Perinatal air pollutant exposures and autism spectrum disorder in the children of nurses’ health study II participants.

Air pollution contains many toxicants known to affect neurological function and to have effects on the fetus in utero. Recent studies have reported associations between perinatal exposure to air pollutants and autism spectrum disorder (ASD) in children. We tested the hypothesis that perinatal exposure to air pollutants is associated with ASD, focusing on pollutants associated with ASD in prior studies. We estimated associations between U.S. Environmental Protection Agency-modeled levels of hazardous air pollutants at the time and place of birth and ASD in the children of participants in the Nurses’ Health Study II (325 cases, 22,101 controls). Our analyses focused on pollutants associated with ASD in prior research. We accounted for possible confounding and ascertainment bias by adjusting for family-level socioeconomic status (maternal grandparents’ education) and census tract-level socioeconomic measures (e.g., tract median income and percent college educated), as well as maternal age at birth and year of birth. We also examined possible differences in the relationship between ASD and pollutant exposures by child’s sex. Perinatal exposures to the highest versus lowest quintile of diesel, lead, manganese, mercury, methylene chloride, and an overall measure of metals were significantly associated with ASD, with odds ratios ranging from 1.5 (for overall metals measure) to 2.0 (for diesel and mercury). In addition, linear trends were positive and statistically significant for these exposures (p < .05 for each). For most pollutants, associations were stronger for boys (279 cases) than for girls (46 cases) and significantly different according to sex. CONCLUSIONS: Perinatal exposure to air pollutants may increase risk for ASD. Additionally, future studies should consider sex-specific biological pathways connecting perinatal exposure to pollutants with ASD.


Estimation of autistic children by metallomics analysis.
Yasuda H, Yasuda Y, Tsutsui T.

Clarification of the pathogenesis and treatment of autism spectrum disorders is one of the challenges today. In this study, we examine scalp hair concentrations of 26 trace elements for 1,967 children with autistic disorders (1,553 males and 414 females). Five-hundred and eighty-four (29.7%), 347 (17.6%) and 114 (5.8%) subjects was found deficient in zinc, magnesium and calcium, respectively, and 2.0% or less in the other essential metals. The incidence rate of mineral deficiency was highly observed in infants aged 0-3 year-old. In contrast, 339 (17.2%), 168 (8.5%) and 94 (4.8%) individuals was found suffering from high burden of aluminium, cadmium and lead, and 2.8% or less from mercury and arsenic burden. These findings suggest that infantile zinc- and magnesium-deficiency and/or toxic metal burdens may epigenetically play principal roles as environmental factors in autistic disorders and that metallomics approach may lead to early screening and prevention of the neurodevelopment disorders. Sci Rep. 2013;3:1199. doi: 10.1038/srep01199. Epub 2013 Feb 4.

Age-dependent decrease and alternative splicing of methionine synthase mRNA in human cerebral cortex and an accelerated decrease in autism.
Muratore CR, Hodgson NW, Trivedi MS, Abdolmaleky HM, Persico AM, Lintas C, De la Monte S, Deth RC.

The folate and vitamin B12-dependent enzyme methionine synthase (MS) is highly sensitive to cellular oxidative status, and lower MS activity increases production of the antioxidant glutathione, while simultaneously decreasing more than 200 methylation reactions, broadly affecting metabolic activity. MS mRNA levels in postmortem human cortex from subjects across the lifespan were measured and a dramatic progressive biphasic decrease of more than 400-fold from 28 weeks of gestation to 84 years was observed. Further analysis revealed alternative splicing of MS mRNA, including deletion of folate-binding domain exons and age-dependent deletion of exons from the cap domain, which protects vitamin B12 (cobalamin) from oxidation. Although three species of MS were evident at the protein level, corresponding
to full-length and alternatively spliced mRNA transcripts, decreasing mRNA levels across the lifespan were not associated with significant changes in MS protein or methionine levels. MS mRNA levels were significantly lower in autistic subjects, especially at younger ages, and this decrease was replicated in cultured human neuronal cells by treatment with TNF-α, whose CSF levels are elevated in autism. These novel findings suggest that rather than serving as a housekeeping enzyme, MS has a broad and dynamic role in coordinating metabolism in the brain during development and aging. Factors adversely affecting MS activity, such as oxidative stress, can be a source of risk for neurological disorders across the lifespan via their impact on methylation reactions, including epigenetic regulation of gene expression. *PLoS One.* 2013;8(2):e56927. doi: 10.1371/journal.pone.0056927. Epub 2013 Feb 20.

