Valproic Acid Exposure and its Impact on CNTNAP2 Gene Expression in the Development of Autism Spectrum Disorder

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Abstract

Autism Spectrum Disorder (ASD) is a neurodevelopmental disorder characterized by both genetic and environmental factors. The CNTNAP2 gene, critical for neural development, has been associated with ASD, particularly when its expression is disrupted. Prenatal exposure to valproic acid (VPA), a teratogenic anticonvulsant, furthers this risk by altering CNTNAP2 expression during pregnancy. This review explores how CNTNAP2 function and VPA exposure interact to influence ASD development, with an emphasis on interactions between the environment and genetics. Potential therapeutic approaches, including gene therapy, HDAC inhibitors, and dietary interventions, offer hope for these effects. However, there are gaps in research, particularly in long-term outcomes and personalized treatments. This paper demonstrates the importance for targeted therapies addressing the intersection between genetics and environmental factors.

Keywords: Autism Spectrum Disorder, CNTNAP2, Valproic Acid, Brain Development, Critical Period, Gene Expression.

Introduction

Autism Spectrum Disorder (ASD) is a neurodevelopmental disorder where individuals experience difficulties in social interaction, communication, repetitive behaviors, and sensory issues. Individuals with ASD also have a broad range of cognitive abilities, ranging from intellectual disability to above-average functioning (American Psychiatric Association, 2013). ASD has a wide range of symptoms causing it to be considered a spectrum.

According to the Centers for Disease Control and Prevention (CDC), approximately 1 in 36 children in the United States are diagnosed with ASD, which is an increase from 1 in 54 children (CDC, 2023). This rise is attributed to several reasons including diagnostic criteria, awareness of the condition, and detection methods (Elsabbagh et al., 2012). This increase in the prevalence of ASD demonstrates the growing need for more understanding about the disorder.

ASD results from genetic and environmental influences (Sandin et al., 2014). Both inherited genetic variants and external environmental factors, such as prenatal exposures to toxicants increase the risk of development of ASD (Sandin et al., 2014). This suggests environmental factors during periods of development can further increase the development of the disorder (i.e., diathesis-stress model; APA Dictionary of Psychology, n.d.). Here, one environmental and one genetic factor are reviewed.

Valproic Acid (VPA)

Valproic Acid (VPA) is a widely prescribed anticonvulsant medication used to treat conditions such as epilepsy, bipolar disorder, and migraines. For example, in 2018 the number of pregnant women taking VPA during pregnancy was 0.16% in the United Kingdom (Gaudio et al., 2022). However, despite its benefits for treating certain conditions, VPA is known to have teratogenic effects when used during pregnancy, putting fetal development at risk (Christensen et al., 2013). Prenatal exposure to VPA has been associated with developmental issues, such as neural tube defects, craniofacial abnormalities, and risk for neurodevelopmental disorders like ASD (Christensen et al., 2013).

CNTNAP2

CNTNAP2 (Contactin Associated Protein-Like 2) is a gene that encodes a protein for neuronal communication and the formation of neural networks. CNTNAP2 belongs to the neurexin family, which is important for synaptic function, including the creation and maintenance of electrical synapses between neurons (Poot, 2015). The protein is essential for development as it influences processes like neuronal migration, axon guidance, and synapse formation.

Changes in CNTNAP2 expression or function can disrupt normal brain development, which contributes to abnormal neural activity associated with disorders like ASD (Peñagarikano et al., 2011). Individuals with CNTNAP2 mutations often display difficulties in social behavior and communication, both of which are features of ASD (Peñagarikano et al., 2011). Additionally, research has shown that changes in CNTNAP2 expression may affect brain regions critical for language, which may be related to communication and language challenges in ASD.

This review aims to explore the interactions between ASD, CNTNAP2 gene expression, and VPA exposure. By exploring existing research, this paper will investigate how exposure to VPA affects CNTNAP2 gene expression in the development of ASD during fetal development. This paper will further focus on gene-environment interactions and the potential long-term impact of VPA on CNTNAP2 and their effect on ASD.

