PEDIATRICS PERSPECTIVES

Considering Toxic Chemicals in the Etiology of Autism

Heather E. Volk, PhD,[a] Jennifer L. Ames, PhD,[b] Aimin Chen, PhD,[c] M. Daniele Fallin, PhD,[a] Irva Hertz-Picciotto, PhD,[d] Alycia Halladay, PhD,[e,f] Deborah Hirtz, MD,[g] Arthur Lavin, MD,[h] Beate Ritz, MD, PhD,[i] Tom Zoeller, PhD,[j] Maureen Swanson, MPA[k]

Diagnoses of autism spectrum disorder (ASD) (also referred to here as autism), a condition that arises during early brain formation, have increased to now affect 1 in 54 children in the United States.[1] People who have autism exhibit a range of social and communication impairments, along with impairing, restricted, or repetitive patterns of behavior or interests and repetitive behaviors, that can impact activities of daily living.[1] Male children are 4 times more likely to be affected than female children.[1]

Scientists long recognized that genetic factors contribute to autism etiology, as indicated in family, twin, and genetic studies.[2] Yet twin studies, from which heritability estimates are primarily derived, may inflate the role of genetics as both gene-only and genetic-x–shared-environment influences are summarized as genetic. This pervasive problem (of identifying genetic contributions and assuming their effects cannot result from genes acting in concert with environmental agents) also applies to a recent analysis of twin and family studies purporting to demonstrate that the environmental component is an unlikely explanation of both ASD risk and the increase in ASD over time.[3] The environment may act in concert with genetic risk pathways or affect the intrauterine environment directly. In addition, the environment may induce similar epigenetic signatures in twins during gestation.[4] Thus, the shared environment is itself complex and not easily disentangled from shared genetics.

EVIDENCE FOR ENVIRONMENTAL INFLUENCE ON ASD RISK

A large body of evidence, including decades of research on lead and child IQ, indicate a link between toxic environmental exposures and poorer neurodevelopmental outcomes.[5] In animal models and human studies, several toxic chemicals have been implicated in ASD and ASD-related traits and biological markers.[2] Specifically, scientists have found that air pollution exposures during pregnancy and early infancy, at levels typically found in large cities, are associated with autism.[6–8] Several studies suggest that gestational exposures to some neurotoxic and endocrine-disrupting

[a]Wendy Klag Center for Autism and Developmental Disabilities Research, Department of Mental Health, Bloomberg School of Public Health, Johns Hopkins University, Baltimore, Maryland; [b]Division of Research, Kaiser Permanente Northern California, Oakland, California; [c]Department of Biostatistics and Epidemiology, Perelman School of Medicine, University of Pennsylvania, Philadelphia, Pennsylvania; [d]Department of Public Health Sciences, MIND (Medical Investigations of Neurodevelopmental Disorders) Institute, University of California Davis, Davis, California; [e]Department of Pharmacology and Toxicology, Rutgers University, Piscataway, New Jersey; [f]Autism Science Foundation, New York, New York; [g]Departments of Pediatrics and Neurological Sciences, School of Medicine, University of Vermont, Burlington, Vermont; [h]Department of Pediatrics, Akron Children's Hospital, Akron, Ohio; [i]Departments of Epidemiology, Environmental Health, and Neurology, Fielding School of Public Health, University of California Los Angeles, Los Angeles, California; [j]Department of Biology, University of Massachusetts, Amherst, Massachusetts; and [k]The Arc of the United States, Washington, District of Columbia

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Address correspondence to Heather Volk, PhD, 624 North Broadway, HH833, Baltimore, MD 21205. E-mail: hvolk1@jhu.edu

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pesticides, including organochlorines, organophosphates, and pyrethroids, increase the chances of an autism diagnosis or autism-related behaviors in children.[9] Evidence is emerging that other toxic chemicals are associated with autism or autism-related behaviors, notably phthalates, ubiquitous chemicals that cause a decrease in testosterone.[10]

Some environmental factors may also reduce the probability of autism. For example, folate, a vitamin involved in the manufacture and methylation of DNA, plays a critical role in neurodevelopment. Folic acid supplementation, at appropriate levels, around the time of conception appears protective for autism and may ameliorate the impact of toxic chemicals.[11,12] In several studies, researchers have found that the associations linking autism with air pollutants, pesticides, and phthalates are stronger among children of women who either did not take folic acid or had higher folate requirements during pregnancy.[10–12]

STUDIES OF GENE-ENVIRONMENT INTERACTION

As with other multifactorial conditions, it is highly likely that the interplay of gene variants and environmental factors contributes to a substantial proportion of autism.[13,14] Although inherited genes are fixed at conception, environmental factors vary over time and place and can be modified. Thus gene-environment interactions can provide crucial biological insights and opportunities for intervention at the individual, community, and policy levels. Though only a handful of epidemiological studies have examined the role of environmental factors in combination with genetic variation in autism (Table 1), early evidence is striking. For example, a meta-analysis revealed that a variant of the MTHFR gene, which reduces the body's ability to convert folate to its active form, contributes to higher rates of autism, especially in countries without folate fortification in food.[15] Other pieces of compelling evidence also underscore the interaction of genetic susceptibility with toxic chemicals as a major contributor to autism. Researchers analyzing 206 genes from an established genomics database against a database that records the activities of >10 000 chemicals on genes and gene-pathways found that 4428 chemicals interacted with one or more of the genes linked to autism.[16] Additionally, rare genetic variants that by themselves have a strong influence on ASD diagnosis have been shown to have greater biological perturbations when considered in the presence of toxic chemical exposures in in vitro studies.[17,18]

FUTURE RESEARCH AND POLICY

Given the emerging evidence and potential impact of work to investigate chemical exposures and autism, federal agencies and foundations must expand research funding for studies on environmental factors and gene-environment interactions. An approach that has been successful in identifying strong interactions is to examine a limited set of environmental exposures in combination with genetic liability (ie, copy number burden and polygenic risk score), high-risk genes, or epigenetic markers selected for their known biological relevance to specific exposures or autism (Table 1). A reverse application of this approach would be to select genetic variants in biological pathways, like the folate pathway, that interact with an array of environmental factors. However, implementation of such work requires interdisciplinary team science that is not yet common. In short, we need a full-scale commitment to identify the possibly vast number of chemicals that interact with genomic variation and influence the likelihood that children develop neurodevelopmental disorders, including autism and its associated impairments.