**Redox metabolism abnormalities in autistic children associated with mitochondrial disease.**


Research studies have uncovered several metabolic abnormalities associated with autism spectrum disorder (ASD), including mitochondrial disease (MD) and abnormal redox metabolism. Despite the close connection between mitochondrial dysfunction and oxidative stress, the relation between MD and oxidative stress in children with ASD has not been studied. Plasma markers of oxidative stress and measures of cognitive and language development and ASD behavior were obtained from 18 children diagnosed with ASD who met criteria for probable or definite MD per the Morava et al. criteria (ASD/MD) and 18 age and gender-matched ASD children without any biological markers or symptoms of MD (ASD/NoMD). Plasma measures of redox metabolism included reduced free glutathione (fGSH), oxidized glutathione (GSSG), the fGSH/GSSG ratio and 3-nitrotyrosine (3NT). In addition, a plasma measure of chronic immune activation, 3-chlorotyrosine (3CT), was also measured. Language was measured using the preschool language scale or the expressive one-word vocabulary test (depending on the age), adaptive behaviour was measured using the Vineland Adaptive Behavior Scale (VABS) and core autism symptoms were measured using the Autism Symptoms Questionnaire and the Social Responsiveness Scale. Children with ASD/MD were found to have lower scores on the communication and daily living skill subscales of the VABS despite having similar language and ASD symptoms. Children with ASD/MD demonstrated significantly higher levels of fGSH/GSSG and lower levels of GSSG as compared with children with ASD/NoMD, suggesting an overall more favourable glutathione redox status in the ASD/MD group. However, compare with controls, both ASD groups demonstrated lower fGSH and fGSH/GSSG, demonstrating that both groups suffer from redox abnormalities. Younger ASD/MD children had higher levels of 3CT than younger ASD/NoMD children because of an age-related effect in the ASD/MD group. Both ASD groups demonstrated significantly higher 3CT levels than control subjects, suggesting that chronic inflammation was present in both groups of children with ASD. Interestingly, 3NT was found to correlate positively with several measures of cognitive function, development and behavior for the ASD/MD group, but not the ASD/NoMD group, such that higher 3NT concentrations were associated with more favourable adaptive behaviour, language and ASD-related behavior. To determine whether difference in receiving medications and/or supplements could account for the differences in redox and inflammatory biomarkers across ASD groups, we examined differences in medication and supplements across groups and their effect of redox and inflammatory biomarkers. Overall, significantly more participants in the ASD/MD group were receiving folate, vitamin B12, carnitine, co-enzyme Q10, B vitamins and antioxidants. We then determined whether folate, carnitine, co-enzyme Q10, B vitamins and/or antioxidants influenced redox or inflammatory biomarkers. Antioxidant supplementation was associated with a significantly lower GSSG, whereas antioxidants, co-enzyme Q10 and B vitamins were associated with a higher fGSH/GSSG ratio. There was no relation between folate, carnitine, co-enzyme Q10, B vitamins and antioxidants with 3NT, 3CT or fGSH. Overall, our findings suggest that ASD/MD children with a more chronic oxidized microenvironment have better development. We interpret this finding in light of the fact that more active mitochondrial can create a greater oxidized microenvironment especially when dysfunctional. Thus, compensatory upregulation of mitochondria which are dysfunctional may both increase activity and function at the expense of a more oxidized microenvironment. Although more ASD/MD children were receiving certain supplements, the use of such supplements were not found to be related to the redox biomarkers that were related to cognitive development or behavior in the
ASD/MD group but could possibly account for the difference in glutathione metabolism noted between groups. This study suggests that different subgroups of children with ASD have different redox abnormalities, which may arise from different sources. A better understanding of the relationship between mitochondrial dysfunction in ASD and oxidative stress, along with other factors that may contribute to oxidative stress, will be critical to understanding how to guide treatment and management of ASD children. This study also suggests that it is important to identify ASD/MD children as they may respond differently to specific treatments because of their specific metabolic profile.


Elevated maternal C-reactive protein and autism in a national birth cohort.

