innate immunity, in which phagocytic cells engulf and process (kill or disassemble) invading pathogens and subsequently jump-start other branches of the immune system for an optimal immune response. Preservation of the gut mucosal barrier to minimize intestinal permeability was another proposed mechanism of GLN supplementation in this study. Fig. 5 summarizes potential pathways for GLN benefits in the critically ill patient.

Interestingly, supplementation of GLN has been reported to demonstrate significant benefit and no added benefit to immunocompetence based on the route of delivery and study design. Hall and coworkers [33] reported that low-dose enteral GLN therapy to critically ill patients resulted in no improvement in the incidence of sepsis, body condition, and mortality compared with unsupplemented controls. There are numerous studies reporting benefit and lack of benefit from parenteral GLN in critically ill patients. Cellular mucosal and peripheral immune cell functions were evaluated in dogs receiving a 2% GLN-fortified parenteral admixture (PA) before and after intestinal resection and anastomosis surgery. PA-GLN resulted in a significantly increased helper and decreased suppressor T-cell population, increased IgM, a mild increase in IgA, significantly less diarrhea days, and a lower hospital cost compared with those parameters in patients receiving a calorie- and GLN-free solution before and after surgery [34]. Again, dosage, timing, and response indices varied and likely influenced the outcome in these studies. In summary, most trials associated with GI disease indicated that larger GLN doses were more beneficial than lower doses of GLN [31]. There is limited availability of studies in critically ill or diseased companion animals supplemented with GLN; therefore, summarizing human studies may support or refute the idea of GLN as a useful immunomodulator in the feeding management of sick pets. The results of a meta-analysis [35], including 14 randomized trials involving 751 patients, of GLN administration in critically ill patients versus standard care indicates a shorter hospital stay, lower rate of infectious complications (risk ratio [RR] = 0.81), and lower mortality rate (RR = 0.78). Subgroup analysis revealed a treatment benefit of high-dose GLN (>0.20 g/kg of body weight [BW] per day) over low-dose GLN (<0.20 g/kg of BW per day) with regard to mortality. Table 3 summarizes only the enteral GLN studies evaluated in the meta-analysis.

GLN is also a nitrogen donor for the synthesis of purines and pyrimidines, a substrate for protein synthesis, and a precursor to glutamate, which is

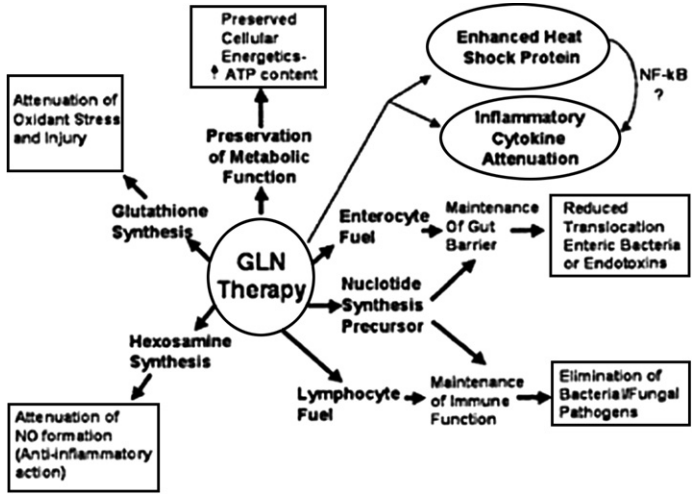

Fig. 5. Potential mechanisms for the beneficial effects of glutamine (GLN) in critically ill patients. ATP, adenosine triphosphate; NF-κB, nuclear factor-κB; NO, nitric oxide. (*Adapted from* Wischmeyer PE. Clinical application of L-glutamine: past, present, and future. Nutr Clin Pract 2003;18(5):378; with permission from the American Society for Parenteral and Enteral Nutrition [ASPEN].)

incorporated into the antioxidant GSH. GSH depletion was associated with diminished IFNγ production. A rise in lymphocyte GSH content was accompanied by an increase in mitogen-induced lymphocyte proliferation and IL-2 production [23]. These studies suggest that GLN promotes a range of cell-mediated and innate immune responses.

