Harnessing MSC-derived exosomes to modulate the pathophysiology of ASD: Recent advances and therapeutic implications (Review)

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Received July 24, 2025; Accepted October 31, 2025

DOI: 10.3892/ijmm.2026.5731

Abstract. Autism spectrum disorder (ASD) is a complex neurodevelopmental condition characterized by marked genetic heterogeneity and diverse environmental influences. Current treatment approaches focus on symptom management, with only a limited number of effective interventions targeting the underlying causes. Recently, mesenchymal stem cells (MSCs) and their derived exosomes (MSC-Exos) have emerged as promising candidates for ASD therapy owing to their potent immunomodulatory, neuroprotective and targeted delivery properties. The present review discusses the functions of MSC-Exos and their potential use in ASD. MSC-Exos improve neuroinflammation, enhance synaptic plasticity and restore neural network function by delivering bioactive molecules. Moreover, MSC-Exos exhibit a low immunogenicity, a favorable safety profile and scalability for clinical production. Despite promising results however, clinical trials continue to face challenges, particularly in standardizing the isolation, characterization, dosing and administration routes of exosomes. In addition, significant challenges persist in production processes, quality control and the elucidation of the mechanisms of action. In conclusion, MSC-Exos represent a groundbreaking, cell-free therapeutic strategy with substantial potential to target the core pathophysiology of ASD. In the future, multicenter randomized controlled trials and interdisciplinary collaborations will be crucial for translating preclinical findings into the development of effective and transformative therapies for ASD.

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Abbreviations: ASD, autism spectrum disorder; CNV, copy number variation; MSC, mesenchymal stem cell; MSC-Exos, mesenchymal stem cell-derived exosomes; NF-κB, nuclear factor κ-light-chain-enhancer of activated B cells; PSD, postsynaptic density; PI3K, phosphatidylinositol 3 kinase; Akt, protein kinase B; RR, relative risk; TGF-β, transforming growth factor β; BMP, bone morphogenetic protein; VPA, valproic acid

Key words: autism spectrum disorder, mesenchymal stem cell-derived exosomes, precision medicine, clinical translation

Contents

1. The concept of autism spectrum disorder and recent progress

Core symptoms and current epidemiological status. Autism spectrum disorder (ASD) encompasses a group of pervasive neurodevelopmental conditions characterized by a broad spectrum of symptoms, broadly categorized into core and associated symptoms. Core symptoms include persistent deficits in social interaction and communication, as well as restricted and repetitive patterns of behavior and interests that significantly impair daily functioning. By contrast, associated symptoms encompass attention deficits, mood disturbances, sensory processing abnormalities, sleep disorders, language impairment, anxiety, epilepsy, mania and self-injurious behaviors. Core symptoms typically emerge in early childhood and

persist throughout life (1-3). In recent years, increasing public awareness of ASD has coincided with a steady increase in its rate of diagnosis, rendering it a significant global public health concern. According to 2022 data from the US Centers for Disease Control and Prevention (CDC), the incidence of ASD in the USA reached ~1 in 31 children. Moreover, the prevalence of ASD among boys is notably higher than that among girls, with a male-to-female ratio of ~3.44:1 (4). Furthermore, advances in early screening and diagnostic tools have enabled the identification of a greater number of individuals with milder ASD phenotypes, thereby enhancing epidemiological research in this field.

Large-scale genome-wide association studies (GWAS) (5) and family-based genetic analyses (6) have revealed that ASD possesses a complex and highly heterogeneous genetic background. Genetic factors are estimated to account for ~81% of the risk of developing ASD, whereas environmental factors contribute to a ~14-22% risk. Of note, >100 ASD-associated risk genes have been identified, including neurexin 1 (*NRXN1)*, Src homology 3 and multiple ankyrin repeat domains 2/3 (*SHANK2, SHANK3*) and chromodomain helicase DNA binding protein 8 (*CHD8*). The principal mutation types include *de novo* truncating mutations, missense mutations and copy number variations (CNVs). These risk-associated genes influence gene expression, neurogenesis, synaptic function and chromatin regulatory networks (6-8).