Autism is a complex neuropsychiatric syndrome with a largely unknown etiology. Inflammation during pregnancy may represent a common pathway by which infections and other insults increase risk for the disorder. Hence, we investigated the association between early gestational C-reactive protein (CRP), an established inflammatory biomarker, prospectively assayed in maternal sera, and childhood autism in a large national birth cohort with an extensive serum biobank. Other strengths of the cohort included nearly complete ascertainment of pregnancies in Finland (N=1.2 million) over the study period and national psychiatric registries consisting of virtually all treated autism cases in the population. Increasing maternal CRP levels, classified as a continuous variable, were significantly associated with autism in offspring. For maternal CRP levels in the highest quintile, compared with the lowest quintile, there was a significant, 43% elevated risk. This finding suggests that maternal inflammation may have a significant role in autism, with possible implications for identifying preventive strategies and pathogenic mechanisms in autism and other neurodevelopmental disorders.


A two-phase study evaluating the relationship between Thimerosal-containing vaccine administration and the risk for an autism spectrum disorder diagnosis in the United States.
Geier DA, Hooker BS, Kern JK, King PG, Sykes LK, Geier MR.

Autism spectrum disorder (ASD) is defined by standardized criteria of qualitative impairments in social interaction, qualitative impairments in communication, and restricted and stereotyped patterns of behavior, interests, and activities. A significant number of children diagnosed with ASD suffer a loss of previously-acquired skills, which is suggestive of neurodegeneration or a type of progressive encephalopathy with an etiological pathogenic basis occurring after birth. To date, the etiology of ASD remains under debate, however, many studies suggest toxicity, especially from mercury (Hg), in individuals diagnosed with an ASD. The present study evaluated concerns about the toxic effects of organic-Hg exposure from Thimerosal (49.55% Hg by weight) in childhood vaccines by conducting a two-phased (hypothesis generating/hypothesis testing) study with documented exposure to varying levels of Thimerosal from vaccinations.

A hypothesis generating cohort study was undertaken to evaluate the relationship between exposure to organic-Hg from a Thimerosal-containing Diphtheria-Tetanus-acellular-Pertussis (DTaP) vaccine in comparison to a Thimerosal-free DTaP vaccine administered, from 1998 through 2000, for the risk of ASD as reported in the Vaccine Adverse Event Reporting System (VAERS) database (phase I). A hypothesis testing case-control study was undertaken to evaluate the relationship between organic-Hg exposure from Thimerosal-containing hepatitis B vaccines administered at specific intervals in the first six months of life among cases diagnosed with an ASD and controls born between 1991 through 1999 in the Vaccine Safety Datalink (VSD) database (phase II).

In phase I, it was observed that there was a significantly increased risk ratio for the incidence of ASD reported following the Thimerosal-containing DTaP vaccine in comparison to the Thimerosal-free DTaP vaccine. In phase II, it was observed that cases diagnosed with an ASD were significantly more likely than controls to receive increased organic-Hg from Thimerosal-containing hepatitis B vaccine administered within the first, second, and sixth month of life.
CONCLUSIONS: Routine childhood vaccination is an important public health tool to reduce the morbidity and mortality associated with infectious diseases, but the present study provides new epidemiological evidence supporting an association between increasing organic-Hg exposure from Thimerosal-containing childhood vaccines and the subsequent risk of an ASD diagnosis. *Transl Neurodegener.* 2013 Dec 19;2(1):25. doi: 10.1186/2047-9158-2-25.

**B-lymphocytes from a population of children with autism spectrum disorder and their unaffected siblings exhibit hypersensitivity to thimerosal.**
Sharpe MA, Gist TL, Baskin DS.

The role of thimerosal containing vaccines in the development of autism spectrum disorder (ASD) has been an area of intense debate, as has the presence of mercury dental amalgams and fish ingestion by pregnant mothers. We studied the effects of thimerosal on cell proliferation and mitochondrial function from B-lymphocytes taken from individuals with autism, their nonautistic twins, and their nontwin siblings. Eleven families were examined and compared to matched controls. B-cells were grown with increasing levels of thimerosal, and various assays (LDH, XTT, DCFH, etc.) were performed to examine the effects on cellular proliferation and mitochondrial function. A subpopulation of eight individuals (4 ASD, 2 twins, and 2 siblings) from four of the families showed thimerosal hypersensitivity, whereas none of the control individuals displayed this response. The thimerosal concentration required to inhibit cell proliferation in these individuals was only 40% of controls. Cells hypersensitive to thimerosal also had higher levels of oxidative stress markers, protein carbonyls, and oxidant generation. This suggests certain individuals with a mild mitochondrial defect may be highly susceptible to mitochondrial specific toxins like the vaccine preservative thimerosal. *J Toxicol.* 2013;2013:801517. doi: 10.1155/2013/801517. Epub 2013 Jun 9.