Taurine

Taurine is another sulfur amino acid that seems to be involved in immune function, and several studies highlighting this relation are summarized by Calder and Yagood [23]. Taurine is derived from the metabolism of methionine and cysteine; thus, it is not considered to be a component of proteins. It is present in high concentrations in cells of the immune system, however, and accounts for 50% of the free amino acid pool within lymphocytes. Although the role of taurine within lymphocytes is not well defined, it is reported that cats fed taurine-deficient diets exhibit atrophy of the lymph nodes and spleen, a decrease in circulating lymphocytes, and impaired oxidative burst by phagocytes. Administration of taurine prevented and reversed adverse T-cell changes in mice of various ages. Taurine chloramine, a complex of taurine with hypochlorous acid (HOCl), protects the host from toxic damage of HOCl derived from oxidative processes and has also been shown to be bacteriocidal in its own right. Because taurine chloramine decreases NO, superoxide, PGE_2, TNFα, and IL-6 production by leukocytes, it has been proposed that taurine may offer a therapeutic approach to acute inflammatory events.

Table 3
Randomized trials of enteral glutamine in critically ill human patients

Study	Patient population (no.)	Dosage L-glutamine (g/kg of BW per day)	Mortality (%)		Infectious complications (%)	
			Expt.	Control	Expt.	Control
Houdijk et al, 1998	CI (78)	>25	4/41 (9.8)	3/39 (7.7)	20/35 (57.1)	26/37 (70.2)
Jones et al, 1999	Mixed (165)	−0.16	10/26 (38.5)	9/24 (37.5)	—	—
Brantley and Pierce, 2000	CI (70)	0.50	−0/31 (0.0)	0/41 (0.0)	—	—

Abbreviations: BW, body weight; CI, critically ill; Expt, experiment; Mixed, ICU/hospital; —, not available.

LIPIDS

Lipid metabolism and use can not only yield a useful energy source but can influence metabolic and immunologic parameters during health and illness. Stored body fat is the major energy reserve for nonstressed starvation-adapted patients; however, under circumstances of stress, the protein-sparing effect of fat oxidation is lost. Administration of relatively high levels of lipid in critically ill patients provides a concentrated energy/calorie source and helps to avoid the complications associated with overfeeding CHO. Conversely, excessive fat dosing can itself lead to complications related to cardiopulmonary dysfunction, platelet dysfunction, and immune function compromise. Decreased clearance of bacteria from phagocytosis of lipid globules, subsequently increasing the risk of bacteremia and sepsis, has been reported in human and animal patients receiving excessive lipid in the form of long-chain triglycerides (LCTs) [36]. Replacement of the same fraction of the intravenous (parenteral)-delivered LCTs with medium-chain triglycerides (MCTs) seemed to protect septic patients from these adverse immune system sequelae [36]. This suggests that lipid content and form can influence immune function and that assessment of the patient's immunologic status is paramount when determining lipid content in nutritional support protocols. Evaluation of specific immune cell functions as indicators of immunologic status in patients is realistically more suited for a research setting. Having said this, evaluation of bleeding time could be performed in a clinical setting and would give some measure of platelet function. With its reported relationship to omega-3 FA lipid content (LCT), it could be utilized as a means to assess adequate lipid content nutritional support protocols. A full spectrum, 'quick and dirty' type of immunologic panel for use in daily practice is not currently available.

