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**Pathophysiology of autism spectrum disorders: Revisiting gastrointestinal involvement and immune imbalance**

Samsam M *et al.* Gastrointestinal and immune involvement in autism

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**Abstract**

Autism spectrum disorders (ASD) comprise a group of neurodevelopmental abnormalities that begin in early childhood and are characterized by impairment of social communication and behavioral problems including restricted interests and repetitive behaviors. Several genes have been implicated in the pathogenesis of ASD, most of them are involved in neuronal synaptogenesis. A number of environmental factors and associated conditions such as gastrointestinal (GI) abnormalities and immune imbalance have been linked to the pathophysiology of ASD. According to the March 2012 report released by United States Centers for Disease Control and Prevention, the prevalence of ASD has sharply increased during the recent years and one out of 88 children suffers now from ASD symptoms. Although there is a strong genetic base for the disease, several associated factors could have a direct link to the pathogenesis of ASD or act as modifiers of the genes thus aggravating the initial problem. Many children suffering from ASD have GI problems such as abdominal pain, chronic diarrhea, constipation, vomiting, gastroesophageal reflux, and intestinal infections. A number of studies focusing on the intestinal mucosa, its permeability, abnormal gut development, leaky gut, and other GI problem raised many questions but studies were somehow inconclusive and an expert panel of American Academy of Pediatrics has strongly recommended further investigation in these areas. GI tract has a direct connection with the immune system and an imbalanced immune response is usually seen in ASD children. Maternal infection or autoimmune diseases have been suspected. Activation of the immune system during early development may have deleterious effect on various organs including the nervous system. In this review we revisited briefly the GI and immune system abnormalities and neuropeptide imbalance and their role in the pathophysiology of ASD and discussed some future research directions.

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**Key words:** Autism spectrum disorders; Gastrointestinal abnormalities; Immune activation; Crohn’s disease; Neuropeptides; Brain-derived neurotrophic factor; *Mycobacterium paratuberculosis*

**Core tip:** According to the March 2012 report released by United States. Centers for Disease Control and Prevention there was a 73% increase in the prevalence of Autism Spectrum Disorders (ASD) during 2002-2008 in the United States. Although several genes causing ASD have been discovered, genetic cause of ASD is about 25% of cases. There was not a significant research focus on environmental factors and ASD-associated co-morbidities in the last two decades. We revisited the gastrointestinal (GI) involvement, GI infection, immune imbalance, maternal infection and immunity, and intestinal microflora in ASD. Some neuropeptides, neurotrophins, their effects, and side effects have also been discussed.

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**INTRODUCTION**

Autism spectrum disorders (ASD) comprise a group of neurodevelopmental abnormalities that begin in early childhood although the first diagnosis may sometimes occur later in life and are characterized by problems in communication and social behavior. According to the 5th edition of the Diagnostic and Statistical Manual of Mental Disorders (DSM-5) diagnostic criteria that has recently been released, major ASD manifestations include impairment in social communication and behavioral problems such as fixated (restricted) interests and repetitive behaviors; delay in language and age of onset are not emphasized in DSM-5 diagnostic criteria[1,2].

According to a report by Centers for Disease Control and Prevention that was released in March 30th 2012, referring to 2008 surveillance year, the prevalence of ASD among 8 years old children in 14 Autism and Developmental Disabilities Monitoring (ADDM) sites in the United States is more than 1% (11.3 per 1,000 or one per 88 children) and that male/female ratio is approximately 4/1 (ASD is more seen among boys, 18.4 per 1000, that is one in 54 boys while in girls the prevalence was 4.0 per 1000, that is one per 252 girls). The report found differences among race and ethnicity as well, although, the latter findings were recommended to be interpreted by caution[3]. Nevertheless, the study shows a 23% increase in the prevalence of ASD from 2006 to 2008 and an overall 78% increase during 2002-2008 among children aged 8 years[3]. The focus of that study by Centers for Disease Control and Prevention on 8 years old children is due to the baseline study by CDC demonstrating this age as identified peak prevalence of the disease. Moreover, ASD evaluation in that report was according to the DSM-IV-Text Revision diagnostic criteria that included the Autistic disorder, Pervasive Developmental Disorder-Not Otherwise Specified (PDD-NOS, that includes the Atypical Autism), or Asperger Disorder. Although DSM-V criteria that was introduced in 2013 has modified the ASD diagnostic criteria[2] and the ASD prevalence estimates will probably be lower under the DSM-V criteria[4], the numbers in the statistics given are high and indicate that more research and effort is needed to investigate the pathomechanism of ASD, it’s treatment, and patient care under new criteria while continue to support the ASD patients who were identified under previous diagnostic criteria.

**PATHOMECHANISM OF ASD**

***Genetic causes***

The exact pathomechanism of ASD is not known so far while several factors have been implicated in its pathogenesis of autistic disorders. Among these, the genetic cause has long been implicated to be a strong evidence-based etiology[5,6] in cases of some co-occurring or associated conditions with ASD such as tuberous sclerosis, fragile X syndrome, Rett syndrome[7] and some other. Siblings of autistic offspring have a higher incidence of autism than general population[8] and twin studies have also indicated strong role for inheritance[9]. There is wide range of phenotype but more genetically homogeneous ASD patients present with less phenotypic heterogeneity[10]. In addition, human genetic investigations and animal models[11] of ASD detected de novo copy number mutations[12,13,14],and rare variant mutations resulting in abnormal alleles in the person or close ancestry that influence neuroanatomical and behavioral traits[15]. These studies have shown dysregulations in genes involved in synapse function[16]. A comprehensive and informative review of several genetic studies by Banerjee and colleagues show abnormal assembly or structure of several transmembrane and scaffolding proteins involved in synaptogenesis and its maintenance, as well as dysregulation of genes involved in the signal transduction mechanism of synapse formation are among the major genetic abnormalities of ASD[17]. Nevertheless, with the discovery of several genes as well as interactions of multiple genes in one individual, epigenetic factors, and effects of environmental modifiers on these genes in ASD, genetic causes including the diagnosable medical conditions, single-gene defect, and cytogenetic problems comprise 25% of the ASD patients so far[18-20]. Therefore, a number of clinical phenotypes and associated co-morbidities have become the characteristic features of ASD[21]. Although some studies indicated a role for mitochondrial DNA mutation in ASD that may possibly lead to impairment of mitochondrial energy metabolism, more research is needed for definitive answers[22]. Mitochondrial dysfunction has been implicated in several neurological disorders[23,24] and it may have a role in ASD. Mitochondria has antibacterial immunity[25] and would be important in case of infections especially that of the GI tract in ASD children.

***Male to female ratio in ASD***

The reason for 4/1 male to female ratio in ASD is not very well understood but it is very important. Recent studies implicate some epigenetic phenomena such as sex-specific effects of Y-linked genes, balanced, as well as skewed X-inactivation, escaping X-inactivation, and parent-of-origin allelic gene among others in the etiology of ASD[26] and heterogeneity in gene regulation at allelic level as well as total gene expression[27]. These sex differences may be due to genetic and hormonal differences that could be initiated during early times of development due to differences in responses to and interactions with various environmental factors such as diet, stress, infection, and drugs. Due to the involvement of many X-linked genes involved in placenta formation and placenta-specific epigenetic processes, placenta plays an important role in sex-specific responses to environmental factors and disease states later in life[28] Internal and external environmental factors have long been implicated in the etiology of ASD. Early maternal immune activation may cause prenatal stress, affecting boys more severely due to a vulnerable genotype[26].

