NUTRITION AND NEURODEVELOPMENTAL DISORDERS IN CHILDREN: AN OVERVIEW

Attention deficit/hyperactivity disorder (ADHD) and autism spectrum disorder (ASD) have been reported as having reached epidemic proportions[1,2]. In response, the medical community has recognized that a comprehensive nutritional supplement program can play a pivotal role in the health and functioning of children diagnosed on this spectrum of neurodevelopmental disorders, from mildly disruptive behavior to the far more severe problems of autism[3].

ADHD is the most common behavioral disorder in children, manifesting as a cluster of abnormalities including attention deficit/inattention, impulsivity, and hyperactivity, all of which impair the child's ability to function[4]. Autism, on the other hand, is characterized by dysfunction in social interaction and communication and is associated with restrictive, repetitive, and stereotypic behaviors. The exact cause of these disorders has not been clearly established; however, ongoing research suggests that biological and/or environmental factors may be associated with their onset[5-7].

The full spectrum of neurodevelopmental disorders include attention deficit disorder (ADD), ADHD, learning disability (LD), pervasive developmental disorder (PDD), and autism. Some clinicians do not see these conditions as discrete entities, recognizing them instead as a continuum of disorders with related features. The work of Greenspan and Wieder, for example, suggests that each of these conditions share similar causes (etiologies), common signs, and common symptoms as well as responsiveness to common treatment approaches[8]. These authors have laid out a framework, represented in Figure 1, which graphically depicts the relationship among these conditions.

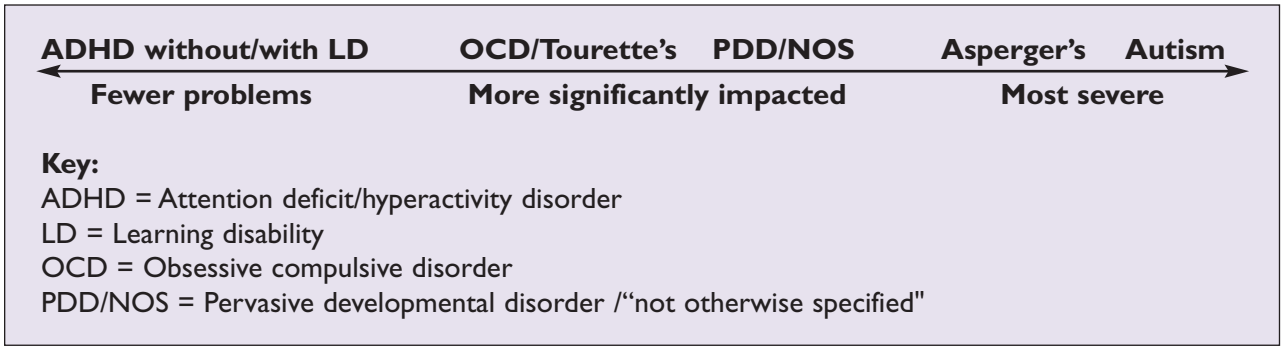

Figure 1: Spectrum of Childhood Behavioral and Neurodevelopmental Disorders

As noted in the figure, ADHD occurs on the lesser end of the spectrum, and PDD and autism are positioned at the more severe end. (The location of each disorder on the continuum depends specifically upon the severity of its associated symptoms.) These conditions are both complex and multi-factorial. At the same time, symptoms of the various conditions overlap in many children; for example, they often share such features as immune disorders, allergies, food sensitivities, and slowing of brain activity as indicated by an electroencephalogram (EEG). Some children share symptoms from both ends of the scale: A child with autism may also be significantly hyperactive or a child with ADHD may show marked obsessive and restricted interest traits as well. **However, it's important to note that each disorder is distinct and experts agree that ADHD is not autism despite some shared features.**

Nutrition and Neurodevelopmental Disorders in Children: An Overview

Treatment Approaches

The traditional medical approach to treating neuro-developmental disorders uses pharmacotherapy to provide symptom control. Unfortunately, many of these medications have provided inconsistent, and in some cases, limited benefit to the individual child. For example, side-effects often limit their usefulness and growth can be impaired by stimulants[9]. In addition, because a growing body of evidence suggests that several possible causes or contributors may trigger or perpetuate these developmental disorders[10-12], there is a growing interest in a multidisciplinary treatment approach. Indeed, clinical studies are showing that such a multifaceted approach may be helpful[13,14].

All these complexities of the spectrum of conditions is challenging the medical community to look at a number of alternative and complementary therapies, referred to as "biomedical treatments," to complement or, in some cases, replace the psychotropic drugs commonly used in these disorders, in conjunction with behavioral modification therapies. Of special interest are recent reports and scientific documentation that indicate that implementing dietary restrictions and nutritional supplementation can bring about a noticeable reduction of symptoms in many of these children[15-17].

The common threads interwoven throughout the spectrum of attention deficit and autism disorders that are alluded to above provide an understanding of what therapeutic approaches may be of help. Clinicians have pointed to the presence of the following: nutrient deficiencies; fatty acid abnormalities; hyper-sensitivities or intolerances to food; adverse responses to food additives, preservatives, artificial colors and flavorings, sulfites, salicylates and phenols; all as co-existing problems in many of these disorders[18-20]. Gastrointestinal issues, including inflammatory and non-specific colitis, have been found in many autistics, and recently, similar intestinal findings have been seen in children with ADHD[21]. In a specific example, researchers suggest that incomplete digestion of wheat and other gluten-containing grains as well as milk/dairy products can be linked to behavioral symptoms recognized in those with developmental problems[10,22].

Many of these ADD, ADHD, and autism conditions appear to respond to the use of biomedical approaches, which include:

- Nutritional supplementation with a full spectrum of supportive nutrients
- Dietary modification with avoidance of casein from dairy foods and gluten from wheat products as well as elimination of potentially reactive foods
- Digestive enzyme supplementation to support documented deficiencies in digestion
- Nutritional support for central nervous system development and function
- Use of detoxification protocols and nutritional support of detoxification pathways
- Nutritional support of the immune system to counteract the immune dysregulation often seen in children with ADHD/ASD
- Correction of intestinal dysbiosis from potentially pathogenic bacteria, yeast, or parasites
- Lessening of exposure to environmental chemicals/ pollutants and neurodevelopmental toxins such as heavy metals

Nutritional intervention, in conjunction with other treatment modalities, offers a safe and clinically effective means of managing and supporting children with neurodevelopmental disorders. Further studies are warranted in order to appropriately evaluate the potential effectiveness of nutritional support in these individuals. However, clinicians implementing comprehensive nutritional protocols have described marked improvements in the function and behavior of many children with attention deficit and/or autism-related conditions.

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The Importance of Nutrition and Neurodevelopmental Disorders in Children

According to numerous studies, the diets of American children do not meet standards set by the US government. In a study of 3307 children, ages 2 to 19, researchers found that only 30% met the recommendations for fruit, grain, meat, and dairy standards, and only 36% met recommendations for vegetables. Only 1% of children's diets met all dietary requirements[1]. In the Bogalusa Heart Study of over 500 young adults, researchers found that intakes of vitamins A, B6, E, D, and C, folacin, magnesium, iron, zinc, and calcium were inadequate compared with the Recommended Dietary Allowances (RDA), with more females than males reporting nutrient intakes less than two-thirds of the RDA[2].

