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Review Article

Gender disparities, environmental pollutants, genetic and epigenetics in autism spectrum disorder: revealing the links

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ABSTRACT

Autism Spectrum Disorder (ASD) is a complex neurodevelopmental disorder characterized by deficits in social interaction, communication challenges, and repetitive behaviors. Its etiology is multifactorial, involving interactions among genetic, epigenetic, and environmental factors. Recent studies have revealed that environmental pollutants, immune dysregulation, mitochondrial dysfunction, oxidative stress, and epigenetic alterations play significant roles in the development of ASD. Exposure to environmental toxins such as heavy metals and air pollutants has been linked to an increased risk of ASD, primarily by inducing oxidative stress, neuroinflammation, and neurotoxicity. Immune dysregulation, marked by abnormal immune responses, may also contribute to neurodevelopmental alterations observed in ASD. Mitochondrial dysfunction, resulting in impaired energy metabolism and elevated oxidative stress, is frequently reported in individuals with ASD. In addition, epigenetic mechanisms such as DNA methylation and histone modifications are crucial for regulating gene expression and can be influenced by environmental exposures. Individuals with ASD exhibit abnormal epigenetic patterns, indicating that environmental factors can influence gene function without modifying the genetic code. The higher prevalence of ASD in males indicates potential sex-specific susceptibilities and points to a complex gene–environment interaction. While genetic predisposition is a major factor, environmental influences may intensify or reduce the risk. Understanding these interconnected mechanisms is vital for developing improved diagnostic tools, early interventions, and effective therapies. Future research should aim to identify specific biomarkers and molecular targets, enabling personalized approaches to ASD management. A deeper understanding of these mechanisms offers promising pathways for prevention, early detection, and treatment of ASD.

Keywords:
Adaptive immunity
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Introduction

Despite their different processes, onset periods, and consequences, neurodevelopmental and neurodegenerative illnesses share certain commonalities, according to recent studies. Recognizing these similarities may expedite research and allow for the repurposing of existing drugs or the collaborative development of innovative therapies (Jęśko et al., 2020). Autism spectrum disorder (ASD) is a neurodevelopmental condition distinguished by difficulties in social interaction, communication impairments, repetitive activities, and reduced motivation. Symptoms typically become apparent before the age of three and range from mild language and communication difficulties to profound developmental disability (Gonçalves and Monteiro 2023; Maenner et al., 2023; Olsson and Nilholm 2023). According to DSM-5, ASD encompasses autistic disorder, childhood disintegrative disorder, pervasive developmental disorder not otherwise specified, and Asperger's syndrome (Association 2000). A consistent gender disparity is observed, with ASD being about 4–5 times more prevalent in males.

Multiple factors contribute to ASD, including complications during childbirth, hypoxia, advanced parental age, maternal diabetes, medication use, elevated glutamine levels, neuroinflammation, oxidative stress, and exposure to environmental contaminants (Tsai et al., 2019). Many individuals with ASD also present with comorbidities such as gastrointestinal disturbances, sleep problems, sensory processing difficulties, anxiety, hyperactivity, and obsessive-compulsive behaviors (Margari et al., 2020). Recent evidence highlights the gut microbiota as a crucial modulator of brain function and behavior. In ASD, where gastrointestinal disturbances are frequently observed, microbiota alterations may represent an important pathway connecting peripheral symptoms with central nervous system processes (Berding et al., 2021). Some individuals, particularly females, may use compensatory strategies or "camouflaging" behaviors to adapt in social environments, which can obscure diagnosis (Cook et al., 2021). Recent studies highlight the importance of gene–environment interactions and epigenetic modifications in ASD pathophysiology. Environmental exposures during pregnancy may alter DNA methylation, histone acetylation, and chromatin remodeling, thereby affecting transcription and contributing to neurodevelopmental changes observed in ASD (Pellicano and den Houting 2022). These molecular alterations help explain the genetic heterogeneity and phenotypic diversity associated with ASD. Communication between the gut and brain is mediated by several mechanisms, including microbial metabolites, immune pathways, neuronal signaling, and metabolic processes. These interactions, influenced by diet and microbiota composition, are increasingly recognized as important in regulating brain and behavior (Berding et al., 2021). Given the frequent gastrointestinal comorbidities reported in ASD, such mechanisms may also be relevant in explaining links between gut dysbiosis, immune function, and neurodevelopment. In addition, accumulating evidence suggests that environmental pollution-including air pollutants, heavy metals, pesticides, and endocrine-disrupting chemicals-may contribute to ASD risk, especially during critical periods of prenatal and early childhood neurodevelopment, further underscoring the role of gene–environment interactions in ASD pathophysiology (Bhattacharya et al., 2025).