Although lipids are an essential component of the body, it seems that the immunomodulation of the specific and nonspecific immune systems is profoundly

influenced through action of the essential fatty acids (EFAs), omega-6 and omega-3 families. Immune cells, including monocytes, macrophages, lymphocytes, and granulocytes, are able to synthesize non-EFAs but must rely on circulating blood lipids as the source of EFAs. Therefore, the lipid composition of immune cells reflects the fatty acid composition of lipids in the diet [1]. There are numerous reviews outlining the metabolism of the essential polyunsaturated fatty acids (PUFAs) linoleic acid (n6) and linolenic acid (n3). Briefly, linoleic acid is converted to AA, which serves as a precursor to prostanoids (particularly PGE_2), thromboxanes of the 2 series, and leukotrienes of the 4 series. These compounds are largely proinflammatory and have been implicated as mediators in the vascular component of septic shock [21]. Conversely, provision of n3 fatty acids, principally from fish oil, leads to production of eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA). These compete with AA for cyclooxygenase (COX) and lipoxygenase enzymes, ultimately to yield the 3 series of prostanoids and 5 series leukotrienes, which are reported not only to be less inflammatory and vasoactive but to possess anti-inflammatory action [37]. This indicates that the type of fatty acid in the diet is another avenue to influence immune function.

Omega-6 Fatty Acids

Vegetable oils, including corn, soy, canola and safflower oils, are a primary source of ω-6 fatty acids in the diets of companion animal. Prostanoids derived from metabolism of n6 PUFAs seem to have a dose effect. Extremely low concentrations induce lymphocytes to differentiate into T cells; however, overproduction of PGE_2 depresses measurements of T-cell function, including response to mitogens, clonal proliferation, production of lymphokines, migration and generation of cytotoxic T cells, and killing activity of phagocytes [1]. The leukotrienes are short-turn-around activators of leukocytes. They can stimulate leukocytes to aggregate and adhere to endothelial cells. They also influence NK cell activity. The n6 fatty acids, with certainty, play a significant role in immunosuppression, tumorigenesis, and enhancing inflammation (Fig. 6) [38].

Omega-3 Fatty Acids

α-Linolenic acid (18:3ω3) is an essential nutrient for companion animals, thereby necessitating its inclusion in the diet to promote normal growth and development. Dietary omega-3 fatty acids are most commonly derived from marine fish oil, although several agricultural sources for n3 oils also exist. In fish oil–derived n3 fatty acids, EPA is the most active component. Although DHA is also derived from α-linolenic acid and possesses anti-inflammatory as well as other properties, EPA is the form of n3 fatty acid that has been researched most extensively. EPA and DHA attenuate the inflammatory response, stabilize the nuclear factor-κB (NF-κB) complexes, decrease platelet adhesiveness, enhance lymphocyte and neutrophil function, and aid in membrane stability and microvascular perfusion, whereas high levels of AA have the inverse effect [39,40]. Clinically, n3 fatty acids have reportedly shown

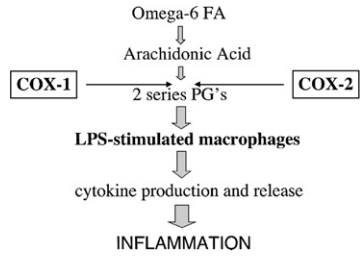

Fig. 6. Overview of the role omega-6 fatty acids play in cell-mediated cytokine production and inflammation. FA, fatty acids; PG, prostaglandin; LPS, lipopolysaccharide; COX-1, cyclooxygenase-1; COX-2, cyclooxygenase 2.

benefit in a variety of disease states, including arthritis, sepsis, cardiac abnormalities, and cancers. The primary mechanism of action underlying the value of n3 fatty acids seems to be, directly and indirectly, their anti-inflammatory focus. Through cell signal cascades, n3 fatty acids influence COX-2 expression and ultimately exhibit COX-2 inhibitor-like action to inhibit PGE_2 production and lessen the inflammatory response. The n3 fatty acids have also been demonstrated to decrease macrophage NF-κB nuclear translocation and subsequently inhibit production of proinflammatory cytokines via NF-κB [40]. It has also been reported that n3 fatty acids alter the proteins and specific nuclear transcription factors in the mitogen-activated protein kinase (MAPK) pathways, including activator protein-1 (AP-1). These pathways ultimately activate or inhibit cell proliferation versus apoptosis (cell death) [40,41]. Numerous commercial (therapeutic and nontherapeutic) diets have focused on the n6:n3 fatty acid ratio in an attempt to maximize immunomodulating properties of the n3 fatty acid family. Currently, there is a range of approximately 12:1 to 1:1 depending on the marketing focus. An overall "optimal" ratio has yet to be established, keeping in mind that the clinical value of dietary n6:n3 may well be disease specific. A second approach is to enhance diets with n3 fatty acids specifically, and thereby focus on the total amount of n3 FA rather than the ratio.