Nutrition plays a direct role in cognition and behavior in children and adolescents. Diet influences every aspect of neurological development, from formation of structural components to neurotransmitter production to neurotransmission activity. Animals exposed to early malnutrition demonstrate lasting changes around emotion, motivation, and anxiety; and these changes are known to profoundly affect all aspects of behavioral functioning, including cognition[3]. While it used to be thought that alterations caused by malnutrition could be reversed, it is now understood that alterations in brain neural receptor function resulting from early malnutrition are long-lasting, if not permanent[4]. A recently published longitudinal study of a birth cohort of 1559 children reported that children categorized as malnourished at the age of 3 demonstrated poorer cognitive function and scholastic ability at age 11, independent of psychosocial adversity[5]. Even if gross malnutrition is not present, subtle changes in diet may modulate brain function[6]. To understand the role chronic, mild malnutrition plays in behavior and development, it is necessary to move beyond protein/calorie deficits[7] to consider the role of the intake of micronutrients such as iron, zinc, and B complex vitamins. Chronic, mild malnutrition appears to be an essential factor in behavioral deficits[8].

Both vitamins and minerals are essential to brain function. Vitamins function in numerous roles, including—but not limited to—antioxidant activity, prosthetic groups bound to apoenzymes, and the production of adenosine triphosphate (ATP). Supplementation has been shown to improve both cognition and test scores[9-12]. Mounting evidence indicates that nutritional supplementation can play a valuable role in the treatment of children who have a diagnosis of attention deficit disorder or autism spectrum disorder. An open survey of parents with autistic children found that parents supplementing their children with vitamin C, folic acid, vitamin B6, magnesium, calcium, zinc, niacin, niacinamide, and dimethylglycine reported improvements in 41-58% of cases as compared to 1-8% responding that their supplemented children had worsened symptoms[13].

In a study of 23 autistic, 12 learning disabled, and 16 control children ages 4 to 13, low circulating levels of **vitamin B6** (pyridoxal phosphate) (<30 pmol/ml) were found in 15% of controls, 27% of learning disabled, and 42% of autistic children[14]. In addition, the many studies published since 1965 on the role of vitamin B6 in autism concur that supplementation with B6 may benefit about one-half of all autistic children and adults[15].

Vitamin C is essential to brain function, both as an antioxidant and as a coenzyme. It plays an essential role in metabolism and enhances the absorption of iron, and it is essential for the production of both serotonin and dopamine[16]. Researchers in the field of autism have demonstrated both *in vitro* and *in vivo* that vitamin C can function as a dopaminergic neuromodulator[17]. Vitamin C supplementation may help reduce certain behaviors in autistic children. One double-blind, placebo-controlled crossover study investigating the effectiveness of therapeutic doses of supplemental vitamin C (8 grams per 70 kg body weight/day) in school-age autistic children reported decreased stereotypic behaviors in children who received ascorbic acid[18].

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Vitamin C, **vitamin A**, **vitamin E**, and **beta-carotene** all function as antioxidants. In the lipid phase, tocopherols, carotenes, vitamin A, and ubiquinols are protective; in the aqueous phase, ascorbate (vitamin C) is protective. Recent clinical trials with autistic subjects have shown that autistic children have low activity levels of blood antioxidant enzyme systems[19] and that increased oxidative stress and altered enzymatic antioxidants may be relevant to the pathophysiology of autism[20].

Iron serves primarily as an oxygen shuttle to all cells and is involved in those enzymes that participate in cellular respiration. Children with iron deficiency anemia have been found to exhibit reduced scholastic performance, sensorimotor competence, attention, learning, and memory[21]; and there is evidence that mental and motor developmental test scores are lower among infants with iron deficiency anemia. Research has shown that children with iron deficiency anemia in infancy continue to have poorer cognition, academic achievement, and more behavioral problems into middle childhood[22,23]. Alterations in attentional processes also may be associated with iron deficiency as well as lower developmental IQ and achievement test scores. Failure to respond to test stimuli, short attention span, unhappiness, increased fearfulness, withdrawal, and increased body tension are also linked to iron deficiency anemia[24,25]. Further, studies show that iron replacement therapy has immediate (within 14 days) and long-term (over 3 months) beneficial effects on behavior and psychomotor development; levels of neurotransmitters such as noradrenaline, serotonin, and dopamine are altered during iron deficiency[26].

Investigation into the utility of **magnesium** and **vitamin B6** supplementation for children with autistic behaviors began over three decades ago. Supplementation with magnesium and vitamin B6 is proven to be efficacious; however, questions exist around dose amounts for these nutrients. Upon systematic analysis of the multiple studies investigating the role of magnesium and vitamin B6 with children with autism, researchers suggest that moderate amounts of magnesium and vitamin B6 are warranted and recommended but mega-doses are not[27,28]. In addition, supplementation with the activated coenzyme form of vitamin B6 (pyridoxal-5′-phosphate) is thought to provide metabolic advantage for optimal utilization.

Zinc-dependent enzymes are essential for the production of several neurotransmitters. For example, glutamate is a primary transmitter among the excitatory neurons in the cerebral cortex, and at least one glutaminergic neuron is known to accumulate zinc at axon terminals and release it into the synapse upon firing. Although the precise role of zinc in synaptic function is unclear, its presence is certain and zinc-binding sites exist[29]. Fatty acid abnormalities have been linked to numerous neurodevelopmental disorders, including attention deficit hyperactivity disorder (ADHD), dyspraxia, dyslexia, and autism[30]. A study to evaluate the relationships between serum free fatty acids (FFA) and zinc, and attention deficit hyperactivity disorders found a statistically significant correlation between zinc and FFA levels in the ADHD group, and researchers speculated that zinc deficiency may play a role in the aetiopathogenesis of ADHD[31]. Zinc deficiencies can result in children who are irritable, tearful, and sullen; these children are not soothed by close body contact and they resent disturbances. Photophobia is also present and gaze aversion is common[32]. In addition, copper levels are often higher in autistic children compared with controls[33], and zinc levels tend to be low in the presence of excess copper[34]. Intake of copper through foods or supplementation needs to be carefully monitored.

Magnesium, **zinc**, **iron**, and **calcium** levels (as assessed in plasma, erythrocyte, urine, and hair) are lower in children diagnosed with hyperactivity[35]. Research indicates that autistic children also have lower levels of magnesium and manganese, as compared to controls[36]. Clinical trials with supplementation have shown positive results. In 75 magnesium-deficient hyperactive children between 7 and 12 years of age, 50 of the patients who received magnesium supplementation (3 mg/lb/day for 6 months) had statistically improved results, while control subjects' behavior worsened over the test period[37]. In a study of 116 children diagnosed with ADHD (68 children were diagnosed with coexisting disorders including disruptive behavior), researchers found magnesium deficiency in 95% of those examined, suggesting that magnesium deficiency in children with ADHD occurs more frequently than in healthy children; a subsequent study indicated that supplementation with magnesium and calcium corrected deficiencies in most of the children[38]. Similarly, lowered serum levels of zinc have been reported in 43 children with ADHD when

compared to controls[29]. And finally, four patients with autism who were hitting or poking their eyes were all found to have hypocalciuria; three of these patients reduced or stopped self-injury after calcium supplementation[39]. Another important and often overlooked essential element is **iodine**. It's important for children to consume adequate iodine, either by supplementation or iodized table salt.