The global prevalence of ASD has increased substantially over the past two decades, emphasizing the urgent need for improved early detection and intervention strategies (Styles et al., 2020). The aim of this study is to better understand the etiology and pathophysiology of ASD and to propose recommendations for future research in this field.

Assessments of ASD Prevalence and Diagnosis

Disparities in the diagnosis of autism are associated with demographic, ethnic, and socioeconomic characteristics, and there are hypotheses proposing potential root explanations for these disparities (Zeidan et al., 2022). The prevalence of autism diagnosis has risen in recent years due to the expansion of diagnostic criteria to include people who were not previously diagnosed in infancy. As a result, formerly distinct diagnostic categories have been combined into a single category called 'autism spectrum disorder' (Gallagher and McGrath 2022). Research investigates the requirements and encounters of individuals with autism throughout their adult years (Crane et al., 2021). Due to the absence of dependable biomarkers, the disorder is diagnosed by observing and describing the key features that affect daily functioning (ICD). Autism affects 1 out of every 69 American and 1 out of every 59 British children, with the majority of cases occurring in western, industrialized nations. While prevalence rates are often higher in

western, industrialized nations, underdiagnosis and limited healthcare or educational resources in many low- and middle-income countries, particularly in Asia and Africa, may contribute to fewer reported cases and insufficient support (Elsabbagh et al., 2012; Zeidan et al., 2022). Additionally, lifestyle variables in more industrialized countries may contribute to the higher prevalence of autism. The frequency of autism is often higher in western, industrialized nations (Hull et al., 2020).

The Phenotype of Female Autism and Camouflage

The male gender plays a prominent role in autism, leading to what is known as the female protective effect (FPE). Women require a higher level of causative factors to exhibit the same impact as men (Lu et al., 2022). Autism diagnoses are often skewed towards males due to the lower likelihood of autism development in females and under-diagnosis. This is because some females with autism exhibit subtle deviations in behavior that are not detected by current diagnostic tools. Additionally, females are more likely to display more pronounced autism characteristics, and a higher percentage of females with autism also have intellectual disabilities compared to boys. Males with autism are often found in the upper intelligence quotient (IQ) range (Baio et al., 2020). The FPE theory suggests that females are more susceptible to genetic and environmental risks compared to males when exhibiting autistic traits to the same extent. Consequently, females are "shielded" from displaying autistic characteristics. This theory is supported by the observation that autistic females possess a higher number of spontaneous, non-inherited mutations linked to autism compared to males. Research indicates that females require a greater genetic impact to meet the diagnostic criteria, implying the presence of an inherent protective factor in females that leads to diminished behavioral manifestations of autistic traits (Jiang et al., 2022). The concept posits that girls are often shielded against autism, indicating that autistic females possess a greater genetic burden than men to manifest the same degree of traits. However, the data supporting this idea is inconclusive, as some research indicates that the close relatives of autistic females exhibit more autistic traits compared to the close relatives of autistic men, while other studies show the opposite outcome (Khramtsova et al., 2023). The extreme male brain hypothesis (EMB) theory suggests that autistic traits, such as heightened systemizing skills and challenges with cognitive empathy and emotional expressiveness, may be linked to higher levels of androgens and other sex hormones that are more prevalent in men. Autistic individuals are often depicted as exhibiting behavioral and psychological characteristics commonly associated with males to an intense degree (Janković-Nikolić et al., 2023). According to the EMB hypothesis, individuals with lower levels of androgen hormones, such as females, also exhibit lower levels of these characteristics, indicating that low androgen levels provide protection against autistic features. Although there is evidence of a connection between autistic traits and elevated androgen levels in females, other studies suggest a limited association between levels of androgens during fetal and early developmental stages with the diagnosis of autism (Weir et al., 2021). A gene on the paternal X chromosome in females increases the expression of autism compared to men, suggesting that the X chromosome may be protective against autism. However, no specific protective gene has been identified. Environmental factors, such as exposure to *in vitro* medications, increase the incidence of autism in boys more than in girls. These factors may interact with genetic risks to contribute to a higher likelihood of autism in males and relative protection against autism in females. Further investigation is needed to elucidate these aspects (Yarger and Redcay 2020). The data highlights relational interest, internalizing issues, and social challenges associated with a female autism phenotype. Camouflaging, a common issue among autistic individuals without intellectual disabilities, may lead to missed diagnoses or inadequate treatment. If females with autism are identified at a lower rate than males, there can be behavioral or genetic variations. The FPE offers insights for current diagnostic methods and research on female autistic patients (Zeidan et al., 2022).

