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Vitamin D and Autism:
Clinical Review

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Abstract

Background: Autism spectrum disorder (ASD) is a complex neurodevelopmental disorder with multiple genetic and environmental risk factors. The interplay between genetic and environmental factors has become the subject of intensified research in the last several years. Vitamin D deficiency has recently been proposed as a possible environmental risk factor for ASD.

Objective: The aim of the current paper is to review the research regarding the possible connection between ASD and vitamin D, and to address the issue of how vitamin D might be involved in the etiology or phenotypical expression of autism.

Results: Systematic data obtained by different research groups provide some support the possible role of vitamin D deficiency in the pathogenesis of ASD. There are two main areas of involvement of vitamin D in the human body, that may have direct impact on the development of ASD: (1) brain homeostasis, brain's own immune system and neurodevelopment, and (2) gene regulation.

Conclusion: Vitamin D deficiency – either during pregnancy or early childhood – may be an environmental trigger for ASD in individuals genetically predisposed for the broad phenotype of autism. On the basis of the results of the present review, we argue for the recognition of this possibly important role of vitamin D in ASD, and for urgent research in the field.

INTRODUCTION

Autism spectrum disorder (ASD) is a complex neurodevelopmental disorder with multiple genetic and environmental risk factors (Cannell 2010, Coleman and Gillberg 2011). Gene-environment interaction has recently become the focus of intensified ASD research (Freitag et al. 2010).

Among several established/proposed epidemiological influences on the development of ASD are the (a) much higher monozygotic (60-90%) than dizygotic (0-10%) twin concordance rates (Muhle 2004, Lichtenstein et al 2010); (b) large variability of phenotypic expression (even among monozygotic twins) (Lundström et al 2012), (c) distinct gender ratio (2-4 males to 1 female) (Nygren et al 2011); (d) relationship between autism and immune dysfunction (Coleman and Gillberg 2011); and (e) much increased rate of ASD among dark skinned children living at Northern latitudes (Cannell 2008, Goodman and Richards 1995; Gillberg 1995; Barnevik Olsson et al. 2008, 2010; Eyles 2010, Keen et al. 2010).

There is no agreement as to whether ASD prevalence is genuinely on the rise or if a higher reported rate in recent years might be secondary to better awareness, changing diagnostic trends and more sensitive diagnostic system (Coleman and Gillberg 2011). However, what is clear, is that the synergistic impact of neurotoxins has been steadily rising, either in the form of the industrial waste polluting soil, rivers and oceans, or in the form of additives in our food or materials of everyday life (Grandjean 2006 Lancet). Also, vitamin D deficiency has become common due to the increasingly urbanized lifestyle, rising rates of obesity, and recommendations to avoid sun exposure promulgated from the 1980s onwards (Holick 2005; Cannell 2006; Schwalfenberg 2007; Bosomworth 2011). Moreover, at northern latitudes, sunlight with the ultraviolet B fraction is available only during a limited period of the summer. Dark skinned individuals require about 5 to 10 times longer exposure to sunlight to produce vitamin D compared to fair skinned individuals (Clemens et al. 1982). Therefore, when moving to northern countries, those with dark skin run the risk of not reaching satisfactory vitamin D levels.

The “group of vitamins D” have a unique role in brain homeostasis, embryogenesis and neurodevelopment, immunological modulation, including the brain´s own immune system, ageing, and also, importantly, in gene regulation (Sigmundsdottir 2011, Ramagopalan et al. 2010, Harms et al. 2011). In addition to this effect, vitamin D is now believed to be involved in numerous other functions in the organism. To date, it has been shown to bind with more than 2,700 genes and to regulate the expression of more than 200 of them. Vitamin D is also known to be involved in healing processes (by reducing the risk of cells becoming malignant (Sigmundsdottir 2011).

Vitamin D deficiency – either during pregnancy or early childhood – has recently been proposed as a possible environmental risk factor for ASD (Cannell JJ 2008, Grant and Soles 2009).

Vitamin D: definition, biosynthesis and role in metabolism?

Vitamin D is not really a vitamin. By definition, vitamins are substances that are essential for body functioning but not produced by the body itself, meaning that they have to be obtained from exogeneous sources. Vitamin D does not fall into this category and should be rather viewed as a powerful steroid hormone,* since it is produced in the body by a cascade of chemical transformations, commencing with a key photochemical reaction in the skin on exposure to the ultraviolet rays of the sun, followed by a series of further chemical transformations. Its receptors have been found in many tissues and organs. Its best known role is to facilitate the calcium and phosphate absorption in the intestine, impacting directly on the formation of the bones and their density. The biosynthesis of calcitriol, the active form of vitamin D of vertebrates, starts from its prime precursor 7-dehydrocholesterol, which in turn is produced by a complex cascade from acetyl coenzyme A via mevalonic acid, farnesyl pyrophosphate, squalene, and lanosterol, and is eventually hydrogenated to produce cholesterol. 7-dehydrocholesterol first undergoes the key photochemical electrocyclization reaction in the skin, producing an intermediate that is spontaneously converted into calciferol (vitamin D3), or cholecalciferol to be precise and to emphasize its chemical relation to cholesterol. Since the first reaction requires

irradiation with UV light (at 290-315 nm), it can only proceed in the skin, i.e., within the reach of the UV rays. Cholecalciferol is then transported to the liver, where it is hydroxylated in the side-chain (at position 25) by the microsomal enzyme 25-hydroxylase to produce calcidiol [25(OH)D, or cholecalcidiol]. Finally, the latter compound is further transported to the proximal tubules of the kidneys, where it is further hydroxylated (at position 1α) by the enzyme 1α-hydroxylase to finally produce calcitriol [i.e., 1,25-dihydroxycalciferol, $1,25(OH)_2D$, or cholecalcitriol], the active compound (vitamin D1). The levels of the enzyme required for the final hydroxylation are controlled by parathyroid hormone, whose secretion is, in turn, triggered by low concentrations of calcium or phosphate. The latter enzymatic hydroxylation reaction, producing calcitriol, has also been found to occur in lymphocytes.