Regular and adequate intake of **essential fatty acids** (EFAs) appear to be extremely important for children with ADHD and/or autism and related disorders[40,41]. EFAs maintain cell membrane structure and are specifically important in maintaining central nervous system function. EFAs are also required for the production of eicosanoids, which are involved in almost every biologically significant process in the body. Rats and monkeys who are deficient in EFAs present behavioral, sensory, and neurologic dysfunction. Children with lower blood levels of EFAs demonstrate more behavioral problems, and temper tantrums as well as learning, health, and sleep problems. Research in children has shown that children with ADHD have lower proportions of key fatty acids in plasma and red blood cells. It is thought that one of the causes of lower EFA status in children with ADHD is impaired conversion of fatty acid precursors[42].

While additional research is needed to shed mechanistic and detailed light on how specific nutrients, and synergistic combinations of nutrients, influence brain biochemistry, there is no doubt that children with a neurodevelopmental disorder will benefit from supplemental nutritional support. Combining multiple nutrient interventions simultaneously seems not only theoretically logical but also has demonstrated efficacy. In a recent study, Canadian researchers elected a combined vitamin/chelated mineral supplementation program to alleviate symptoms in two boys with mood instability, obsessive compulsive problems, and rage behaviors. No toxicities or side-effects were noted, and near total relief of symptoms were observed in both boys. However, symptoms returned after cessation of the supplementation program[43].

Because most North American children in general do not receive adequate nutrition through diet alone and children diagnosed with ADHD or autism spectrum disorder appear to have additional nutritional needs, adding supplemental nutrition to the treatment protocol appears warranted.

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Digestion and Neurodevelopmental Disorders in Children

Digestive enzymes play a crucial role in nutrient absorption, metabolism, elimination, and detoxification[1]. Digestive enzymes work individually as well as complementarily to exert a powerful influence in supporting digestive processes and in maintaining intestinal health. Comprehensive enzyme replacement therapy includes utilization of a full spectrum of protease, peptidase, amylase, cellulase, lipase, phytase, lactase, sucrase, and maltase enzymes, which are recognized to maintain optimal support of digestion[1-4]. Enhanced digestion reduces exposure to potentially allergenic macromolecules such as casein and gluten[5-7], helps alleviate intestinal inflammation[8], and optimizes nutrient uptake[1].

A growing body of evidence indicates that digestive enzyme replacement therapy may have a unique role in supporting the complex gastrointestinal conditions recognized to exist in children with neurodevelopmental disorders[9,10]. Children diagnosed with attention deficit/hyperactivity disorder (ADHD) or autism spectrum disorder (ASD) have been observed to share similar symptoms and behaviors, as well as common etiologies[11]. Of interest is the reported connection between a number of gastrointestinal symptoms recognized to coexist with the developmental, cognitive, and sensory problems seen in these children[5,12,13].

Mounting evidence suggests a possible gut-brain connection associated with children with ASD[7,14,15]. Gershon, in his book *The Second Brain*, established a framework for understanding the connection between the gastrointestinal tract and the immune and central nervous systems, providing support for understanding the basis of a gut-brain connection[16]. Again, and very recently, a link between intestinal pathophysiology and its relationship to autism was identified[17]. This comprehensive overview presents the growing body of evidence suggesting the influence that impaired gastrointestinal function can exert in the abnormal brain function seen in autism.

The work of Goodwin provided early documentation of a connection between the brain and the intestinal tract when he reported that malabsorption problems (bulky, odiferous, loose stools and/or diarrhea) and cerebral dysfunction were present in 40% of the autistic children participating in his study[18]. A number of investigators have reported that a significant percentage of autistic children present with gastrointestinal symptoms including indigestion, diarrhea, constipation, abdominal discomfort, reflux, gaseousness, abdominal bloating, and/or foul-smelling stools[12,13,19,20]. Other researchers have reported that previously unrecognized gastrointestinal disorders, including reflux esophagitis and disaccharide malabsorption, might contribute to the behavioral problems of non-verbal autistics[12]. The work of Wakefield *et al* led to the observation that primary gastrointestinal pathology may play an important role in the inception and clinical expression of some childhood neurodevelopmental disorders, including autism[13].

Clinical research has demonstrated the presence of gastrointestinal disturbances including inflammation[13,21,22]; intestinal permeability defects (leaky gut)[15,18,23]; enzyme deficiencies[12,19]; dysbiosis[24]; malabsorption/ maldigestion[13,15,25-27]; immune dysfunction/ dysregulation[28]; and food allergies and/or hypersensitivities[23,24,29] in children with neurodevelopmental disorders.

Researchers have recognized varying degrees of gastrointestinal inflammation in autistic children[12-14]. Horvath undertook a study involving histological examinations of 36 autistic children with gastrointestinal symptoms. The results indicated the presence of reflux esophagitis in 69.4%, chronic inflammation of the gastric mucosa in 16%, and chronic duodenal inflammation (duodentitis) in 66.6% of the children[12,15]. Further studies have demonstrated chronic inflammatory and immune responses in the colon and ileum of autistic children, referred to as autistic enterocolitis, which is now documented to exist in regressive autism[13,16,21].

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Intestinal permeability defects (leaky gut) are recognized to exist in these children as well. D'Eufemia *et al* determined that 43% of autistic children had gut mucosal damage, documented by altered intestinal permeability markers, despite the fact that they had no previous clinical or laboratory findings consistent with known intestinal disorders[18]. The authors speculated that altered intestinal permeability could represent a mechanism for the increased passage through the gut mucosa of peptides derived from foods, resulting in behavioral abnormalities[18]. An inflamed, porous mucosal membrane allows toxins, food allergens, and undigested proteins to pass into the circulatory system where they can trigger a cascade of neurological and systemic symptoms[7,12,18]. These intestinal responses may be reduced with the use of digestive enzymes. Digestive enzymes, recognized to break down a wide range of foods, lessen the overall inflammation of the intestinal membrane, digest exorphin peptides, and support greater bioavailability of important minerals[2,10].

Early recognition of digestive enzyme insufficiency in autism came from the work of Horvath, which indicated that 58% of the children examined had disaccharide/glucoamylase enzyme deficiencies below the normal range as well as low lactase activity[12]. Another clinical investigator has conducted over 400 endoscopy evaluations and enzyme assays and found similar gastrointestinal pathology, with disaccharide/glucoamylase enzyme levels below normal and some 55% of the children having lactase as well as sucrase deficiencies[20]. Clinicians have also reported anecdotal evidence that digestive enzyme supplementation, inclusive of a combination of carbohydrases (including amylase) as well as disaccharidases (including lactase, sucrase and maltase) have provided marked clinical improvement in these gastrointestinal symptoms[30].

