www.directivepublications.org

Advances in Behavioral Neuroscience

DIRECTIVE PUBLICATIONS

Review Article

Pathogenetic Understandings, Genetic Threads, And Therapeutic Prospects Of Neuroglial Dysregulation In Autism Spectrum Disorder.

Nikola Ilic, Bdrijan Larajlija.

University of Teramo, 64100 Teramo, Italy.

Abstract

Background/Goals: Autism Spectrum Disorder (ASD) is a complicated neurodevelopmental disorder characterized by repetitive behaviors, limited interests, and difficulties with social communication. Recent research has demonstrated the critical functions that neuroglial cells, including oligodendrocytes, microglia, and astrocytes, play in neuroinflammation, synaptic function, and brain connection. These results provide a new understanding of the pathophysiology of ASD. The present understanding of neuroglial dysfunction in ASD is summarized in this review, with a focus on the pathophysiological causes, genetic factors, and possible therapeutic approaches.
Methods: Using knowledge from clinical research, neuroscience, and molecular biology, we carried out an extensive literature study. Particular attention was paid to the regulation of synaptic plasticity, glial-mediated neuroinflammatory processes, and the effects of genetic mutations on neuroglial signaling and homeostasis.
Results: Defective oligodendrocyte-driven myelination, poor trocytic glutamate modulation, and aberrant synaptic pruning by microglia all contribute to neuronal architecture disruption in ASD. New treatments that target these pathways, such as cell-based methods, microglial modulators, and anti-inflammatory medications, have the potential to reduce important symptoms of ASD. A thorough foundation for comprehending the neuroglial contributions to ASD is established by this review. It advances our knowledge of the pathophysiology of ASD and opens the door for innovative therapeutic approaches that target neuroglial pathways by combining insights from several disciplines.

Keywords : *Innovative Therapy; Neurogenetics; Neuroglia; Autism Spectrum Disorder (Asd).*

INTRODUCTION

About 1 in 36 children worldwide suffer from autism spectrum disorder (ASD), a neurodevelopmental disorder that is becoming more common as a result of better diagnostic methods and greater awareness [1,2]. With a roughly 4:1 ratio, males are diagnosed with ASD more often than girls. Nonetheless, there is evidence that supports underdiagnosing in males may result from softer symptom presentations or concealing behaviors [3,4]. There is clinically significant heterogeneity in ASD. Repetitive behavior, limited interests, and trouble communicating socially are core signs [5]. These fundamental characteristics frequently coexist with a range of related comorbidities, including intellectual disability, global developmental delay, different types of epilepsy, gastrointestinal problems, anxiety, and sleep disorders [6,7]. Although mild symptoms may be seen as early as infancy, the clinical indications of ASD usually appear by the time a child is three years old. Early warning signs include abnormal social interactions, restricted eye contact, delayed speech and language development, and repetitive motor movements including spinning, rocking, or flapping of the hands [9]. Aversion to particular noises, textures, or lights is one example of a sensory sensitivity that is frequently observed [10,11]. The clinical course varies greatly; some kids show noticeable improvement after receiving early intervention, while others have symptoms that don't go away or get worse [12].Clinical criteria outlined in the Diagnostic and Statistical Manual of Mental Disorders (DSM-5) [13] are used to diagnose ASD. Important diagnostic criteria include limited, repetitive patterns of behavior, interest, or activity, as well as ongoing deficiencies in social communication and interaction in a variety of circumstances [14]. Standardized tests like the Autism Diagnostic Observation Schedule (ADOS)

***Corresponding Author:** Bdrijan Larajlija, University of Teramo, 64100 Teramo, Italy.
Received: 06-Jan-2025, **; Editor Assigned:** 07-Jan-2025 **; Reviewed:** 19-Jan-2025, **; Published:** 27-Jan-2025.
Citation: Bdrijan Larajlija. Pathogenetic Understandings, Genetic Threads, and Therapeutic Prospects of Neuroglial Dysregulation in Autism Spectrum Disorder. Advances in Behavioral Neuroscience. 2025 January; 1(1).

and structured interviews like the Autism Diagnostic Interview-Revised (ADI-R) are frequently utilized diagnostic techniques [12,15].As part of their evaluations, a multidisciplinary team of pediatricians, neurologists, psychologists, and speech therapists frequently performs behavioral, cognitive, and developmental tests [14]. Although they cannot diagnose conditions, genetic, laboratory, and neuroimaging testing can offer supporting data [5,16]. Children with ASD frequently have structural and functional issues in their brains, as seen by MRI scans. These issues include reduced gray matter in the prefrontal cortex and amygdala, as well as altered connections in the default mode network [17, 18].

The diagnostic process for ASD includes a number of genetic tests. The goal of testing is to identify syndromic forms of autism, usually in accordance with recognized clinical recommendations. Well-established genetic alterations that contribute to ASD symptoms and other related characteristics are what define these illnesses [19].

Targeted diagnostics like FMR1 gene analysis are frequently the first choice for suspected monogenic disorders, such as fragile X syndrome, that present clinically as ASD [20].