CARBOHYDRATES

Polysaccharides possess immunomodulatory properties as noted by the incorporation of LPS as a stimulating agent in various models of immunity. LPS-associated polysaccharides are not the common CHO source in diets; rather, they are the soluble CHO component of the antigen (eg, CHO, lipid, protein) associated with endotoxins. These endotoxins are localized on the outer surface of gram-negative bacteria and elicit specific antibody responses. Although the three components of endotoxin should potentially elicit antigenic diversity, evidence supports a primary immunodominant role for the specific polysaccharide component of LPS [42]. In the advent of bacterial translocation through compromised GI mucosa, the polysaccharide component of LPS-associated

bacterial endotoxins could indeed influence immunomodulation of the GALT. On a different note, Melis and coworkers [43] reported that intake of a CHO-rich beverage before surgery can prevent surgery-induced immunodepression. This is a potential avenue to reduce the risk of infectious complications and maintain barrier and GALT functions in stressed and potentially malnourished patients.

Dietary CHO can be viewed from the perspective of glycemic control, particularly in stressed, critically ill, diabetic, or obese patients. Associated risk factors for increasing the incidence of infection include improving preoperative nutrition, choosing the optimal route of nutrient delivery and type of nutritional supplementation, and tight glycemic control in patients through altering CHO intake. A landmark human study of 1548 patients requiring mechanical ventilation [44] showed that euglycemic patients had fewer episodes of acute renal failure, fewer transfusions, and less polyneuropathy. Compared with a hyperglycemic patient group, infectious complications were 46% lower and the mortality rate was decreased by 42%. Studies have repeatedly indicated the adverse effect of modest hyperglycemia on neutrophil function, including decreased chemotaxis, phagocytosis, oxidative burst, and bacteriocidal capacity [45]. Additionally, modest hyperglycemia has been shown to promote the proinflammatory state through increasing levels of the inflammatory mediator TNFα and activating NF-κB, which promotes production of TNF [46]. The article in this issue covering critical illness provides more details on dietary CHO control, especially during parenteral feeding.

MINERALS AND VITAMINS

These nutrients, although required in small concentrations in the diet, can be considered to be the metabolic glue that is behind nearly all anabolic and catabolic processes in the mammalian system. Electrolytes play key roles in maintaining cell structure, and thus flow of cell regulators. Trace minerals and vitamins alike facilitate complex metabolic reactions and are key components of antioxidant activities, which currently are receiving attention in the scientific arena as modulators of the immunologic components of health and disease.

Copper, Zinc, and Iron

Cu has a profound effect on many aspects of the immune system. Animal models of Cu deficiency have been concisely reported by Lukasewycz and Prohaska [47] and indicate that Cu is important for antibody generation (IgG), for cell-mediated immunity, and for the generation of the inflammatory response. Laboratory animals on a low-Cu diet have demonstrated an increased susceptibility to bacterial infections. Domestic animals with insufficient Cu intake show decreased bactericidal activity, impaired macrophage phagocytosis and MHC class II expression [48], and a higher susceptibility to infection [47]. Mice receiving a Cu-deficient diet showed a decreased reactivity to T- and B-cell mitogens, a low proliferative and delayed type hypersensitivity (DTH) response to stimulation, a redistribution of lymphocyte subsets, a decrease in

NK cell activity [47], and a reduced ability of T cells to produce sufficient quantities of IL-2 to allow cells to progress to the S phase [49]. Many of these aspects of altered immunocompetency have reportedly been reversed by the addition of adequate Cu. Ceruloplasmin, an acute-phase protein, and superoxide dismutase (SOD), an endogenous antioxidant that scavenges damaging ROS in immune cells, are Cu-dependent enzymes whose activity and concentrations reflects dietary Cu intake [47]. These enzyme functions help to define Cu as an integral component of phagocytic cell function.