Panksepp initially postulated that dietary peptides, derived from casein and gluten, could be causative in autism[31]. Researchers made additional observations suggesting that individuals with psychiatric disorders, including autistic children, have elevated levels of urinary peptides[32-34]. These peptides are recognized to form following the incomplete breakdown of gluten and casein, resulting in the development of opioid or morphine-like compounds called exorphins. These neuro-active peptides, referred to as beta-casomorphins (from casein in dairy products) and gluteomorphin and gliadorphin peptides (from gluten grains including wheat, oat, barley and rye)[35], are known to function as false neurotransmitters adversely affecting the central nervous system[7,26,36]. Once in the bloodstream, they stimulate undesirable neurological, immune, and inflammatory responses, triggering a number of adverse neurological and physiological consequences recognized in autism[5,26,31,32,33,37]. It has been reported that these peptides stimulate opioid receptors in the brain, which can significantly affect behavior, emotions, and cognitive ability as well as pain threshold and other symptoms commonly associated with autism[5,31,32].

Dipeptidyl peptidase IV (DPP-IV) is an intestinal brush-boarder enzyme recognized to breakdown/degrade exorphin peptides. Because of the particular formation of these peptides, they are known to be highly resistant to breakdown by general peptidase enzymes[38]. Researchers have made important observations regarding DPP-IV and its role in autism and have identified specific exorphin and other opioid peptides in the urine and serum of autistic children[7,31,33]. While more clinical research is needed, there is growing evidence that the activity of the DPP-IV enzyme may play a significant role in supporting autism-related intestinal issues[10,33,37]. Studies indicate that avoidance of dietary casein and gluten with reduction of exorphins can help to improve symptoms of autism[7,10,37,39]. A recent clinical trial evaluated the response to a plant-based enzyme formula utilizing high protease-, peptidase-, and DPP-IV-containing enzymes in 29 children with attention deficit disorder (ADD)[10]. Study results documented the enzymes' safety and reported measurable improvement in 13 parameters of function and behavior in the test subjects.

Enzymes function as biological catalysts, facilitating optimal breakdown of foods, proper absorption, and utilization. By definition, enzymes assist or accelerate digestive activity and when performing its function, the enzyme is not used up, changed, or destroyed[1,2]. Each enzyme has a specific and individual function that is not performed by other enzymes. Enzymes used for enzyme replacement therapy can be obtained from different sources including animal-derived pancreatic enzymes and non-animal sources, such as microbial and plant-derived enzymes.

Digestion and Neurodevelopmental Disorders in Children

Highly concentrated and naturally-derived microbial and plant-based enzymes (such as those derived from *Aspergillus oryzae*, *Aspergillus niger*, *Rhizopus oryzae*, and *Hordeum vulgare*) are effective in supporting diverse gastrointestinal conditions. Enzyme activity is best measured in standardized activity units according to the Food and Chemical Codex (FCC), United States Pharmacopeia, or other approved compendial methods[40]. Most digestion occurs in the alkaline environment of the small intestine. Therefore, replacement enzymes with demonstrated activity in neutral to alkaline pH provide the greatest clinical efficacy at promoting healthy digestive function[2,4]. For example, proteolytic enzymes measured in USP units and FFC PC units possess demonstrated activity in neutral to alkaline pH whereas enzymes measured in HUT units (assayed at Ph 4.7) may be inactive in the small intestine.

Compared to animal-sourced pancreatic enzymes, including pepsin, plant-derived enzymes are recognized to be active and stable in both the acidic environment of the stomach as well as the more alkaline conditions found in the small intestine[2,4,41,42]. With the ability to breakdown a wider variety of dietary substrates, under a broader range of pH conditions (pH of 2 to 12), these enzymes are ideal for supporting the gastrointestinal needs of these children[2,4].

Microbial and plant-derived enzymes not only have a history of safety and tolerance when studied in children with ASD[10], but they are also recognized as non-toxic and approved for use by the FDA in foods and dietary supplements[43]. Digestive enzyme activity is measured by assaying the quantity of digestion that occurs under the following specific conditions: enzyme concentration, enzyme quantity, pH, temperature, and substrate. For optimal digestive function, a comprehensive blend of different proteases, peptidases, amylases, lipases, and other enzymes will provide the broadest possible range of enzyme activity for the child with autism or related conditions[2]. In addition, the inclusion of peptidases containing DPP-IV activity, providing additional digestion of casein, gluten and soy, will further enhance efficacy.

Clinical work and investigation has lead to a deeper understanding of the complex gastrointestinal pathology recognized to exist in children with neurodevelopmental disorders[5,7,12,13,18,21,31,32]. Evidence for the utility of protease- and peptidase-containing enzymes in addressing overall protein digestion, as well as opioid peptide problems is increasing[10,36]. Clinicians have reported favorable responses to many gastrointestinal conditions with the use of broad-spectrum microbial and plant-derived enzyme supplementation that include a full range of protease-, peptidase-, and DPP-IV-containing enzymes[29].

The growing body of evidence indicates that digestive enzyme replacement therapy may have a unique role in supporting the complex gastrointestinal conditions recognized to exist in children with neurodevelopmental disorders[9,10].

Digestion and Neurodevelopmental Disorders in Children

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Detoxification and Neurodevelopmental Disorders in Children

Over the past several decades, concerns have been raised in regard to unacceptable levels of environmental toxins. There are between 50,000 and 100,000 synthetic chemicals in commercial production, and new synthetics enter commerce at an average rate of three per day[1]. How these chemicals affect humans in the developmental stage of life has not been adequately studied; however, data suggests that exposure to neurotoxic compounds at levels believed to be safe for adults could adversely affect brain function if it occurs during a critical period of brain development. It is plausible that children with pre-existing neurological conditions such as attention deficit/hyperactivity disorder (ADHD) or autism spectrum disorder (ASD) might be more vulnerable to certain low-level neurotoxic exposures[2].

It is widely thought that detoxification problems play an important role in child-onset neurodevelopmental disorders. Such dysfunction may be linked to nutritional deficits, impaired metabolism, endotoxemia of gastrointestinal origin, and/or an increased exposure to environmental toxins. For example, studies have found that children with autism often have abnormal sulfate metabolism leading to excess urinary excretion of sulfate and very low plasma levels of sulfate[3,4], one of the amino acids required in the detoxification process. In one such study, 100% of 20 autistic children showed abnormal liver detoxification profiles, including abnormal distribution patterns for glucaric acid, a marker for contamination by xenobiotics[5]. And recently, it has been discovered that the families of children with ASD often present with dysregulated amino-acid metabolism[6].

Detoxification is accomplished through two mechanisms: Phase I and Phase II detoxification. The primary Phase I enzyme is known as the cytochrome P450 monooxygenase system; its highest concentrations are found in the liver, the site of the most active metabolism. Phase I detoxification changes nonpolar chemicals into polar compounds by adding a polar group or a reactive group (known as biotransformation). Phase II detoxification conjugates chemical groups to the chemical for excretion through the kidneys; major conjugation reactions include acetylation, acylation, glucuronidation, sulfonation, and methylation. Both Phase I and II detoxification pathways must function properly for detoxification to occur.