Chromosome microarray analysis (CMA), which is generally suggested as a first-tier test because of its capacity to detect submicroscopic chromosomal deletions or duplications that are commonly linked to ASD [21], is typically the first step in the testing procedure. Furthermore, whole-exome sequencing (WES) and panel testing of genes linked to autism are two examples of next-generation sequencing (NGS) that is being utilized more and more to find mutations in individual genes associated with ASD [22]. ASD may be largely caused by chromosomal abnormalities, single-gene mutations, and copy number variations (CNV), many of which have an effect on synaptic function, neurogenesis, and neuronal migration [16,19]. An increased incidence of ASD has been associated with environmental variables, like as exposure to toxins such valproic acid, prenatal illnesses, and advanced parental age [24–26].

Histone acetylation and DNA methylation are two examples of epigenetic changes that offer a useful connection between environmental triggers and genetic susceptibility [25–27]. These changes enable dynamic reactions to environmental stimuli by controlling gene expression without changing the underlying DNA sequence. According to new research, cytokine-driven neuroinflammatory pathways may change the development of the fetal brain when the mother's immune system is activated during pregnancy. During crucial developmental windows, these immune-related disturbances are especially worrisome because they may alter synaptic pruning and myelination, which could have long-term neurodevelopmental repercussions [31].Crucial elements of the central nervous system, oligodendrocytes, microglia, and astrocytes aid in the brain's anatomical and functional development. In order to create and preserve brain architecture, these neuroglial cells control basic functions such as immunological homeostasis, synaptogenesis, and neuronal support [32]. Because of their varied functions, neuroglial dysfunction can have a major effect on brain development and connection [33, 34].

As the central nervous system's principal immune effector cells, microglia are essential for preserving immunological homeostasis. Synaptic pruning, a developmental process that improves neuronal networks by removing superfluous or redundant synapses, depends on them [33, 37]. Conversely, oligodendrocytes are necessary to maintain the structural integrity and functionality of axons. They accomplish this by creating myelin sheaths, which promote axon metabolism and enable quick action potential propagation [38].

Higher cognitive and behavioral functions are based on the dynamic processes that shape brain architecture, which are provided by the coordinated actions of these neuroglial cells [32, 39]. Once thought to be only supporting components for neurons, neuroglia are now thought to play a significant role in determining neuronal connections, controlling synaptic dynamics, and preserving homeostatic balance in the central nervous system [40]. A new perspective on glial abnormalities in ASD has been made possible by recent advancements in imaging and molecular biology, which have illuminated their roles in dysregulated synapses, neuroinflammation, and altered brain circuitry. These discoveries contribute to our knowledge of the pathophysiology of ASD and suggest that neuroglia may be interesting targets for new treatment approaches.

NEUROGLIA'S FUNCTION IN NORMAL BRAIN DEVELOPMENT

By delivering glucose and lactate to neurons via specific transport systems, astrocytes maintain metabolic support during crucial stages of brain growth and plasticity [42]. Furthermore, trophic substances such brain-derived neurotrophic factor (BDNF), which is necessary for synaptic maturation, neuronal survival, and differentiation, are released by astrocytes [43]. Through their modulation of calcium signals within astrocytic networks, these cells influence synaptic plasticity and allow for coordinated responses to neuronal activity [35].

Astrocytes

Astrocytes are crucial for preserving homeostasis in the neuronal environment and for promoting healthy brain growth and operation. By controlling neurotransmitter uptake, especially glutamate, and converting it to glutamine to avoid excitotoxicity—a process that can seriously harm neurons because of an excessive buildup of glutamate in

the synaptic cleft—they control synaptic development and function. In the early phases of development, when synaptic overactivity might interfere with network formation, this modulation is essential [35, 36].The blood-brain barrier (BBB), which controls nutrition exchange and shields the brain from dangerous chemicals, is aided by astrocyte endfeet, which encircle blood vessels [44]. Astrocytic dysfunction can result in neurovascular deficiencies and alterations in synaptic connection during development, both of which are connected to neurodevelopmental disorders [35, 36].

Microglia constantly scan the surroundings of the brain. They use phagocytosis to find and remove superfluous synapses in response to molecular cues from neurons and astrocytes [46]. Complement proteins (e.g., C1q and C3) and other signaling pathways carefully control this activity by marking synapses for elimination [47]. By releasing cytokines and growth hormones like insulin-like growth factor-1 (IGF-1) and interleukin-1β (IL-1β), it also promotes brain growth [37]. By eliminating apoptotic cells and cellular detritus, microglia are essential for preserving brain homeostasis. Proper neurodevelopment requires this process [48].

Oligodendrocytes

The main cells in charge of myelination in the central nervous system are oligodendrocytes. Electrical signals travel along neural axons quickly and effectively when proper myelination is present [38]. The maturation and proper operation of neuronal networks depend on the dynamic and strictly controlled process of myelination, which occurs during brain development. In response to neuronal activity, oligodendrocyte precursor cells (OPCs) move in the direction of axons that need myelination. Once there, they develop into fully grown oligodendrocytes and cover axons in concentric myelin sheaths [49].