By contrast, environmental factors primarily influence early fetal development through maternal health conditions and external exposures. A previous large-scale study involving 22,156 cases of ASD across three Nordic countries underscored the crucial role of perinatal and prenatal factors. Maternal gestational hypertension (odds ratio, 1.4), preeclampsia [relative risk (RR), 1.3], overweight or obesity (RR, 1.3) and an advanced maternal age (≥35 years; RR, 1.3) were all shown to be significantly associated with an increased risk of offspring developing ASD. Moreover, an advanced paternal age (each 10-year increase corresponding to a 21% rise in the risk of ASD), interpregnancy intervals that are either too short (<12 months) or too long (≥72 months) and exposure to specific medications, such as valproic acid (VPA) during pregnancy, were also closely linked to elevated risk of developing ASD (5). A previous study demonstrated that the absolute risk of developing ASD reached 4.4% in the VPA-exposed group as compared to the unexposed group with 1.5%. Thus, beyond genetic predisposition, aberrations in the intrauterine environment and maternal metabolic dysregulation may play a critical role in disrupting fetal neurodevelopment and represent important triggers for ASD (9).

Current treatment strategies and key challenges. Early diagnosis and intervention for ASD are deemed pivotal for improving the quality of life of affected children. Such early interventions can enhance language abilities, social communication skills and behavioral regulation, particularly when administered during the rapid phase of neurodevelopment (<3 years of age). This timing is critical for optimizing long-term outcomes, helping to maximize the cognitive, linguistic and social capacities of children (8,10). The American Academy of Pediatrics recommends autism screening at 18 and 24 months of age (https://www.aap.org/en/patient-care/autism). Currently, the primary diagnostic tools include the Modified Checklist for Autism in Toddlers Revised (M-CHAT-R) (11), the Autism Diagnostic Observation Schedule, Second Edition (ADOS-2) and the Autism Diagnostic Interview-Revised (ADI-R) (12). Children who screen positive require comprehensive evaluations, including behavioral observation and an in-depth analysis of developmental history (3,8). However, as the symptoms of ASD are heterogeneous and vary greatly among individuals, early diagnosis remains challenging, particularly in cases with atypical or mild presentations.

At present, treatment for ASD primarily focuses on behavioral interventions and symptom management, as there is no universally accepted cure (3). The behavioral intervention remains the first-line approach, commonly implemented through Early Intensive Behavioral Intervention (EIBI) and Naturalistic Developmental Behavioral Interventions (NDBI) (13).

EIBI is grounded in the principles of Applied Behavior Analysis (ABA) and is typically designed for children <5 years of age. It involves intensive therapy, usually at least 25 h per week over a minimum period of 1 year, to strengthen social, language, cognitive and adaptive behaviors. Among NDBI, the Early Start Denver Model (ESDM) (14) is the most representative approach. It integrates multiple developmental domains, including cognition, language, social interaction, motor skills and imitation, into behavioral interventions. ESDM emphasizes creating meaningful activities for children, fostering interactive relationships and emotional engagement, and promoting the generalization of learning. Motivating activities are incorporated into daily routines to help children acquire new skills, improve language, social and cognitive functioning, and reduce disruptive behaviors (15). These behavioral strategies currently constitute the cornerstone of ASD therapy.

Pharmacological interventions are mainly used to address comorbid symptoms, such as risperidone and aripiprazole, which effectively reduce irritability and aggressive behaviors (16). Methylphenidate, atomoxetine and guanfacine may alleviate hyperactivity and attention deficits. For sleep disturbances, melatonin is commonly prescribed (8).

