Neutrophils, one of the three types of granulocytic white blood cells (leukocytes), are the hallmark of acute inflammation. They serve as key components in the defense against infection, and are the most abundant of white blood cells, accounting for ~60% of all leukocytes. They are primarily associated with acute bacterial inflammation1 and their reaction time is immediate, typically within one hour of tissue injury. Neutrophils are phagocytes, capable of ingesting microorganisms or particles. Each phagocytic event results in the formation of a phagosome into which reactive oxygen species and hydrolytic enzymes are secreted. Due to the consumption of oxygen during this event, a “respiratory burst” ensues. This “respiratory burst” in turn activates the enzyme NADPH oxidase, resulting in the production of large amounts of superoxide. The superoxide is subsequently converted to hypochlorous acid (HOC) via the enzyme myeloperoxidase, which is presumed to result in the elimination of the phagocytized bacteria.

The other two classes of granular leukocytes are the eosinophils and basophils. Eosinophils are activated during allergic diseases, infections, or other medical conditions, and elevated levels are associated with an allergic response or with parasitic infestation. They contain coarse, cytoplasmic granules of uniform size, and make up 1-3% of the total circulating leukocytes. Basophils have a nuclei size similar to eosinophils, however, when activated they degranulate and release cellular components, including histamine and proteoglycans, stored within the granules, along with proteolytic enzymes.2 They represent approximately 0.01-0.3% of the circulating leukocytes.

All leukocytes have a minimal life span, thus have a high turnover rate. As such they are extremely vulnerable to mineral, vitamin and antioxidant deficiencies. The complex interplay between multiple cell types and intercellular messengers makes nutritional status both subtle and far-reaching, particularly when it involves immune activation.

Minerals

Chronic low-grade type infections, associated with neutrophil influx, are characteristic of number of disease processes, ranging from periodontal infections and asthma to cardiovascular disease, pulmonary complications and rheumatoid arthritis. Certain minerals play important roles in supporting optimal immune function. Zinc, copper and calcium are especially important in this role.

Zinc (as zinc gluconate): As an essential cofactor for over 70 enzymes, zinc is a vital component in immunocompetence. Zinc is particularly important for highly proliferating cells, including those of the immune system. Overt signs of zinc deficiency are numerous and have been well documented. They include atrophy of the thymus, spleen and lymph nodes; decreased or delayed hypersensitivity response and allograft rejection; lowered production of B and T-lymphocytes; decreased natural killer cell activity, decreased phagocytosis, as well as decreased thymic hormone activity.3,4 A deficiency in zinc results in a rapid and extensive effect on the immune system, including decreased function of monocytes and macrophages, decreased phagocytosis of the neutrophil granulocytes, and decreased cytotoxicity of natural killer cells.6 Additionally, the number and activity of NK cells have been shown to be dependent upon the level of serum zinc.7 Zinc malabsorption is evidenced by poor wound healing, as well as an increased susceptibility to infections. Short periods of zinc supplementation have shown to substantially improve immune defenses, particularly in children and the elderly, and in individuals with certain diseases, including chronic gastrointestinal disorders.8

Copper (as copper gluconate): Anemia, and neutropenia are two known hematological manifestations of copper deficiency. The phagocytic capacity of the Neutrophil is markedly modified in copper insufficiency.9 Neurological manifestations of copper deficiency have also been observed, a typical presentation being myelopathy.10 Deficiencies in copper, in conjunction with other vitamin and mineral deficiencies, have been correlated with a loss of thymic cellularity, which results in diminished T lymphocyte differentiation. Subsequently, a maturational defect in T lymphocytes ensues, observed by a decrease in both total T cells (T3 and rosette-forming T cells), and in T4 helper/inducer cells. In mice deficient in copper, iron and zinc, cytotoxic T lymphocyte (CTL) activity is impaired. In vitro studies have also reported a reduced number and function of T cells with experimental deficiencies in copper, zinc, iron, vitamin E and vitamin A.11

Calcium (as calcium glycerophosphate): Calcium mobilization plays a critical role in the activation of cytokine gene expression in helper T cells, as it is an integral part of calcineurin, the calcium-dependent phosphatase, which is essential for the activation of cytokine gene expression in helper T cells. Calcineurin activation is required for lytic granule exocytosis in cytotoxic T lymphocytes.12 Intracellular calcium signals (Ca²⁺) also play an essential role in the signaling of Interleukin-8 (IL-8), which is an important constituent in neutrophil activation.13