Several nutrients are known to play important roles in Phase I and Phase II detoxification pathways. **Milk thistle** (*Silybum marianum*) is used clinically as a liver protectant to lessen damage from potentially hepatotoxic drugs and for treating liver disorders including toxic liver damage caused by chemicals. Clinical trials show that when standardized to contain 70-80% of the constituent silymarin, milk thistle improves liver function tests in patients with hepatotoxicity resulting from long-term exposure to organic solvents[7]. In a study of 166 children (under the age of 17) with chronic liver disease, 70% showed improvement upon using milk thistle, 26% stabilized, and only 4% showed no improvement or stabilization; among those with chronic active hepatitis, 32% showed improvement; 44% stabilized, and 24% had no benefit[8].

Calcium-D-glucarate is the calcium salt of D-glucaric acid, which occurs in human tissues and body fluids and is found, in small amounts, in fruits and vegetables. Calcium-D-glucarate has been shown to inhibit beta-glucuronidase, an enzyme produced by colonic microflora and involved in the Phase II liver detoxification process. Elevated beta-glucuronidase activity is associated with an increased risk of various cancers[16].

Calcium d-glucarate is metabolized in the acidic environment of the stomach into an equilibrium of D-glucaric acid, D-glucaro-1,4-lactone (GL), and D-glucaro-6,3-lactone. GL is a direct inhibitor of beta-glucuronidase. By inhibiting beta-glucaronidase activity, GL allows for increased net elimination of toxins and steroid hormones via glucuronidation[9,10], which is considered the most important Phase II

detoxification conjugation reaction as it is the primary means by which many hormones and toxins are excreted.

N-acetyl-L-cysteine (NAC) is the N-acetyl derivative of the amino acid L-cysteine, a precursor of glutathione, which has been repeatedly observed to be low in the plasma of children with autism[11]. The glutathione sulfur-transferases (GSTs) catalyze the conjugation of xenobiotics with glutathione, an initial step in the formation of mercapturic acids, the ultimate excretory form of many xenobiotics[5]. NAC produces a dramatic acceleration of urinary methylmercury excretion in animals[12]. In addition, NAC treatment has been shown to reduce liver damage after paracetamol (acetaminophen) overdose[13].

Alpha-ketoglutarate (AKA) helps detoxify ammonia, synthesized from urea in the colon[14]. Moderately elevated ammonia is often found in children diagnosed with Rett Syndrome, and children with autism spectrum disorder often present with gut dysbiosis[15,16], leading to enhanced ammonia in the GI tract. AKA is also a precursor of glutamine[17], a conditionally essential nutrient well recognized for its importance in maintaining healthy gut mucosal function[18,19]. Researchers have studied antioxidative properties of various concentrations of alpha-keto acids, including pyruvate, alpha-ketoglutarate, and others; they have found that alpha-keto acids protect against oxidative damage induced by hydrogen peroxide[20]. Orally, AKA is used for treating chronic kidney and gastrointestinal dysfunction, bacterial overgrowth, intestinal toxemia, liver dysfunction, and chronic candidiasis[21].

Methyl-sulfonyl-methane (MSM) is a naturally occurring sulfur compound found in human diets. Sulfur is an essential component of the sulfur-bearing amino acids methionine, cystine, and cysteine, and sulfur donors support sulfonation, part of the Phase II detoxification reactions.

Taurine is a conditionally essential sulfonic amino acid found in high concentrations in the human brain. Taurine is involved in central nervous system neuromodulation and appears to inhibit catecholamine oxidation[22]. In addition, taurine has physiological roles with osmoregulation, antioxidation, and stimulation of glycolysis and glycogenesis[23]. Taurine is also required for the formation of bile salts, an important mode of toxin elimination[24,25].

Methylation is another important Phase II detoxification reaction. Nutritional methyl donors, such as the amino acid methionine as well as choline and betaine anhydrous, aid in methylation conjugation reactions.

Methionine is a sulfur-bearing essential amino acid found in animal proteins. Methionine assists both sulfonation and methylation reactions in the Phase II detoxification by removing heavy metals from the body and adding a methyl group to xenobiotics, which aids in their excretion from the body. A dietary deficiency of methyl donors such as methionine and choline has been shown to enhance the activity of hepatocarcinogens in animal studies. In one such study, researchers found that supplementation with methionine and choline resulted in longer survival rates after carcinogenic doses of aflatoxin B1, and such supplementation increased levels of several of the cytochrome P450 enzymes. Increased levels of methionine and choline appear to favor activation of oxidization mechanisms[26].

Choline is an important nutrient for several metabolic pathways, including normal membrane function, acetylcholine synthesis (acetylcholine acts as a neurotransmitter), and methyl group metabolism[27]. Choline is required to make the essential structural phospholipid, phosphatidylcholine[28]. The cytochrome P450 enzymes of Phase I detoxification are choline dependent[29]. Individuals who consume a choline-deficient diet develop hepatic dysfunction[28,30].

Betaine anhydrous, also known as trimethylglycine, is the major metabolite of choline and has an important role as a methyl donor to form the amino acid methionine[27]. Betaine anhydrous, not to be confused with betaine hydrochloride, occurs naturally in the body and in small amounts in some foods (e.g., beets, spinach, and seafood)[31].

Finally, the essential mineral **selenium** is required for the synthesis of glutathione peroxide[10], a vital antioxidant enzyme that helps to detoxify hydrogen peroxide produced within cells. Selenium helps detoxify mercury

by forming selenium-mercury complexes, which can be safely excreted from the body[32]. Animal studies have shown that supplementation with taurine, selenium, and glutathione are beneficial for maintaining endogenous antioxidant systems[33].

Based on research findings linking autism and other neurodevelopmental disorders with physiological issues such as exposure to environmental toxins and gastrointestinal irregularities, supplementation with appropriate nutrients and botanicals is proving valuable in supporting the health of these individuals.

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Cognitive Function and Neurodevelopmental Disorders in Children

Children diagnosed with neurodevelopmental disorders such as attention deficit hyperactivity disorder (ADHD) or autism spectrum disorder (ASD) struggle with cognitive function and learning. The direct influence of nutrition on both the structure and the function of the human brain is rapidly being identified and elucidated among investigative research and clinical communities. Research over the past several decades provides a scientific framework for the importance of therapeutic nutrition in children with neurodevelopmental disorders[1,2]. Further, research clearly suggests a role for supplementation with special nutrients and botanicals to help these children to optimize cognitive processing and calm abnormal behaviors. Although it is imperative that basic nutritional needs are met, it is also important to provide additional key nutrients proven to promote a healthy nervous system and to boost cognitive function.

Acetyl-L-carnitine has been used in treating a variety of neurological and cognitive conditions. Acetyl-L-carnitine occurs naturally in the body within the inner membrane of the mitochondria. Structurally related to acetylcholine, it may act as a cholinergic-enhancing agent by serving as a mitochondrial precursor to acetyl coenzyme A (acetyl CoA). Acetyl-L-carnitine participates in the production of cellular energy by acting as a shuttle between the cytoplasm and the mitochondria for long-chain fatty acids. It may also enhance choline acetyltransferase activity, facilitate actions on serotonergic pathways, and enhance synaptic transmission. Acetyl-L-carnitine appears to prevent ATP depletion and buffer oxidative stress[3]. In clinical studies, acetyl-L-carnitine has been shown to reduce hyperactivity behavior in fragile X patients[4] (the fragile X condition presents with an autistic-like constellation of symptoms).