By shielding axons from metabolic stress, myelination contributes to preserving axonal integrity in addition to accelerating signal transmission [50]. Additionally, oligodendrocytes ensure the long-term survival of axons by transferring lactate to them, which supports metabolism [51].

The development of cognition and motor skills depends on the timing and pattern of myelination. Learning, memory, and behavior deficiencies can arise from disruptions in oligodendrocyte activity or myelination during early development, which can affect neuronal network connectivity [52]. It is becoming more widely acknowledged that myelination supports adaptive changes in brain connections throughout life by modulating neuronal plasticity [38]. Neural circuits can adjust their operation in response to environmental cues, educational experiences, and behavioral demands thanks to activity-dependent myelination [53]. This dynamic process emphasizes how crucial oligodendrocytes are for both early development and preserving cognitive flexibility in later life [54, 55].Widespread white matter integrity abnormalities linked to aberrant myelination and compromised oligodendrocyte function have been found in neuroimaging investigations in ASD [17]. The social and cognitive impairments typical of ASD have been connected to decreased fractional anisotropy in important white matter pathways, such as the corpus callosum and the superior longitudinal fasciculus [56,57]. Additionally, there is experimental evidence that these abnormalities are made worse by genetic variations that affect oligodendrocyte development, such as those involving OLIG2 and CNTNAP2.

Interactions between neurons and glia

Synaptic plasticity, neuronal migration, and general brain maturation are all synchronized by the collaborative relationship between neurons and glial cells [40]. For instance, thrombospondins and hevin are released by astrocytes, which encourage the development of new synapses [35]. By eliminating unnecessary synapses through complimentary system activity, microglia improve these connections [47].

This intricate interaction is further modulated by neural activity, which gives glial cells feedback and guides their activities to high-demand regions. For example, astrocytic support for ion buffering and metabolic exchange or signal oligodendrocytes to enhance myelination in active circuits might be recruited by enhanced neuronal activity [52,60].

During learning and development, adaptive plasticity depends on these feedback systems. Impaired synapse formation, decreased myelination, or increased inflammation are just a few of the cascade impacts that disruptions in neurone–glia interactions can have on brain development. The pathophysiology of several neurodevelopmental disorders, including ASD, is influenced by these deficiencies [61].

FINDINGS

The main conclusions about neuroglial dysfunction in ASD are outlined in this part, with an emphasis on the cellular, molecular, and genetic changes that underlie the disorder.

DYSREGULATION OF NEUROGLIA IN AUTISM SPECTRUM DISORDER

In ASD, neuroglial function dysregulation has a significant impact on brain activity and development. Core characteristics of ASD, such as cognitive impairments, repetitive behaviors, and social communication deficiencies, are influenced by impaired glial cell activity [39]. Reactive astrogliosis, hyperactive microglial pruning, and poor myelination are highlighted in recent studies as important glial abnormalities that contribute to the pathogenesis of ASD [62]. Impaired cognitive and behavioral results result from these glial alterations, which also modify neuronal signaling and network

dynamics [39,62].

Maintaining synaptic homeostasis, supporting neurons, and ensuring overall brain balance all depend on astrocytes [36, 42]. These pathways are disturbed in ASD by astrocytic dysfunction, which adds to the pathophysiological alterations seen in the disorder [36,62].

A cytoskeletal protein that indicates the reactive state of astrocytes is called Glial Fibrillary Acidic Protein (GFAP) [63]. Elevated GFAP levels are found in post-mortem examinations of ASD brains, which is in line with chronic astrogliosis and related neuroinflammation [64]. Because of their altered calcium signaling, reactive astrocytes are unable to react to synaptic action in a proper manner. There is growing evidence that the pathophysiology of ASD may be influenced differently by different reactive astrocyte subtypes, such as A1 (neurotoxic) and A2 (neuroprotective).

These modifications worsen the disruption of neural connections, synaptic plasticity, and network stability in general [65]. By transforming synaptic glutamate into glutamine, astrocytes contribute significantly to the glutamate–glutamine cycle, avoiding excitotoxicity and preserving synaptic homeostasis [66]. Reduced expression of excitatory amino acid transporter 2 (EAAT2), the main astrocytic glutamate transporter, has been linked to abnormalities in glutamate uptake in ASD. Excitotoxic neuronal damage and decreased neural circuit functionality are the results of this decline, which raises extracellular glutamate levels. Core characteristics of ASD, like as repetitive behaviors and impairments in sensory processing, have been linked to this dysregulation of glutamate transmission.

Trocytes' synchronized activity effectively mediates neurovascular coupling. In order to control cerebral blood flow and satisfy metabolic demands, they mediate communication between neurons and blood vessels [42, 44]. Astrocytic dysfunction lowers cerebral blood flow in ASD, especially in areas important for higher order cognitive and social processes [69, 70]. Neural circuit dysregulation is exacerbated by this compromised neurovascular coupling, which jeopardizes metabolic support to neurons [60]. Key regions linked to executive function and social cognition, such as the prefrontal cortex and anterior cingulate cortex, are frequently shown to have hypoperfusion in neuroimaging studies [71]. In addition to astrocytic dysfunction, ASD is associated with significant abnormalities in microglia [33]. Although these cells are essential for immunological surveillance, synaptic pruning, and inflammatory control, they frequently exhibit chronic activation and excessive complement-mediated synapse removal in ASD [33, 45]. ASD neuropathology is characterized by aberrant neural circuit development, excessive synaptic connection, and persistent neuroinflammation, all of which may be influenced by microglial hyperactivity, according to an expanding body of literature [33].