ASD-Causing Environmental and Prenatal Factors

The prevalence of ASD has risen in recent years, with environmental variables such as metallic components having a substantial influence on impairing neurodevelopmental pathways. The implementation of regulations such as the Clean Air Act, as well as worldwide initiatives like those taken by Sweden and the USA, have spurred a research project to investigate the impact of ambient metallic components on the advancement of ASD (Fiłon et al., 2020). Neuronal pathways and neuro-

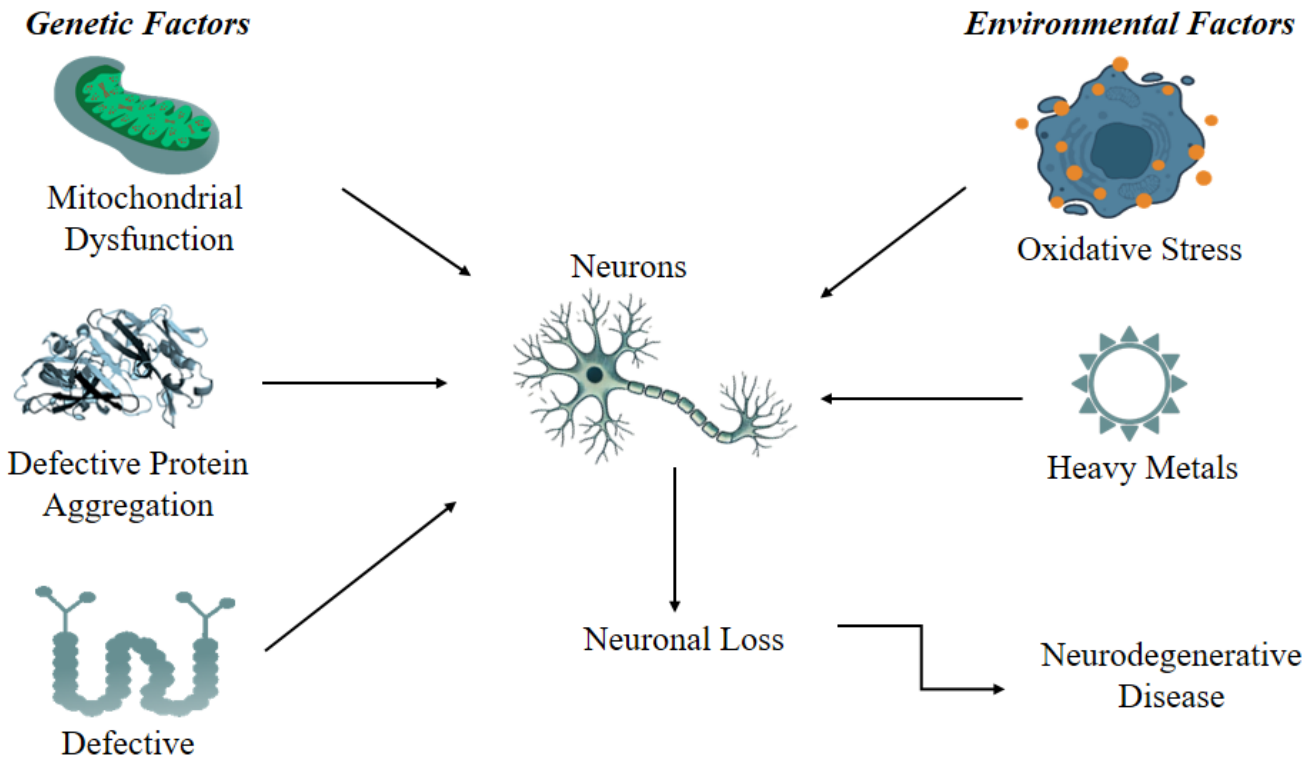

FIGURE 1. Genetic and environmental factors in neurodegeneration. Mitochondrial dysfunction, defective protein aggregation, and gene defects, along with oxidative stress and heavy metal exposure, contribute to neuronal loss and the development of neurodegenerative diseases.

logical functions may be drastically altered and degraded by metallic contaminants present in the environment. The consequences mentioned include the activation of inflammatory mediators, vascular endothelial growth factors, microglial activation, oxidative stress, aberrant astrogliogenesis, overexpression of glutathione, disruption of cell organelles, and autoimmune reactions. Both genetic and environmental variables may have detrimental impacts on brain tissue during early development, possibly triggering and exacerbating genetic abnormalities that lead to changes in neurons. Several metals have been linked to ASD; hence, it is crucial to investigate their prevalence in the environment. This analysis highlights the main metals that have a notable impact on the prevalence of ASD, including aluminum, lead, and mercury. Figure 1 illustrates the proportion of metals that contribute to the overall health index. Arsenic and mercury are the main contributors (Kaur et al., 2021).