**NEUROPATHOLOGICAL CHANGES OF ASD**

Both postmortem and neuroimaging studies and animal models of ASD show abnormalities in different brain regions such as the frontal cortex, cerebellum, hippocampus, and the amygdaloid nucleus and cerebello-thalamo-cortical pathways[29]. One of the neuropathological findings in ASD includes the presence of focal cortical dysplasias due to possibly the heterochronic division of germinal cells leading to abnormal migration of daughter cells to their target regions[30,31]. Abnormal neuronal migration leads to circumscribed foci of thin cortical areas in ASD human brain especially in frontal lobe containing smaller pyramidal neurons and interneurons. These pathological findings have been attributed to the sensory and motor deficits as well as the epileptic seizures seen in ASD[30]. Autism-epilepsy phenotype has recently been shown to be associated with macrocephaly, a pathologic condition due to accelerating brain growth in early development leading to ASD[32]. There are evidences that the overall size of the brain is increased in some cases of ASD[29].

**NON-GENETIC FACTORS IMPLICATED IN ASD**

Several other conditions such as GI abnormalities, inflammation, environmental factors, infection, toxins, diet, and drugs have been associated with ASD[8,28].

**GASTROINTESTINAL ABNORMALITIES IN ASD**

Several studies have indicated a higher prevalence of gastrointestinal problems such as abdominal pain, constipation, chronic diarrhea, vomiting, and gastroesophageal reflux disease (GERD) in ASD patients[8], but a nested-case control study using United Kingdom database indicated that there was not a considerable association between GI abnormalities and ASD[33].

A number of other conditions such as GI immune/inflammation-mediated pathology in ASD or a leaky gut referring to increased intestinal permeability have not been established due to limitations and speculations in those studies[34]. Similarly, studies referring to leaky intestinal epithelium and damaged tight junctions and passage of dietary gluten or casein or digestion product through intestinal barrier into the blood stream causing immunogenic responses in the brain were not conclusive[35-38,8]. Other studies that indicated more frequent diarrhea and other GI symptoms in autistic children reported inconclusive results[39] and that intravenous secretin administration to autistic patients with GI symptoms didn’t improve their language problem in contrast to other reports[40]. Therefore experts suggested the needs for properly powered investigations[34] in these issues.

GI abnormalities are often seen to correlate with the severity of the ASD behavioral problemsand current literature favors a gut-brain interaction where GI abnormalities may be involved in the pathogenesis or severity of ASD[41].

There is high degree of disability when these patients grow up and studies show only 15% of ASD children may have a favorable life in adulthood while a great majority of them will have poor or very poor outcomes when becoming adult[42,43].

***Gut mucosa, absorption, barrier function, permeability, immune response, and oral tolerance***

One of the well-established functions of the GI tract is to break down the structure of the food particles by digestion and converting them to the smallest molecules. . This digested material will be absorbed by the luminal surface of intestinal epithelium through various types of transport mechanisms, and transported into the blood or lymph capillaries on the other side of the intestinal mucosal cells[44].

GI tract works very closely with the immune system to maintain homeostasis and protects our body against microorganisms and foreign antigens.

Intestinal mucosa is continuously challenged by huge amount of foreign antigens and microorganisms from environment. The organized regulation of the intestinal barrier maintains the mucosal immune function and prevents inflammation[45]. In spite of the microorganisms of the gut flora, various antigens from digested food, and pathogenic microorganisms, the response of the mucosal immune system is a controlled physiologic inflammation that regulates the population of (T helper) Th2 versus Th1 responses[46]. Intestinal epithelial mucosal cells express classical and non-classical MHC molecules and activate specific regulatory T cells (Tregs) and therefore, serve as non-professional antigen-presenting cells[46]. Different elements of our intestinal barrier include the epithelial cell integrity, mucus production, epithelial paracellular permeability, and innate immune response. Abnormal changes in these components may lead to inflammatory diseases of the intestine[45].

There are other cells in the intestinal mucosa, the microfold (M) cells that are able to engulf bigger molecules[47]. These cells belong to a group of cells forming the Gut Associated Lymphoid Tissue (GULT, which comprise the intestinal lymphoid follicles, the Peyer’s patches as well) in the mucosa. M cells can pass their engulfed material to the antigen presenting cells such as macrophages and dendritic cells in the subepithelial tissue that are in cross talk with lymphocytes, the B cells, for antibody production (i.e.: intestinal IgA) and are also responsible for oral tolerance towards the ingested material through other classes of immunoglobulins and cytokines [44].

Although larger molecules may get into the circulatory system[48] the amount of material that M cells take in under normal conditions seems to be small compared to the epithelial lining of the intestine. Nevertheless, when single unites of lipids are being absorbed by the intestinal cells they can reassemble and bound to lipoproteins, forming large molecules such as chylomicrons that leave the intestinal cells through vesicular transport to the extracellular space into the lymph capillaries (due to having larger fenestration) in the subepithelial tissue to larger lymphatic vessels and finally into circulation[49,44]. A similar path for other larger molecule is possible.

We have shown that small proteins such as green fluorescent protein (GFP) bound to cholera toxin-B (CTB) subunit is able to get into intestinal epithelial cells in large amounts “by binding to ganglioside M1 (GM1) receptor[50]“ and find its way into the blood stream and be found in the liver and the spleen[51]. In a series of studies we aimed to introduce large amount of bigger peptides (such as proinsulin) to induce oral tolerance towards the protein and treat autoimmune diseases such as diabetes, by converting the Th1 response to Th2 response with its associated cytokines[52].

The cholera toxin-A subunit which is the toxic part attaches to the intestinal cells by means of its CTB subunit, increasing the permeability of the intestinal mucosa that eventually disrupts the Cl- transport and other ionic and water transport disturbances leading to diarrhea[53]. Several reports indicate GI infection in ASD patients. GI infections can increase intestinal permeability.

E-coli bacteria is able to enter the intestinal cells, change the actin dynamics, modulate the immune response and disrupt the tight junctions, leading to a compromised barrier and increased intestinal permeability resulting in diarrhea[54]. Interferon-beta (INFβ) has been shown to protect the intestinal barrier while tumor necrosis factor-alpha (TNF-α) disrupts such barrier through inhibition of INFβ by another molecule[54]. Other inflammatory conditions such as Crohn’s disease are also able to increase intestinal permeability[55] but also the increased baseline permeability in some at risk individuals and exaggeration to environmental stimuli may increase the chance of Crohn’s disease[56]. Frequent intestinal infections in ASD patients have been reported.

Several factors have been implicated in that pathogenesis of Crohn’s disease. Micobacterium Paratuberclosis (MAP) has been found in the milk, blood and surgical tissue samples of individuals suffering from Crohn’s disease[57,58,59]. MAP due to having a molecular mimicry to heat shock proteins has been postulated to be involved in the pathogenesis of ASD by stimulating antibodies that may cross react with the nervous system myelin basic protein[60].

Sutterella species have recently been found in the ileum of ASD patients with GI abnormalities while no control patients with GI disturbances had the bacteria[61]. Clostridium bolteae, a bacterium that was shown to be immunogenic in rabbits, is often found in the intestine of the ASD children and was proposed to possibly be aggravating the GI symptoms in ASD patient [62]. The first reported case of enterovirus encephalitis linked to or possibly causing ASD in a 32-month-old child has been recently published[63].

As mentioned earlier, there are several reports about the increased permeability or leaky intestine in ASD patients but more research and convincing data is needed therefore, we think this area of research deserves more work due to various GI symptoms in ASD patients. However, it is well known that infections can lead to increased permeability and GI symptoms and beyond. Since ASD children are often reported to have GI infection and diarrhea and that the immune system is imbalanced in ASD patients due to a direct relation of the GI mucosa with the immune system (see the following section) it is necessary to do more research to better understand the GI mucosal environment and barrier activity, subepithelial tissue, susceptibility to infection, causative agents, and the immune response in ASD patients in order to treat them more effectively.

**IMMUNE SYSTEM IMBALANCE IN ASD**

Other co-morbid conditions in ASD such as inflammation, inflammatory response, and immune activation have long been implicated in the pathogenesis of ASD but studies so far were not conclusive[64,65,21]. A number of studies reveal abnormalities of the peripheral immune system supporting the ides of immune involvement in ASD however, immune abnormalities such as activation of microglial cells and innate neuroimmune system are also found in the brain and cerebrospinal fluid (CSF) of ASD patient, the neuroinflammation[66].