**Activation of the maternal immune system during pregnancy alters behavioral development of rhesus monkey offspring.**
Bauman MD, Iosif AM, Smith SE, Bregere C, Amaral DG, Patterson PH.

Maternal infection during pregnancy is associated with an increased risk of schizophrenia and autism in the offspring. Supporting this correlation, experimentally activating the maternal immune system during pregnancy in rodents produces offspring with abnormal brain and behavioral development. We have developed a nonhuman primate model to bridge the gap between clinical populations and rodent models of maternal immune activation (MIA). A modified form of the viral mimic, synthetic double-stranded RNA (polyinosinic:polycytidylic acid stabilized with poly-L-lysine) was delivered to two separate groups of pregnant rhesus monkeys to induce MIA: 1) late first trimester MIA (n = 6), and 2) late second trimester MIA (n = 7). Control animals (n = 11) received saline injections at the same first or second trimester time points or were untreated. Sickness behavior, temperature, and cytokine profiles of the pregnant monkeys confirmed a strong inflammatory response to MIA. Behavioral development of the offspring was studied for 24 months. Following weaning at 6 months of age, MIA offspring exhibited abnormal responses to separation from their mothers. As the animals matured, MIA offspring displayed increased repetitive behaviors and decreased affiliative vocalizations. When evaluated with unfamiliar conspecifics, first trimester MIA offspring deviated from species-typical macaque social behavior by inappropriately approaching and remaining in immediate proximity of an unfamiliar animal. CONCLUSIONS: In this rhesus monkey model, MIA yields offspring with abnormal repetitive behaviors, communication, and social interactions. These results extended the findings in rodent MIA models to more human-like behaviors resembling those in both autism and schizophrenia. *Biol Psychiatry.* 2014 Feb 15;75(4):332-41. doi: 10.1016/j.biopsych.2013.06.025. Epub 2013 Sep 5.
Antipurinergic therapy corrects the autism-like features in the poly(IC) mouse model.

Autism spectrum disorders (ASDs) are caused by both genetic and environmental factors. Mitochondria act to connect genes and environment by regulating gene-encoded metabolic networks according to changes in the chemistry of the cell and its environment. Mitochondrial ATP and other metabolites are mitokines-signaling molecules made in mitochondria-that undergo regulated release from cells to communicate cellular health and danger to neighboring cells via purinergic signaling. The role of purinergic signaling has not yet been explored in autism spectrum disorders. We used the maternal immune activation (MIA) mouse model of gestational poly(IC) exposure and treatment with the non-selective purinergic antagonist suramin to test the role of purinergic signaling in C57BL/6J mice. We found that antipurinergic therapy (APT) corrected 16 multisystem abnormalities that defined the ASD-like phenotype in this model. These included correction of the core social deficits and sensorimotor coordination abnormalities, prevention of cerebellar Purkinje cell loss, correction of the ultrastructural synaptic dysmorphology, and correction of the hypothermia, metabolic, mitochondrial, P2Y2 and P2X7 purinergic receptor expression, and ERK1/2 and CAMKII signal transduction abnormalities. CONCLUSIONS: Hyperpurinergia is a fundamental and treatable feature of the multisystem abnormalities in the poly(IC) mouse model of autism spectrum disorders. Antipurinergic therapy provides a new tool for refining current concepts of pathogenesis in autism and related spectrum disorders, and represents a fresh path forward for new drug development. PLoS One. 2013;8(3):e57380. doi: 10.1371/journal.pone.0057380. Epub 2013 Mar 13.

Effectiveness of methylcobalamin and folinic Acid treatment on adaptive behavior in children with autistic disorder is related to glutathione redox status.
Frye RE1, Melnyk S, Fuchs G, Reid T, Jernigan S, Pavliv O, Hubanks A, Gaylor DW, Walters L, James SJ.

Treatments targeting metabolic abnormalities in children with autism are limited. Previously we reported that a nutritional treatment significantly improved glutathione metabolism in children with autistic disorder. In this study we evaluated changes in adaptive behaviors in this cohort and determined whether such changes are related to changes in glutathione metabolism. Thirty-seven children diagnosed with autistic disorder and abnormal glutathione and methylation metabolism were treated with twice weekly 75 µg/Kg methylcobalamin and twice daily 400 µg folinic acid for 3 months in an open-label fashion. The Vineland Adaptive Behavior Scale (VABS) and glutathione redox metabolites were measured at baseline and at the end of the treatment period. Over the treatment period, all VABS subscales significantly improved with an average effect size of 0.59, and an average improvement in skills of 7.7 months. A greater improvement in glutathione redox status was associated with a greater improvement in expressive communication, personal and domestic daily living skills, and interpersonal, play-leisure, and coping social skills. Age, gender, and history of regression did not influence treatment response. The significant behavioral improvements observed and the relationship between these improvements to glutathione redox status suggest that nutritional interventions targeting redox metabolism may benefit some children with autism. Autism Res Treat. 2013;2013:609705. doi: 10.1155/2013/609705. Epub 2013 Oct 12.