Phosphorous (as calcium glycerophosphate): Cellular phosphorous is closely linked to calcium, typically referenced as being between the ratio of 1:2 - 2:1, calcium to phosphorous. Phosphorous is an essential constituent of nucleic acids, ATP and phospholipids. Studies have demonstrated an association of the phospholipid phosphatidylinositol with the activated neutrophil.14 In animal studies the effect of dietary phosphorous on inflammation has also been demonstrated. As an example, in a study with pigs, the effect on cellular and humoral immune response was assessed when associated with an inflammatory challenge. A linear increase in the average daily gain (P<0.02) was associated with an increase in dietary phosphorous, which was correlated to enhanced cell-mediated immune response, with a corresponding reduction in humoral response.15

Vitamins

Vitamin A (as palmitate and mixed carotenoids). Vitamin A provides immunosupport via its action on cellular immunity in response to challenges, as well as by means of its role in the support of mucosal cell surfaces.16 It also aids in maintaining

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the integrity of lymphatic tissues, and the level of antibodies, especially that of secretory IgA. Antibody production is affected by hypovitaminosis A, evidenced by a 55% reduction in NK cell activity (\(P<0.05\)) in animals presented with an immune challenge. Vitamin A repletion restored function either partially or completely. Additionally, when spleen cells were assessed, the deficiency in vitamin A was correlated to significantly less interferon production (\(P<0.05\)), theoretically implicating a decreased immune response and an increased susceptibility to disease.\(^{15}\) In pediatric patients with irritable bowel disease (IBD), low vitamin A status (<20 mcg/dL) was determined to be a common occurrence (16% of the population), which was correlated to the severity of the disease.\(^{16}\) In adults with IBD, in addition to other deficiencies a 26% inadequacy in Vitamin A was observed.\(^{17}\) Deficiency, however, appears to be only partially alleviated by the by the consumption of dark green, leafy vegetables,\(^{20}\) which has been correlated to a low bioavailability of vitamin A in fruits and vegetables.\(^{21}\) Natural mixed carotenoids containing alpha and beta carotene, lutein, zeaxanthin seem to be more readily absorbed and to be more effective antioxidants than synthetic (all trans) beta carotene.\(^{22}\)

### Vitamin C (as mixed ascorbates)

Vitamin C is a potent water-soluble antioxidant and functions as an active electron donor and acceptor.\(^{23}\) It dynamically participates in immune support, as it acts as both an anti-inflammatory mediator and an immunomodulator. The neutrophil is a known concentrator of vitamin C, increasing its intracellular concentration as much as 10-fold upon activation.\(^{24,25}\) Neutrophils have also been designated as ascorbate recyclers, having the capability to enhance their intracellular vitamin C concentration as much as 30-fold in the presence of microorganisms, which was correlated to extracellular ascorbate concentration.\(^{26}\) Ascorbic acid (2g/d for 5 days) supplementation has demonstrated to significantly increase the cellular ascorbic acid content of both granulocytes and platelets.\(^{27}\) Conversely, with a deficiency in ascorbate a defect in the clearance and apoptosis of macrophages has been observed, which was correlated to macrophage recognition inability, implicated as a “novel and important function for vitamin C in inflammatory cells”. The mechanism ascribed was the upregulation of the hypoxia-inducible factor-1\(\alpha\) (HIF-1\(\alpha\)).\(^{28}\) This mechanism has also been attributed to the depletion of ascorbate by nickel(II) and cobalt(II).\(^{29}\)

### Vitamin E (as d-alpha tocopheryl acetate & mixed tocopherols)

Vitamin E acts as a potent oxygen free-radical scavenger, and has shown to be protective against injury to the gut mucosa, useful in minimizing oxidative stress, and has been shown to provide significant therapeutic benefit to the elderly.\(^{30,31}\) It is also the major membrane and lipid antioxidant of the body. In healthy individuals, short-term vitamin E supplementation was shown to improve immune responsiveness, as evidenced by a decrease in lipid-peroxidation products, including PGE2.\(^{32}\) Supplemental vitamin E has also been associated with a reduced incidence of respiratory tract infections in the elderly,\(^{33}\) and has shown to significantly reduce both the incidence and number of common colds in the elderly.\(^{34}\) Animals deficient in vitamin E were shown to have a 90-fold depletion in alpha-tocopherol, which was correlated to a significant decline in antioxidants, as well as to the accumulation of lipid peroxidation products, both of which were associated with a greater incidence of inflammation.\(^{35}\)