For the sake of simplicity and to avoid confusion, which is widespread in the literature, we will use the following nomenclature for the vitamin D family originating from 7-dehydrocholesterol: **calciferol** for cholecalciferol (vitamin D3), **calcidiol** for 25-hydroxy-cholecaciferol (vitamin D2), and **calcitriol** for 1,25-dihydroxy-cholecalciferol (vitamin D1). Where the literature does not discriminate, we will refer to **vitamin D** in general. Analogues originating from ergosterol would then be specifically highlighted as **ergocaciferol**, etc.

Vitamin D in the body follows first-order mass action kinetics (Holick 2005), which means that at serum levels lower than 50 nmol/L the majority of ingested or sun-derived vitamin D is immediately diverted to metabolic needs leaving nothing to its higher functions within the brain, immune system, or gene regulation.

Estrogens have the ability to increase the levels of calcitriol while testosterone does not. Vitamin D deficiency during gestation has been linked with dysregulation of many proteins involved in brain development. Thus, rat pups developed increased brain size and enlarged ventricles – both abnormalities reported in a minority of autism cases (Cannell 2008; Lainhart et al. 2006). Calcitriol also has the capacity to down-regulate inflammatory cytokines, which, according to a few studies, have been found to be elevated in children with autism (Singh 1997; 2002). Some families of children with autism have also been reported to experience clusters of autoimmune disorders (Atladóttir et al. 2009).

Deficiency in calcitriol may stem from the inherited polymorphisms in cytochrome P450 gene CYP27B1, which is required for the hydroxylation of calcidiol (Cannell 2008).

There are two main areas of involvement of vitamin D in the human body, which may have direct impact on the development of ASD: (1) brain homeostasis, brain's own immune system and neurodevelopment, and (2) gene regulation.

The aim of the current paper is to review research findings regarding the connection between ASD and vitamin D.

METHODS

A literature search covering the period January 1 1995 through October 31 2011 was made in PubMed, the Web of Knowledge, EBSCO OVID, MEDLINE, PsycARTICLES, Psychology and Behavioral Sciences Collection, PsycINFO, SocINDEX databases with Full Text Number of Hits.

The search strategy was as follows: vitamin d or vitamin D or ergocalciferol or vitamin d2 or vitamin D2 or vitamin d 2 or vitamin D 2 or cholecalciferol or vitamin d3 or vitamin D3 or vitamin d 3 or vitamin D 3 or calcitriol or vitamin 1,25 D3 or vitamin 1,25 d3 or vitamin 1,25 D 3 or vitamin 1,25 d 3 or calcidiol or vitamin 25 D or vitamin 25D or 25 hydroxy vitamin d or 25-OHD or 25- hydroxyvitamin D or 25 hydroxyvitamin D or 25 hydroxy vitamin d or 25 hydroxyvitamin D AND autism or autism spectrum disorder or ASD or Asperger.

RESULTS

A systematic literature search yielded 35 articles from PubMed that dealt with autism and vitamin D in one way or another. Only four studies have looked at vitamin D serum levels in individuals with diagnosed ASD.

The search was then supplemented by articles reporting on vitamin D and its relationship to brain function.

Results were grouped under seven subheadings, relating to different aspects of vitamin D and autism*: (1) D-vitamin in individuals with autism, (2) Genetics and gene regulation, (3) Brain homeostasis, neurodevelopment and brain's own immune system, (4) Epilepsy/seizures, (5) Medication during pregnancy, (6) Nutritional status during pregnancy and early childhood, and (7) Latitudinal effects and ethnicity.*

1) ***Autism – vitamin D: research studies documenting vitamin D-levels in individuals with autism and their family members HERE YOU MUST PUT a brief table with the few studies that have ever looked at actual vitamin D-levels in autism and describe and conclude on the basis of that table in a brief text section!***
2) ***Autism – vitamin D: genetics and gene regulation***

Previous genetic findings in ASD indicated that the likely causes of ASD involve mutations or common variants in genes, which are involved in (1) cell-cell interaction and synaptic function, including development of dendritic spines, (2) neuronal migration and growth, or (3) excitatory and inhibitory neurotransmission (Freitag*). Animal research has demonstrated that Vitamin D plays a role, to some extent, in all of these brain processes (Eyles el at. 2011)

Vitamin D exerts its effects on genes through the vitamin D receptor (VDR), which binds to specific locations of the genome to influence gene expression. Employing the ChIP-seq technique, **Ramagopalan et al (2010)** isolated fragments of genomic DNA

bound to the VDR before and after treatment of cells with calcitriol, and then sequenced the DNA fragments. By mapping the sequences back to the genome, the group identified more than 2,700 sites of VDR binding, a number showing the importance of vitamin D in human body and also the wide variety of biological pathways in which vitamin D plays a role in. These findings support the hypothesis that vitamin D interacts with genes in the pathogenesis of diseases and underscores the serious risks of vitamin D deficiency (Ramagopalan SV 2010; Kalueff 2006).

Prenatal vitamin D deficiency or defects in its metabolism can disrupt normal neuro-development as vitamin D plays a role in neuronal growth (Ciu et al. 2007) and in regulating cell proliferation in the developing brain. This role is corroborated by detecting calcitriol and its receptors in a variety of brain tissues early in embryogenesis (McGrath et al. 2001).

Recent studies portray the role of signalling pathways in the brain and the synapse structure as crucial to the development of ASD. Most plausible explanations consider an interplay between underlying genetic and biological factors. The importance of this hypothesis is that risk factors for autism involve a combination of genetic susceptibilities and environmental exposures. Interestingly, cytogenetic aberrations in autistic individuals have been located in nearly every chromosome (Gillberg 1998).