Doshi-Velez F, Ge Y, Kohane I.

The distinct trajectories of patients with autism spectrum disorders (ASDs) have not been extensively studied, particularly regarding clinical manifestations beyond the neurobehavioral criteria from the Diagnostic and Statistical Manual of Mental Disorders. The objective of this study was to investigate the patterns of co-occurrence of medical comorbidities in ASDs. International Classification of Diseases, Ninth Revision codes from patients aged at least 15 years and a diagnosis of ASD were obtained from electronic medical records. These codes were aggregated by using phenotype-wide association studies categories and processed into 1350-dimensional vectors describing the counts of
the most common categories in 6-month blocks between the ages of 0 to 15. Hierarchical clustering was used to identify subgroups with distinct courses. Four subgroups were identified. The first was characterized by seizures (n = 120, subgroup prevalence 77.5%). The second (n = 197) was characterized by multisystem disorders including gastrointestinal disorders (prevalence 24.3%) and auditory disorders and infections (prevalence 87.8%), and the third was characterized by psychiatric disorders (n = 212, prevalence 33.0%). The last group (n = 4316) could not be further resolved. The prevalence of psychiatric disorders was uncorrelated with seizure activity (P = .17), but a significant correlation existed between gastrointestinal disorders and seizures (P < .001). The correlation results were replicated by using a second sample of 496 individuals from a different geographic region. CONCLUSIONS: Three distinct patterns of medical trajectories were identified by unsupervised clustering of electronic health record diagnoses. These may point to distinct etiologies with different genetic and environmental contributions. Additional clinical and molecular characterizations will be required to further delineate these subgroups. Pediatrics. 2014 Jan;133(1):e54-63. doi: 10.1542/peds.2013-0819. Epub 2013 Dec 9.

Gastrointestinal Problems in Children with Autism, Developmental Delays or Typical Development.
Chaidez V, Hansen RL, Hertz-Picciotto I.

To compare gastrointestinal (GI) problems among children with: (1) autism spectrum disorder (ASD), (2) developmental delay (DD) and (3) typical development (TD), GI symptom frequencies were obtained for 960 children from the Childhood Autism Risks from Genetics and Environment (CHARGE) study. We also examined scores on five Aberrant Behavior Checklist (ABC) subscales comparing ASD children with high versus low frequency GI symptoms. Compared to TD children, those with ASD [aOR 7.92 (4.89-12.85)] and DD [aOR 4.55 (2.51-8.24)] were more likely to have at least one frequent GI symptom. Restricting to ASD children, those with frequent abdominal pain, gaseousness, diarrhea, constipation or pain on stoolsing scored worse on irritability, social withdrawal, stereotypy, and hyperactivity compared with children having no frequent GI symptoms. Frequent GI problems affect young children with ASD and DD more commonly than those with TD. Maladaptive behaviors correlate with GI problems, suggesting these comorbidities require attention. J Autism Dev Disord. 2013 Nov 6. [Epub ahead of print]

Identification of unique gene expression profile in children with regressive autism spectrum disorder (ASD) and ileocolitis.
Walker SJ, Fortunato J, Gonzalez LG, Krigsman A.