Neuroimmune abnormalities have been recently reviewed elsewhere[43]. Blood brain barrier (BBB) is an important regulator of the brain homeostasis[67]. There are evidences that the BBB function is altered in ASD children due to neurological inflammation, immune dysregulation and increased inflammatory cytokines in the brain[68]. Immune response abnormalities are seen in the GI tract and other tissues, the peripheral blood, and in the central nervous system (CNS) of the ASD patients. On the other hand, maternal infection or inflammation, and autoimmune diseases of the family of ASD children have also been shown to cause immune problems in the offspring. We discuss that briefly as maternal immune activation in the following section.

Significantly lower subpopulation of CD4+ and CD8+ lymphocyte as well as imbalance between Th1 and Th2-like cytokines have been observed in autism[69]. Several interleukins (IL) and IFN-gamma imbalance has been reported in the peripheral blood of ASD children with increased activation of both Th1 and Th2 pathways leaning more towards Th2 arm[70]. The immunoglobulins are also reported to be imbalanced in the serum of ASD children. Total serum protein was significantly increased in autistic patients referring to increased albumin and gamma globulin, as well as increased serum IgG, IgG2 and IgG4 that was attributed to possibly an underlying autoimmune disorder and/or increased vulnerability to infections[71]. A number of other immune abnormalities have also been reported in ASD[24]. The immune system is activated in many neurological and psychiatric disorders including those with genetic abnormalities and growing evidence shows that these neurological disorders are aggravated by the immune system activation leading to worsening of the initial disease[72,73] although immune cells have beneficial effects as well[74].

Immune system has several beneficial effects protecting us from microorganisms and helps destroying the tumor cells or disrupting their growth. Immunotherapy has been an effective approach that uses molecules of the body’s own immune system to interfere with the growth of cancer cells and is being used in the treatment of brain tumors such as metastatic melanomas[67,75]. Nevertheless, what we see in case of neurological and psychiatric disorders is mostly the deleterious effect of the immune activation against brain tissue and its related structures.

Since the Th2 pathway produces more immunosuppressory cytokines compared to Th1 arm that favors more the pro-inflammatory cytokines, and both arms are reportedly activated with a predominant Th2 arm in ASD patients, it might help the body tolerate and not to react towards many antigens (possibly penetrated through GI tract), but these antigens can have deleterious effect on other tissues such as brain. More research is needed to understand the role of immune system in ASD.

**MATERNAL IMMUNE ACTIVATION AND ASD**

Many studies show alteration of immune system and an imbalance of various cytokines in ASD children[76,77]. A number of studies show a link between ASD and a family history of autoimmune diseases or those families with altered inflammatory cytokines or other immune problems[78,76]. The autoimmune hypothesis and development of the mental disorders has long existed[79].

When antibodies developed in immune-mediated disorders were introduced to pregnant monkeys, the offspring showed behavioral changes and CNS pathology[80]. Perinatal exposure to infection has been implicated in the pathogenesis of ASD and schizophrenia[81]. Activation of the immune system in pregnant mice leads to the activation of macrophages in the offspring[82].

A recent study investigating the role of maternal autoimmune disease, asthma, and allergy on developmental disorders looked at 560 ASD patients and 168 cases of developmental delay without autism (DD) has found a significant modest increase in both the ASD and DD combined (the ASD alone data was not significant) in the children of sick mothers during pregnancy[83]. Anti-phospholipid antibodies have been linked with psychological problems such as cognitive malfunction, repetitive behavior and anxiety. Increased levels of anti-cardiolipin, bate 2-glycoprotein 1, and anti-phospholipid antibodies were found in the blood plasma of the ASD children compared to their age matched typically developing children and the DD children[84].

Results of the animal studies indicate that the behavioral and maternal immune activation are different among different mice species referring to the possibility that a subpopulation of human might be more vulnerable to particular environmental agents[85].

Maternal immune activation due to infections, inflammatory diseases and autoimmune diseases can have a deleterious effect on the fetus by affecting fetal tissue and its consequences during postnatal period. This area deserves more focus. Animal models of ASD especially the infection models can be very informative.

**MICROBIOTA IN ASD**

Microbiota is an emerging topic that has attracted several researchers to look for the possible connection between the GI microflora and behavioral abnormalities. Earlier repot of deficient disaccharidase enzymatic activity in ASD children and GI symptom[86] prompted investigations looking for intestinal mucosal microbiota involved in carbohydrate metabolism. Abnormal carbohydrate digestion and transport and mucosal dysbiosis (imbalance in the intestinal microbial ecosystem) was reported in the ASD children[87].

Gut dysbiosis was proposed to be involved in the pathogenesis of several diseases[88]. Reduced level of fermenters has been found in the intestinal microflora of the ASD patients[89]. The microbiota-gut-brain axis refers to the ability of gut microbiota to communicate with brain and regulate behavior[90]. Fecal microbiota transplantation has been used in treating several GI disorders but increase knowledge and control trials are needed before it can be used broadly in clinic[91].

Nevertheless, other studies didn’t find a difference in GI microbiota of ASD children with and without GI disturbances [92]. Imbalance in gut microbiota population may render the intestinal mucosa susceptible to injuries, infections, inflammation, abnormal digestion, immune imbalance, immune reaction and cross reaction in other tissues including the brain. More research is emerging in this area.

**VASOACTIVE INTESTINAL PEPTIDE AND OTHER NEUROPEPTIDES IN ASD**

Elevated levels of several neuropeptide including vasoactive intestinal peptide (VIP), calcitonin gene-related peptide (CGRP), brain-derived neurotrophic factor (BDNF) and neurotrophin 4/5 (NT4/5) in the blood of 60 neonates who were just born compared to 56 age-matched control group[93] opened many lines of investigation. Those 60 neonates developed ASD or mental retardation later in life. Nelson and colleagues looked at their blood in the archived neonatal blood samples that are usually drawn at birth.

VIP and pituitary adenylate cyclase-activating peptide (PACAP) are members of the VIP-glucagon-secretin family of peptides[94]. VIP has several functions in the digestive tract, cardiovascular system, lungs, kidney, and endocrine system[95] and is involved in cerebral growth and neurogenesis and astrocytogenesis[8]. VIP is a parasympathetic neurotransmitter and neuromodulator that has been implicated in the pathogenesis of cluster headaches[96].

VIP is involved in activation of brainstem reflexes, and its infusion was shown to decrease the blood velocity in the middle cerebral artery. VIP/PACAP have been implicated in homeostasis of the immune system and are believed to have anti-inflammatory effect on innate and adaptive immunity, promote Th2 responses and decrease the proinflammatory Th1 response[97,98]. Several properties of VIP including those mentioned above and its function on intestinal tight junctions and permeability, as well as therapeutic use have been recently reviewed[99].

BDNF is a well-known neurotrophic factor of the nerve growth factor (NGF) family together with other neurotrophins that acts mainly on high affinity tyrosine kinase (Trk)B receptor but also like all other family members acts on the low affinity neurotrophin receptor, the p75 receptor.

Low concentration of BDNF is able to excite neurons in cerebral cortex, cerebellum, and the hippocampus[100]. BDNF and NT4/5 were able to depolarize brain neurons as rapidly as glutamate (one of the strongest excitatory neurotransmitters) at 1,000-fold lower concentrations[100]. BDNF deficiency has been implicated in a number of psychiatric disorders and ASD including their animal models[101,102].

Although neurotrophic factors are important for neuronal survival, they can have adverse effects as well. It is not known why BDNF is increased in the blood of infants in Nelson’s study. Although the trophic effects of the neurotrophic factors is very well known and low levels of BDNF has been reported in some psychiatric diseases and ASD[103,104] a recent study on mice overexpressing BDNF shows that the animals found abnormal behavior by age[105]. As mentioned above, if BDNF is 1000 fold stronger than glutamic acid in exciting neurons[100] the adverse effect of overexpression might be due to excitotoxicity and possible neuronal degeneration; those mice suffered from epilepsy[105].