3) Autism – vitamin D: neurodevelopment, brain homeostasis and the brain's own immune system

Table I provides an overview of some of the most important papers published in recent years on the role of vitamin D for brain development and brain functions.

In view of the presence of the vitamin D system, its activating enzymes, and vitamin D receptors (VDR) in the brain, and assuming that they are involved in normal brain functioning, an argument can be made for re-classifying the vitamin D group as neuro-steroids. In addition, the presence of high levels of VDR in the developing brain from very early stages and its increase with gestational age, suggests a role for vitamin D in neurodevelopment. It seems reasonable to speculate that vitamin D deficiency during development could be a risk-modifying factor in relation to other factors, such as maternal infection, stress, or neurotoxicity (**Cannell 2008**; Grant 2009; Bodnar 2007).

Various animal studies have demonstrated the role of Vitamin D in mammalian brain development. For example in the down-regulation of neurologically harmful cytokines in rat?? brain (Moore et al. 2005), is it this reference about age-related inflammatory changes in the rat hippocampus? If so, maybe it is not needed here./ef the partial reversal of brain damage in rats (Burne et al. 2004), and to increase cellular levels of glutathione (Garcion et al. 2002), which is capable of removing free radicals and to chelate heavy metals, including mercury (Kern et al. 2006). Vitamin D also plays a trophic role in the central nervous system (CNS) and also protects cultured cortical neurons from glutamate excitotoxicity via up regulation of the VDR **(Blaylock 2009).** In children with autism, Vitamin D deficiency has been shown to dysregulate 36 proteins involved in brain development (Almeras 2007). Maybe this paragraph is a bit too far from autism?/ef THIS IS NOT AT ALL WHAT THE ALMERAS PAPER SHOWS – INSTEAD THEIR PAPER IS ALL ABOUT ADULT RATS!!! I WONDER IF THIS WHOLE SECTION SHOULD BE DELETED OR AT LEAST REPHRASED IN A MAJOR WAY. THE OVERENTHUSIASTIC TONE IS

STILL PERVASIVE IN THIS NEW VERSION, AND ALL THE CLAIMS HAVE TO BE TONED DOWN. SEVERAL OF THE REFERENCES THAT I HAVE LOOKED AT DO NOT AT ALL SUPPORT THE VERY STRONG CLAIMS BEING MADE IN THIS TEXT.

Many studies show that vitamin D plays a major role in brain homeostasis. For example, Taniura et al. (2006) has demonstrated a very high level of activity of the vitamin D specific DNA-response element (VDRE) in the cerebellum, a brain region that has been frequently linked to autism and also other neurodevelopmental disorders. Other areas of the brain, e.g hippocampus, limbic system, pituitary, substantia nigra, white matter, diencephalon and cerebral cortex also have higher concentrations of VDR and the enzyme necessary for conversion of calcitriol (Eyles 2005)..

Vitamin D affects numerous cellular functions and animal studies have demonstratred a specific rol in cell differentiation, neurotrophic factor expression, cytokine regulation, neurotransmitter synthesis, intracellular calcium signalling, anti-oxidant activity, and the expression of genes and proteins involved in neuronal differentiation, structure and metabolism **(Eyles 2011)**.

Animal studies showed that Vitamin D deficiency has a negative effect on embryonic neurodevelopment, manifested, e.g., in decreased neurotrophic factor levels, increased mitosis, and decreased apoptosis, enhanced proliferation, and some changes in the brain morphology and altered behaviour (Eyles et al 2006; **Harms et al 2011**). It is

known that microglia contain vitamin D receptors (VDR) and when they are activated, they can actually synthesise calcitriol (Neveu et al. 1994; Lefebvre et al. 2003).

A very high activity was observed for vitamin D specific DNA-response element (VDRE) binding in the cerebellum of a rat (Taniura 2006). In human brain, there are higher concentrations of VDR in the cerebellum, hippocampus, limbic system, pituitary, substantia nigra, white matter diencephalon, and cerebral cortex, as well as of the enzyme 1α-hydroxylase (encoded by the CYP27B1 gene), which is essential for the synthesis of the active form, i.e., calcitriol (Eyles 2005).

In addition, calciferol induces the production of anti-inflammatory cytokines IL-10, IL-4 and TGF-β1. Its deficiency has been shown to suppress NOS has NOS been explained? and to result in elevation of glutathione levels. Furthermore, calciferol significantly stimulates the production of glial-derived neurotropic factor (GDNF) and thus serves as a neuroprotective agent (Cantorna MT 1996).

3) Autism – vitamin D and epilepsy/seizures

Animal and clinical studies have demonstrated the neuroprotective role of vitamin D in epilepsy, multiple sclerosis, Parkinson's disease, and chronic stress. As epilepsy and convulsions are common in autism, it is most interesting to note calcitriol's neuroprotective effects via its inhibitory effect on Ca^{2+} influx. Calcitriol up-regulates the expression of the Ca^{2+}-binding proteins calbindin and parvalbumin in motor neurons (Alexianu 1998), which chelate intracellular Ca^{2+} and thus limit the excitotoxicity (Harms 2011).

Vitamin D deficiency thus might lead to disruption in calcium signalling and homeostasis, which might account for some pathologies and disrupted information processing typical for autism. Severe vitamin D deficiency (calcidiol) has been shown to be associated with seizures caused by disruption of calcium levels (Mehrotra 2009). Vitamin D has also been shown to increase the electroconvulsive threshold for seizures, to decrease the severity of seizures, and to enhance the effect of the antiepileptic medication (Siegel 1984; Kalueff 2005; Borowicz 2007). The protection against excitotoxicity might also explain the protective action of vitamin D against seizures (Harms 2011).

However, antiepileptic drugs are known to decrease vitamin D levels, which further complicate research of the potential link between vitamin D deficiency and increased risk of seizures (Bergquist 2007).