These interventions mainly aim to alleviate symptoms and minimize the effect on daily life and quality of life, rather than addressing the underlying causes of ASD. To date, to the best of our knowledge, no medication has been shown to effectively treat the core symptoms of ASD, and treatment outcomes vary widely among individuals. Consequently, there is an urgent need to explore novel treatment strategies, particularly those targeting the underlying pathophysiological mechanisms, which have become a central focus of contemporary ASD research.

Future directions for early diagnosis and intervention. The future of early ASD diagnosis and intervention hinges on precision, personalization and enhanced accessibility. First, personalized intervention plans are crucial for addressing the heterogeneity of ASD (17). By integrating assessments of the behavioral, cognitive and language abilities of a child, along with data-driven analytical methods, more tailored interventions can be designed to meet individual needs (18). In addition, using biomarkers (e.g., miRNAs or exosomal components) to aid in diagnosis and treatment offers a potential

avenue for refining interventions (19), such as identifying neuroinflammatory features, which may enable more targeted immunomodulatory therapies (20).

The integration of emerging technologies presents new opportunities for the treatment of ASD. For instance, mesenchymal stem cells (MSCs) exhibit tremendous promise owing to their multilineage differentiation potential, immunomodulatory properties, and capacity to secrete various neuroprotective factors (21,22). Beyond enhancing the neuroinflammatory microenvironment and supporting neuroplasticity *in vivo*, MSCs also release exosomes that transport essential proteins, nucleic acids, and miRNAs to repair damaged neurons and glial cells (23). These molecules, in turn, modulate gene expression and cell function, mitigate neuroinflammation and repair synaptic structures (24). Exosomes possess low immunogenicity, the ability to cross the blood-brain barrier, and the capacity for targeted delivery of specific bioactive molecules (25,26). A number of studies have focused on the use of MSC-derived exosomes (MSC-Exos) in ASD interventions, exploring their potential mechanisms and translational applications for improving social, communication and cognitive functions (22,27-29).

Moreover, developing novel evaluation metrics is crucial for assessing the efficacy of interventions. Researchers can more comprehensively track the progress of a child in detection and intervention by integrating behavioral data, neuroimaging findings (30,31) and biomarker changes (32). Long-term follow-up assessments further elucidate how specific strategies affect social adaptation and overall quality of life in adulthood (33).

Lastly, enhancing the social awareness of ASD and strengthening policy support are also required. Public education campaigns can help reduce societal bias, and training programs for parents and educators can improve the effectiveness of early diagnosis and intervention (34). In this regard, additional funding from government research agencies and related organizations can expand the reach of innovative research and healthcare services for individuals with ASD.

Overall, the advancements in ASD diagnosis and intervention hinge on technological progress, cross-disciplinary collaboration, and strategic resource allocation. By harmonizing scientific breakthroughs with robust policy support, current treatment barriers can be addressed and this may help foster renewed optimism for children with ASD and their families.

2. Molecular genetics and neuropathology of ASD

With deepening insight into ASD, research on its etiology and pathogenesis has expanded from early, straightforward genetic associations to a comprehensive framework encompassing molecular pathology, neural network disruptions and interdisciplinary approaches. In recent years, emerging technologies, such as high-throughput single-cell sequencing (35) and brain organoid models (36), have rapidly evolved, enabling a more in-depth analysis of gene-environment interactions and neurodevelopmental abnormalities in ASD. These advances also provide novel approaches to potential interventions and therapies. Against this backdrop, the following sections provide a systematic review of current research on ASD, focusing on genetic landscapes, synaptic imbalances, inflammatory regulation, glial cell activation and immune dysregulation (37).