Dietary Cu deficiency is uncommon in small animal nutrition when feeding most commercially formulated rations. Research strongly suggests that many nutrients, including Cu, may be required in excess of current dietary recommendations during critical illness to compensate for losses and increased use during acute or chronic disease challenge.

The discovery of nutritional zinc (Zn) deficiency in the pathogenesis of disease provided the focus for studies to elucidate the mechanism of Zn-induced changes in immunity. Zn is an important cofactor for numerous enzymes involved in cell metabolism, but deficiency of this mineral can also result in a profound immunodeficiency state. Several common hallmarks of Zn deficiency are lymphopenia and thymic atrophy. Along with atrophy, a decrease in thymic hormone activity is also observed. Deficiency of Zn results in depressed DTH reactions, decreased CD4 (Th) and increased CD8 (T-suppressor) cell populations, a reduced proliferative response to mitogens and NK cell activity, and decreased chemotaxis of monocytes and neutrophils [50]. Lymphopoiesis can likewise be influenced by Zn deficiency. Fraker and King [51] reported that a 30-day period of suboptimal Zn intake in adult mice caused a 40% to 90% depletion of marrow cells in the B-cell compartment. Zn is also a component of the antioxidant enzyme, SOD, which scavenges damaging ROS in immune cells, defining Zn as an integral component of phagocytic cell function and viability [52].

Fe is probably one of the most important trace elements in the body. It plays an important role in the transport of oxygen and in the oxidation-reduction pathways of many systems. Fe is intimately associated with malnutrition as a result of poor intake, poor absorption, or excessive losses. A negative Fe balance can initially lead to decreased tissue stores, a decreased blood hemoglobin concentration, and, finally, the appearance of microcytic hypochromic anemia. Lymphoid cells require Fe for cell division, electron transport, and oxidation-reduction reactions. Fe bound to transferrin is the mode of transport into immune cells. It has been reported that abnormalities in cellular morphology and function of red and white blood cells because of a negative Fe status are associated with decreased DTH reactions, reduced T-cell mitogen response, reduced lymphokine production by T cells, reduced antibody production and phagocytic activity, and enhanced susceptibility to infection [15,53].

Selenium and Vitamin E

Together selenium (Se) and vitamin E share a unique relation through interactions involving the antioxidant enzyme, glutathione-peroxidase (GSH-Px).

These interactions directly and indirectly influence immunity through their combined antioxidant functions. An in-depth discussion of antioxidants in health and disease is presented in another article in this issue. Aside from the antioxidant capacity of Se-dependent GSH-Px, this enzyme has been shown to alter immunity through its impact on lymphocyte differentiation; signal transduction; and regulation of proinflammatory cytokines, such as leukotrienes, thromboxanes, and prostaglandins [10,54]. There are five different forms of GSH-Px, which exhibit their protective functions in unison but at different sites in the body.

Based on recently reported studies, the form of dietary Se should perhaps be considered. The commonly used inorganic sodium selenite is actually capable of promoting $O_2^{-}\cdot$ formation, and ultimately enhancing oxidative stress [54]. Selenomethionine (Se-Met), an organic Se compound, is nontoxic and noncatalytic and does not produce the superoxide free radical [55]. Lymphocytes treated with Se-Met inhibited peroxyl radical formation in a dose-dependent manner [56]. Seo and coworkers [57] showed that Se-Met induced repair of damaged DNA and protected normal fibroblasts from oxidative damage at easily attainable in vivo diet supplementation levels. Synergistic effects of Se-Met with other antioxidants (ie, ascorbic acid, GSH, mannitol) were not appreciated in enhanced immune cell protection [58,59]. Waters and colleagues [60] reported that Se-Met or Se-enriched yeast lowered the DNA damage in prostate cells and peripheral blood lymphocytes of dogs; however, interestingly, the total activity of plasma glutathione peroxidase was not associated with DNA alterations.