Coenzyme Q10 (also known as CoQ10 or ubiquinone) provides antioxidant benefits as well as functioning as a membrane stabilizer and cofactor in many metabolic pathways. It is present in the human body in both plasma and lymphocytes. CoQ10 is used in the production of cellular energy (adenosine triphosphate) in oxidative respiration. Although CoQ10 is found in animal foods, the amounts ingested in foods do not approach therapeutically effective doses. Supplemental CoQ10 protects DNA from oxidative damage (prevents DNA strand-break formation) and enhances DNA repair enzyme activity[5]. Children diagnosed with ASD are more susceptible to systemic candida overgrowth, and candida may prevent dietary ubiquinone absorption, requiring supplementary doses to achieve a therapeutic effect[6].

L-theanine (5-N-ethylglutamine), an amino acid commonly found in green tea, is an analog of glutamate, an excitatory neurotransmitter. Levels of glutamate increase and cause neuronal death during periods of cerebral ischemia, and theanine appears to protect the brain by decreasing ischemic neuronal death, as evidenced in animal models[7]. Plasma levels of glutamic acid and other amino acids tend to be higher in autistic and Asperger syndrome patients[8]. L-theanine has been investigated for its ability to promote a restful, relaxed state without diminishing daytime alertness[9]. Theanine has been studied in its role in modulating neurotransmitter activity in the brain. Animal studies show that theanine is incorporated into the brain through the blood brain barrier via the leucine-preferring transport system. While levels of norepinephrine are unaffected by theanine administration, theanine appears to cause significant increases in serotonin and/or dopamine concentrations in the brain, particularly the striatum, hypothalamus, and hippocampus[10]. Studies have been mixed, however; some studies show that while brain tryptophan (a precursor of serotonin) content is increased following theanine administration, serotonin and 5-hydroxyindole acetic acid (5HIAA) are decreased[11].

Carnosine is a natural substance, formed of two amino acids (ß-alanyl-L-histidine). It is often called a neuropeptide due to its brain-protective properties[12,13]. Carnosine is found naturally in healthy muscles, heart,

brain, liver, kidneys, and other tissues. The muscles contain about 20 µmol/g dry weight. Carnosine can enhance frontal lobe function and is neuroprotective; it can also correlate with gamma-aminobutyric acid (GABA) with possible anticonvulsive effects. In a study of 31 children with ASD, 800 mg of L-carnosine resulted in statistically significant improvements on the Gilliam Autism Rating Scale (total score and the Behavior, Socialization, and Communication subscales) and the Receptive One-Word Picture Vocabulary Test[14]. In addition to antioxidant capabilities, carnosine reacts with deleterious aldehydes to protect susceptible macromolecules, particularly those found in brain and other nerve tissues. *In vitro* studies have indicated that the dipeptide inhibits nonenzymic glycosylation and cross-linking of proteins induced by aldose and ketose sugars and other molecules[15,16]. Carnosine also appears to be able to extend the lifespan of cultured cells; rejuvenate senescent cells; inhibit the toxic effect of amyloid peptide (A beta), malondialdehyde, and hypochlorite to cells; inhibit glycosylation of proteins and protein-DNA and protein-protein cross-linking; and maintain cellular homeostasis[17]. In addition, literature suggests that carnosine may possess neurotransmitter activity, modulate enzymatic activities, and chelate heavy metals[18].

Choline is an essential nutrient in methylation, acetylcholine and phospholipids biosynthesis, and in cell signaling. Dimethylaminoethanol or **DMAE** (deanol), a precursor to choline, may enhance central acetylcholine formation. In a study of 74 children referred for problems with learning disabilities, including hyperactivity, significant improvements were achieved in learning and behavior disorders after DMAE supplementation[19,20]. Some studies also show weak anticholinesterase activity[21], and numerous studies in Alzheimer patients have shown a significant boost in cognitive activity from medications that act by stimulating anticholinesterase function[22]. Similar effects are observed in autism[23].

Botanicals have also been used in the treatment of ASD and attention deficits. In a study of 36 children who fit the diagnostic criteria for ADHD that used a combination botanical product containing **American ginseng** (200 mg) and ***Ginkgo biloba* extract** (50 mg), improvements were noted in hyperactive-impulsive attribute, anxious-shy attribute, and in social problems[24]. And in a study of 20 healthy young adult volunteers who received graded doses of a combination of *Ginkgo biloba* and *Panax ginseng*, researchers found a dose-dependent improvement in performance on the quality-of-memory factor for the highest dose. There was also a dose-dependent decrease in performance of the speed-of-attention factor at the mid-dosage range[25].

Ginkgo biloba is active as a free-radical scavenger but may have direct effects on the cholinergic system, which may explain its acute and chronic cognitive-enhancing effects[26]. Ginkgo appears to modulate genetic expression, modulate the effects of neurotransmitters[27], protect against neuronal death, increase hippocampal high-affinity choline uptake, inhibit the down-regulation of hippocampal glucocorticoid receptors, enhance neuronal plasticity, and counteract the cognitive deficits that follow stress or traumatic brain injury[28]. In adults, clinical studies have demonstrated that daily doses of 120 to 240 mg of *Ginkgo biloba* can improve symptoms associated with cerebral insufficiency such as memory loss, depression, and tinnitus[29].

According to the German Commission E[30], the following pharmacological effects have been established experimentally for *Ginkgo biloba*:

- Improvement of hypoxic tolerance, especially in cerebral tissue
- Inhibition in age-related reduction of muscrinergic cholinoceptors and alpha-adrenoceptors, as well as stimulation of choline uptake in the hippocampus
- Increased memory performance and learning capacity
- Improvement in the compensation of disturbed equilibrium
- Improvement of blood flow, particularly in the region of microcirculation
- Inactivation of toxic oxygen radicals (flavonoids)
- Neuroprotective effect

The flavonoids found in ginkgo may enhance the release of catecholamines and other neurotransmitters, inhibit biogenic amine uptake, protect catechol-O-methyltransferase and monoamine oxidase, and protect endothelial-derived relaxing mechanisms in the brain[31].

Cognitive Function and Neurodevelopmental Disorders in Children

American ginseng (*Panax quinquefolius*) has been a revered botanical for several centuries. Saponins (particularly the ginsenosides) are considered the primary biologically active components of the Panax species. Reported pharmacological actions of saponins include weak CNS stimulant activity, anti-fatigue action, and a slight increase in motor activity[32]. Ginsenoside Rbl has CNS-depressant activity; is anticonvulsant, analgesic, antipyretic, antipsychotic, and ulcer-protective; inhibits conditioned avoidance response; has weak anti-inflammatory activity and an antihemolytic action; and increases gastrointestinal motility. Since autistic children are often severely constipated, increases in gastrointestinal motility could be helpful in restoring normal elimination. In addition, *Panax ginseng* has been shown to have beneficial effects on the immune cells of individuals with chronic fatigue syndrome, an area of concern for children with ADHD or ASD as well[33].