Gastrointestinal symptoms are common in children with autism spectrum disorder (ASD) and are often associated with mucosal inflammatory infiltrates of the small and large intestine. Although distinct histologic and immunohistochemical properties of this inflammatory infiltrate have been previously described in this ASD(GI) group, molecular characterization of these lesions has not been reported. In this study we utilize transcriptome profiling of gastrointestinal mucosal biopsy tissue from ASD(GI) children and three non-ASD control groups (Crohn's disease, ulcerative colitis, and histologically normal) in an effort to determine if there is a gene expression profile unique to the ASD(GI) group. Comparison of differentially expressed transcripts between the groups demonstrated that non-pathologic (normal) tissue segregated almost completely from inflamed tissue in all cases. Gene expression profiles in intestinal biopsy tissue from patients with Crohn's disease, ulcerative colitis, and ASD(GI), while having significant overlap with each other, also showed distinctive features for each group. Taken together, these results demonstrate that ASD(GI) children have a gastrointestinal mucosal molecular profile that overlaps significantly with known inflammatory bowel disease (IBD), yet has distinctive features that further supports the presence of an ASD-associated IBD variant, or, alternatively, a prodromal phase of typical inflammatory bowel disease. Although we report qPCR confirmation of representative differentially expressed transcripts determined initially by microarray, these findings may be considered preliminary to the extent that they require further confirmation in a validation cohort. PLoS One. 2013;8(3):e58058. doi: 10.1371/journal.pone.0058058. Epub 2013 Mar 8.
Microbiota modulate behavioral and physiological abnormalities associated with neurodevelopmental disorders.
Hsiao EY1, McBride SW2, Hsien S2, Sharon G2, Hyde ER3, McCue T3, Codelli JA4, Chow J2, Reisman SE4, Petrosino JF3, Patterson PH5, Mazmanian SK6.

Neurodevelopmental disorders, including autism spectrum disorder (ASD), are defined by core behavioral impairments; however, subsets of individuals display a spectrum of gastrointestinal (GI) abnormalities. We demonstrate GI barrier defects and microbiota alterations in the maternal immune activation (MIA) mouse model that is known to display features of ASD. Oral treatment of MIA offspring with the human commensal Bacteroides fragilis corrects gut permeability, alters microbial composition, and ameliorates defects in communicative, stereotypic, anxiety-like and sensorimotor behaviors. MIA offspring display an altered serum metabolomic profile, and B. fragilis modulates levels of several metabolites. Treating naive mice with a metabolite that is increased by MIA and restored by B. fragilis causes certain behavioral abnormalities, suggesting that gut bacterial effects on the host metabolome impact behavior. Taken together, these findings support a gut-microbiome-brain connection in a mouse model of ASD and identify a potential probiotic therapy for GI and particular behavioral symptoms in human neurodevelopmental disorders. PAPERCLIP: Cell. 2013 Dec 19;155(7):1451-63. doi: 10.1016/j.cell.2013.11.024. Epub 2013 Dec 5

Markers of Celiac Disease and Gluten Sensitivity in Children with Autism.
Lau NM, Green PH, Taylor AK, Hellberg D, Ajamian M, Tan CZ, Kosofsky BE, Higgins JJ, Rajadhyaksha AM, Alaedini A.

Abstract
Gastrointestinal symptoms are a common feature in children with autism, drawing attention to a potential association with celiac disease or gluten sensitivity. However, studies to date regarding the immune response to gluten in autism and its association with celiac disease have been inconsistent. The aim of this study was to assess immune reactivity to gluten in pediatric patients diagnosed with autism according to strict criteria and to evaluate the potential link between autism and celiac disease. Study participants included children (with or without gastrointestinal symptoms) diagnosed with autism according to both the Autism Diagnostic Observation Schedule (ADOS) and the Autism Diagnostic Interview, Revised (ADI-R) (n = 37), their unaffected siblings (n = 27), and age-matched healthy controls (n = 76). Serum specimens were tested for antibodies to native gliadin, deamidated gliadin, and transglutaminase 2 (TG2). Affected children were genotyped for celiac disease associated HLA-DQ2 and -DQ8 alleles. Children with autism had significantly higher levels of IgG antibody to gliadin compared with unrelated healthy controls (p<0.01). The IgG levels were also higher compared to the unaffected siblings, but did not reach statistical significance. The IgG anti-gliadin antibody response was significantly greater in the autistic children with gastrointestinal symptoms in comparison to those without them (p<0.01). There was no difference in IgA response to gliadin across groups. The levels of celiac disease-specific serologic markers, i.e., antibodies to deamidated gliadin and TG2, did not differ between patients and controls. An association between increased anti-gliadin antibody and presence of HLA-DQ2 and -DQ8 alleles was not observed. CONCLUSIONS: A subset of children with autism displays increased immune reactivity to gluten, the mechanism of which appears to be distinct from that in celiac disease. The increased anti-gliadin antibody response and its association with GI symptoms points to a potential mechanism involving immunologic and/or intestinal permeability abnormalities in affected children. PLoS One. 2013 Jun 18;8(6):e66155. Print 2013.

Methylomic analysis of monozygotic twins discordant for autism spectrum disorder and related behavioural traits.
Wong CC, Meaburn EL, Ronald A, Price TS, Jeffries AR, Schalkwyk LC, Plomin R, Mill J.