Valproic Acid (VPA): Mechanisms and its Role in ASD Development

Valproic acid (VPA) is an anticonvulsant and mood-stabilizing medication which is used for conditions like epilepsy, bipolar disorder, and migraines. It inhibits GABA transaminase, which leads to increased levels of gamma-aminobutyric acid (GABA) in the brain (Harden, 2013). GABA is the primary inhibitory neurotransmitter in the central nervous system, and increased availability helps to stabilize neuronal activity, leading to reduction of seizures and mood fluctuations (Poot, 2015;Smith & Brown, 2014).

VPA is also known to function as a histone deacetylase (HDAC) inhibitor, which impacts gene expression through epigenetic mechanisms (Bromley et al., 2013). Histones are proteins that help package DNA into a compact, organized structure within the nucleus, and their acetylation status determines how tightly or loosely the DNA is wrapped (Geschwind, 2012). HDAC inhibitors like VPA increase histone acetylation, causing increase of the transcription of certain genes, such as those involved in neural development. (Bromley et al., 2013).

Studies have shown a strong correlation between in utero VPA exposure and an increased risk of ASD in offspring (Pinto et al., 2015). This association has been observed in animal models, where prenatal VPA exposure resulted in behaviors like those of individuals with ASD (Smith & Brown, 2014). For instance, mice exposed to VPA in utero are more likely to exhibit core ASD traits such as reduced social interaction and repetitive behaviors,(Poot, 2015; Geschwind, 2012).

VPA affects multiple molecular pathways that are critical for brain development (Geschwind, 2012). These include ion channels and signaling pathways such as Wnt signaling, which regulate cellular differentiation, migration, and synaptic formation (Penagarikano & Geschwind, 2012). Disruption of these pathways by VPA exposure leads to functional abnormalities (Penagarikano et al., 2011). Furthermore, oxidative stress induced by VPA has been shown to contribute to cellular damage and altered neural growth (St George-Hyslop et al., 2022).

Effect of CNTNAP2 on ASD

CNTNAP2 is essential for neuronal development and function. The structure of CNTNAP2 includes multiple extracellular domains, such as laminin G and epidermal growth factor-like domains, which allow it to interact with other neuronal proteins. This is crucial for synaptic stability, the formation of neuronal circuits, and synaptic plasticity, which enable the brain to adapt (Mohammad-Rezazadeh et al., 2016; Lu et al., 2014). CNTNAP2 serves as a bridge, facilitating communication between neurons, particularly in the regulation of myelinated axons (Rodenas-Cuadrado et al., 2014; Penagarikano & Geschwind, 2012).

During brain development, CNTNAP2 expression is highly regulated, with peak expression occurring during periods of heightened neuronal activity such as synaptogenesis and neuronal migration. These stages are important for the formation of neural circuits that support cognitive, motor, and social functions (Rodenas-Cuadrado et al., 2014). Dysregulation of CNTNAP2 during these periods has been linked to ASD (Lu et al., 2014; Poot, 2015).

The synaptic formation process involves the clustering of voltage-gated potassium channels at juxtaparanodal regions of myelinated axons, ensuring proper signal transmission along neuronal pathways. This process is vital for electrical signals between neurons, which controls various cognitive and motor functions (St George-Hyslop et al., 2022). When CNTNAP2

is deficient or its expression is altered there are once again disruptions in the neural circuits that regulate behaviors associated with ASD (Scott-Van Zeeland et al., 2010).

Studies involving CNTNAP2 knockout models (i.e., mice genetically engineered to lack the CNTNAP2 gene) demonstrate that disruptions in this gene's function lead to ASD-like behaviors (Penagarikano et al., 2011). For example, CNTNAP2 knockout mice show behaviors such as impaired social interactions, repetitive actions, and communication deficits (Rodenas-Cuadrado et al., 2014). Additionally, analyses of individuals with ASD have identified both common and rare variants of CNTNAP2, suggesting the significant role of this gene (Mohammad-Rezazadeh et al., 2016).