Role of Metallic Pollutants in ASD

Metallic toxins such as aluminum, cadmium, arsenic, lead, mercury, manganese, and iron are responsible for causing alterations in living organisms. Studies have shown a connection between ASD and toxic metals, highlighting the need to examine their effects and prevalence. Neurotoxicity is induced by toxic metals through several mechanisms including oxidative stress, activation of microglia, disruption of metalloproteins, endoplasmic reticulum stress, autoimmune responses, and inhibition of glycolysis (Mabrouk et al., 2021). These mechanisms impede the growth of the nervous system, speed up the degeneration of nerve cells, and cause toxic consequences, cell death, and inflammation in the nervous system. Research has shown a comparative analysis between persons with and without ASD by using biomarkers to identify the presence of metals in urine, red blood cells, hair (Fiłon et al., 2020), and teeth (Luhach et al., 2024). Metallotoxicity may have detrimental effects on living organisms, necessitating the use of therapy methods such as chelating agents, dietary adjustments, glutamate antagonists, anti-inflammatory drugs, and antioxidant treatments (Gorzi et al., 2021). These therapies may boost the process of detoxification

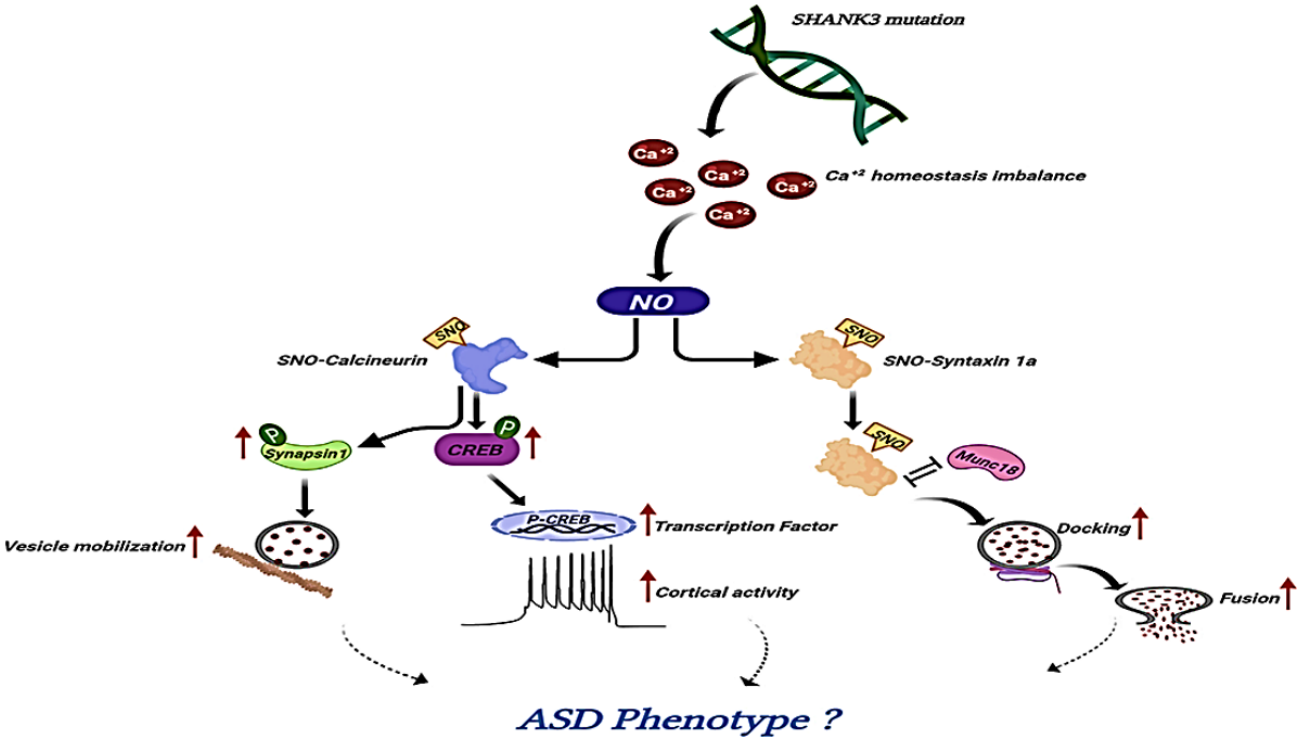

FIGURE 2. Nitric oxide (NO) signaling in autism spectrum disorder (ASD). Schematic representation of the role of NO in ASD. Mutation in the SHANK3 gene may disrupt Ca^{2+} homeostasis, leading to increased intracellular NO production and subsequent S-nitrosylation of multiple proteins. S-nitrosylation of calcineurin inhibits its phosphatase activity, resulting in elevated phosphorylated (P) synapsin-1 and phosphorylated CREB (P-CREB). P-synapsin-1 promotes synaptic vesicle mobilization, while P-CREB enhances the recruitment of transcriptional co-activators and cortical activity. Additionally, S-nitrosylation of syntaxin-1a impairs its interaction with Munc-18, thereby facilitating vesicle docking and fusion. Collectively, these NO-mediated alterations may contribute to synaptic dysfunction associated with ASD.