Animal models show that behavioral rigidity and sensory hypersensitivity—trade-offs frequently seen in individuals with ASD—are caused by abnormalities in microglial activation [73]. Further connecting preclinical and clinical findings, it's interesting to note that human postmortem and imaging studies have similarly documented changes in microglial activation patterns in ASD [74].

The myelinating cells of the central nervous system, oligodendrocytes, in addition to astrocytes and microglia, also show notable malfunction in ASD [75]. Effective neural transmission depends on proper oligodendrocyte maturation and myelination; however, research suggests that ASD is linked to delayed oligodendrocyte precursor maturation, poor myelin formation, and decreased white matter integrity [38,49,52].

One notable characteristic seen in the brains of people with ASD is hypomyelination [50,62]. Research shows that oligodendrocyte differentiation and myelination are either delayed or decreased in important brain areas like the prefrontal cortex and corpus callosum [77]. According to longitudinal neuroimaging research, compromised white matter integrity frequently manifests early in ASD and continues to exist throughout development [78]. Our knowledge of how neuroglial failure leads to aberrant brain connectivity has grown as a result of the convergence of neuroimaging and genetic investigations, which reveal a crucial role for defective myelination in ASD. These deficiencies link anatomical abnormalities to the fundamental behavioral characteristics of ASD and correlate with problems in executive function, social behavior, and sensory processing. Additionally, reduced fractional anisotropy (FA), a crucial indicator of white matter integrity that implies microstructural abnormalities in brain connections, is regularly reported in neuroimaging investigations of people with ASD. The corpus callosum, which is essential for interhemispheric communication, and the superior longitudinal fasciculus, which promotes long-range connection between frontal, parietal, and temporal regions, are two key white matter tracts where these changes are most noticeable. Deficits in social communication, executive function, and sensory processing have been connected to reduced FA in these pathways, highlighting the part white matter abnormalities play in the pathogenesis of ASD. These results support the more general theory that abnormal information processing and cognitive function are caused by disturbed brain connection, which is a fundamental characteristic of ASD.

INTEGRATIVE PERSPECTIVES: THE GENETIC FOUNDATION AND DYSREGULATED PATHWAYS OF NEUROGLIAL DYSFUNCTION IN ASD

Neuroglial dysfunction in ASD is shaped by a complex

interaction between genetic and epigenetic variables, persistent neuroinflammation, and dysregulated signaling pathways. These interrelated processes contribute to the defining characteristics of ASD by upsetting synaptic homeostasis, neuronal–glial communication, and general brain connection [27,62,65]. Of them, the wingless-related integration site/β-catenin (Wnt/β-catenin) signaling cascades and the mammalian target of rapamycin (mTOR) signaling route stand out as essential modulators of neurodevelopmental processes that are frequently disrupted in ASD [82,83].The mammalian target of rapamycin (mTOR) pathway is extremely sensitive to genetic and environmental cues that influence early brain development. It is a key regulator of cellular metabolism, homeostasis, and the integration of signals from nutrients, growth factors, energy availability, and synaptic plasticity. Mammalian target of rapamycin complexes 1 and 2 (mTORC1 and mTORC2) are two separate complexes that it uses to carry out its physiological activities. By phosphorylating downstream effectors such S6 kinase (S6K) and 4E-binding protein 1 (4E-BP1), mTORC1 regulates protein synthesis, lipid metabolism, and autophagy, fostering anabolic processes essential for neuronal and glial function.

Parallel to this, the Wnt/β-catenin pathway is crucial for coordinating neurodevelopmental processes such as myelination, glial differentiation, and synapse construction [89,90]. Wnt proteins, a class of secreted glycoproteins, start the pathway when they attach to the cell membrane's Frizzled receptors and low-density lipoprotein receptor-related proteins (LRP5/6). The β-catenin destruction complex, which consists of axin, glycogen synthase kinase-3β (GSK-3β), and adenomatous polyposis coli (APC), is inhibited by this interaction. Consequently, β-catenin builds up in the cytoplasm and moves into the nucleus, where it interacts with transcription factors for T-cell factor/lymphoid enhancer-binding factor (TCF/LEF) to control the expression of genes related to cell survival, differentiation, and proliferation [91,92].

Wnt/β-catenin signaling regulates the equilibrium between precursor cell proliferation and maturation, which is crucial for oligodendrocyte differentiation and myelination in the central nervous system. It ensures the appropriate formation of cortical layers and functional neural networks by influencing axonal guidance and neuronal migration during early development. Furthermore, dendritic arborization depends on Wnt/β-catenin signaling, which controls synaptic stability and flexibility in response to external stimuli. This route also affects astrocyte function because Wnt/β-catenin signaling regulates astrocyte-mediated glutamate uptake, ion homeostasis, and neuronal trophic support. Additionally, microglial activity is regulated by Wnt/β-catenin signaling, which helps maintain synaptic efficiency and circuit refinement, especially in synaptic pruning and neuroinflammatory responses [85,93].

