Stress and Folate Impact Neurodevelopmental Disorders

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Abstract

Autism Spectrum Disorder (ASD) is one of several developmental disabilities that can create significant communication and behavioral challenges in affected individuals. Several studies have found that children with ASD have high levels of Folate Receptor Antibody (FRA), which blocks the transport of folate across the Blood-Brain Barrier (BBB) and leads to Cerebral Folate Deficiency (CFD). Supplementation with folate in its reduced form, such as with folinic acid, has been found to improve communication in autistic children with folate receptor antibodies. Here, we provide an overview of the role of folate in nervous system development, effects of FRA on brain folate levels, and clinical trials that have examined the efficacy of folate supplementation in reducing the symptoms of developmental disabilities. Further, we highlight the importance of prenatal folate supplementation in reducing the risk and severity of developmental disorders and the need for additional research to explore optimal dietary interventions to aid in managing them. The results suggest that supplementing with reduced folate may offer a promising treatment approach for individuals with neurodevelopmental disorders, particularly those with FRA.

Folate in Nervous System Development

Folate Role in Development:

Folate (Vitamin B-9) is a key component in nervous system development [1,2]. Bio-available folate is present in many foods, including legumes, leafy greens, and fruits. The naturally occurring form is methylated, while the synthetic version in many vitamin supplements is the oxidized form, folic acid, which is stable for a much longer time than the methylated, reduced form of folate found in foods. Folate is necessary for neural tube formation and its closure in the human embryo and plays an essential role in fetal brain development [3-5]. Low cerebral folate levels is causative of many developmental conditions, including spina bifida in the newborn [6,7]. Because of the key neurodevelopmental role of folate and to reduce spina bifida that results in pregnancies with insufficient vitamin B-9, prenatal vitamins contain folic acid, the oxidized and stable form of folate, rather the reduced form. Additionally, the US since 1998 adds folic acid (the oxidized version of vitamin B-9) to grain products to ensure all pregnancies have sufficient folate. This is credited with a 70% reduction in spina bifida. Most individuals

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convert sufficient folic acid to folate in the gut. However, 5-10% of the population is limited in absorbing folate into the brain due to the presence of an autoantibody for the folate receptor [8], as measured with an antibody test (FratNow, Philadelphia, PA).

Folate Receptor Antibody:

Folate Receptor Antibody (FRA) blocks the high affinity folate receptor alpha, found at the choroid plexus and responsible for the elevated folate levels in brain relative to blood. FRA thus prevents folate from crossing the blood brain barrier [8,9]. Clinical studies have shown that blood levels of FRA can be reduced by changes in diet, particularly elimination of dairy [10]. If FRA levels are reduced or eliminated, this may allow sufficient folate to enter the brain. While FRA is present in a small subsection of the population, clinical tests show that 70% of ASD children have FRA [11]. Meta-analysis from multiple studies reveal that ASD children are 20 times more likely to have FRA [12], indicating a likely genetic aspect that restricts brain absorption of Vitamin B-9. While FRA can be lowered with significant change in diet [10], a symptom of ASD is resistance to dietary change. Thus, the dietary factors contributing to FRA are self-perpetuating due to this ASD-related change resistance, as FRA reduces folate in the brain, creating Cerebral Folate Deficiency (CFD). The CFD can be countered by supplementation with elevated levels of the natural version of folate, in the form of L-methyl-folate or L-folinic acid (but not with folic acid) [8,9,13]. These reduced forms of folate cross the blood brain barrier via a low-affinity transport, necessitating larger blood levels to obtain sufficient cerebral levels of folate.