by increasing the levels of glutathione, optimizing the use of antibiotics during pregnancy, and supplying protective substances such as curcumin, magniferin, selenium, and chelating agents (Ramazani et al., 2024). In general, the results of studies conducted in recent years have shown that the ASD group's levels of Cd, Pb, arsenic, and Hg were greater than those of the healthy control group. Also, exposure to toxic heavy metals can trigger the generation of reactive oxygen species (ROS) and exert an epigenetic influence, potentially contributing to the emergence of ASD (Ding et al., 2023). Given the increased likelihood of ASD in today's society, it is essential to carefully examine the presence of metallic elements in the environment and implement measures to prevent neurological developmental problems in children. This will help create a safe and efficient environment for future generations (Cable et al., 2022).

The Role of Oxidative Stress and Nitric Oxide (No) in ASD

Neurodegenerative disorders, such as Parkinson's disease (PD), Alzheimer's disease (AD), and Amyotrophic lateral sclerosis (ALS), are characterized by the excessive generation of reactive oxygen species (ROS), a form of oxidative stress. This is observed in the brain tissues of deceased individuals who were diagnosed with these disorders, including the presence of nitric oxide (NO) (van Maarschalkerweerd et al.,).

The SHANK3 mutation is a very promising mutation related to ASD, as several investigations have shown abnormalities in biochemical, electrophysiological, and cellular pathways. Amal et al. (Amal et al., 2020) suggested that the Shank3 mutation increases Ca2+ influx, activating neuronal nitric oxide synthase (nNOS or NOS1) activity, leading to NO production and molecular alterations like S-nitroso glutathione (GSNO), 3-nitrotyrosine (Ntyr), and S-nitrosylation (SNO). SNO selectively affects many intracellular proteins, thereby modifying signaling pathways and possibly resulting in impairments in synapse function, neuronal activity, and behavioral performance. The examination of both wild type and Shank3 knockout (KO) mice using sys-

tem biology techniques showed a 9-fold alteration in the SNO level of proteins related to the synaptic vesicle cycle in the brain of KO mice. The investigation of gene ontology and KEGG in 6-week-old KO mice revealed an abundance of proteins associated with neurodevelopment and ASD. The data provide compelling evidence of a correlation between the Shank3 mutation and NO (Amal et al., 2020).

The research discovered that the cortex of KO mice has a network of S-nitrosylated proteins that play a role in the synaptic vesicle cycle, neurotransmission, and the control of glutamate. The cortex of KO mice, both 6 weeks and 4 months old, had common proteins that displayed indications of enhanced processes impacted by ASD, such as the synaptic vesicle cycle. The research further observed elevated amounts of 3-nitrotyrosine in various cortical areas, increased GSNO levels in both the KO and wild-type (WT) groups, and the cortex showed SNO of calcineurin, which hindered its phosphatase function. The inhibition resulted in an elevation of p-Synapsin-1 and p-CREB protein levels (Fig.2) (Tripathi et al., 2020). The results indicate that knockout mice may have enhanced mechanisms that are influenced by ASD.

The research demonstrates that calcineurin, which is a target of calcineurin, is accountable for the enhanced movement of vesicles in mutant mice. The elevated phosphorylation of Synapsin-1 in the cortex of these animals suggests that SNO of calcineurin is the cause of enhanced vesicle mobilization (Chi et al., 2003). In the mutant mice, it was observed that the levels of p-CREB, which is another substrate of calcineurin, were elevated. Syntaxin1a, which promotes the assembly of the SNARE complex, was also discovered to be SNO in the mutant mice. The research proposes that SNO of metabotropic glutamate receptor 7 (mGluR7) might enhance the entry of calcium ions (Ca2+) into presynaptic neurons, thereby promoting the fusion of vesicles (Palmer et al., 2008). The results indicate that NO plays a significant role in ASD, and this discovery may have implications for a wider range of individuals with different genetic backgrounds but similar underlying causes (He et al., 2015).

Several post-mortem investigations in people diagnosed with ASD have shown that the affected parts of the brain display increased cell death when exposed to oxidative stress (Thorsen 2020).

Lopez-Hurtado and Prieto (López-Hurtado and Prieto 2008) found that individuals with ASD have a greater concentration of lipofuscin, a combination of lipids and proteins formed due to oxidative damage, in specific regions of the cortical brain that are responsible for communication. This occurrence often leads to the loss of speech and language abilities during regression.