Along with its well-defined role as an antioxidant, it has been reported that vitamin E may indirectly enhance immune factors. This effect is thought to be via inhibition of macrophage secretion of PGE_2, which suppresses IL-1 production, mitogen- and antigen-induced lymphocyte blastogenesis, antibody production, and cytotoxic T-cell activity [61]. A limited number of companion animal studies have evaluated the relation between dietary vitamin E concentrations and immunity. The studies reported by Meydani and coworkers [62] suggested that higher (280-IU/kg diet) dietary vitamin E helped to maintain lymphocyte proliferative activity as compared with lower (27-IU/kg diet) levels in Beagles. Diets supplying vitamin E at a rate of 250 to 500 IU/kg of diet seemed to provide immunologic benefit to older healthy cats [61].

Based on recent reports, perhaps the form of vitamin E supplied in the diet should be a consideration. γ-Tocopherol is the major form of vitamin E in many plants, but α-tocopherol is the predominant form of vitamin E in tissues and the form primarily used in supplements. Recent studies suggest that γ-tocopherol has unique physiologic features that may justify its importance in health and disease. The γ-form seems to be more effective in trapping lipophilic electrophiles compared with α-tocopherol. It is reported to be well absorbed and stored in appreciable concentrations in tissues. Urine is the major route of γ-tocopherol metabolite excretion, and this water-soluble metabolite seems to exhibit natriuretic activity [63]. Unlike the α-form, γ-tocopherol and its major metabolite (γ-carboxyethyl-hydroxychromans [CEHC]) inhibit COX activity and subsequently influence anti-inflammatory properties. Jiang and

coworkers [64] reported that γ-tocopherol and γ-CEHC inhibit PGE_2 synthesis in LPS-stimulated macrophages and in Il-1β–activated epithelial cells at significantly lower concentrations than α-tocopherol. The COX-2 inhibitory effects of the γ-form versus the α-form of vitamin E indicate a nonantioxidant property of γ-tocopherol that can be important in the prevention and management of disease, such as autoimmune disease, cancer, and type-1 diabetes. Based on reported properties of α- and γ-tocopherol as well as on the understanding that large doses of α-tocopherol deplete plasma and tissue γ-tocopherol, possible immunomodulatory benefits of γ-tocopherol, the dietary tocopherol form, should be given consideration in nutritional support programs.

Vitamin C

This vitamin was mentioned previously for its role in recycling and reactivating vitamin E. It has more direct implications in immunity as well. Decreased vitamin C (ascorbic acid) is known to be associated with depressed cell-mediated immunity, poor bactericidal activity, and impaired macrophage mobilization. Supplementation with vitamin C enhances T- and B-cell proliferation and bacterial phagocytosis by macrophages [65].

Vitamin A

There are numerous well-documented vitamin A deficiency symptoms, including immune function abnormalities. Deficiency of this vitamin impairs secretory IgA production, decreases mucus production (a component of the innate immune system), and leads to keratinization of secretory epithelia. When vitamin A is bound to RBP and chylomicron remnants, it modulates normal B-cell activation, cytokine production, antibody production, and cell differentiation. β-Carotene, a precursor to vitamin A, has been shown to enhance T-cell and B-cell generation in animals [65].

B-Vitamins

The B-vitamins are generally considered as a "complex," (Table 4) except in specific disease-associated deficiencies, such as small intestinal bacterial overgrowth (SIBO). They all are equally necessary to drive intermediary metabolism in the body; however, with regard to having a specific impact on immune function, there is one B-vitamin in the complex that should be mentioned, pyridoxine (B_6). Deficiency of this B-vitamin is associated with impaired cell-mediated and humoral immune responses. Human beings with poor B_6 intake have been reported to exhibit a decrease in circulating lymphocytes, reduced IgD levels, and a decreased percentage of Th cells [65]. In the human literature, it is suggested that B_6 may be needed in excess of the recommended daily allowances to maintain adequate immune function in compromised patients.