### Nucleotides and Nutrition

**RNA:** Nucleotides along with their metabolites are important to many bodily processes, and have documented efficiency in optimizing function. Nucleotides are considered essential for both cell-mediated immunity and T-lymphocyte function.\(^{36,37}\) The need for dietary nucleotides is particularly evident in times where there is a high physiological demand, such as rapid growth, metabolic stress, recovery from a major surgery or trauma, or with inadequate liver function.\(^{38}\) An adequate supply of nucleotides in the form of purines and pyrimidines, which comprise RNA and DNA, allows for rapid cell proliferation and protein synthesis. In tissue culture studies coculturing nucleotides with specific antigens was demonstrated to have an influence on both immune cell growth and cytokine secretion.\(^{39}\) Other studies have indicated that cellular immunity is significantly depressed when animals are maintained on a nucleotide-free diet.\(^{40}\) When reversed a nucleotide-supplemented diet was shown to upregulate Th1 immune response via the enhancement of IL-12 production, with a noted corresponding suppression of antigen-specific IgE response. This was correlated to a significantly higher production of antigen-specific interferon-\(\gamma\) by spleen cells.\(^{41}\)

### Botanical Support

**Maitake mushroom (Grifola frondosa) (aerial part):**

Maitake, an edible mushroom, is a source of complex carbohydrates (glucans), polysaccharides, and minerals. A standardized beta-glucan polysaccharide (beta-1,6 glucan and beta 1,3 glucan) from maitake, termed the D-fraction, has been extensively studied. In these studies administration has demonstrated immunomodulatory effects, including enhanced humoral immunity, increased production in nitric oxide, interleukin (IL) 10,\(^{42}\) and IL-12, which resulted in enhanced cytotoxicity of NK cells.\(^{43}\) Others have noted an increased NK cell activity with maitake intake.\(^{44}\) In a separate study animals given a 20% maitake fortified diet were observed to have an altered lipid metabolism, which was attributed to both inhibition of lipid accumulation and deterrence of lipid elevation.\(^{45}\) These observations correlated with previous studies, which also noted a beneficial effect of maitake on lipid metabolism.\(^{46,47}\) Maitake has also shown favorable outcomes in animals studies of hypertension,\(^{48}\) diabetes mellitus,\(^{49,50,51}\) and diabetes mellitus.\(^{49,50,51}\) In one animal study administration of maitake, along with vancomycin resulted in macrophage activation and a 2.7 fold increase in the production of IL-1\(\beta\). Enhanced bactericidal activity of splenic T cells was also observed, denoting a 2.6 fold increase in activity with maitake intake, as compared to nontreated cells.\(^{52}\) Both observations indicate maitake’s potent action on immunocompetent cells.

**Chrysanthemum morifolium (flower) (extract):**

Although traditionally used as a component in drink, specifically tea, the flower portion of Chrysanthemum has documented medicinal benefits. The flower contains flavonoids, amino acids, vitamins and trace elements, as well as caffeoylquinic acids,\(^{53}\) luteolin and apigenin, the latter two being ascribed as the primary bioactive components.\(^{54}\) Evidence of the beneficial attributes of *Chrysanthemum morifolium* (Cm) was demonstrated in one *in vitro* study, in which cardioprotective effects were observed in isolated rat heart following ischemia/anoxia and reperfusion/reoxygenation. *Cm* was shown to have a protective effect on the ventricular myocytes by virtue of its attenuation of the reduction of left ventricular pressure and...
coronary flow caused by ischemia/reperfusion. The Chinese literature documents Cm for prevention of sore throat and promoting fever reduction, when drunk as a tea. The Chinese Materia Medica also indicates its effectiveness against Staphylococcus aureus, B-hemolytic Streptococcus and Shigella sonnet.

Loquat (Eriobotrya japonica) (leaf) (extract): In an animal study administration of Eriobotrya japonica (Ej) was demonstrated to exert a significant hypoglycemic effect, as evidenced by a lowered blood glucose level in both normal or/ and alloxan-diabetic mice. Ej is known to contain triterpene acids, and in a separate study the inflammatory response of experimentally induced chronic bronchitis was investigated. Animals given Ej were noted to have a significantly decreased level of inflammatory cytokines, including TNF-α, IL-1, NF-kB, PGE2 and {leukotriene B4 LT(B4)} expression, as compared to the control group. The investigators concluded that Ej “inhibited NF-kB activation” and “led to downregualtion of TNF-alpha, IL-1, PGE(2) and LT(B4) expression” in a dose dependent manner, thus demonstrating immunosupportive properties.