In a recent study, children with ADHD were given both gingko and *Panax ginseng* and found to have significant lessening of their ADHD symptoms. The side-effects were infrequent and only rarely attributable to the botanicals (2 of 36 children)[25]. Since ASD shares some features with ADHD, it is reasonable to presume a beneficial effect in the autistic population as well, although clinical studies are underway to more carefully establish this link.

Neurodevelopmental disorders, such as ADHD or ASD, are complex conditions that benefit from multi-nutritional and botanical intervention. When combined with an adequate diet and supplementary nutritional support, research supports important and efficacious roles for select special nutrients and botanicals as listed above to positively impact the cognitive and nervous system function in affected children.

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IMMUNE RESPONSE AND NEURODEVELOPMENTAL DISORDERS IN CHILDREN: A CLOSER LOOK

Many clinicians and researchers believe changes in the immune system play a pivotal role in the symptoms of attention deficit/hyperactivity disorder (ADHD) or autism spectrum disorder (ASD)[1-3]. Despite the immune-system differences intrinsic to each disorder, commonalties exist in the area of allergy and food sensitivity[4-6]. This often manifests itself as inflammation of the lymph nodes in the intestinal tract[7]; swelling of these glands is nonspecific and is often observed in food allergy patients. Researchers at Georgetown observed these changes in ADHD children[8], and another group at the Royal Free Hospital in London found similar but worse changes in children with autism[9].

Sometimes these changes, considered insults to the immune system, occur in the womb, but more frequently they occur post-birth and may involve exposure to toxic metals and/or persistent systemic infections. We now know there is a unique tapestry of interwoven etiologic factors that play a role in the development of neurodevelopmental disorders including ADHD and ASD. Researchers have documented a number of different immunologic mechanisms, each one providing another piece of the larger puzzle[10].

Clinicians and researchers have suggested a genetic predisposition in initiating these conditions[10,11]. However, a growing body of evidence[2,12-15] suggests that environmental insults (chemical toxins, heavy metals, vaccinations, viral exposures, chronic fungal infections, etc.) may initiate immune system dysregulation in children with neurodevelopmental disorders. Singh *et al* summarized it well when they suggested that a number of factors are implicated including immune, neurochemical, genetic susceptibility, and environmental factors[16]. Evidence from Johns Hopkins University has indicated that the MMR vaccine may induce a switch in the immune system tending toward allergy[17]. And Geier and Geier found a link between the mercury preservative in vaccines and ASD[18]; mercury is a known allergy stimulator. Thus, the function of the immune system in neurodevelopmental disorders is complex and altered by multiple different harmful events.

Studies also indicate that ASD children exhibit numerous immune-system dysfunctions, including immunoglobulin deficiency (IgA)[9,19]; abnormal lymphocyte balance (imbalanced in a TH-2 direction)[20]; excess Tumor Necrosis Factor (TNF)-alpha (a chemical messenger that turns on inflammation and destroys healthy cells)[6]; autoimmunity with brain antibodies to myelin basic protein (MBP)[16,21]; higher persistence of measles virus (MV) in the GI tract[22,23]; inflammatory gut disorders[15]; and extreme reactivity to proteins in grains, dairy foods, and other foods[24-26].

GENETIC FACTORS IN PREDISPOSITIONS FOR ASD AND ADHD

Researchers have studied the C4B gene, one of those that control the function and regulation of the immune system in autism. It was recognized that a deficient form of this gene, known as the C4B null allele, encodes a product that is involved in eliminating pathogens such as viruses and bacteria from the body. These researchers found an increased frequency of the C4B null allele in autism[27]. Interestingly, however, in a second study, they confirmed an increased incidence of C4D null allele ADHD and dyslexia as well[28]. They suggested that these significantly lower C4B levels may provide a marker for ADHD as well as an etiologic factor for this condition.

GENERAL IMMUNE DYSFUNCTION

It has been demonstrated that children with autism have an immunological shift that leans them toward allergy[19]. The results of this shift predispose to autoimmunity as well as chronic atypical viral infections and/or candida/yeast infections, all documented to exist in these children. This is supported by the work of Wakefield *et al* who initially reported intestinal abnormalities along with evidence of a shifted immune response in a subset of children with autism[9].

Research on the biological pathophysiology of autism has found some evidence that immune alterations may play a pivotal role[29]. One study of 40 autistic boys with age-matched controls found increased concentrations of

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total serum protein (TSP) characterized by increased serum albumin and IgG subclasses (including IgG2 and IgG4)[19]. The authors suggested that the increased serum concentrations of IgGs in autism may point towards an underlying autoimmune disorder and/or enhanced susceptibility to infections resulting in chronic viral infections. This theory is also supported by the work of Gupta *et al*[30].

Research also indicates that children with ASD produce higher levels of pro-inflammatory cytokines than do children without ASD. In a study at the University of Minnesota, 83 children with ASD were compared with a group of healthy matched controls. Three-quarters of the ASD children produced higher levels of at least one pro-inflammatory cytokine compared with controls, and from 75-80% of the ASD children were reactive to dietary proteins[31].

Autoimmunity with Immune-Mediated Responses/Antibodies Against the Brain

Research has demonstrated that immune factors such as autoimmunity may play a role in the etiology of autism[3]. This involves the development of antibodies to myelin basic protein (anti-MBP) which suggests an association with the development of this condition[32]. Although not all studies have concurred, one study using human immunoglobulin (IVIG) to modify immune reactions demonstrated favorable improvement in many autistic children[30]. This has been confirmed clinically by Bradstreet and El-Dahr, who reported marked improvement in the symptomotology of autoimmune children with anti-MBP antibodies (myelin basic protein) following the use of IVIG therapy[33].

Altered Immune Response With Leaky Gut

Researchers studying the actual mechanism of how inflammation in the gut influences the brain have developed several theories. One serious concern is the way in which the immune system activates self-digesting proteins called matrix metalloproteinases (MMP). These enzymes have been recognized as key intermediaries in most autoimmune disorders and they are activated in all immune-activated inflammatory bowel diseases[34-36]. It is also known that many children with ADHD or ASD have immune activation and/or inflammatory bowel disease.

Once the gut is wounded in this way, it becomes more susceptible to food proteins, and this MMP activation is the likely link to food allergy reactions. So it appears that the MMP keeps the process going and creates a self-perpetuating wound, unless the cycle is broken. Pathogenic strains of bacteria and yeast may also promote this immune reaction. It is also known that MMP is an intermediary in the destruction caused by viral infection of the brain[37,38].

For these reasons the key to recovery of a healthy immune system is systematically addressing the cycle of gut immune activations by:

1. Providing probiotics to get healthy bacteria to replace the unhealthy ones[39].
2. Digesting foods with enzymes to reduce adverse immune effects[40].
3. Promoting better natural immunity through immune support as described herein.
4. Decreasing inflammatory reactions with essential fatty acids, and through reduced sugar and starch intake[41].
5. Reducing autoimmunity through specific medical regimens, e.g., IVIG, steroids, salicylates, etc.

Nutritional Support

It is imperative that parents become proactively involved in support of the immune function of their child with either ADHD or ASD.