To do so, researchers must begin new cross-cutting collaborations that leverage advances in phenotype assessment, biostatistics, and environmental epidemiology and toxicology, overcoming existing disciplinary silos. In addition, gene-environment studies are often limited in statistical power. Increased evaluation and investigation of quantitative trait phenotypes with high correspondence to a traditional diagnosis or that map onto biological underpinnings can improve statistical power and biological relevance. Also, alternative measures of exposures to environmental chemicals coming from advances in use of noninvasive or clinically discarded biospecimens and epigenomic signatures of exposure can expand research opportunities. Analytic techniques to leverage biological database knowledge and focus on discovery of genetic variants in the context of the environment need to be developed and used more broadly. Finally, increased translational research efforts across in vivo, in vitro, and epidemiological efforts would further strengthen the base of knowledge regarding not only the role of genomics in the context of the environment but also the chemical environment itself, in brain development.

It is not necessary to discover all the toxic chemicals that contribute to autism specifically, or to uncover all

TABLE 1 Epidemiologic Studies on Specific Gene-By-Environment Interactions in Autism Risk, Etiologic Mechanisms, Or Severity

Author(s), Year	Study Design	Outcome	Genetic	Environmental	Interaction
Schmidt et al, 2011	Case-control	ASD, odds ratio	Maternal or child one-carbon metabolism genes[a]: genotypes for inefficient metabolism or transmethylation	Prenatal vitamin supplement use during periconception	Highest ASD risk for child if mother took no prenatal vitamin supplements in the periconception and had high-risk (inefficient metabolism or transmethylation) genotypes for MTHFR677, COMT472, CBS, TCN2, and FOLR2
Schmidt et al, 2012	Case-control	ASD, odds ratio	Maternal and child MTHFR677A	Amount of intake of folic acid from food and supplements in first month of pregnancy	Either mother or child or both lacking a T allele, in combination with maternal low folic acid intake in month 1 of pregnancy, conferred elevated child ASD risk
Volk et al, 2013	Case-control	ASD, odds ratio	MET "C" genotype	Air pollutants[b] during pregnancy and the first and second year of life	Elevated risk in children homozygous for the C allele whose prenatal exposures were in the top 25% for each of traffic-related air pollution, PM10, and NO_2.[c]
Kim et al, 2017	Case-control	ASD, odds ratio	Quartiles of total genome-wide copy number variants	Quartiles of air pollutants[b] during pregnancy, and first and second year of life	Elevated risk in children for whom their total length of SNP duplications and their prenatal or first or second year of life ozone exposures were both in the top quartiles of their respective distributions in controls.
Carter and Blizzard, 2012	Database linkage: Autworks' ASGs with chemical activity in Comparative Toxicogenomics Database	Intersections of the 2 databases	206 ASG[d]	Ten environmental chemicals/compound classes; 6 drugs; and 7 endogenous chemicals	Examined ∼1 million chemical x gene interactions involving ∼10 000 chemicals in Comparative Toxicogenomics Database. More than 4000 ASGs were selectively targeted by exogenous chemicals (eg, pesticides, heavy metals, and endocrine disruptors). A total 750 chemicals affected ≥5 ASG's.

TABLE 1 Continued

Author(s), Year	Study Design	Outcome	Genetic	Environmental	Interaction
Webb et al, 2017	Case-only	Severity of symptoms	Presence of ASD-associated CNVs	First trimester ultrasound	Among male children with ASD and CNVs of interest, exposure to ultrasound in the first trimester was associated with lower nonverbal IQ and more severe repetitive behaviors than those without ultrasound. A significant interaction of CNVs and ultrasound was found for both verbal and nonverbal IQ in male children with ASD.
Mazina et al, 2015	Case-only	Severity score	Presence of ASD-associated CNVs	Maternal infection or febrile episode during pregnancy	Children with ASD and ASD-associated CNVs whose mother had an infection or high fever during pregnancy had more severe restricted or repetitive/ stereotyped behaviors and greater deficits in communication, and reciprocal social interactions.

ASG, autism susceptibility gene; CNV, copy number variant; NO_2, nitrogen dioxide; SNP, single-nucleotide polymorphism.
[a] MTHFR677, MTR66, BHMT716, COMT472, CBS, FOLR2, and TCN2.
[b] CALINE model estimates for traffic pollution, NO_2, ozone, PM2.5, and PM10.
[c] Reference group were those in the bottom 75% of the distribution.
[d] Autworks database.

molecular mechanisms underlying autism before we act to protect future generations from known or suspected developmental neurotoxicants. Governments should not allow new chemicals to enter the market unless they are tested for developmental neurotoxicity. We need robust regulatory action now to protect women and children from toxic chemicals, including classes of toxic chemicals like phthalates and pesticides, which have been implicated in other brain-based and behavioral problems, and a long-term strategy to further reduce air pollution, with priority on the most heavily impacted populations. Limiting toxic environmental exposures has the potential to reduce the risk of disabilities associated with autism for future generations.

ABBREVIATION

ASD: autism spectrum disorder

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