4) Autism – vitamin D and medication during pregnancy

Bromley et al. (2009) reported on an increased incidence of autism among infants of mothers taking antiepileptic medication, especially sodium valproate, during pregnancy. This finding may suggest low vitamin D levels as a possible mechanism deserving further investigation. An inverse relationship between the antiepileptic drugs and 25-hydroxyvitamin D levels have been (Borowicz et al. 2007) demonstrated in animal research.

5) Autism – vitamin D and nutritional status

One of the suggested mediating roles of vitamin D in aetiology of autism is its involvement in absorption of magnesium (Mg) **(Johnson 2000)**, one of several very important micronutrients, known for several crucial roles in brain development, e.g.,

preventing oxidative damage. The absorption of magnesium requires adequate levels of parathyroid hormone and vitamin D and may thus be dependent on nutrition as well as season and sun exposure. There are many studies examining a possible relationship between month and season of birth and risk for autism spectrum disorders. However, results are diverging as to potential risk period and some have found no support at all for a seasonality hypothesis (Gillberg 1990, Landau et al. 1999, Kolevzon et al. 2006, Hebert et al. 2010, Zerbo et al. 2011).

It has been suggested that supplementation with zinc, magnesium, ascorbic acid and the vitamins necessary for magnesium metabolism, namely vitamin D and B6, started before pregnancy, may prevent some cases of autism, schizophrenia, epilepsy and Parkinson's disease (Johnson 2000, 2011?). which year for the reference?/ef

Is this the reference? Johnson DD, Wagner CL, Hulsey TC, McNeil RB, Ebeling M, Hollis BW. Vitamin D deficiency and insufficiency is common during pregnancy. Am J Perinatol 2011;28(1):7-12.

Lindsay et al. (2006) used a quantitative Food Frequency Questionnaire (FFQ) to prospectively study the nutritional intake of 20 children (5-13 years old) with autism. The results of this questionnaire study suggested that fifty percent of these children with autism were likely to have inadequate vitamin D intake.

Several observational and clinical studies reported vitamin D deficiency in children with autism as a consequence of their highly selective nutritional intake. Individuals with autism have been reported very often to have selective nutritional manners, which can potentially exacerbate their vitamin D deficiency/insufficiency status. Several studies reported an association between the vitamin D status and nutrition in children or young people with autism. Thus, **Shamberger (2010)** in an ecological

study across the 50 United States, showed that infants who were solely breast-fed had diets low in Vitamin D and other nutrients and that in States with high rates of exclusive breast-feeding, autism rates were also higher. This implies the need for vitamin supplementation during lactation period as nutrition may be important in the development of autism.

Herndon et al. (2008) found few differences in average nutritional intake between children with autism and typically developing children in Colorado, USA. However, a large proportion of children in both groups did not meet the national recommendations for daily intake of fibre, calcium, iron, vitamin E and vitamin D. Very similar results have been reported recently by **Sadowska J and Cierebiej M (2011)** in Poland.

There are 4 case studies reporting an extreme form of the inadequate nutritional status due to the selectivity with food and food avoidant-behaviours in children or young people with autism, leading to the symptomatic nutritional rickets or painful leg weakness. In all cases a severe vitamin D deficiency was diagnosed and then successfully treated with adequate diet and vitamin D supplementation **(Noble et al 2006; Weig SG 2009; Stewart C and Latif A 2007; Clark JH 1993)**. This reflects high rates of acquired hypo-calcemia and hypo-vitaminosis D in subgroups of children and adolescents with autism and suggests the importance of comprehensive history-taking, careful diet assessment, and when appropriate, screening for vitamin deficiencies as an integral part of every child with autism's medical care (Noble et al 2006).

Three studies investigated plasma levels of vitamin D in individuals with autism. **Humble et al (2010)** tested Vitamin D levels in adult outpatients with a range of psychiatric disorders and found that those with a diagnosis of autism or schizophrenia had significantly lower levels than other groups. **This study demonstrated a considerable improvement in several patients of some of their psychiatric symptoms, e.g. psychosis and depression. However, there are some limitations of this study as there is very little information on details of the treatment and there was no control group. Molloy et al. (2010)** explored the comparison of the actual plasma 25(OH)vitamin D_3 levels in a cohort of Caucasian boys with ASD diagnosis (4-8 years old) and a group of age-matched typically developing comparison boys having intravenous catheters placed for outpatient tonsillectomies. There were no differences observed in the levels between participants with ASD and controls, however the majority of all children in this cohort (61%) had low vitamin D levels (<20 ng/ml or <50 nmol/L). This is the minimum concentration recommended by the American Academy of Paediatrics to ensure adequate bone health. This supports major health concerns regarding the use of sun blocks, increased indoor activities with limited sun exposure among children and inadequate dietary sources. In addition the limitation of this study has to be taken into account -

Meguid et al. (2010) reported a cohort of Egyptian children with ASD having significantly lower levels of both 25(OH) vitamin D_3 (28.5 ng/ml) and 1,25$(OH)_2$ vitamin D_3 (27.1 ng/ml) levels as well as lower calcium serum values compared to healthy controls. No significant difference was found for the month and season of birth among either group.

Although these results regarding vitamin D levels in children and adults with autism appear conflicting, of the three studies, two are subject to significant methodological problems. The Molloy study suffered from the fact that the control children were likely to have had some degree of inflammation, which could possibly have affected the vitamin D levels. The Humble study did not have a control group and studied a range of disorders, not only autism. In contrast, in the Meguid study, vitamin D was measured in an appropriate manner and the recruitment of cases and controls minimised bias. Taking these studies together, they are suggestive that there may be an association between low Vitamin D and autism and this will be an important direction for future research.

6) Autism – vitamin D, clinical studies; latitudinal effects and ***ethnicity***

Grant et al. (2009) has found a strong effective latitudinal (related to wintertime solar UVB radiation) increase in infantile autism prevalence. These finding are consistent with the hypothesis of maternal vitamin D deficiency being a risk factor for autism,..