Increased oxidative stress indicators have been discovered in brain areas linked to symptoms of ASD in studies. The orbitofrontal cortex, Wernicke's area, cerebellar vermis, cerebellar hemisphere, and pons had the greatest concentrations of 3-nitrotyrosine (3-NT), which is linked to speech processing, sensory coordination, emotional behavior, and memory.

Comparing individuals with ASD to control individuals, Sajdel-Sulkowska et al (Sajdel-Sulkowska et al., 2011). discovered elevated levels of neurotrophin-3 (NT-3) and 3-NT, which are indicators of oxidative stress, in the cerebellum of folks diagnosed with ASD.

Evans et al (Evans et al., 2008) discovered increased levels of oxidative stress indicators in the brains of individuals who have been diagnosed with ASD. A distinctive thread-like pattern was discovered in the brains of individuals with ASD, which was absent in the brains of neurotypical individuals employed as controls. This discovery emphasizes the need to comprehend the oxidative stress response in individuals with ASD. Chauhan et al. examined the process of DNA oxidation and the balance of glutathione redox in postmortem brain tissues obtained from people diagnosed with ASD and compared them to a control group of individuals of the same age. The researchers discovered a significant rise in DNA oxidation levels in the frontal brain, temporal cortex, and cerebellum of individuals with ASD, as compared to the control group. In addition, the levels of reduced glutathione were decreased and the levels of oxidized glutathione were elevated in samples taken from the cerebellum and temporal cortex of individuals with ASD. Chauhan et al (Chauhan et al., 2012) observed a significant increase in lipid hydroperoxide levels in the cerebellum and temporal cortex of people with ASD, as compared to those without any behavioral or disciplinary issues.

8-oxo-guanosine (8OHdG), an RNA marker used for evaluating oxidative stress in the brain, has shown effect in quantifying brain damage and degeneration in neurodegenerative disorders. It exhibits a noteworthy asso-

ciation with serum S100beta levels, which are already employed for measuring brain damage (Michalska and León 2020). Sajdel-Sulkowska et al (Sajdel-Sulkowska et al., 2009) used 8OHdG levels to assess oxidative damage in the cerebellum of people diagnosed with ASD. The findings indicated a significant rise of 63.4% in cerebellar 8OHdG levels compared to the control group.

Levels of Vitamin D and ASD

Vitamin D is crucial for brain homeostasis, DNA protection, and neurotransmission. It plays a role in calcium signaling, proliferation, differentiation, neurotrophic and neuroprotective actions, and synaptic plasticity. However, 95% of patients have Vitamin D deficiencies. Adolescents with ASD have lower levels of 25-OHD compared to those under 11. In a group of 100 diagnosed with ASD, average blood 25-OHD levels were lower and alkaline phosphatase levels were higher. A correlation exists between vitamin D and ASD in children, making it essential to monitor vitamin D levels in autistic children for preventive measures and early treatment (Şengenç et al., 2020).

Immune Dysregulation in ASD

The immunological hypothesis posits that the development of autism is affected by immune malfunction, which is associated with behavioral characteristics in individuals with autism. The analysis of cerebrospinal fluid and peripheral blood reveals distinct immunophenotypes and associated behavioral symptoms in different subgroups of individuals with autism. This provides valuable information on the severity of the disorder and its clinical manifestations (Robinson-Agramonte et al., 2022).

The Major Histocompatibility Complex (MHC) is a collection of genes that exhibits a high degree of genetic variation, resulting in a wide range of different alleles in the genome. It facilitates both adaptive and innate immune responses. Various studies have shown a connection between the development of ASD and certain genes, such as human leukocyte antigen (HLA) alleles A2, Death Receptor (DR)4, and DR11 (Hughes et al., 2022). These genes are connected with a weakened response from lymphocytes and a significant vulnerability to ASD. A deficiency of the C4B complement allele in the HLA class III region, caused by duplications of C4A, increases the chance of developing ASD by a factor of 4.3 (Hashem et al., 2020). The PRKCB1 gene, which is a serine and threonine kinase C gene, is associated with both B-cell activation and neural function, and has also been connected to ASD. Studies analyzing RNA expression in peripheral blood have shown an increase in the activity of genes associated with the activation of the innate immune system via the natural killer (NK) pathway in patients diagnosed with ASD (Hughes et al., 2022; Mapelli et al., 2022). Several genetic variations that increase vulnerability to altered gene expression, specifically in relation to the activation of the innate immune system and the impairment of adaptive immunological control, have been proposed.