In the adult brain, Wnt/β-catenin signaling is still active beyond its developmental roles, supporting synaptic maintenance, plasticity, and injury-repair pathways. Its importance in both neurodevelopment and continuous neuronal activity is shown by the fact that proper modulation of this route is essential for preserving white matter integrity and overall brain connectivity [94].

According to diffusion tensor imaging (DTI) studies, which show disturbed connectivity in important associative pathways including the corpus callosum and superior longitudinal fasciculus, altered Wnt/β-catenin activity in ASD has been linked to reduced white matter integrity [80]. Astrocytic function is also affected by dysfunctional Wnt/β-catenin signaling, which reduces their capacity to control synapse formation and plasticity while also affecting microglial activation states [95]. ASD's structural and functional abnormalities are further exacerbated by dysregulation of brain progenitor cell fate determination, which has been linked to disturbances in Wnt/β-catenin signaling [96].Chronic neuroinflammation is another intricate pathophysiological component of ASD, in addition to disruption of the major signaling pathways. The pathogenesis of the illness is further complicated by the complex interactions between immunological signaling and neuroglial function [99,100]. An increasing amount of research indicates that abnormal synaptic pruning, decreased neural plasticity, and disturbances in excitatory/inhibitory balance—all of which are characteristics of ASD—are caused by continuous activation of microglia and astrocytes [101]. In addition to impairing neuroglial communication, elevated levels of pro-inflammatory cytokines such IL-6, TNF-α, and IL-1β can worsen synaptic dysfunction by changing glutamatergic and GABAergic transmission, which results in cognitive and behavioral deficits [65,102].Research has indicated that abnormalities in genes such as PTEN, MECP2, and CNTNAP2 impair vital functions like myelination, inflammation control, and synaptic pruning [105]. The intricacy of ASD pathology cannot be entirely explained by these genetic predispositions [106]. There is mounting evidence that implies epigenetic mechanisms fine-tune neuroglial gene expression by acting as a crucial interface between genetic predispositions and environmental influences [107]. The long-term effects of prenatal exposures on neuroglial integrity may be mediated by changes like DNA methylation and histone acetylation, which dynamically control transcription in response to developmental signals. Prenatal exposures are linked to long-term neuroglial impairments through epigenetic mechanisms such DNA methylation and histone acetylation, which alter gene expression in response to environmental stimuli [29]. A crucial epigenetic change that usually results in transcriptional silence by changing chromatin structure and

inhibiting transcription factor binding is DNA methylation, which mostly occurs in CpG islands. On the other hand, by neutralizing the positive charge of histone proteins, histone acetylation relaxes chromatin and promotes gene transcription, increasing the accessibility of regulatory regions. During crucial stages of neurodevelopment, these changes are extremely dynamic and sensitive to outside stimuli, such as maternal stress, nutritional variables, infections, inflammations, and exposure to environmental pollutants. Long-lasting changes in the structure and function of the brain can result from disruptions in these epigenetic processes, which can cause persistent changes in the gene expression involved in synaptic plasticity, glial differentiation, and neuroinflammation. Knowing the intricate relationship between neuroglial integrity and epigenetic regulation offers important insight into how early environmental factors affect neurodevelopmental trajectories.

Thanks to developments in genome-editing techniques, researchers can now precisely examine these genetic contributions and analyze the functional effects of particular epigenetic changes as well as their role in neurodevelopmental processes. Unprecedented control over gene regulation is made possible by methods like base editing and Clustered Regularly Interspaced Short Palindromic Repeats-Cas9 (CRISPR-Cas9), which make it easier to create tailored treatment approaches. Precision medicine approaches for neurological illnesses could be made possible by these techniques, which improve our capacity to modify epigenetic landscapes and have the potential to rectify aberrant gene expression patterns linked to neuroglial dysfunction.

CONVERSATION

The results above explained the complex role of neuroglial dysfunction in ASD, showing how anomalies in oligodendrocytes, microglia, and astrocytes lead to neuroinflammation, altered synaptic connection, and decreased neural communication. These pathophysiological abnormalities imply that ASD is a syndrome in which glial cells play a critical regulatory role in determining brain growth and function, rather than just a disorder of neuronal dysfunction [62]. Numerous therapy modalities have been investigated as a result of the intricate interaction between neuroglial dysfunction and ASD [111]. These therapies focus on several facets of glial disease, such as myelination support, synaptic remodeling, and neuroinflammation [112]. The main biological and pharmacological approaches that have demonstrated promise in modifying neuroglial activity in ASD are covered here.

Minocycline, a tetracycline antibiotic with significant anti-inflammatory and neuroprotective qualities, is another substance that has drawn interest. Apart from its antibacterial properties, minocycline has been shown to inhibit matrix metalloproteinases (MMPs), decrease microglial activation, and alter neuroinflammatory pathways linked to ASD [116].