In 1995 **Gillberg et al. (1995**) reported on 3 boys with autism, born in one area of Sweden and with mothers coming from Uganda and discussed possible reasons for the high autism rate in this particular ethnic subgroup.

Due to an increased prevalence of autism in children of Somali origin living in Sweden, and the evidence that low vitamin D impacts adversely on brain development, serum levels of 25-hydroxyvitamin D were analysed in mothers of Somali origin with and without a child with autism **(Fernell et al. 2010).** Both groups of mothers of Somali origin had significantly lower values of 25-hydroxyvitamin D

compared to Swedish mothers. The difference in the levels of 25-hydroxyvitamin D between mothers of Somali origin with and without a child with autism was not statistically significant, but the lowest values were found in mothers with a child with autism. The authors concluded that the findings regarding very low vitamin D levels generally in Somali mothers have considerable consequences in a public health perspective.

A review by **Dealberto (2010)** found several studies reporting increased rates of autism among dark-skinned immigrant mothers, especially those who moved to high latitudes. The results of this review are consistent with the hypothesis that the maternal deficiency (or insufficiency) levels of vitamin D may be associated with autism.

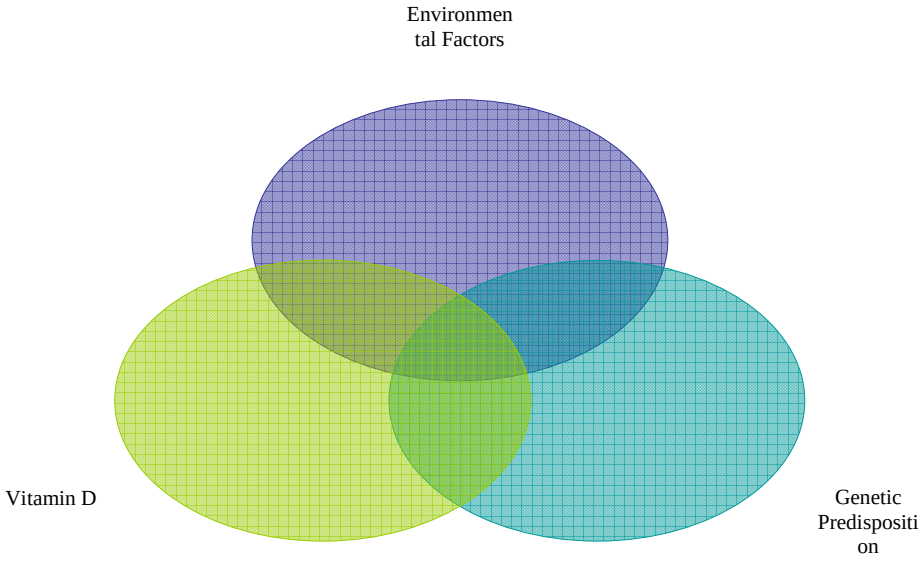
Although the prevalence rates of autism in African and Arabic countries have not yet been clearly established, **Bakare et al. (2011)** in his review reports autism and hypomelanotic disorders (such as albinism) can co-occur. This association between autism and oculo-cutaneous albinism led Bakare (2011) to formulate a hypothesis of individuals with hypo-melanotic skin types being genetically more predisposed to autism with an acknowledged modulatory role of vitamin D deficiency. Aetiological factors postulated included post-encephalitic infection, genetic and auto-immune factors, and vitamin D deficiency.

A list of clinical studies of vitamin D and autism are presented in Table 2.

DISCUSSION

Vitamin D deficiency – either during pregnancy or early childhood – has recently been proposed as a possible environmental risk factor for ASD. A large number of studies support the role of vitamin D in numerous cellular functions; in particular cell differentiation, neurotrophic factor expression, cytokine regulation, neurotransmitter synthesis, intracellular calcium signalling, anti-oxidant activity, to partially reverse brain damage (Burne et al. 2004), and to increase cellular levels of glutathione (Garcion et al. 2002) and the expression of genes and proteins involved in neuronal differentiation, structure and metabolism (Eyles 2011). Results of these studies have been translated into theories and hypotheses regarding a possible involvement and role of vitamin D in aetiology and/or phenotype expression of autism. However, most of these theories are still only hypotheses, some more plausible than others. Most of these hypotheses support the notion that neither genetic nor environmental exposure operate alone. Risk factors involve a combination of genetic susceptibilities and environmental exposures, Figure 1.

Fig 1 Vitamin D role of a key modulator in a complex interplay of genetic predispositions and environmental factors in the aetiology of autism



Latitudinal effects, skin pigmentation and some medications play a significant role in pregnancy and support the hypothesis of vitamin D deficiency involvement in etiology of autism. There is now evidence to recommend that all pregnant women should be screened for vitamin D deficiency and the supplement dosage should reflect the needs of both mother and foetus, especially during the third trimester and lactation.

Interest in ongoing research in schizophrenia might provide possible clues to the research of autism as there are some parallels between aetiology of both disorders and hypothetically both may be related to prenatal and developmental vitamin D deficiency (Grant et al. 2009; Humble et al. 2010).

When children with ASD were studied as to level of vitamin D status they were repeatedly shown to have inadequate intake of vitamin D and other nutrients due to

their highly selective intake of food and thus children with autism are one of the populations that are at a great risk of being deficient in vitamin D. Thus, patients with ASD should have their diet carefully assessed and, when appropriate, screened for vitamin deficiencies as an integral part of the child/patient's medical care (Noble et al 2006).

CONCLUSION

The findings obtained over the past 15 years - including in animal studies, human molecular, cellular and physiologic research, post-mortem brain studies, neuro-imaging studies, and genetic studies - support the unique role of the D vitamins in numerous human body processes. Although a very limited literature provides only weak support for the hypothesis of the modulatory role of vitamin D in the aetiology and phenotype of autism specifically, there is an urgent need for intensified research in this important area.