Dyer’s-Woad (Isatis indigotica) (root) (extract): Isatis has noted antibacterial actions, demonstrating effectiveness against strains of staphylococci, pneumococci and meningococci. It is also considered an effective agent against viruses, including the influenza virus. Its actions are described as antipyretic, anti-inflammatory, and it demonstrates choleretic actions.

Prickley Ash (Zanthoxylum americanum) (bark) (extract): In traditional Chinese medicine, Zanthoxylum is utilized to increase blood flow, and to promote the circulation of qi. Its actions are considered warming and stimulating, thus benefiting circulation. The native Indians of North American have long utilized it for rheumatism and toothaches (odontalgic). The bark is considered an irritant and demonstrates antirheumatic properties. Both the roots and bark have been utilized as a tonic in debilitating conditions of the stomach and digestive organs.

Thyme (Thymus vulgaris) (leaf) (extract): Thyme’s medicinal properties are associated with its actions as a bronchial antispasmodic, an antibacterial agent and an expectorant. The chief components of thyme are thymol (20-55%) and carvacrol, along with other minor ingredients. Carvacrol has demonstrated effectiveness as both an antimicrobial, as well as an antifungal agent.

Mullein (Verbascum thapsus) (leaf) (extract): Verbascum’s use is correlated to its effectiveness as an expectorant, in stimulating the expulsion of phlegm, and in association with reducing mucus formation. Its classical use is in the management of tracheitis and bronchitis. The leaves, along with the flowers are claimed to be anodyne, anti-inflammatory, antiseptic, antispasmodic, astringent, demulcent, diuretic, emollient, expectorant and vulnerary. Extracts contain various derivatives, including arabinogalactans, iridoids as (catalpol derivatives), saponins (verbasosaponin), flavonoids (kaempferol, luteolin, rutin, apigenin) and phenolic acids (caffeic acid, ferulic acid). In folk medicine Verbascum has been utilized in supporting wound healing, bronchial function, and immune responses, perhaps as a function of the antioxidant activity of the flavonoids and phenolic acids.

Phragmites communis (root) (extract): Common compounds isolated from Phragmites, also called Reed Herb, include tricine, luteolin, chrysosorol, rutin and isocherizin. The root properties are noted as being beneficial in asthma, as an aide for nausea and vomiting (antiemetic), as an aide in reducing fevers, as a cough suppressant (antitussive), as a cleansing agent, as a diuretic, as a fever reducer (febrifuge), as an aide in the preventing stone formation (lithotriptic agent), as an aide in increasing the flow of saliva (sialogogue), as a stomach toner (stomachic), and as a sedative. It is traditionally used for diarrhea, fevers, cough, vomiting, coughs with phlegm, urinary tract infections and food poisoning, especially from sea foods.

Barberry (Berberis vulgaris) (bark) (extract): Berberis is a source of vitamin C and isooquinoline alkaloids, including among others berberine, berbamine, and oxyacanthine. It is documented to have antipyretic and cholangine effects, and the root bark has been shown to aide in normalizing blood pressure. Other properties attributed to the bark include antiseptic, astrigent, cholangine, hepatic, purgative, stomachic and tonic qualities. Berberine has also shown effectiveness as antiadrialler agent.

Oregon Grape Root (Mahonia aquifolium) (root) (extract): The chemical components of Mahonia aquifolium consists of isooquinolin and isooquinoline derivates, including among others berberine, berbamine, and oxyacanthine. The traditional use of Ma by North American Indian tribes was for the treatment of appetite loss and debility. Its use has also been found to be effective in treating gastroenteritis, stomachic and antiseptic. Its immunosupportive properties have been attributed to its synergistic antibacterial, anti-inflammatory and bile-stimulating characteristics. Earlier studies demonstrated that Ma exhibited a strong ability to quench the superoxide anion radical, contrary to a strong radical scavenging ability. Recently, Ma was demonstrated to have an inhibitory effect on lipoygenase, indicating its potential role in modulating inflammatory processes.

The combination of the above mentioned components in Neutrophil Plus are intended to provide general support for the immune system, designated specifically for upper respiratory tract symptoms. It is specifically designed to support the immune response.

Contraindications: Not recommended for pregnant or lactating women.

References
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