Nutrition plays a fundamental role in building and supporting the strength of the immune system. Vitamins, minerals, and botanicals have been shown to positively impact the immune system. Some of the specific and easily available resources naturally available to support the immune system are included here.

The functions of **vitamin C** (ascorbic acid) are extensive and well documented in the literature. Sufficient quantities of ascorbic acid are required for the synthesis of neurotransmitters (specifically dopamine), for the synthesis of adrenal hormones, for the synthesis of L-carnitine, and for tyrosine catabolism. Vitamin C functions as an antioxidant and, in this role, acts as a chemical reducing agent in intracellular and extracellular reactions. Ascorbic acid protects DNA from oxidant damage and wields other influences on genetic expression[42]. When researchers looked at excretion levels of vitamin C in children with developmental disabilities,

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compared to children classified as normal, they observed that excretion levels varied widely. Investigators found that following a load dose of vitamin C (mean value 204% of recommended levels), all of the children classified as normal were high excreters. Conversely, the excretion levels of the children with developmental disabilities varied from very low to high; two were classified as deficient in vitamin C, based on fasting serum and urinary levels[43].

Vitamin C also promotes resistance to infection by supporting the immunologic activity of leukocytes and the production of interferon, by enhancing the inflammatory reaction, and by helping maintain the integrity of the mucous membranes[44]. Ascorbic acid status is compromised by acute and chronic immune system challenges. A preliminary 30-week double-blind, placebo-controlled study looking at the effectiveness of therapeutic doses of vitamin C as a supplemental treatment for autism was conducted in 18 autistic school-age children. Behaviors were rated using the Ritvo-Freeman scale. Investigators concluded that vitamin C supplementation was effective as changes in test scores and symptom reductions were significant[45].

A common symptom of **zinc** deficiency includes impaired wound healing and altered immune function. Zinc is required in over 200 enzymes, including those that are directly involved in antioxidant activities, notably superoxide dismutase[46,47]. Zinc deficiency impairs immune function and is associated with an overall loss in lymphocytes (B and T cells) of the peripheral immune system, possibly because of atrophy of the thymus and the loss of the zinc-dependent hormone thymulin[48]. In addition, zinc-deficient diets induce oxidative stress in brain tissues as well as in the peritoneal macrophages, and such diets are associated with glutathione depletion and enhanced production of reactive oxygen species[49]. The common cold is associated with over 200 viruses. *In vitro* studies demonstrate that zinc appears to interfere with viral replication and retards the ability of the virus to penetrate the host cell[45].

Inositol hexaphosphate (IP-6 or phytic acid) is an inositol molecule with six phosphate groups attached. It is ubiquitous in the plant kingdom and abundant in cereals and legumes. When combined with additional inositol (part of the vitamin B complex), IP-6 supports healthy cell development and increased natural killer cell activity[50-52]. IP-6 also regulates cellular proliferation[53], enhances natural killer-cell activity while enhancing natural-killer cell cytotoxicity in a dose-dependent manner[54], and inhibits the formation of liver cancers and regresses pre-existing liver cancer growth[55].

Botanical medicines have been used to support immune system function. **Cat's claw** or standardized *Uncaria tomentosa* (Saventaro®) is an herb from the Central Peruvian rain forest that has been used as a traditional medicine by the Ashaninka Indians to enhance natural immunity and modify the acquired immune system. The root of the plant contains pentacyclic oxindole alkaloid (POA) and isomitraphylline[56]. Studies have shown that these POAs enhance the natural immune system by increasing the rate of phagocytosis. It is important that *Uncaria tomentosa* be free of TOAs (tetracyclic oxindole alkaloids) because the presence of TOAs, even in minor amounts, can counteract the proven immune effectiveness of the POA rich *Uncaria tomentosa*. POAs also influence the acquired immune system through the endothelial lymphocyte-proliferation regulating factor, which causes an increase in the proliferation of resting and weakly activated lymphocytes (B, T4, and T8) and inhibits the proliferation of highly activated lymphoblasts (B and T)[57]. Extracts of *Uncaria tomentosa* also increase interleukin production by macrophages in a dose-dependent manner[58], and cat's claw acts as an anti-inflammatory via suppression of TNF-alpha synthesis[59].

Another revered botanical with both experiential and clinical efficacy is aloe vera. **Aloe vera** has immune enhancing properties and promotes healing[60]. These attributes are specifically associated with constituents found in the inner leaf of the plant.

Bioflavonoids exert a positive influence on the immune system, such as stimulating the activities of mast cells, basophils, neutrophils, eosinophils, T & B lymphocytes, macrophages, platelets, smooth muscle hepatocytes, and others[61]. Specifically, **citrus flavonoids** induce apoptosis in human leukemia cells while simultaneously producing no cytotoxicity against human peripheral blood mononuclear cells[62].

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Grape seed is a free radical quencher extracted from the seeds of *Vitis vinifera*. Procyanidins from grape seed extract are a group of polyphenolic bioflavonoids that possess antioxidant activities that surpass the antioxidant capabilities of vitamins E and C singly and in combination[63,64].

Pycnogenol, extracted from the French Maritime Pine bark (*Pinus maritima*), is rich in polyphenols and is a powerful antioxidant[65]. Pycnogenol protects against oxidative stress in several cell systems by doubling the intracellular synthesis of antioxidative enzymes and acting as a scavenger of free radicals. It plays a role in the regeneration and protection of vitamins C and E, and it has conferred anti-inflammatory benefits both *in vitro* and *in vivo*[66]. Pycnogenol has been used successfully in the treatment of attention-deficit hyperactivity disorders (ADHD)[67]. Improvements in cognitive function have been observed in animal studies, confirming anecdotal reports in humans[68]. Oral supplementation of pycnogenol in an animal model demonstrated improvement in immune function (T and B cell)[69]. Clinicians and parents both have reported positive effects using pycnogenol, citing improvement in EEG, handwriting, school performance, and continuous-performance testing improvements[70]. Further, they report the most significant improvements in areas relating to sustained attention and distractibility, not so much with hyperactivity and impulsivity. Few to no side effects were reported.

Monolaurin is a monoglyceride used to destroy lipid-coated viruses such as HIV, herpes, cytomegalovirus, influenza, various pathogenic bacteria including listeria monocytogenes and *helicobacter pylori*, and protozoa such as *Giardia lamblia*[71,72]. Food sources of monolaurin include coconut and human breast milk. Monolaurin from human milk was found to inhibit the growth of cytomegalovirus (CMV) and decreased the growth of some rhinoviruses[73] *in vitro*. It is effective in blocking or delaying the production of exotoxins by pathogenic gram-positive bacteria[74], and it appears preferential to gram-positive bacteria[75]. *In vitro* studies show that monolaurin is effective against *helicobacter pylori*[76].

Supporting the immune system of children who are immuno-compromised or who have been diagnosed with a neurodevelopmental disorder requires a multidisciplinary approach. Adequate nutrition is the first requirement, provided by a balanced diet and augmented by supplementary nutrition to address nutrient deficits. Both clinical and research work supports the use of vitamins and minerals, special nutrients, and botanicals for immune system support.

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