Some references

Ey E, Leblond CS, Bourgeron T. Behavioral profiles of mouse models for autism spectrum disorders. Autism Res 2011;4(1):5-16.

Cui X, McGrath JJ, Burne TH, Mackay-Sim A, Eyles DW. Maternal vitamin D depletion alters neurogenesis in the developing rat brain. Int J Dev Neurosci 2007;25(4):227-32.

McGrath J, Feron F, Eyles D, Mackay-Sim A. Vitamin D: the neglected neurosteroid? Trends Neurosci. 2001;24(10):570-2.

Gillberg C. Chromosomal disorders and autism. J Autism Dev Disord. 1998;28(5):415-25. Review.

Kinney DK, Barch DH, Chayka B, Napoleon S, Munir KM. Environmental risk factors for autism: do they help cause de novo genetic mutations that contribute to the disorder? Med Hypotheses 2010;74(1):102-6.

Cannell JJ. Autism and vitamin D. Med Hypotheses. 2008;70(4):750-9.

Grant WB, Soles CM. Epidemiologic evidence supporting the role of maternal vitamin D deficiency as a risk factor for the development of infantile autism. Dermatoendocrinol 2009;1(4):223-8.

Ramagopalan SV, Heger A, Berlanga AJ, Maugeri NJ, Lincoln MR, Burrell A, Handunnetthi L, Handel AE, Disanto G, Orton SM, Watson CT, Morahan JM, Giovannoni G, Ponting CP, Ebers GC, Knight JC. A ChIP-seq defined genome-wide map of vitamin D receptor binding: associations with disease and evolution. Genome Res. 2010;20(10):1352-60.

Harms LR, Burne TH, Eyles DW, McGrath JJ. Vitamin D and the brain. Best Pract Res Clin Endocrinol Metab 2011;25(4):657-69.

Sigmundsdottir H. From the bench to the clinic: New aspects on immunoregulation by vitamin D analogs. Dermatoendocrinol 2011;3(3):187-192.

Gillberg C, Schaumann H, Gillberg IC. Autism in immigrants: children born in Sweden to mothers born in Uganda. J Intellect Disabil Res 1995;39 (Pt 2):141-4.

Fernell E, Barnevik-Olsson M, Bågenholm G, Gillberg C, Gustafsson S, Sääf M. Serum levels of 25-hydroxyvitamin D in mothers of Swedish and of Somali origin who have children with and without autism. Acta Paediatr 2010;99(5):743-7.

Dealberto MJ. Prevalence of autism according to maternal immigrant status and ethnic origin. Acta Psychiatr Scand 2011:123:339–348

Humble MB, Gustafsson S, Bejerot S. Low serum levels of 25-hydroxyvitamin D (25-OHD) among psychiatric out-patients in Sweden: relations with season, age, ethnic origin and psychiatric diagnosis. J Steroid Biochem Mol Biol. 2010;121(1-2):467-70.

Goodman R, Richards H. Child and adolescent psychiatric presentations of second-generation Afro-Caribbeans in Britain. Br J Psychiatry. 1995;167(3):362-9.

Barnevik-Olsson M, Gillberg C, Fernell E. Prevalence of autism in children born to Somali parents living in Sweden: a brief report. Dev Med Child Neurol. 2008;50(8):598-601.

Barnevik-Olsson M, Gillberg C, Fernell E. Prevalence of autism in children of Somali origin living in Stockholm: brief report of an at-risk population. Dev Med Child Neurol. 2010;52(12):1167-8.

Eyles DW. Vitamin D and autism: does skin colour modify risk? Acta Paediatr. 2010;99(5):645-7.

Keen DV, Reid FD, Arnone D. Autism, ethnicity and maternal immigration. Br J Psychiatry. 2010;196(4):274-81.

Clemens TL, Adams JS, Henderson SL, Holick MF. Increased skin pigment reduces the capacity of skin to synthesise vitamin D3. Lancet. 1982;1:74-76.

Bakare MO, Munir KM, Kinney DK. Association of hypomelanotic skin disorders with autism: links to possible etiologic role of vitamin-D levels in autism? Hypothesis (Tor). 2011;9(1). pii: e2.

Zerbo O, Iosif AM, Delwiche L, Walker C, Hertz-Picciotto I. Month of conception and risk of autism. Epidemiology 2011;22(4):469-75.

Hebert KJ, Miller LL, Joinson CJ. Association of autistic spectrum disorder with season of birth and conception in a UK cohort. Autism Res 2010;3(4):185-90.

Kolevzon A, Weiser M, Gross R, Lubin G, Knobler HY, Schmeidler J, Silverman JM-Reichenberg A. Effects of season of birth on autism spectrum disorders: fact or fiction? Am J Psychiatry 2006;163(7):1288-90.

Landau EC, Cicchetti DV, Klin A, Volkmar FR. Season of birth in autism: a fiction revisited. J Autism Dev Disord 1999;29(5):385-93.

Gillberg C. Do children with **autism** have March birthdays? Acta Psychiatr Scand. 1990;82(2):152-6.

References[1;2]

Reference List

(1) Moan J, Juzeniene A. Solar radiation and human health. *Journal of Photochemistry and Photobiology B: Biology 101 (2) (pp 109-110), 2010 Date of Publication: November 2010* 2010;(2):November.

(2) Muhle R, Trentacoste SV, Rapin I. The genetics of autism. [Review] [222 refs]. *Pediatrics* 2004; 113(5):e472-e486.

1 Almeras L, Eyles D, Benech P, Laffite D, Villard C, Patatian A et al. Developmental vitamin D deficiency alters brain protein expression in the adult rat: Implications for neuropsychiatric disorders. *Proteomics 7 (5) (pp 769-780), 2007 Date of Publication: Mar 2007*(5):Mar.