BDNF acting on p75 receptor causes axonal degeneration[106] which is used during normal development when some sprout should be eliminated. Another study shows that BDNF has increased the spasticity in spinal cord injured mice[107]. BDNF has been reported to activate glutamate receptors[108] and this might cause excitotoxicity[109]. Moreover, BDNF mediates nociceptive plasticity[110] and was found to initiate and maintain a chronic pain state[111] and that BDNF enhances the excitability of small diameter trigeminal ganglion neurons[112].

Nuerotrophic factors may not be stable at normal body temperature, are expensive as recombinant proteins and therefore several animal studies have introduced neurotrophic factor genes through injection of viral vectors into animal tissue which make the amount of gene product (protein) thus the dosage difficult to predict, while immunity of the host against the virus is a major problem usually in gene therapy.

CGRP is a neuromodulator/neurotransmitter peptide that is found in tissues including the nervous tissue. It is a sensory neuropeptide that coexists usually with glutamate and substance P and neurokinin A (NKA) in sensory neurons[113]. It is a vasodilator peptide and causes hyperemia in some pathological conditions and has been implicated in the pathogenesis of migraine[114]. Its receptors are found on blood vessels and axons and neurons in several tissues including the central nervous system.

CGRP has been found elevated in the peripheral blood of migraine patients[115]. CGRP receptor antagonists are the gepant family of drugs such as Olcegepant and Telcagepant, and B144370TA, are some of the newest antimigraine drugs that lack vasoconstrictive activity and were thought to overcome some of the adverse effect of the Triptan family of drugs (serotonin 1B/D receptor agonists), but elevated liver enzymes and other side effects have been observed and drugs are still in clinical trials phase II and III [116,117,114]. CGRP potently enhanced BDNF release from trigeminal ganglion neuronal culture indicating that BDNF might be a mediator of nociceptive plasticity[110,98].

It is not known precisely why neuropeptide levels are increased in newborn children that later develop ASD. Elevated neuropeptide levels might be due to inflammation, or compensation and restoration of neuronal or other tissues homeostasis in ASD infants. This merits a thorough investigation to see their role in ASD.

**CONCLUSION**

Although ASD has a strong genetic base, GI abnormalities and immune imbalance as part of the environmental factors have been implicated in the pathogenesis of ASD. A number of GI abnormalities such as abdominal pain, diarrhea, constipation, gastroesophageal reflux, and GI infections have been reported in ASD patient but a clear and convincing link of these symptoms to ASD has not yet been found.

An expert panel of American Academy of Pediatrics[34] has strongly recommended further investigation in the role of GI abnormalities in the pathophysiology of ASD. As we discussed, the GI disturbances are directly related to the immune system. Immune imbalances are common in ASD patients. GI infections can also activate the immune system in ASD patients. Maternal autoimmune disease and infections can also cause damage to the embryonic/fetal tissues and aggravate a genetic problem in ASD if not causing the damage to the nervous system. The significant increase in the prevalence of ASD (at least 73% from 2002-2008) in the United States reported by the Center for Disease Control and Prevention in 2012 is a strong indication for revisiting critically every possible factor and look for possible clues in order to slow down or prevent this sharp increase in the prevalence of ASD in our young population and find more therapeutic opportunities to treat ASD more efficiently.

Increased VIP and other neuropeptides in ASD newborns were discussed briefly but this area certainly deserves more exploration and conclusive studies on how to proceed with the findings. Decreased BDNF has been shown later in life in ASD and other mental disorder patients, but its administration although may have positive effects, may also have adverse effects and neurotoxicity on the already compromised nervous tissue in ASD and other diseases that should be taken into consideration. We have not discussed several other factors that have been linked to ASD such as impairment in gut development[8] or autonomic dysfunction[43] or other factors.

**REFERENCES**

1 **Lauritsen MB**. Autism spectrum disorders. *Eur Child Adolesc Psychiatry* 2013; **22** Suppl 1: S37-S42 [PMID: 23300017 DOI: 10.1007/s00787-012-0359-5]

2 **Grzadzinski R**, Huerta M, Lord C. DSM-5 and autism spectrum disorders (ASDs): an opportunity for identifying ASD subtypes. *Mol Autism* 2013; **4**: 12 [PMID: 23675638 DOI: 10.1186/2040-2392-4-12]

3 **Center for Disease Control and Prevention.** Prevalence of Autism Spectrum Disorders-Autism and Developmental Disabilities Monitoring Network, 14 sites, United States, 2008. *MMWR* 2008; **61**: 3

4 **Maenner MJ**, Rice CE, Arneson CL, Cunniff C, Schieve LA, Carpenter LA, Van Naarden Braun K, Kirby RS, Bakian AV, Durkin MS. Potential impact of DSM-5 criteria on autism spectrum disorder prevalence estimates. *JAMA Psychiatry* 2014; **71**: 292-300 [PMID: 24452504 DOI: 10.1001/jamapsychiatry.2013.3893]

5 **Shen Y**, Dies KA, Holm IA, Bridgemohan C, Sobeih MM, Caronna EB, Miller KJ, Frazier JA, Silverstein I, Picker J, Weissman L, Raffalli P, Jeste S, Demmer LA, Peters HK, Brewster SJ, Kowalczyk SJ, Rosen-Sheidley B, McGowan C, Duda AW, Lincoln SA, Lowe KR, Schonwald A, Robbins M, Hisama F, Wolff R, Becker R, Nasir R, Urion DK, Milunsky JM, Rappaport L, Gusella JF, Walsh CA, Wu BL, Miller DT. Clinical genetic testing for patients with autism spectrum disorders. *Pediatrics* 2010; **125**: e727-e735 [PMID: 20231187 DOI: 10.1542/peds.2009-1684]

6 **Silver WG**, Rapin I. Neurobiological basis of autism. *Pediatr Clin North Am* 2012; **59**: 45-61, x [PMID: 22284792 DOI: 10.1016/j.pcl.2011.10.010]

7 **Betancur C**, Sakurai T, Buxbaum JD. The emerging role of synaptic cell-adhesion pathways in the pathogenesis of autism spectrum disorders. *Trends Neurosci* 2009; **32**: 402-412 [PMID: 19541375 DOI: 10.1016/j.tins.2009.04.003]

8 **White JF**. Intestinal pathophysiology in autism. *Exp Biol Med (Maywood)* 2003; **228**: 639-649 [PMID: 12773694]

9 **Bailey A**, Le Couteur A, Gottesman I, Bolton P, Simonoff E, Yuzda E, Rutter M. Autism as a strongly genetic disorder: evidence from a British twin study. *Psychol Med* 1995; **25**: 63-77 [PMID: 7792363]

10 **Bruining H,** de Sonneville L, Swaab H, de Jonge M, Kas M, van Engeland H, Vorstman J. Dissecting the clinical heterogeneity of autism spectrum disorders through defined genotypes. *PLoS One* 2010; **5:** e10887 [DOI: 10.1371/journal.pone.0010887]

11 **Shinoda Y**, Sadakata T, Furuichi T. Animal models of autism spectrum disorder (ASD): a synaptic-level approach to autistic-like behavior in mice. *Exp Anim* 2013; **62**: 71-78 [PMID: 23615300]

12 **Sebat J**, Lakshmi B, Malhotra D, Troge J, Lese-Martin C, Walsh T, Yamrom B, Yoon S, Krasnitz A, Kendall J, Leotta A, Pai D, Zhang R, Lee YH, Hicks J, Spence SJ, Lee AT, Puura K, Lehtimäki T, Ledbetter D, Gregersen PK, Bregman J, Sutcliffe JS, Jobanputra V, Chung W, Warburton D, King MC, Skuse D, Geschwind DH, Gilliam TC, Ye K, Wigler M. Strong association of de novo copy number mutations with autism. *Science* 2007; **316**: 445-449 [PMID: 17363630]