OTHERS

Nucleotides

Nucleotides, which are the precursors of DNA and RNA, can be considered to be the building blocks of life. The supply of nucleotides needed for biochemical

Table 4
Summary of immunomodulatory capabilities of minerals and vitamins

Nutrient		Immunomodulation
Minerals	Iron	Necessary for optimum neutrophil and lymphocyte function; free iron is necessary for bacterial growth
	Copper	Deficiency associated with increased rate of infections, depressed RES and microbicidal activity of granulocytes, impaired antibody response, and depressed thymic hormone; component of the antioxidant enzyme SOD, scavenges immune cell-damaging ROS
	Zinc	Deficiency associated with susceptibility to infection, abnormal cell-mediated immunity, depressed circulating thymic hormones, and altered complement and phagocytic function; component of the antioxidant enzyme SOD, scavenges immune cell-damaging ROS
	Selenium	Deficiency reduces antibody responses; component of the antioxidant enzyme GSH-PX, scavenges immune cell-damaging ROS
	Iodine	Decreased microbicidal activity of neutrophils in hypothyroid patients, and a reversal seen with treatment
	Magnesium	Deficiency causes thymic hyperplasia, impaired humoral and cell-mediated immunologic responsiveness, depressed immunoglobulins (IgG1, IgG2, IgA)
	Manganese	Required for normal antibody synthesis/secretion; excess inhibits antibody formation and chemotaxis and increases susceptibility to pneumococcal infection
	Sodium	Brush border cells in the gastrointestinal tract are dependent on sodium for transport of glutamine, which is pivitol in maintaining an intact gut barrier
Vitamins	A	Deficiency reduces lymphocyte response to mitogens and antigens; β-carotene and retinoids stimulate immune responses
	B complex (B_6)	Deficiency associated with decreased antibody response and impaired cellular immunity
	C	Extreme deficiency impairs phagocyte function and cellular immunity
	D	Deficiency causes anergy in the delayed hypersensitivity skin test
	E	Deficiency decreases antibody response to T-cell–dependent antigens; effect is compounded by a Se deficiency; supplementation has been shown to enhance immune responses

Abbreviations: GSH-PX, glutathione peroxidase; RES, reticuloendothelial system; ROS, reactive oxygen species; Se, Selenium; SOD, superoxide dismutase.

processes is mainly supplied through de novo synthesis of purines and pyrimidines. Dietary intake is a secondary source during health but can become of major importance during illness. Animal studies have indicated that a dietary requirement of preformed purine or pyrimidine bases may be required for normal development. Babineau [21] has summarized numerous studies of Van

Buren, Kulkarni, and others in investigating the role of nucleotides in immune function. Uracil seems to be the most important nucleic acid influencing the immune response. Delayed cutaneous hypersensitivity, mitogen-stimulated lymphocyte proliferation, graft rejection, and graft-versus-host disease have all been reported as being suppressed by a diet that lacks nucleotides. Van Buren and coworkers demonstrated that dietary nucleotides were able to reverse malnutrition- and starvation-induced immunosuppression in rats. They also showed that helper/inducer T lymphocytes require exogenous nucleotides to respond normally after immune stimulation. Another study demonstrated that dietary nucleotide restriction negatively influenced the phagocytic cell response to a bacterial sepsis challenge in mice. These studies specifically emphasize the role of nucleotides in lymphocyte and macrophage function and metabolism, although dietary sources are equally as important to support the optimal growth and function of other metabolically active cells, such as intestinal cells.