Genetic characteristics and polygenic complexity of ASD. Epidemiological studies in recent years have underscored the dominant role of genetic factors in ASD, which is recognized as a complex disorder driven largely by heritability. Multiple twin studies consistently demonstrate much higher concordance for ASD in monozygotic (MZ) than dizygotic (DZ) twins. In a population-based study using standardized ADI-R/ADOS assessments, probandwise concordance ranged from ~58-77% for MZ twins and ~21-36% for DZ twins across strict-autism and broader ASD definitions; rare sex-specific DZ subgroups can be lower (38). A previous meta-analysis further indicates near-perfect MZ correlations (~98%) with DZ correlations that vary by the assumed prevalence threshold, yielding substantial heritability (~64-91%) and showing that estimates of the shared environmental component are sensitive to modelled prevalence (39). Through GWAS and familial genetic analyses comparing large-scale datasets of individuals with ASD and healthy controls, numerous ASD-associated susceptibility genes and variations have been identified, including *CHD8*, *NRXN1*, *SHANK2*, *SHANK3* and contactin associated protein 2 (*CNTNAP2*) (6-8,40). Among these genes, *CHD8* encodes a chromatin remodeler that regulates a wide array of neurodevelopmental genes by binding to their promoters and enhancers. Previous studies have reported that *CHD8* haploinsufficiency results in dysregulated transcriptional programs, aberrant neurogenesis, and epigenetic abnormalities (41). *NRXN1* encodes the presynaptic adhesion molecule neurexin-1, which interacts with postsynaptic ligands such as neuroligins to form trans-synaptic connections and is essential for synapse formation and homeostasis. In ASD mouse models carrying *NRXN1* mutations, CNVs have been shown to cause synaptic connectivity defects, impairments in neural circuit development and synaptic transmission, ultimately leading to deficits in social interaction and communication (42). The *SHANK* gene family, particularly *SHANK3*, functions as a core scaffolding and signaling component of the postsynaptic density (PSD) at excitatory synapses, orchestrating the organization of α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid and N-methyl-D-aspartate receptors, cytoskeletal structures, and intracellular signaling pathways. *SHANK* deficiency leads to PSD disassembly, impaired synaptic plasticity, altered dendritic spine morphology and excitatory/inhibitory imbalance, thereby contributing to ASD-related behavioral abnormalities. Both animal models and patient-derived induced pluripotent stem cells studies have provided strong evidence supporting these mechanisms (43,44). The *CNTNAP2*, a member of the neurexin superfamily, is involved in axon-myelin interactions, synaptic function and neuronal positioning. Early candidate gene studies identified *CNTNAP2* as being associated with ASD, language impairment and epilepsy. The loss or mutation of *CNTNAP2* has been linked to abnormal neuronal migration during development, defects in axonal and myelin function, and disrupted excitatory neuronal activity (45).

The analysis of the Simons Simplex Collection (SSC) has identified six key risk loci (1q21.1, 3q29, 7q11.23, 16p11.2, 15q11.2-13 and 22q11.2). These large CNVs likely harbor multiple risk genes of moderate effect, primarily involving upstream regulators of gene expression during neural development, as well as synapse-related genes that directly impact neuronal structure and function (6).

Exome sequencing data, when integrated with functional annotations of different types of genetic variants (e.g., rare single-nucleotide variants, insertions/deletions and CNVs) and selection-pressure metrics, e.g., Loss of Function Observed and Expected Upper Bound Fraction (LOEUF) scores, indicate that the risk of developing ASD is strongly associated with rare coding variants and *de novo* mutations. Among the genes significantly linked to ASD (false discovery rate ≤0.001), *de novo* protein-truncating variants, damaging missense variants and CNVs account for 57.5, 21.1 and 8.44% of the genetic risk, respectively. Notably, CNVs carry a markedly high relative risk since they alter the copy number of large genomic segments, potentially affecting multiple genes and leading to cumulative effects (7).