13 **Mikhail FM**, Lose EJ, Robin NH, Descartes MD, Rutledge KD, Rutledge SL, Korf BR, Carroll AJ. Clinically relevant single gene or intragenic deletions encompassing critical neurodevelopmental genes in patients with developmental delay, mental retardation, and/or autism spectrum disorders. *Am J Med Genet A* 2011; **155A**: 2386-2396 [PMID: 22031302 DOI: 10.1002/ajmg.a.34177]

14 **Ronemus M**, Iossifov I, Levy D, Wigler M. The role of de novo mutations in the genetics of autism spectrum disorders. *Nat Rev Genet* 2014; **15**: 133-141 [PMID: 24430941 DOI: 10.1038/nrg3585]

15 **Malhotra D**, Sebat J. CNVs: harbingers of a rare variant revolution in psychiatric genetics. *Cell* 2012; **148**: 1223-1241 [PMID: 22424231 DOI: 10.1016/j.cell.2012.02.039]

16 **Zoghbi HY**, Bear MF. Synaptic dysfunction in neurodevelopmental disorders associated with autism and intellectual disabilities. *Cold Spring Harb Perspect Biol* 2012; **4**: [PMID: 22258914 DOI: 10.1101/cshperspect.a009886]

17 **Banerjee S**, Riordan M, Bhat MA. Genetic aspects of autism spectrum disorders: insights from animal models. *Front Cell Neurosci* 2014; **8**: 58 [PMID: 24605088 DOI: 10.3389/fncel.2014.00058]

18 **Muhle R**, Trentacoste SV, Rapin I. The genetics of autism. *Pediatrics* 2004; **113**: e472-e486 [PMID: 15121991]

19 **Betancur C**. Etiological heterogeneity in autism spectrum disorders: more than 100 genetic and genomic disorders and still counting. *Brain Res* 2011; **1380**: 42-77 [PMID: 21129364 DOI: 10.1016/j.brainres.2010.11.078]

20 **Miles JH**. Autism spectrum disorders--a genetics review. *Genet Med* 2011; **13**: 278-294 [PMID: 21358411 DOI: 10.1097/GIM.0b013e3181ff67ba]

21 **Jyonouchi H**, Geng L, Streck DL, Toruner GA. Immunological characterization and transcription profiling of peripheral blood (PB) monocytes in children with autism spectrum disorders (ASD) and specific polysaccharide antibody deficiency (SPAD): case study. *J Neuroinflammation* 2012; **9**: 4 [DOI: 10.1186/1742-2094-9-4]

22 **Álvarez-Iglesias V**, Mosquera-Miguel A, Cuscó I, Carracedo Á, Pérez-Jurado LA, Salas A. Reassessing the role of mitochondrial DNA mutations in autism spectrum disorder. *BMC Med Genet* 2011; **12**: 50 [PMID: 21470425 DOI: 10.1186/1471-2350-12-50]

23 **Knott AB**, Bossy-Wetzel E. Impairing the mitochondrial fission and fusion balance: a new mechanism of neurodegeneration. *Ann N Y Acad Sci* 2008; **1147**: 283-292 [PMID: 19076450 DOI: 10.1196/annals.1427.030]

24 **Randolph-Gips M**, Srinivasan P. Modeling autism: a systems biology approach. *J Clin Bioinforma* 2012; **2**: 17 [PMID: 23043674 DOI: 10.1186/2043-9113-2-17]

25 **West AP**, Shadel GS, Ghosh S. Mitochondria in innate immune responses. *Nat Rev Immunol* 2011; **11**: 389-402 [PMID: 21597473 DOI: 10.1038/nri2975]

26 **Schaafsma SM,** Pfaff DW.Etiologies underlying sex differences in Autism Spectrum Disorders. *Front Neuroendocrinol* 2014; [Epub ahead of print] [PMID: 24705124 DOI: 10.1016/j.yfrne.2014.03.006]

27 **Ben-David E,** Shohat S, Shifman S.Allelic expression analysis in the brain suggests a role for heterogeneous insults affecting epigenetic processes in autism spectrum disorders. *Hum Mol Genet* 2014; [Epub ahead of print] [PMID: 24659497]

28 **Gabory A**, Roseboom TJ, Moore T, Moore LG, Junien C. Placental contribution to the origins of sexual dimorphism in health and diseases: sex chromosomes and epigenetics. *Biol Sex Differ* 2013; **4**: 5 [PMID: 23514128 DOI: 10.1186/2042-6410-4-5]

29 **Santangelo SL**, Tsatsanis K. What is known about autism: genes, brain, and behavior. *Am J Pharmacogenomics* 2005; **5**: 71-92 [PMID: 15813671]

30 **Casanova MF**, El-Baz AS, Kamat SS, Dombroski BA, Khalifa F, Elnakib A, Soliman A, Allison-McNutt A, Switala AE. Focal cortical dysplasias in autism spectrum disorders. *Acta Neuropathol Commun* 2013; **1**: 67 [PMID: 24252498 DOI: 10.1186/2051-5960-1-67]

31 **Casanova MF.** Autism as a sequence: From heterochronic germinal cell divisions to abnormalities of cell migration and cortical dysplasias. *Med Hypotheses* 2014; [Epub ahead of print] [PMID: 24780284 DOI: 10.1016/j.mehy.2014.04.014]

32 **Marchese M**, Conti V, Valvo G, Moro F, Muratori F, Tancredi R, Santorelli FM, Guerrini R, Sicca F. Autism-epilepsy phenotype with macrocephaly suggests PTEN, but not GLIALCAM, genetic screening. *BMC Med Genet* 2014; **15**: 26 [PMID: 24580998 DOI: 10.1186/1471-2350-15-26]

33 **Black C**, Kaye JA, Jick H. Relation of childhood gastrointestinal disorders to autism: nested case-control study using data from the UK General Practice Research Database. *BMJ* 2002; **325**: 419-421 [PMID: 12193358 DOI: 10.1136/bmj.325.7361.419]

34 **Buie T,** Campbell DB, Fuchs GJ 3rd, Furuta GT, Levy J, Vandewater J, Whitaker AH, Atkins D, Bauman ML, Beaudet AL, Carr EG, Gershon MD, Hyman SL, Jirapinyo P, Jyonouchi H, Kooros K, Kushak R, Levitt P, Levy SE, Lewis JD, Murray KF, Natowicz MR, Sabra A, Wershil BK, Weston SC, Zeltzer L, Winter H. Evaluation, diagnosis, and treatment of gastrointestinal disorders in individuals with ASDs: a consensus report. *Pediatrics* 2010; **125** Suppl 1: S1-18 [DOI: 10.1542/peds.2009-1878C]

35 **D'Eufemia P**, Celli M, Finocchiaro R, Pacifico L, Viozzi L, Zaccagnini M, Cardi E, Giardini O. Abnormal intestinal permeability in children with autism. *Acta Paediatr* 1996; **85**: 1076-1079 [PMID: 8888921 DOI: 10.1111/j.1651-2227.1996.tb14220.x]

36 **Millward C,** Ferriter M, Calver S, Connell-Jones G. Gluten- and casein-free diets for autistic spectrum disorder. *Cochrane Database Syst Rev* 2008; **2**: CD003498 [DOI: 10.1002/14651858.CD003498.pub3]

37 **Goodwin MS**, Cowen MA, Goodwin TC. Malabsorption and cerebral dysfunction: a multivariate and comparative study of autistic children. *J Autism Child Schizophr* ; **1**: 48-62 [PMID: 5172439 DOI: 10.1007/BF01537742]

38 **Dohan FC**. Hypothesis: genes and neuroactive peptides from food as cause of schizophrenia. *Adv Biochem Psychopharmacol* 1980; **22**: 535-548 [PMID: 6994444]

39 **Lightdale JR**, Hayer C, Duer A, Lind-White C, Jenkins S, Siegel B, Elliott GR, Heyman MB. Effects of intravenous secretin on language and behavior of children with autism and gastrointestinal symptoms: a single-blinded, open-label pilot study. *Pediatrics* 2001; **108**: E90 [PMID: 11694674 DOI: 10.1542/peds.108.5.e90]