2 Atladottir HO, Pedersen MG, Thorsen P, Mortensen PB, Deleuran B, Eaton WW et al. Association of family history of autoimmune diseases and autism spectrum disorders. *Pediatrics 124 (2) (pp 687-694), 2009 Date of Publication: August 2009*(2):August.

3 Bakare MO, Munir KM. Autism spectrum disorders (ASD) in Africa: A perspective. *African Journal of Psychiatry (South Africa) 14 (3) (pp 208-210), 2011 Date of Publication: 2011* 2011;(South Africa):2011.

4 Barnevik-Olsson M, Gillberg C, Fernell E. Prevalence of autism in children of Somali origin living in Stockholm: brief report of an at-risk population. *Developmental Medicine and Child Neurology* 2010; 52(12):1167-1168.

5 Bejerot S, Humble M. [Increased occurrence of autism among Somali children--does vitamin D deficiency play a role?]. *Tidsskrift For Den Norske L+ªgeforening: Tidsskrift For Praktisk Medicin, Ny R+ªkke* 2008; 128(17):1986-1987.

6 Blaylock R, Strunecka A. Immune-Glutamatergic Dysfunction as a Central Mechanism of the Autism Spectrum Disorders. *Current Medicinal Chemistry* 2009; 16(2):157-170.

7 Borowicz KK, Morawska M, Furmanek-Karwowska K, Luszczki JJ, Czuczwar SJ. Cholecalciferol enhances the anticonvulsant effect of conventional antiepileptic drugs in the mouse model of maximal electroshock. *European Journal of Pharmacology* 2007; 573(1-3):111-115.

8 Cannell J. Vitamin D "advertisement". *American Journal of Obstetrics & Gynecology* 202(5):e9-10.

9 Cannell JJ, Hollis BW. Use of vitamin D in clinical practice. [Review] [97 refs]. *Alternative Medicine Review* 2008; 13(1):6-20.

10 Cannell JJ. Autism and vitamin D. *Med Hypotheses* 2008; 70(4):750-759.

11 Cannell JJ, Vieth R, Willett W, Zasloff M, Hathcock JN, White JH et al. Cod liver oil, vitamin A toxicity, frequent respiratory infections, and the vitamin D deficiency epidemic. *Annals of Otology, Rhinology and Laryngology 117 (11) (pp 864-870), 2008 Date of Publication: November 2008* 2008;(11):November.

12 Cannell JJ. On the aetiology of autism. *Acta Paediatrica* 2010; 99(8):1128-1130.

13 Cannell JJ. Autism and vitamin D. *Med Hypotheses* 2008; 70(4):750-759.

14 Clark JH, Rhoden DK, Turner DS. Symptomatic Vitamin-A and Vitamin-D Deficiencies in An 8-Year-Old with Autism. *Journal of Parenteral and Enteral Nutrition* 1993; 17(3):284-286.

15 Dealberto M. Ethnic origin and increased risk for schizophrenia in immigrants to countries of recent and longstanding immigration. *Acta Psychiatr Scand* 2010; 121(5):325-339.
16 Eyles D, Burne T, Mcgrath J. Vitamin D in fetal brain development. *Seminars in Cell and Developmental Biology 22 (6) (pp 629-636), 2011 Date of Publication: August 2011*(6):August.
17 Eyles D, Brown J, Mackay-Sim A, Mcgrath J, Feron F. Vitamin D3 and brain development. *Neuroscience 118 (3) (pp 641-653), 2003 Date of Publication: 2003*(3):2003.
18 Eyles D, Burne T, Mcgrath J. Vitamin D in fetal brain development. *Seminars in Cell and Developmental Biology 22 (6) (pp 629-636), 2011 Date of Publication: August 2011* 2011;(6):August.
19 Eyles DW. Vitamin D and autism: does skin colour modify risk? *Acta Paediatrica* 2010; 99(5):645-647.
20 Eyles DW, Burne THJ, Alexander S, Cui X, McGrath JJ. The developmental vitamin D (DVD) model of schizophrenia. *Animal Models of Schizophrenia and Related Disorders Neuromethods 59 (pp 113-125), 2011 Date of Publication: 2011* 2011;(pp 113-125):2011.
21 Fernell E, Barnevik-Olsson M, Bagenholm G, Gillberg C, Gustafsson S, Saaf M. Serum levels of 25-hydroxyvitamin D in mothers of Swedish and of Somali origin who have children with and without autism. *Acta Paediatrica* 2010; 99(5):743-747.
22 Goodman R, Richards H. Child and adolescent psychiatric presentations of second-generation Afro-Caribbeans in Britain. *British Journal of Psychiatry* 1995; 167(3):362-369.
23 Grandjean P, Landrigan P. Developmental neurotoxicity of industrial chemicals. *Lancet 368 (9553) (pp 2167-2178), 2006 Date of Publication: 16 Dec 2006*(9553):16.
24 Harms LR, Burne THJ, Eyles DW, McGrath JJ. Vitamin D and the brain. *Best Practice and Research: Clinical Endocrinology and Metabolism 25 (4) (pp 657-669), 2011 Date of Publication: August 2011* 2011;(4):August.
25 Herndon AC, DiGuiseppi C, Johnson SL, Leiferman J, Reynolds A. Does nutritional intake differ between children with autism spectrum disorders and children with typical development? *Journal of Autism & Developmental Disorders* 2009; 39(2):212-222.
26 Humble MB, Gustafsson S, Bejerot S. Low serum levels of 25-hydroxyvitamin D (25-OHD) among psychiatric out-patients in Sweden: relations with season, age, ethnic origin and psychiatric diagnosis. *Journal of Steroid Biochemistry & Molecular Biology* 2010; 121(1-2):467-470.
27 Johnson S. Micronutrient accumulation and depletion in schizophrenia, epilepsy, autism and Parkinson's disease? *Med Hypotheses* 2001; 56(5):641-645.
28 Lindsay RL, Eugene AL, Aman MG, Vitiello B, Posey DJ, McDougle CJ et al. Dietary status and impact of risperidone on nutritional balance in children with autism: a pilot study. *Journal of Intellectual & Developmental Disability* 2006; 31(4):204-209.
29 Mcgrath J, Feron F, Eyles D, Mackay-Sim A. Vitamin D: the neglected neurosteroid? *Trends in Neurosciences* 2001; 24(10):570-572.
30 Meguid NA, Hashish AF, Anwar M, Sidhom G. Reduced serum levels of 25-hydroxy and 1,25-dihydroxy vitamin D in Egyptian children with autism. *Journal of Alternative & Complementary Medicine* 2010; 16(6):641-645.
31 Molloy CA, Kalkwarf HJ, Manning-Court, Mills JL, Hediger ML. Plasma 25(OH)D concentration in children with autism spectrum disorder. *Developmental Medicine & Child Neurology* 2010; 52(10):969-971.
32 Muhle R, Trentacoste SV, Rapin I. The genetics of autism. [Review] [222 refs]. *Pediatrics* 2004; 113(5):e472-e486.
33 Nakamura K, Takahashi T, Sasaki Y, Tsuyuoka R, Okuno Y, Kurino M et al. 1,25-dihydroxyvitamin D3 differentiates normal neutrophilic promyelocytes to monocytes/macrophages in vitro. *Blood* 1996; 87(7):2693-2701.
34 Noble JM, Mandel A, Patterson MC. Scurvy and rickets masked by chronic neurologic illness: revisiting "psychologic malnutrition". *Pediatrics* 2007; 119(3):e783-e790.
35 Sadowska J, Cierebiej M. Evaluation of the nutrition manner and nutritional status of children with autism. Preliminary investigations. *Pediatria Wspolczesna 13 (3) (pp 155-160), 2011 Date of Publication: 2011*(3):2011.
36 Sakurai T, Ramoz N, Barreto M, Gazdoiu M, Takahashi N, Gertner M et al. Slc25a12 disruption alters myelination and neurofilaments: a model for a hypomyelination syndrome and childhood neurodevelopmental disorders. *Biological Psychiatry* 2010; 67(9):887-894.