According to research in animal models, administering minocycline may alleviate social interaction deficits, increase synaptic remodeling, and lower neuroinflammatory markers [119].

However, due to worries about long-term safety, mitochondrial toxicity, and the possibility of antibiotic resistance with prolonged use, its clinical applicability is still unknown despite these encouraging preclinical findings. Cytokine modifying biologics, such as monoclonal antibodies that block pro-inflammatory cytokines including interleukin-6 (IL-6) and tumor necrosis factor-alpha (TNF-α), have been used in more focused strategies to treat neuroinflammation. Since some people with ASD have been found to have elevated levels of these cytokines, it has been suggested that inhibiting their activity could reduce neuroinflammation and enhance behavioral results. Although these treatments have demonstrated significant efficacy in the management of inflammatory and autoimmune diseases, their use in ASD is currently in the experimental phase [114,121]. One of the main worries is that widespread immunosuppression could disrupt vital neurodevelopmental processes, especially in young infants. For this reason, methods that specifically target pathological inflammation while maintaining physiological immune functions must be developed.

Notwithstanding the possible advantages of anti-inflammatory therapies, a number of significant obstacles prevent their broad clinical use. Since not everyone with ASD exhibits the same level of neuroinflammation, this heterogeneity is one of the biggest obstacles [99]. This diversity hinders the creation of standardized therapeutic protocols and makes it challenging to determine which subgroups may benefit the most from anti-inflammatory medications. Furthermore, a number of currently available anti-inflammatory drugs have systemic effects on peripheral immune function, raising the risk of infections, metabolic disorders, and gastrointestinal issues over time [116]. Since the majority of currently available medications influence inflammation broadly without specifically impacting microglial or astrocytic activity, another significant drawback is the lack of selectivity in targeting neuroglial cells.

It has been demonstrated that experimental treatments, like the sushi domain protein SRPX2, selectively disrupt C1q function, preventing complement-mediated synapse loss and maintaining synaptic homeostasis [126]. Anti-inflammatory modulatory treatments and microbiota-targeted interventions are complementary approaches that converge on shared pathophysiological pathways since immune dysregulation and gut microbiota imbalance are closely related in ASD. By changing neuroimmune interactions, gut-brain axis signaling,

and metabolic pathways, gut microbiota modification affects ASD [127]. ASD-related behavioral and cognitive symptoms can be exacerbated by changes in intestinal permeability, inflammation, and neurotransmitter levels caused by changes in microbiota composition. Through a variety of therapies, therapeutic approaches focusing on gut microbiota in ASD seek to enhance neurobehavioral outcomes and restore microbial balance. Beneficial bacterial strains like Lactobacillus and Bifidobacterium are introduced by probiotics and may improve gut integrity, lower inflammation, and alter the generation of neurotransmitters. Prebiotics, such as dietary fibers and oligosaccharides, promote the development of good bacteria and have an impact on the synthesis of short-chain fatty acids, which has neuroprotective properties.

By directly restoring a more varied and healthy gut microbiome, fecal microbiota transplantation (FMT), however still in the experimental stage, has demonstrated encouraging outcomes in reducing gastrointestinal and behavioral problems. 6.3. Treatments Based on Cannabinoids Cannabidiol (CBD), a non-psychoactive cannabinoid produced from the Cannabis sativa plant, has gained great interest for its possible neuroprotective and anti inflammatorypropertiesinASD. CB1 and CB2 receptors, which are extensively expressed in both neurons and glial cells, are the main way that CBD interacts with the endocannabinoid system. CBD has been demonstrated to influence the excitatory–inhibitory balance, control neurotransmitter release, and lessen neuroinflammation via affecting these pathways—all of which are frequently disturbed in ASD.

Numerous clinical research have investigated how CBD affects behavioral symptoms associated with ASD; some have found that it improves social interaction, decreases repetitive behaviors, and eases anxiety and agitation [131]. Furthermore, early findings indicate that CBD may reduce excessive microglial activation, which would lessen chronic neuroinflammation, which is a contributing factor to synaptic dysfunction. Children with ASD who received CBD-enriched treatments in a recent study showed improved social responsiveness and a reduction in the frequency of self-stimulatory behaviors, confirming the idea that cannabinoids may have positive effects on the neural circuits linked to the symptoms of ASD. remains constrained by a number of outstanding issues. The absence of extensive randomized controlled trials to determine conclusive efficacy and safety profiles is one of the main issues. Although small-scale research indicates some advantages, data interpretation is made more difficult by variations in dosage, purity, and composition. Furthermore, it is still unknown how long-term CBD administration in juvenile populations would affect neurodevelopmental processes in particular. The ideal dosage schedule presents another difficulty since CBD has a biphasic impact, meaning that distinct physiological reactions can be elicited by low and high dosages [133]. The absence of consistent guidelines for cell preparation, delivery, and dosage is one of the main challenges in converting MSC therapy into a standardized treatment for ASD.