40 **Erickson CA**, Stigler KA, Corkins MR, Posey DJ, Fitzgerald JF, McDougle CJ. Gastrointestinal factors in autistic disorder: a critical review. *J Autism Dev Disord* 2005; **35**: 713-727 [PMID: 16267642 DOI: 10.1007/s10803-005-0019-4]

41 **Hsiao EY.** Gastrointestinal issues in autism spectrum disorder. *Harv Rev Psychiatry* 2014; **22**: 104-111 [DOI: 10.1097/HRP.0000000000000029]

42 **Seltzer MM**, Shattuck P, Abbeduto L, Greenberg JS. Trajectory of development in adolescents and adults with autism. *Ment Retard Dev Disabil Res Rev* 2004; **10**: 234-247 [PMID: 15666341 DOI: 10.1002/mrdd.20038]

43 **Goyal DK,** Miyan JA. Neuro-Immune Abnormalities in Autism and Their Relationship with the Environment: A Variable Insult Model for Autism. *Front Endocrinol* (Lausanne) 2014; **5**: 29 [PMID: 24639668]

44 **Samsam M.** Functionally Oriented Regional Anatomy, 2nd edition, Plymouth, MI: Hayden McNeil publishing, 2013; 209-302

45 **Pastorelli L,** De Salvo C, Mercado JR, Vecchi M, Pizarro TT. Central Role of the Gut Epithelial Barrier in the Pathogenesis of Chronic Intestinal Inflammation: Lessons Learned from Animal Models and Human Genetics. *Front Immunol* 2013; **4**: 280 [PMID: 24062746]

46 **Rabinowitz K**, Mayer L. Working out mechanisms of controlled/physiologic inflammation in the GI tract. *Immunol Res* 2012; **54**: 14-24 [PMID: 22466933 DOI: 10.1007/s12026-012-8315-5]

47 **Kernéis S**, Bogdanova A, Kraehenbuhl JP, Pringault E. Conversion by Peyer's patch lymphocytes of human enterocytes into M cells that transport bacteria. *Science* 1997; **277**: 949-952 [PMID: 9252325 DOI: 10.1126/science.277.5328.949]

48 **Hemmings WA**, Williams EW. Transport of large breakdown products of dietary protein through the gut wall. *Gut* 1978; **19**: 715-723 [PMID: 680603 DOI: 10.1136/gut.19.8.715]

49 **Dixon JB**. Mechanisms of chylomicron uptake into lacteals. *Ann N Y Acad Sci* 2010; **1207** Suppl 1: E52-E57 [PMID: 20961306 DOI: 10.1111/j.1749-6632.2010.05716.x]

50 **Lencer WI**. Microbes and microbial Toxins: paradigms for microbial-mucosal toxins. V. Cholera: invasion of the intestinal epithelial barrier by a stably folded protein toxin. *Am J Physiol Gastrointest Liver Physiol* 2001; **280**: G781-G786 [PMID: 11292584]

51 **Limaye A**, Koya V, Samsam M, Daniell H. Receptor-mediated oral delivery of a bioencapsulated green fluorescent protein expressed in transgenic chloroplasts into the mouse circulatory system. *FASEB J* 2006; **20**: 959-961 [PMID: 16603603 DOI: 10.1096/fj.05-5134fje]

52 **Ruhlman T**, Ahangari R, Devine A, Samsam M, Daniell H. Expression of cholera toxin B-proinsulin fusion protein in lettuce and tobacco chloroplasts--oral administration protects against development of insulitis in non-obese diabetic mice. *Plant Biotechnol J* 2007; **5**: 495-510 [PMID: 17490448 DOI: 10.1111/j.1467-7652.2007.00259.x]

53 **Muanprasat C**, Chatsudthipong V. Cholera: pathophysiology and emerging therapeutic targets. *Future Med Chem* 2013; **5**: 781-798 [PMID: 23651092 DOI: 10.4155/fmc.13.42]

54 **Long TM,** Nisa S, Donnenberg MS, Hassel BA. Enteropathogenic Escherichia coli inhibits type I interferon- and RNase-L-mediated host defense to disrupt intestinal epithelial cell barrier function. *Infect Immun* 2014; [Epub ahead of print] [PMID: 24733098]

55 **Wyatt J**, Oberhuber G, Pongratz S, Püspök A, Moser G, Novacek G, Lochs H, Vogelsang H. Increased gastric and intestinal permeability in patients with Crohn's disease. *Am J Gastroenterol* 1997; **92**: 1891-1896 [PMID: 9382060]

56 **Meddings JB**. Review article: Intestinal permeability in Crohn's disease. *Aliment Pharmacol Ther* 1997; **11** Suppl 3: 47-53; discussion 53-6 [PMID: 9467978 DOI: 10.1111/j.1365-2036.1997.tb00808.x]

57 **Naser SA**, Schwartz D, Shafran I. Isolation of Mycobacterium avium subsp paratuberculosis from breast milk of Crohn's disease patients. *Am J Gastroenterol* 2000; **95**: 1094-1095 [PMID: 10763975 DOI: 10.1111/j.1572-0241.2000.01954.x]

58 **Naser SA**, Ghobrial G, Romero C, Valentine JF. Culture of Mycobacterium avium subspecies paratuberculosis from the blood of patients with Crohn's disease. *Lancet* ; **364**: 1039-1044 [PMID: 15380962 DOI: 10.1016/S0140-6736(04)17058-X]

59 **Romero C**, Hamdi A, Valentine JF, Naser SA. Evaluation of surgical tissue from patients with Crohn's disease for the presence of Mycobacterium avium subspecies paratuberculosis DNA by in situ hybridization and nested polymerase chain reaction. *Inflamm Bowel Dis* 2005; **11**: 116-125 [PMID: 15677904 DOI: 10.1097/00054725-200502000-00004]

60 **Dow CT**. Mycobacterium paratuberculosis and autism: is this a trigger? *Med Hypotheses* 2011; **77**: 977-981 [PMID: 21903338 DOI: 10.1016/j.mehy.2011.08.024]

61 **Williams BL**, Hornig M, Parekh T, Lipkin WI. Application of novel PCR-based methods for detection, quantitation, and phylogenetic characterization of Sutterella species in intestinal biopsy samples from children with autism and gastrointestinal disturbances. *MBio* 2012; **3**: [PMID: 22233678 DOI: 10.1128/mBio.00261-11]

62 **Pequegnat B**, Sagermann M, Valliani M, Toh M, Chow H, Allen-Vercoe E, Monteiro MA. A vaccine and diagnostic target for Clostridium bolteae, an autism-associated bacterium. *Vaccine* 2013; **31**: 2787-2790 [PMID: 23602537 DOI: 10.1016/j.vaccine.2013.04.018]

63 **Marques F**, Brito MJ, Conde M, Pinto M, Moreira A. Autism spectrum disorder secondary to enterovirus encephalitis. *J Child Neurol* 2014; **29**: 708-714 [PMID: 24782421 DOI: 10.1177/0883073813508314]

64 **Ashwood P**, Wills S, Van de Water J. The immune response in autism: a new frontier for autism research. *J Leukoc Biol* 2006; **80**: 1-15 [PMID: 16698940 DOI: 10.1189/jlb.1205707]

65 **Singh VK**. Phenotypic expression of autoimmune autistic disorder (AAD): a major subset of autism. *Ann Clin Psychiatry* 2009; **21**: 148-161 [PMID: 19758536]

66 **Pardo CA**, Vargas DL, Zimmerman AW. Immunity, neuroglia and neuroinflammation in autism. *Int Rev Psychiatry* 2005; **17**: 485-495 [PMID: 16401547 DOI: 10.1080/02646830500381930]

67 **Samsam M**. Editorial: central nervous system drugs in the treatment of neurological disorders. *Cent Nerv Syst Agents Med Chem* 2012; **12**: 153-157 [PMID: 22894608]