Prior research has shown a reduced immune response of lymphocytes to mitogens in children diagnosed with ASD, where certain types of lymphocytes contribute to the development of ASD (Rose et al., 2020). As a result, there is an unequal proportion of helper/suppressor cells, characterized by a reduction in helper-inducer T cells and cells that are responsive to interleukin-2R during mitogenic stimulation. There is a negative correlation between the intensity of autistic characteristics and these cells. Autism has been shown to be linked with a notable rise in CD4+ memory cells and a decline in CD4+ naïve T cells, particularly in those with HLA A2-DR11 (Chua et al., 2021). Furthermore, there exists a disparity in the cytokines generated by CD4+ and CD8+ T cells, with a bias towards Th2 response. As a result, there is a decrease in the percentage of CD4+ and CD8+ T cells that produce IFN-γ and IL-2, in comparison to suppressor cytokines such as IL-4. T regulatory cells (Tregs) have a crucial function in regulating immunological responses, and autistic children have a reduced population of CD4+ CD25 high Tregs (Brynge et al., 2022). The primary difficulty in ASD is in comprehending the correlation between immunological abnormalities and neurodevelopmental difficulties, as well as formulating efficacious therapy techniques. This will mitigate disparities across diverse groups and provide a fresh clinical and experimental outlook, facilitating the proposition of more appealing resolutions and innovative therapies.

Association between Mitochondrial Malfunction and ASD

It is possible that there is a fundamental cellular process responsible for ASD. One area of interest is mitochondria, which play a role in various cellular process-

es, particularly metabolism. Recently, researchers have been considering mitochondria-encoded genes because they are inherited from the mother, have their own genetic material, and are involved in embryonic development. Several studies have linked mitochondrial problems, such as oxidative stress, production of reactive oxygen species (ROS), and variations in mitochondrial DNA copy number, to autism. Autistic children tend to have lower activity levels in mitochondria-encoded bioenergetic complexes I, III, IV, and V (Balachandar et al., 2021).

Epigenetic Mechanisms and Autism Development

Epigenetic mechanisms can modify DNA without altering its sequence, thereby contributing to neurodevelopmental disorders such as autism spectrum disorder (ASD). Patients with ASD show significant gene expression disparities associated with epigenetic alterations, underlining the crucial role of these mechanisms. Among them, DNA methylation and histone modification play central roles in regulating gene expression, while noncoding RNAs influence chromatin structure and gene expression. DNA methylation is the most extensively studied epigenetic process in ASD research. Post-mortem brain tissues have provided essential evidence linking environmental exposures with gene changes in ASD (Mapelli et al., 2022).

Methylation array analyses have revealed variations in specific CpG sites and differentially methylated regions, although no global differences in methylation patterns were detected between ASD and control brains (Yoon et al., 2020).

Wong et al. performed research on the methylome analyses of ASD in the cerebral cortex. They discovered many comethylation modules that are strongly linked to the diagnosis of ASD. The study used DNA samples extracted from the prefrontal and temporal cortex of post-mortem individuals (Wong et al., 2019). The gene ontology analysis of comethylation modules identified pathways that are enriched in immune functions, synaptic signaling, and postsynaptic density. These findings indicate the combined effects of immunological and neural dysregulation that are linked with ASD. Vogel Ciernia et al. identified gene pathway enrichments in the brain of individuals with ASD using a technique called whole-genome bisulfite sequencing (WGBS). Other methylomic investigations that use arrays have also discovered comparable results (Vogel Ciernia et al., 2020). Ladd-Acosta et al. identified three differentially methylated regions (DMRs) in the temporal cortex of individuals with ASD. These DMRs are located near genes that play a role in neuronal and immunological pathways, as well as in sustaining imprinting patterns (Ladd-Acosta et al., 2014). Nardone et al. (Nardone et al., 2014) discovered distinct patterns of methylation CpGs in samples of the prefrontal cortex of individuals with ASD. They observed that hypomethylated CpGs were more prevalent in genes associated with immunological activities, whereas hypermethylated CpGs were more prevalent in genes associated with synaptic activity between neurons. Additional investigation of ASD methylome identified certain areas with a high concentration of comethylation, which were shown to be associated with synaptic, neuronal, GABAergic, and immunological processes. Analysis of the methylome in the cerebellum has revealed similar overall findings, but with variations in the specific genetic locations where differences related to ASD are observed. Corley et al. conducted a study on DNA methylation in the subventricular zone, which is a part of the cerebellum responsible for producing neuronal stem cells. They identified specific locations in the DNA where methylation patterns differed significantly in samples from individuals with ASD (Corley et al., 2019). There are 18 HDAC enzymes in humans that are categorized into the HDAC family, which consists of class I, class II, class IV, and class III. HDACs catalyze the removal of acetyl groups from lysine residues on histones, resulting in alterations to the structure of chromatin and a decrease in the transcription of genes. Nevertheless, several HDAC paralogs exhibit extranuclear functionality. Histone acetyltransferases, in contrast, loosen the coiling of DNA around histones, hence enhancing gene transcription (Fig.3) (Tseng et al., 2022). HDACs and histone acetyltransferases modulate gene expression by altering acetylation levels, leading to many downstream consequences, such as alterations in cognitive processes. These impacts are recognized to have a substantial influence on gene expression (Berding et al., 2021).