Autism spectrum disorder (ASD) defines a group of common, complex neurodevelopmental disorders. Although the aetiology of ASD has a strong genetic component, there is considerable monozygotic (MZ) twin discordance indicating a role for non-genetic factors. Because MZ twins share an identical DNA sequence, disease-discordant MZ twin pairs provide an ideal model for examining the contribution of environmentally driven epigenetic factors in disease. We performed a genome-wide analysis of DNA methylation in a sample of 50 MZ twin pairs (100 individuals) sampled from a
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representative population cohort that included twins discordant and concordant for ASD, ASD-associated traits and no autistic phenotype. Within-twin and between-group analyses identified numerous differentially methylated regions associated with ASD. In addition, we report significant correlations between DNA methylation and quantitatively measured autistic trait scores across our sample cohort. This study represents the first systematic epigenomic analyses of MZ twins discordant for ASD and implicates a role for altered DNA methylation in autism. Mol Psychiatry. 2013 Apr 23. doi: 10.1038/mp.2013.41. [Epub ahead of print]

Effectiveness of methylcobalamin and folinic Acid treatment on adaptive behavior in children with autistic disorder is related to glutathione redox status.

Treatments targeting metabolic abnormalities in children with autism are limited. Previously we reported that a nutritional treatment significantly improved glutathione metabolism in children with autistic disorder. In this study we evaluated changes in adaptive behaviors in this cohort and determined whether such changes are related to changes in glutathione metabolism. Thirty-seven children diagnosed with autistic disorder and abnormal glutathione and methylation metabolism were treated with twice weekly 75 µg/Kg methylcobalamin and twice daily 400 µg folinic acid for 3 months in an open-label fashion. The Vineland Adaptive Behavior Scale (VABS) and glutathione redox metabolites were measured at baseline and at the end of the treatment period. Over the treatment period, all VABS subscales significantly improved with an average effect size of 0.59, and an average improvement in skills of 7.7 months. A greater improvement in glutathione redox status was associated with a greater improvement in expressive communication, personal and domestic daily living skills, and interpersonal, play-leisure, and coping social skills. Age, gender, and history of regression did not influence treatment response. The significant behavioral improvements observed and the relationship between these improvements to glutathione redox status suggest that nutritional interventions targeting redox metabolism may benefit some children with autism. Sci Rep. 2013;3:1199. doi: 10.1038/srep01199. Epub 2013 Feb 4.


Despite the fact that seizures are commonly associated with autism spectrum disorder (ASD), the effectiveness of treatments for seizures has not been well studied in individuals with ASD. This manuscript reviews both traditional and novel treatments for seizures associated with ASD. Studies were selected by systematically searching major electronic databases and by a panel of experts that treat ASD individuals. Only a few anti-epileptic drugs (AEDs) have undergone carefully controlled trials in ASD, but these trials examined outcomes other than seizures. Several lines of evidence point to valproate, lamotrigine, and levetiracetam as the most effective and tolerable AEDs for individuals with ASD. Limited evidence supports the use of traditional non-AED treatments, such as the ketogenic and modified Atkins diet, multiple subpial transections, immunomodulation, and neurofeedback treatments. Although specific treatments may be more appropriate for specific genetic and metabolic syndromes associated with ASD and seizures, there are few studies which have documented the effectiveness of treatments for seizures for specific syndromes. Limited evidence supports l-carnitine, multivitamins, and N-acetyl-L-cysteine in mitochondrial disease and dysfunction, folinic acid in cerebral folate abnormalities and early treatment with vigabatrin in tuberous sclerosis complex. Finally, there is limited evidence for a number of novel treatments, particularly magnesium with pyridoxine, omega-3 fatty acids, the gluten-free casein-free diet, and low-frequency repetitive transcranial magnetic simulation. Zinc and l-carnosine are potential novel treatments supported by basic research but not clinical studies. This review demonstrates the wide variety of treatments used to treat seizures in individuals with ASD as well as the striking lack of clinical trials performed to support the use of these

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treatments. Additional studies concerning these treatments for controlling seizures in individuals with ASD are warranted. *Front Public Health*. 2013 Sep 13;1:31. eCollection 2013.