Immunological Impacts in Fetal Development:

Antibodies are immunoproteins produced by B cells of the adaptive immune system in response to foreign agents. Antibodies bind to target antigens, which are regions of the foreign agents, and to neutralize them, such as by blocking binding or active sites. Typically, B cells that would produce antibodies to bind host proteins are eliminated, thus avoiding an attack on host cells by the immune system. However, in some instances, misdirected attacks occur in which antibodies are generated against host tissue. These autoantibodies typically do not cause harm to the adult brain as the blood-brain barrier (BBB) prevents their entry in the healthy state. Nonetheless, autoantibodies from the mother can cross the placenta during development and have been shown to be reactive against fetal brain proteins [14].

The clinical finding of ASD diagnosis linked to maternal autoantibodies is known as Maternal Autoantibody-Related (MAR)-ASD. Previous research has identified autoantibody targets that are common in mothers whose children were subsequently diagnosed with ASD. Specifically, one study found that maternal autoantibodies against LDHA, LDHB, CRMP1, and STIP1 were 23 times higher in mothers who went on to have children diagnosed as autistic, compared to mothers who had neurotypical children. Within the cohort of ASD children studied, the presence of these circulating maternal autoantibodies was also associated with a greater incidence of stereotypical behaviors [14].

Maternal stress, such as infection or exposure to environmental toxins, has been shown to trigger the immune response that results in the production of autoantibodies that can cross the placenta and react with fetal brain proteins. The resulting neurodevelopmental changes have been implicated in the pathogenesis of disorders such as autism spectrum disorder (ASD) [15-17]. The link between maternal stress and the development of ASD has been observed in both human and animal studies. For instance, maternal immune activation induced by viral or bacterial infection during pregnancy has been associated with an increased risk of ASD in the offspring. Similarly, exposure to environmental toxins such as polychlorinated biphenyls (PCBs) and organophosphate pesticides during pregnancy has been linked to increased autoantibody production and the development of ASD in the offspring. Overall, maternal stress appears to play a critical role in triggering the immune response that may result in neurodevelopmental changes leading toward a neuropsychiatric diagnosis such as ASD [16,18,19].

Clinical Trials to overcome CFD:

In clinical trials with ASD children, those who have

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FRA have improvement in their communication when given daily folate supplements (in the form of L-folinic acid, available as L-leucovorin, Aprofol AG, Zurich, Switzerland) for three months [20,21]. It is clear that supplementation with reduced folate can help overcome CFD, as the reduced form crosses the BBB via a low affinity transport unaffected by FRA. While this transport does not elevate brain levels of folate relative to blood levels, providing medicinal food levels of folate as a supplement is successful at restoring brain levels of this vitamin. Once CFD is reduced or eliminated, the ASD symptoms are lessened. We propose that it may then be possible to revise the diet of ASD children to reduce production of FRA, to provide a long-term means in reducing ASD symptoms. The key elements in such a dietary treatment are to reduce or remove foods that can stimulate FRA production and to provide food sources of natural folate. These two things, elimination of autoantibody stimulating foods and consumption of the reduced, natural form of folate, are key conditions for reducing severity of ASD symptoms. Diets that are richer in natural folate include the Mediterranean diet that focuses on olive oil, fresh vegetables and fruits, nuts, legumes and fish.

Prenatal origins of CFD:

A challenging fact of FRA contributing to CFD is the observation that this CFD likely starts early in development. Ramaekers [9,22] documented that when either or both parents have FRA, their child has higher odds of being ASD. It appears that elevated risk of childhood ASD may be related to elevated prenatal FRA, more so when the FRA is present in the mother, but also when it is present in the father. One study found FRA was present in 75.6% of autistic children, while the FRA prevalence was 34% in their mothers and 29% in their fathers, as compared to 3% FRA positivity in healthy controls. [23] Another study found FRA prevalence of 76% in autistic children, 75% in unaffected siblings, 69% in fathers and 59% in mothers, while the prevalence of FRA in unrelated normal controls was 29% [24].