Conclusion and Future Perspective

ASD is influenced by multifaceted prenatal and postnatal factors including genetic, epigenetic, and environmental factors. Our review highlighted the important

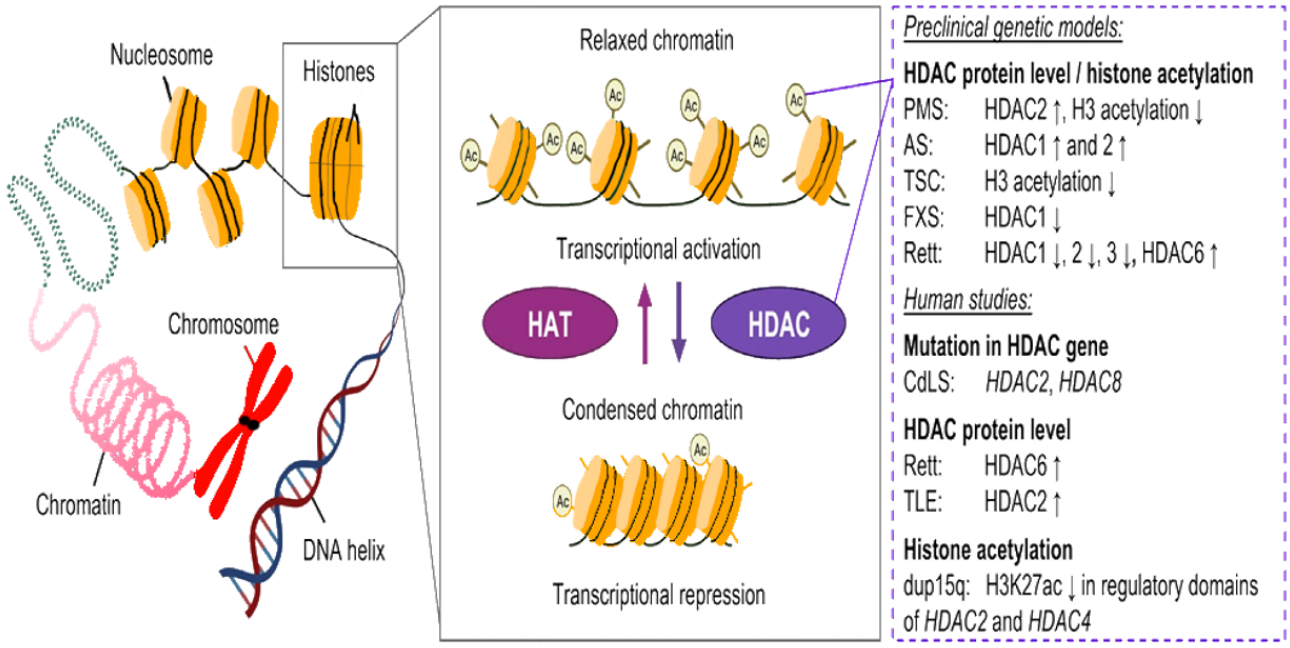

FIGURE 3 . The actions of HDACs and HATs alter chromatin structure and, as a result, gene transcription. HDAC alterations have been found in preclinical and human studies of ASD-associated disorders. AS, Angelman syndrome; CdLS, Cornelia de Lange syndrome; dup15q, duplication 15q syndrome; FXS, fragile X syndrome; HAT, histone acetyltransferase; HDAC, histone deacetylase; PMS, Phelan-McDermid syndrome; Rett, Rett syndrome; TLE, temporal lobe epilepsy; TSC, tuberous sclerosis complex.

role of environmental contaminants in the neurodevelopmental disruption of ASD and the contribution of immune dysregulation, mitochondrial dysfunction, and oxidative stress focusing on NO. In addition, we discussed epigenetic modification including DNA methylation and histone modification. We also shed light on the unique presentation of ASD in females. Due to camouflaging behavior, most ASD females are not diagnosed. Therefore, future studies should focus on developing more accurate and sensitive criteria for ASD diagnosis with specific attention to female protective factors. We recommend that other researchers work on interdisciplinary research to link environmental, genetic, and epigenetic factors to identify new biomarkers and therapeutic targets.

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