**Optimal outcome in individuals with a history of autism.**

Although autism spectrum disorders (ASDs) are generally considered lifelong disabilities, literature suggests that a minority of individuals with an ASD will lose the diagnosis. However, the existence of this phenomenon, as well as its frequency and interpretation, is still controversial: were they misdiagnosed initially, is this a rare event, did they lose the full diagnosis, but still suffer significant social and communication impairments or did they lose all symptoms of ASD and function socially within the normal range? The present study documents a group of these optimal outcome individuals (OO group, n=34) by comparing their functioning on standardized measures to age, sex, and nonverbal IQ matched individuals with high-functioning autism (HFA group, n=44) or typical development (TD group, n=34). For this study, 'optimal outcome' requires losing all symptoms of ASD in addition to the diagnosis, and functioning within the nonautistic range of social interaction and communication. Domains explored include language, face recognition, socialization, communication, and autism symptoms. Optimal outcome and TD groups' mean scores did not differ on socialization, communication, face recognition, or most language subscales, although three OO individuals showed below-average scores on face recognition. Early in their development, the OO group displayed milder symptoms than the HFA group in the social domain, but had equally severe difficulties with communication and repetitive behaviors. CONCLUSIONS: Although possible deficits in more subtle aspects of social interaction or cognition are not ruled out, the results substantiate the possibility of OO from autism spectrum disorders and demonstrate an overall level of functioning within normal limits for this group. *J Child Psychol Psychiatry*. 2013 Feb;54(2):195-205. doi: 10.1111/jcpp.12037.

**Prevalence and incidence rates of autism in the UK: time trend from 2004-2010 in children aged 8 years.**
Taylor B, Jick H, Maclaughlin D.

To update UK studies begun in the early 1990s on the annual prevalence and incidence rates of autism in children; undertaken in response to a March 2012 press release, widely covered by the media, from the US Centre for Disease Control (CDC) reporting that the autism prevalence rate in 2008 in 8-year-old US children was 1 in 88, a 78% increase from a CDC estimate in 2004. This finding suggested a continuation of the dramatic increase in children diagnosed as autistic, which occurred in the 1990s. DESIGN: Population study using the UK General Practice Research Database (GPRD). METHODS: Annual autism prevalence rates were estimated for children aged 8 years in 2004-2010 by dividing the number diagnosed as autistic in each or any previous year by the number of children active in the study population that year. We also calculated annual incidence rates for children aged 2-8 years, by dividing the number newly diagnosed in 2004-2010 by the same denominators. RESULTS: Annual prevalence rates for each year were steady at approximately 3.8/1000 boys and 0.8/1000 girls. Annual incidence rates each year were also steady at about 1.2/1000 boys and 0.2/1000 girls. CONCLUSIONS: Following a fivefold increase in the annual incidence rates of autism during the 1990s in the UK, the incidence and prevalence rates in 8-year-old children reached a plateau in the early 2000s and remained steady through 2010. Whether prevalence rates have increased from the early 2000s in the USA remains uncertain. *BMJ Open*. 2013 Oct 16;3(10):e003219. doi: 10.1136/bmjopen-2013-003219.
The incidence of diagnosed autism spectrum disorders in Finland.

Background: Previous reports indicate an increase in incidence of autism spectrum disorders (ASD). Aims: First, to assess the incidence of diagnosed ASD in children born between 1996 and 1998, based on nationwide inpatient and outpatient register information. Second, to investigate the incidence rate over time of diagnosed ASD and specifically childhood autism, Asperger's syndrome and pervasive developmental disorder (PDD-NOS) in children born between 1987 and 1998. Methods: This is population-based cohort study with children born in Finland between 1 January 1987 and 31 December 2005; a total of more than 1.2 million children. Children were identified in the Finnish Hospital Discharge Register, and the reported diagnoses were based on the International Statistical Classification of Diseases (ICD-10, ICD-9). Results: The annual incidence rate of diagnosed ASD based on inpatient and outpatient register data was 53.7 per 10,000 (95% CI 50.4-57.2). Incidence was 82.6 per 10,000 in boys and 23.6 per 10,000 in girls, yielding a sex ratio (boys:girls) of 3.5:1. We report an eightfold increase in the incidence rates in children of diagnosed ASD and specifically in childhood autism, Asperger's syndrome and PDD-NOS and born between 1987 and 1992 based on inpatient register information. Conclusions: Increased awareness of ASD, more precise diagnostic criteria and changes in practice for diagnosing autism may have had a substantial effect on the increased incidence of inpatient treated ASD cases from 1987 to 1992. Between 1992 and 1998, the incidence rate based on inpatient and outpatient service use remained rather stable. *Nord J Psychiatry. 2013 Dec 20. [Epub ahead of print]*