68 **Noriega DB**, Savelkoul HF. Immune dysregulation in autism spectrum disorder. *Eur J Pediatr* 2014; **173**: 33-43 [PMID: 24297668 DOI: 10.1007/s00431-013-2183-4]

69 **Gupta S**, Aggarwal S, Rashanravan B, Lee T. Th1- and Th2-like cytokines in CD4+ and CD8+ T cells in autism. *J Neuroimmunol* 1998; **85**: 106-109 [PMID: 9627004 DOI: 10.1016/S0165-5728(98)00021-6]

70 **Molloy CA**, Morrow AL, Meinzen-Derr J, Schleifer K, Dienger K, Manning-Courtney P, Altaye M, Wills-Karp M. Elevated cytokine levels in children with autism spectrum disorder. *J Neuroimmunol* 2006; **172**: 198-205 [PMID: 16360218 DOI: 10.1016/j.jneuroim.2005.11.007]

71 **Croonenberghs J**, Wauters A, Devreese K, Verkerk R, Scharpe S, Bosmans E, Egyed B, Deboutte D, Maes M. Increased serum albumin, gamma globulin, immunoglobulin IgG, and IgG2 and IgG4 in autism. *Psychol Med* 2002; **32**: 1457-1463 [PMID: 12455944 DOI: 10.1017/S0033291702006037]

72 **Kobsar I**, Berghoff M, Samsam M, Wessig C, Mäurer M, Toyka KV, Martini R. Preserved myelin integrity and reduced axonopathy in connexin32-deficient mice lacking the recombination activating gene-1. *Brain* 2003; **126**: 804-813 [PMID: 12615640 DOI: 10.1093/brain/awg072]

73 **Samsam M.** Role of inflammation in neurological and psychiatric disorders. Editorial, AIAA-MC, 2010; **3:** 166-169

74 **Berghoff M**, Samsam M, Müller M, Kobsar I, Toyka KV, Kiefer R, Mäurer M, Martini R. Neuroprotective effect of the immune system in a mouse model of severe dysmyelinating hereditary neuropathy: enhanced axonal degeneration following disruption of the RAG-1 gene. *Mol Cell Neurosci* 2005; **28**: 118-127 [PMID: 15607947 DOI: 10.1016/j.mcn.2004.09.001]

75 **Allen T**, Gundrajakuppam L. A role of immunotherapy in metastatic malignant melanoma. *Cent Nerv Syst Agents Med Chem* 2012; **12**: 182-188 [PMID: 22697295]

76 **Gesundheit B,** Rosenzweig JP, Naor D, Lerer B, Zachor DA, Procházka V, Melamed M, Kristt DA, Steinberg A, Shulman C, Hwang P, Koren G, Walfisch A, Passweg JR, Snowden JA, Tamouza R, Leboyer M, Farge-Bancel D, Ashwood P. Immunological and autoimmune considerations of Autism Spectrum Disorders. *J Autoimmun* 2013; **44**: 1-7 [DOI: 10.1016/j.jaut.2013.05.005]

77 **Goines PE**, Ashwood P. Cytokine dysregulation in autism spectrum disorders (ASD): possible role of the environment. *Neurotoxicol Teratol* ; **36**: 67-81 [PMID: 22918031 DOI: 10.1016/j.ntt.2012.07.006]

78 **Sweeten TL**, Bowyer SL, Posey DJ, Halberstadt GM, McDougle CJ. Increased prevalence of familial autoimmunity in probands with pervasive developmental disorders. *Pediatrics* 2003; **112**: e420 [PMID: 14595086]

79 **Sakić B**, Szechtman H, Denburg JA. Neurobehavioral alterations in autoimmune mice. *Neurosci Biobehav Rev* 1997; **21**: 327-340 [PMID: 9168268]

80 **Libbey JE**, Fujinami RS. Role for antibodies in altering behavior and movement. *Autism Res* 2010; **3**: 147-152 [PMID: 20589715 DOI: 10.1002/aur.144]

81 **Meyer U**, Feldon J, Dammann O. Schizophrenia and autism: both shared and disorder-specific pathogenesis via perinatal inflammation? *Pediatr Res* 2011; **69**: 26R-33R [PMID: 21289540 DOI: 10.1203/PDR.0b013e318212c196]

82 **Onore CE**, Schwartzer JJ, Careaga M, Berman RF, Ashwood P. Maternal immune activation leads to activated inflammatory macrophages in offspring. *Brain Behav Immun* 2014; **38**: 220-226 [PMID: 24566386 DOI: 10.1016/j.bbi.2014.02.007]

83 **Lyall K,** Ashwood P, Van de Water J, Hertz-Picciotto I. Maternal Immune-Mediated Conditions, Autism Spectrum Disorders, and Developmental Delay. *J Autism Dev Disord* 2013; [Epub ahead of print] [PMID: 24337796]

84 **Careaga M**, Hansen RL, Hertz-Piccotto I, Van de Water J, Ashwood P. Increased anti-phospholipid antibodies in autism spectrum disorders. *Mediators Inflamm* 2013; **2013**: 935608 [PMID: 24174712 DOI: 10.1155/2013/935608]

85 **Schwartzer JJ**, Careaga M, Onore CE, Rushakoff JA, Berman RF, Ashwood P. Maternal immune activation and strain specific interactions in the development of autism-like behaviors in mice. *Transl Psychiatry* 2013; **3**: e240 [PMID: 23481627 DOI: 10.1038/tp.2013.16]

86 **Horvath K**, Papadimitriou JC, Rabsztyn A, Drachenberg C, Tildon JT. Gastrointestinal abnormalities in children with autistic disorder. *J Pediatr* 1999; **135**: 559-563 [PMID: 10547242 DOI: 10.1016/S0022-3476(99)70052-1]

87 **Williams BL**, Hornig M, Buie T, Bauman ML, Cho Paik M, Wick I, Bennett A, Jabado O, Hirschberg DL, Lipkin WI. Impaired carbohydrate digestion and transport and mucosal dysbiosis in the intestines of children with autism and gastrointestinal disturbances. *PLoS One* 2011; **6**: e24585 [PMID: 21949732 DOI: 10.1371/journal.pone.0024585]

88 **Petrof EO**, Claud EC, Gloor GB, Allen-Vercoe E. Microbial ecosystems therapeutics: a new paradigm in medicine? *Benef Microbes* 2013; **4**: 53-65 [PMID: 23257018 DOI: 10.3920/BM2012.0039]

89 **Kang DW**, Park JG, Ilhan ZE, Wallstrom G, Labaer J, Adams JB, Krajmalnik-Brown R. Reduced incidence of Prevotella and other fermenters in intestinal microflora of autistic children. *PLoS One* 2013; **8**: e68322 [PMID: 23844187 DOI: 10.1371/journal.pone.0068322]

90 **Montiel-Castro AJ**, González-Cervantes RM, Bravo-Ruiseco G, Pacheco-López G. The microbiota-gut-brain axis: neurobehavioral correlates, health and sociality. *Front Integr Neurosci* 2013; **7**: 70 [PMID: 24109440 DOI: 10.3389/fnint.2013.00070]

91 **Aroniadis OC,** Brandt LJ.Fecal microbiota transplantation: past, present and future. *Curr Opin Gastroenterol* 2013; **29**: 79-84 [PMID: 23041678 DOI: 10.1097/MOG.0b013e32835a4b3e]

92 **Gondalia SV,** Palombo EA, Knowles SR, Cox SB, Meyer D, Austin DW. Molecular characterisation of gastrointestinal microbiota of children with autism (with and without gastrointestinal dysfunction) and their neurotypical siblings. *Autism Res* 2012; **5**: 419-427 [PMID: 23041678 DOI: 10.1002/aur.1253]

93 **Nelson KB**, Grether JK, Croen LA, Dambrosia JM, Dickens BF, Jelliffe LL, Hansen RL, Phillips TM. Neuropeptides and neurotrophins in neonatal blood of children with autism or mental retardation. *Ann Neurol* 2001; **49**: 597-606 [PMID: 11357950 DOI: 10.1002/ana.1024]