Effect of VPA on CNTNAP2 Expression

VPA exposure has been shown to downregulate CNTNAP2 expression during crucial periods of brain development (Pinto et al., 2015; Geschwind, 2012). By increasing histone acetylation at the CNTNAP2 gene, VPA alters the expression of this critical gene, leading to long-term changes in brain function and connectivity. This downregulation hinders synaptic formation, increasing the risk of ASD (Poot, 2015; Rodenas-Cuadrado et al., 2014). Studies using CNTNAP2 knockout mice treated with VPA show these animals exhibiting ASD-like behaviors compared to controls (Penagarikano et al., 2011).

Animal studies have shown that exposure to VPA during early to mid-gestation has the most effect on brain development. Specifically, exposure during embryonic day 12 in rodent models corresponds to a period of cortical development, during which neurons are forming connections (Harden, 2013). This timing aligns with the peak periods of CNTNAP2 expression emphasizing the connection.

Implications

Building on these findings, One promising intervention is gene therapy: it aims to restore normal CNTNAP2 function by either correcting the mutation or enhancing the gene's expression. Another approach involves using histone deacetylase (HDAC) inhibitors to normalize the gene expression patterns disrupted by VPA exposure. HDAC inhibitors can potentially reverse the negative effects of VPA. (Peñagarikano et al., 2011).

Further, researchers are investigating pathway-specific therapies. For example, CNTNAP2-deficient mice have shown hyperactive Akt-mTOR signaling in the hippocampus (Han et al., 2019). Studies using inhibitors to suppress this signaling pathway have successfully repressed social deficits in these mice. However other behaviors such as hyperactivity and repetitive actions are not able to be treated. The findings suggest that targeting the Akt-mTOR pathway could help certain social impairments, but the pathway may not address all of the symptoms (Han et al., 2019).

Another area of interest is dietary interventions, such as the ketogenic diet (KD). Studies suggest that the KD may improve behavioral deficits (Ahn et al., 2014). In a pilot study, where pregnant mice adhered to a KD for four weeks, they saw reductions in behavioral deficits. The reason could be that it increases GABA levels, a neurotransmitter associated with inhibitory signaling in the brain (Castro et al., 2016). Additionally, some studies report reductions in ASD severity when combined with gluten- and casein-free diets, particularly in mice with comorbid conditions like epilepsy (Castro et al., 2016).

Despite the progress in understanding the role of CNTNAP2 and VPA in ASD, several gaps remain. While individual studies have investigated the effects of CNTNAP2 mutations or

VPA exposure on ASD, comprehensive studies that explore the interaction between the two are lacking. Additionally, longitudinal studies that track the long-term neurodevelopmental outcomes of individuals exposed to VPA prenatally are necessary to create therapeutic interventions. The studies examined here investigated the role of CNTNAP2 and VPA on ASD in small samples, which limits generalizability of effects. Further, there is large genetic variability between individuals with ASD, and this review only examined the role of one gene. The variability in ASD can be explained by how certain genes respond to specific environmental factors at different developmental stages. Not all individuals with the same genetic mutation will present with ASD, and not all who are exposed to VPA will develop the disorder. This demonstrates the importance of larger studies, which will help develop personalized treatments to reduce risk of ASD onset in a broad range of individuals.

Future research should also focus on personalized medicine for ASD. Given the nature of the disorder and the variability in genetic and environmental influences, personalized approaches could be made based on an individual's specific genetic makeup and environmental exposure history. For example, individuals with specific CNTNAP2 mutations might benefit from therapies that enhance the expression of this gene, whereas those exposed to VPA could benefit from therapies that counteract the negative effects of prenatal exposure. By using the growing understanding of ASD, researchers may be able to develop more effective individualized treatments.

Conclusion

This review shows the critical role of CNTNAP2 in neuronal development and its interaction with prenatal exposure to VPA. The genetic influence of CNTNAP2, with environmental influences such as VPA, demonstrate the complexity of gene-environment interactions in ASD. Current studies research how disruptions in CNTNAP2 expression, specifically during critical periods of brain development, contribute to ASD-like behaviors. Therapeutic approaches, such as gene therapy and pathway-specific interventions, offer promising interventions. However, significant gaps remain, particularly in understanding how CNTNAP2 and VPA influence long-term outcomes. This review emphasizes the need for continued research into these interactions, with a focus on individualized treatment approaches tailored to genetic and environmental factors and larger studies in general.

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