Different MSC sources, growth strategies, and administration systems may produce different therapeutic results. Moreover, although MSCs are widely regarded as safe, little is known about the long-term consequences of stem cell transplantation, which raises questions regarding possible tumorigenicity, immunological rejection, and unintentional differentiation into non-neuronal cell types. The development of biomarkers that can predict treatment responsiveness is also necessary because it is uncertain which patient subgroups may benefit the most from stem cell therapy due to the heterogeneity of ASD. creation of biomarkers capable of forecasting response to treatment [137]. In addition to MSCs, glial progenitor cell transplantation has shown promise as a therapy option for ASD, especially when it comes to addressing the myelination deficiencies and white matter abnormalities frequently seen in those with the disorder. Restoring damaged neural connections is made easier by glial progenitor cells' ability to develop into functional oligodendrocytes, the central nervous system's myelinating cells. According to preliminary research in animal models, glial progenitor cell transplantation may improve behavioral outcomes, encourage brain healing, and increase axonal conduction velocity. Although early human research has indicated some advantages, glial progenitor cell therapy has similar difficulties to MSC transplantation, such as the requirement for long-term safety monitoring and optimal delivery techniques.

New avenues for precise genetic modifications to address mutations linked to neuroglial dysfunction in ASD have been made possible by advances in gene editing [140]. The creation of CRISPR-Cas9 technology has transformed molecular medicine by making it possible to precisely alter particular genetic sequences with never-before-seen precision. This approach enables the correction of harmful mutations, the introduction of protective genetic variants, or the control of gene expression in impacted brain circuits. It does this by using a guide RNA to target the Cas9 endonuclease to a specific DNA area.

Preclinical research has shown that by repairing abnormalities in genes related to neural connections, excitatory inhibitory communication, and glial homeostasis, CRISPR-based therapies can restore synaptic balance. Long-term clinical trials are also required to evaluate the long-term neurodevelopmental effects of newly developed neuroglial-targeted treatments. Although a number of experimental treatments have demonstrated promise in preclinical animals, little is known about their long-term safety and effectiveness. Future research should use thorough longitudinal designs to assess long-term cognitive, behavioral, and functional

outcomes in people with ASD in addition to acute symptom alleviation [151].

The development of human-based models that more accurately depict neuroglial interactions in the pathophysiology of ASD is another essential step.

In order to address the complex and multifaceted character of ASD, multimodal therapy approaches has to be investigated concurrently. In order to improve therapeutic success, future interventions should incorporate anti-inflammatory, neurotrophic, and synaptic-modulating strategies. Pharmacological drugs, immunotherapies, and neuromodulation techniques may also be combined. Combinatorial approaches could be especially helpful in adjusting interventions to the unique glial dysfunction patterns found in various subgroups of ASD [144].

Lastly, it is critical to create ethical and regulatory frameworks that direct the responsible application of stem cell-based and gene-editing therapies in ASD as the science advances toward more sophisticated and customized treatments.

By methodically tackling these issues, the discipline is progressing toward a more accurate and successful neuroglial-targeted therapeutic environment, providing individuals with ASD with fresh hope through innovative therapy approaches that are clinically applicable, morally sound, and scientifically rigorous.

FINAL THOUGHTS

New opportunities for therapeutic intervention have been made possible by the increasing understanding that neuroglial dysfunction is a crucial aspect of the pathophysiology of ASD. The intricate nature of ASD and the requirement for interdisciplinary treatment approaches are highlighted by the interaction of astrocytes, microglia, and oligodendrocytes in determining brain connections, synaptic plasticity, and neuroimmune control. Even while recent studies have shown a number of potential molecular targets, including as the mTOR, Wnt/β-catenin, and NF-κB pathways, much effort needs to be done to improve these strategies in order to guarantee long-term efficacy, safety, and specificity [96,97].

Precision medicine's introduction into the field of ASD signifies a fundamental change in the way we diagnose and treat the condition. Beyond the one-size-fits-all approach, the combination of biomarkers, neuroimaging technologies, and AI-driven analytics may allow for the customization of interventions for specific patients. There is potential for focused therapies that address the scientific mechanisms underlying ASD rather than just its symptoms because to developments in stem cell treatment, gene editing, and pharmacological manipulation of neuroglial activity [134,139]. However, overcoming ethical, practical, and scientific obstacles will be necessary to translate these advancements into broad clinical application. To close the gap between bench-side discoveries and practical implementations, cooperation between researchers, physicians, business executives, and legislators will be crucial. ensuring that emerging therapies are accessible to everybody.

N.I. and A.S. contributed the following

ideation, methodology, literature search, original manuscript draft writing, illustration preparation, editing, review, and manuscript finalization. The published version of the manuscript has been read and approved by all authors.

Funding

No outside funding was obtained for this study.

Statement from the Institutional Review Board

Not relevant.

Statement of Informed Consent

Every participant in the study gave their informed consent.

Data Availability Statement

The article includes the data.

No conflicts of interest have been disclosed by the writers.

REFERENCES

1. Fombonne, E. Epidemiology of pervasive developmental disorders. Pediatr. Res. 2009.

2. Elsabbagh, M.; Divan, G.; Koh, Y.J.; Kim, Y.S.; Kauchali, S.; Marcín, C.; Montiel-Nava, C.; Patel, V.; Paula, C.S.; Wang, C.; et al. Global prevalence of autism and other pervasive developmental disorders. Autism Res. 2012.