94 **Hautmann M**, Friis UG, Desch M, Todorov V, Castrop H, Segerer F, Otto C, Schütz G, Schweda F. Pituitary adenylate cyclase-activating polypeptide stimulates renin secretion via activation of PAC1 receptors. *J Am Soc Nephrol* 2007; **18**: 1150-1156 [PMID: 17360952 DOI: 10.1681/ASN.2006060633]

95 **Burian B**, Ortner A, Prassl R, Zimmer A, Mosgoeller W. Clinical potential of VIP by modified pharmaco-kinetics and delivery mechanisms. *Endocr Metab Immune Disord Drug Targets* 2012; **12**: 344-350 [PMID: 23094831 DOI: 10.2174/187153012803832594]

96 **Edvinsson L**. Sensory nerves in man and their role in primary headaches. *Cephalalgia* 2001; **21**: 761-764 [PMID: 11595008 DOI: 10.1046/j.1468-2982.2001.00245.x]

97 **Gonzalez-Rey E**, Varela N, Chorny A, Delgado M. Therapeutical approaches of vasoactive intestinal peptide as a pleiotropic immunomodulator. *Curr Pharm Des* 2007; **13**: 1113-1139 [PMID: 17430175 DOI: 10.2174/138161207780618966]

98 **Samsam M,** Coveñas R, Ahangari R, Yajeya J, Narváez JA. Role of neuropeptides in migraine; where do they stand in the latest expert recommendations in migraine treatment? *Drug Devel Res* 2007; **68**: 298- 314 [DOI: 10.1002/ddr.20193]

99 **Chandrasekharan B**, Nezami BG, Srinivasan S. Emerging neuropeptide targets in inflammation: NPY and VIP. *Am J Physiol Gastrointest Liver Physiol* 2013; **304**: G949-G957 [PMID: 23538492 DOI: 10.1152/ajpgi.00493.2012]

100 **Kafitz KW**, Rose CR, Thoenen H, Konnerth A. Neurotrophin-evoked rapid excitation through TrkB receptors. *Nature* 1999; **401**: 918-921 [PMID: 10553907 DOI: 10.1038/44847]

101 **Ray MT**, Shannon Weickert C, Webster MJ. Decreased BDNF and TrkB mRNA expression in multiple cortical areas of patients with schizophrenia and mood disorders. *Transl Psychiatry* 2014; **4**: e389 [PMID: 24802307 DOI: 10.1038/tp.2014.26]

102 **Scattoni ML**, Martire A, Cartocci G, Ferrante A, Ricceri L. Reduced social interaction, behavioural flexibility and BDNF signalling in the BTBR T+ tf/J strain, a mouse model of autism. *Behav Brain Res* 2013; **251**: 35-40 [PMID: 23270976 DOI: 10.1016/j.bbr.2012.12.028]

103 **Hashimoto K**. Brain-derived neurotrophic factor as a biomarker for mood disorders: an historical overview and future directions. *Psychiatry Clin Neurosci* 2010; **64**: 341-357 [PMID: 20653908 DOI: 10.1111/j.1440-1819.2010.02113.x]

104 **Taurines R**, Segura M, Schecklmann M, Albantakis L, Grünblatt E, Walitza S, Jans T, Lyttwin B, Haberhausen M, Theisen FM, Martin B, Briegel W, Thome J, Schwenck C, Romanos M, Gerlach M. Altered peripheral BDNF mRNA expression and BDNF protein concentrations in blood of children and adolescents with autism spectrum disorder. *J Neural Transm* 2014; [Epub ahead of print] [PMID: 24500031]

105 **Weidner KL**, Buenaventura DF, Chadman KK. Mice over-expressing BDNF in forebrain neurons develop an altered behavioral phenotype with age. *Behav Brain Res* 2014; **268**: 222-228 [PMID: 24768643 DOI: 10.1016/j.bbr.2014.04.025]

106 **Singh KK**, Park KJ, Hong EJ, Kramer BM, Greenberg ME, Kaplan DR, Miller FD. Developmental axon pruning mediated by BDNF-p75NTR-dependent axon degeneration. *Nat Neurosci* 2008; **11**: 649-658 [PMID: 18382462 DOI: 10.1038/nn.2114]

107 **Fouad K,** Bennett DJ, Vavrek R, Blesch A. Long-term viral brain-derived neurotrophic factor delivery promotes spasticity in rats with a cervical spinal cord hemisection. *Front Neurol* 2013; **4**: 187 [DOI: 10.3389/fneur.2013.00187]

108 **Jänisch W**, Engel U, Leonhardt T. [Diffuse primary leptomeningeal gliomatosis]. *Zentralbl Pathol* 1991; **137**: 523-530 [PMID: 1805932 DOI: 10.1038/sj.bjp.0707509]

109 **Behrens MM**, Strasser U, Lobner D, Dugan LL. Neurotrophin-mediated potentiation of neuronal injury. *Microsc Res Tech* 1999; **45**: 276-284 [PMID: 10383120]

110 **Buldyrev I**, Tanner NM, Hsieh HY, Dodd EG, Nguyen LT, Balkowiec A. Calcitonin gene-related peptide enhances release of native brain-derived neurotrophic factor from trigeminal ganglion neurons. *J Neurochem* 2006; **99**: 1338-1350 [PMID: 17064360 DOI: 10.1111/j.1471-4159.2006.04161.x]

111 **Melemedjian OK**, Tillu DV, Asiedu MN, Mandell EK, Moy JK, Blute VM, Taylor CJ, Ghosh S, Price TJ. BDNF regulates atypical PKC at spinal synapses to initiate and maintain a centralized chronic pain state. *Mol Pain* 2013; **9**: 12 [PMID: 23510079 DOI: 10.1186/1744-8069-9-12]

112 **Takeda M,** Takahashi M, Kitagawa J, Kanazawa T, Nasu M, Matsumoto SBrain-derived neurotrophic factor enhances the excitability of small-diameter trigeminal ganglion neurons projecting to the trigeminal nucleus interpolaris/caudalis transition zone following masseter muscle inflammation. *Mol Pain* 2013; **9**: 49 [DOI: 10.1186/1744-8069-9-49]

113 **Samsam M**, Coveñas R, Ahangari R, Yajeya J, Narváez JA, Tramu G. Simultaneous depletion of neurokinin A, substance P and calcitonin gene-related peptide from the caudal trigeminal nucleus of the rat during electrical stimulation of the trigeminal ganglion. *Pain* 2000; **84**: 389-395 [PMID: 10666545 DOI: 10.1016/S0304-3959(99)00240-7]

114 **Samsam M**. Central nervous system acting drugs in treatment of migraine headache. *Cent Nerv Syst Agents Med Chem* 2012; **12**: 158-172 [PMID: 22533510 DOI: 10.2174/187152412802430147]

115 **Gallai V**, Sarchielli P, Floridi A, Franceschini M, Codini M, Glioti G, Trequattrini A, Palumbo R. Vasoactive peptide levels in the plasma of young migraine patients with and without aura assessed both interictally and ictally. *Cephalalgia* 1995; **15**: 384-390 [PMID: 8536297 DOI: 10.1046/j.1468-29821995.1505384.x]

116 **Olesen J**, Diener HC, Husstedt IW, Goadsby PJ, Hall D, Meier U, Pollentier S, Lesko LM. Calcitonin gene-related peptide receptor antagonist BIBN 4096 BS for the acute treatment of migraine. *N Engl J Med* 2004; **350**: 1104-1110 [PMID: 15014183 DOI: 10.1056/NEJMoa030505]

117 **Ho TW**, Mannix LK, Fan X, Assaid C, Furtek C, Jones CJ, Lines CR, Rapoport AM. Randomized controlled trial of an oral CGRP receptor antagonist, MK-0974, in acute treatment of migraine. *Neurology* 2008; **70**: 1304-1312 [PMID: 17914062 DOI: 10.1212/01.WNL.0000286940.29755.61]

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