37 Shamberger RJ. Autism linked to nutrition. *FASEB Journal Conference: Experimental Biology 2010, EB Anaheim, CA United States Conference Start: 20100424 Conference End: 20100428 Conference Publication: (var pagings) 24 , 2010 Date of Publication: April, 2010* 2010;(var.pagings):April.
38 Shamberger RJ. Autism Rates Associated with Nutrition and the WIC Program. *Journal Of The American College Of Nutrition* 2011; 30(5):348-353.
39 Singh B. New media campaign highlights dangers of maternal vitamin D deficiency. *Life Extension* 2009; 15(6):19.
40 Stewart C, Latif A. Symptomatic nutritional rickets in a teenager with autistic spectrum disorder. *Child: Care, Health & Development* 2008; 34(2):276-278.
41 Taniura H, Ito M, Sanada N, Kuramoto N, Ohno Y, Nakamichi N et al. Chronic vitamin D3 treatment protects against neurotoxicity by glutamate in association with upregulation of vitamin D receptor mRNA expression in cultured rat cortical neurons. *Journal of neuroscience research 83 (7) (pp 1179-1189), 2006 Date of Publication: 15 May 2006*(7):15.
42 Weig SG, Henerson FW, Wills-Gallagher J, Bhavaraju-Sanka RK, Howard JF, Thorne LB et al. A 6-year-old boy with autism and painful leg weakness. *Journal of Clinical Neuromuscular Disease Conference: 31st Annual Carrell-Krusen Symposium Dallas, TX United States Conference Start: 20090219 Conference End: 20090220 Conference Publication: (var pagings) 10 (3) (pp 160-161), 2009 Date of Publicatio* 2009;(var.pagings):March.

Table 2 maybe this should be table 1

Possible Vitamin D involvement in autism	First author	Type of study	Year
Brain	Harms	Review	2011
	Blaylock	Review	2009
	Lucas	Review	2008
	Lawrence	Original article	2008
Foetal brain development	Eyles	Review	2011
Genetics	Currenti	Review	2010
	Ramagopalan	Original article	2010
	Freitag	Review	2009
De novo genetic mutations	Kinney	Review	2009
Immune system	Goines	Review	2010
Autoimmunity	Dow	Hypothesis	2011
	Carega	Review	2010
Hypomelanosis	Bakare	Hypothesis	2011

Table 2. Clinical Studies of Vitamin D and Autism

Vitamin D and Autism	Author	Type of study	Year
Strong effect of latitudinal increase on Childhood Autism prevalence	Grant et al.		2009
Vitamin D and micronutrients (Magnesium metabolism)	Johnson S	Clinical study	2010
Nutrition – inadequate vitamin D intake due to the selectivity with food	Lindsay	Clinical	2006
	Schamberg	studies	2010

	Herndon		2008
	Sadowska		2011
Extreme cases of inadequate nutritional intake resulting in co-morbidity of autism and rickets	Nobel et al	Clinical studies	2006
	Weig		2009
	Stewart and Latif		2007
	Clark		1993
Medication in pregnancy – antiepileptic drugs	Bromley	Clinical study	2009
Ethnicity Observed low vitamin D in Somali mothers of children with ASD	Fernell	Clinical study	2010
Increased rates of ASD among dark-skinned immigrant mothers at high latitudes	Dealberto	Review	2010
Clinical studies Treatment of children with verbal apraxia (65% with co-morbid ASD) with omega 3 acids and vitamin D- low levels of vitamin D in 57% of children	Morris and Agin	Clinical study	2009
Plasma levels in children with ASD	Molloy	Clinical studies	2010
	Meguid		2010
Plasma levels of vitamin D measured in a group of adult psychiatric out-patients with various psychiatric illnesses – lowest levels in groups with ASD and schizophrenia	Humble	Clinical study	2010