3. Lai, M.C.; Lombardo, M.V.; Auyeung, B.; Chakrabarti, B.; Baron-Cohen, S. Sex/gender differences and autism: Setting the scene for future research. J. Am. Acad. Child Adolesc. Psychiatry 2015.

4. Hull, L.; Petrides, K.V.; Allison, C.; Smith, P.; Baron-Cohen, S.; Lai, M.C.; Mandy, W. "Putting on My Best Normal": Social camouflaging in adults with autism spectrum conditions. J. Autism Dev. Disord. 2017.

5. Masi, A.; DeMayo, M.M.; Glozier, N.; Guastella, A.J. An overview of autism spectrum disorder, heterogeneity, and treatment options. Neurosci. Bull. 2017.

6. Simonoff, E.; Pickles, A.; Charman, T.; Chandler, S.; Loucas, T.; Baird, G. Psychiatric disorders in children with autism spectrum disorders: Prevalence, comorbidity, and associated factors in a population-derived sample. J. Am. Acad. Child Adolesc. Psychiatry 2008.

7. Khachadourian, V.; Mahjani, B.; Sandin, S.; Kolevzon,

A.; Buxbaum, J.D.; Reichenberg, A.; Janecka, M. Comorbidities in autism spectrum disorder and their etiologies. Transl. Psychiatry 2023.

8. Waizbard-Bartov, E.; Fein, D.; Lord, C.; Amaral, D.G. Autism severity and its relationship to disability. Autism Res. 2023.

9. Webb, S.J.; Jones, E.J. Early identification of autism: Early characteristics, onset of symptoms, and diagnostic stability. Infants Young Child. 2009.

10. Ben-Sasson, A.; Hen, L.; Fluss, R.; Cermak, S.A.; Engel-Yeger, B.; Gal, E. A meta-analysis of sensory modulation symptoms in individuals with autism spectrum disorders. J. Autism Dev. Disord. 2009.

11. Rogers, S.J.; Ozonoff, S. Annotation: What do we know about sensory dysfunction in autism? A critical review of the empirical evidence. J. Child Psychol. Psychiatry 2005.

12. Gotham, K.; Pickles, A.; Lord, C. Trajectories of autism severity in children using standardized ADOS scores. Pediatrics 2012.

13. AmericanPsychologicalAssociation. Diagnostic and Statistical Manual of Mental Disorders (DSM-5), 5th ed.; American Psychological Association: Washington, DC, USA, 2013.

14. Johnson, C.P.; Myers, S.M. Identification and evaluation of children with autism spectrum disorders. Pediatrics 2007.

15. Rutter, M.; Le Couteur, A.; Lord, C. Autism Diagnostic Interview-Revised (ADI-R); Western Psychological Services: Los Angeles, CA, USA, 2003.

16. Barton, K.S.; Tabor, H.K.; Starks, H.; Garrison, N.A.; Laurino, M.; Burke, W. Pathways from Autism Spectrum Disorder Diagnosis to Genetic Testing. Genet. Med. 2018

17. Ecker, C.; Bookheimer, S.Y.; Murphy, D.G. Neuroimaging in autism spectrum disorder: Brain structure and function across the lifespan. Lancet Neurol. 2015.

18. Amaral, D.G.; Schumann, C.M.; Nordahl, C.W. Neuroanatomy of autism. Trends Neurosci. 2008.

19. Geschwind, D.H. Genetics of autism spectrum disorders. Trends Cogn. Sci. 2011.

20. Salcedo-Arellano, M.J.; Hagerman, R.J.; Martínez-Cerdeño, V. Fragile X syndrome: Clinical presentation, pathology and treatment. Gac. Méd. Méx. 2020.

21. Martin, C.L.; Ledbetter, D.H. Chromosomal Microarray Testing for Children With Unexplained Neurodevelopmental Disorders. JAMA2017.

22. Sandal, S.; Verma, I.C.; Mahay, S.B.; Dubey, S.; Sabharwal, R.K.; Kulshrestha, S.; Saxena, R.; Suman, P.; Kumar, P.; Puri, R.D. Next-Generation Sequencing in Unexplained Intellectual Disability. Indian J. Pediatr. 2024.

23. Colvert, E.; Tick, B.; McEwen, F.; Stewart, C.; Curran, S.R.; Woodhouse, E.; Gillan, N.; Hallett, V.; Lietz, S.; Garnett, T.; et al. Heritability of autism spectrum disorder in a UK population-based twin sample. JAMA Psychiatry 2015.

24. Sandin, S.; Schendel, D.; Magnusson, P.; Hultman, C.; Surén, P.; Susser, E.; Grønborg, T.; Gissler, M.; Gunnes, N.; Gross, R.; et al. Autism risk associated with parental age and with increasing difference in age between the parents. Mol. Psychiatry 2016.

25. Christensen, J.; Grønborg, T.K.; Sørensen, M.J.; Schendel, D.; Parner, E.T.; Pedersen, L.H.; Vestergaard, M. Prenatal valproate exposure and risk of autism spectrum disorders and childhood autism. JAMA 2013.