Critical Periods in Neurological Development:
These findings are consistent with the presence of critical periods in development. [19] The presence of critical periods, first shown for the visual system, is the developmental time when sensory stimulation is necessary for visual perception to become established. If an animal is deprived of the sensory input during the critical period, the functional perception of visual images is impaired throughout its life. [25] Similarly, depletion of cerebral folate during an infant's fetal development may be similar to a critical period that could lead to increased severity of autism. We postulate that ASD may arise from a modification of critical periods, where CFD impairs neurological development, increasing the probability of a later diagnosis of ASD.

Pregnancy and FRA:

In pregnancy, the presence of FRA in the mother blocks folate delivery to the developing fetus. These FRAs are common in pregnancies that have births with spina bifida or ASD. [6] Identifying pregnancies where FRA is present and providing the pregnant mother with supplemental L-folinic acid or L-methyl-folate may permit sufficient folate to reach the fetus, in turn lowering the risk for developmental disorders, including ASD [26].

There may be an additional advantage to diagnosis of FRA in prospective parents. A recent report documented that supplementation with vitamin B-12 and reduced vitamin B-9 increased pregnancy and live birth in women who have experienced difficulty in conceiving, [27] indicating an essential role of these vitamins in pregnancy and healthy fetal development. Their observation that folic acid decreased fertility while reduced B-9 restored it reveals a key role of reduced folate in conception.

Folate and ASD:

Multiple clinical studies have shown that when FRA is present, supplementation with folate in its reduced form and with B-12 can overcome the FRA to permit sufficient folate for a child's brain development to continue its normal course 11,21].

Folate is a key vitamin in neural health. Evidence shows that most autistic people produce the autoantibody FRA, which blocks folate absorption into the brain, resulting in CFD. Stress may exacerbate this

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deficiency, worsening communication difficulties in ASD. [28] Diets that are rich in natural forms of folate, such as the Mediterranean diet, may help alleviate CFD, and when coupled with reduction in stress may improve communication in autistic people.

Folate and Critical Periods:

We postulate there are critical periods in the development of ASD, with the first period *in utero* and a later one in the first five years of life. These periods can predispose a child to be more likely to develop as ASD, and begin to set the conditions for ASD development (ages 2-5). Nutritional supplementation along with psychological counseling was effective for children under five in one study, [29] suggesting therapy and nutrition should be provided in the first years of life to maximize positive outcomes. Preliminary data from V. Stephanyshyn (personal communication) finds that treatment of FRA positive ASD children prior to age 5 is substantially more effective than later, substantially reducing communication issues in these young children after receiving L-folinic acid supplementation. Additionally, reports indicating that the presence of FRA can predispose a child for ASD [5,12] leads us to recommend that individuals presenting with FRA who may become pregnant be advised to take a prenatal supplement that includes a reduced form of folate (such as L-methyl-folate or L-folinic acid), and that children born to parents with FRA or children who have FRA would be advised to have nutritional supplementation to ensure sufficient levels of bioavailable vitamin B-9 for brain development.

Early Diagnosis:

Thus, for optimal treatment, early diagnosis may be essential, including making a prognostic diagnosis early in pregnancy. There is support for using the presence of FRA in either biological parent as a predictor for this antibody in the newborn and an increased probability of ASD development in childhood [23,24]. The test for FRA (FratNow, Philadelphia, PA) requires a blood sample, and if a non-invasive test were to become available for widespread screening, the FRA test could be used as confirmation and point to the therapeutic method that would be successful.

Several types of screening tests are under development for ASD. Lai et al [30] are using retinal images to rapidly screen children for autism. Duan et al [31] are using computer algorithms to assess children based on behavioral phenotypes. Elbattah et al [32] have a machine learning method to make more rapid diagnoses of autism in young children. We propose use of a simple diagnostic assessment screening based on the presence of ASD, depression or spina bifida in any immediate family member of the birth parents, followed by a FRA test. Any of these systems that allow a quick screening that can identify potential future ASD cases should be effective.

Conflict of Interest

The author has read and approved the final version of the manuscript. The author has no conflicts of interest to declare.

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