GUIDELINES WITH A DISCLAIMER

Nutrient guidelines are available for feeding the healthy animal during various life stages and phases of production and/or reproduction. To date, a comprehensive set of recommendations for nutrient support of the immune system does not exist for small or large animals. It may be possible to extrapolate from human or animal studies, but overfeeding a specific nutrient can just as easily diminish one or more aspects of immune function as enhance it. In many circumstances of immune function challenge or malnutrition, increasing a specific nutrient over maintenance recommendations is beneficial to the immune response. The problem lies in exactly how much to enhance. At present, a wiser and safer approach may be to prevent deficiency of identified immunomodulating nutrients in your nutritional support plan. That said, some general guidelines for specific nutrients have been summarized for consideration based on extrapolation from the current literature. The reader should consider these guidelines to be generic, however, because the optimal dose remains controversial. Nutrition support plans need to be individualized based on a thorough and continual assessment of each patient.

Suggestions for clinical use of GLN in the human literature include two categories: (1) critically ill patients can be supplemented with GLN at a rate of 30 to 50 g in addition to the standard enteral diet, with a goal to achieve GLN at a rate of 0.35 to 0.65 g/kg of BW per day, and (2) pre- or postsurgical patients can be supplemented with GLN at a rate of 25 to 50 g in addition to the standard enteral diet, with a goal to supply GLN at a rate of 0.30 to 0.65 g/kg of BW per day. The dose of a powder formula for the small animal patient is reported to be 10 mg/kg/d. A suggested dose of GLN supplementation to PAs for immunomodulation is 2% of the total daily PA as L-GLN. The suggested goal for dietary arginine intake of small animal patients with cancer would be approximately 500 to 600 g per 100 kcal/d. Although values are not available for managing patients with severe sepsis or inflammation, studies suggest a 1.5- to 2.5-fold increase in dietary intake over the standard enteral diet.

Current recommended oral doses for vitamin E in the α-tocopherol isomeric form are in a range of 400 to 500 IU/d for inflammatory disease. This is a dose 2 to 10 times greater than the daily requirement for dogs. Earlier studies suggested that a diet supplying vitamin E at a rate of approximately 250 to 500 IU/kg of diet may help to maintain lymphocyte proliferative activity in healthy cats and dogs. The suggested dose of γ-tocopherol, based on scientific literature, is 50 mg/kg. There are no specific recommendations reported regarding the choice of isomeric form (α or γ) for immunomodulation during critical illness or disease states in small animals.

Reported levels of total n3 fatty acids in commercial diets range between 0.20% and 7.3% dry matter basis (DMB). An initial total dose of n3 fatty acids at 50 to 250 mg/kg of BW per day seems to be effective in a large number of studies for its anti-inflammatory effect. Studies suggest a dietary n3 fatty acid concentration of 7.3% DMB (1348 mg per 100 kcal) as most beneficial for overall management of the canine cancer patient. A dietary n6:n3 fatty acid ratio of 1:1 or less is reported to reduce tumor marker expression significantly and to inhibit tumor growth in feline models.

SUMMARY

The complexity of the immune system allows for a multitude of potential avenues for nutrient modulation, but this also increases the challenge of producing a predictable in vivo response. Numerous studies have attempted to evaluate the clinical usefulness of specific nutrient supplementation as well as the benefit(s) of nutrient-enriched diets in modulating immunity. Because the immune response is a cascade of biologic events, development of nutritional support paradigms cannot and should not be made in a vacuum or with the expectation of a singular response. It is absolutely imperative that the clinician/nutritionist understand the differences in metabolic and physiologic responses to disease states (ie, shock, trauma, organ-specific dysfunction) so as to maximize immunocompetence through specialized feeding practices.

More is not always better, especially when it come to immune enhancement. Timing, dosage, and duration criteria of nutritional immunomodulation need to be identified for specific disease states rather than making blanket recommendations. This takes further evaluation of nutrients, diets, and disease scenarios. Much of the human nutrition research takes this stepwise approach, and although progress is sometimes slow, nutrition support recommendations for disease states and health seem to be mechanism based. This level of understanding is invaluable, especially when considering the possible benefit of nutrient combinations for immunomodulation.

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