CNVs frequently occur in gene-dense, functionally important regions critical for neurodevelopment, synaptic plasticity and chromatin regulation. Abnormalities in these regions may lead to extensive functional disruption (46). In addition, CNVs can alter gene dosage (e.g., an increase or decrease in gene copies), and dosage-sensitive genes can exhibit significant biological effects when copy numbers deviate from the normal (47,48). A number of CNVs also exhibit pleiotropy, where a single gene may influence multiple biological processes or phenotypes, so its dysregulation can affect various systems concurrently (49). Moreover, CNVs can disrupt regulatory elements (e.g., enhancers and promoters) in non-coding regions, thereby altering gene expression and regulatory networks (48).

The Genes associated with ASD exhibit a higher expression in mature neurons, aligning with their roles in neuronal maturation and synaptic connectivity. By contrast, genes linked to developmental delay are predominantly active in immature neurons and progenitor cells. This suggests that different gene networks affect the nervous system at specific developmental stages (7).

Core neuropathological mechanisms of ASD. A primary pathological hallmark of ASD is abnormal synaptic development and plasticity, primarily manifested as disruptions in synapse formation and pruning. This leads to excessive or insufficient synaptic connections, compromising the precise establishment of neuronal networks (50-52). The excitatory and inhibitory ratio, a core mechanism in synaptic signaling, is critically disrupted in a number of cases, typically manifesting as excessive excitatory activity [e.g., elevated glutamate receptor function (53)] or diminished inhibitory signaling (e.g., reduced gamma-aminobutyric acid receptor levels), both of which may lead to neuronal circuit instability, driving hyperexcitability or network dysregulation (54). Either scenario can result in hyperactive or imbalanced neuronal networks. Furthermore, mutations in ASD-associated genes, such as neuroligin 4 (*NLGN4*) (55) and *SHANK3* (56) can impair presynaptic neurotransmitter release and postsynaptic receptor responsiveness, thereby reducing synaptic transmission efficiency. Abnormalities in synaptic plasticity processes, including long-term potentiation (LTP) and long-term depression, further exacerbate these dysfunctions, as reflected by impaired modulation of synaptic strength in key brain regions, such as the hippocampus, prefrontal cortex and amygdala (57).

Additionally, synaptic protein misfolding and the activation of endoplasmic reticulum stress can lead not only to a loss of synaptic function, but also to heightened neuroinflammation via the unfolded protein response, thereby aggravating ASD pathophysiology (58). Collectively, these disruptions in synaptic development and plasticity underlie prominent deficits in social interaction, cognitive function, and behavioral patterns in ASD, providing essential clues for understanding its molecular mechanisms and for developing potential therapeutic approaches (59).

Developmental abnormalities in specific brain regions are a crucial aspect of the neurobiological features of ASD. Research indicates that both structure and function are significantly affected in the prefrontal cortex (60), hippocampus (61) and basal ganglia (62). Alterations in causal connectivity within the prefrontal cortex are closely linked to deficits in social behaviors and cognitive functions, typified by an excitatory/inhibitory signaling imbalance and diminished synaptic plasticity (60). In the central region of the hippocampus, which is critical for learning and memory, reduced synaptic density and impaired LTP are commonly observed in ASD and directly correlate with decreases in spatial memory (61). Moreover, abnormalities in basal ganglia circuitry, particularly those involved in gating motor output, have been implicated in repetitive and stereotypic behaviors; changes in synaptic transmission within these pathways may affect behavior by modulating motor control and reward mechanisms (62).

Such developmental anomalies manifest at the micro-level of neuronal connections and alter interregional network communication, thereby contributing to the diverse and complex behavioral phenotypes of ASD. In-depth research into these brain regions' developmental perturbations is instrumental in elucidating the pathological mechanisms of ASD and lays the groundwork for precision-targeted therapies.

Cytological basis of neuroinflammation in ASD. Neuroinflammation and aberrant immune regulation play pivotal roles in the pathogenesis of ASD, with the activation of microglia and astrocytes constituting a core cellular foundation of this neuroinflammatory process. Microglia and astrocytes exhibit distinct polarization states in the neuroinflammatory microenvironment of ASD, with M1/A1 subtypes promoting injury and M2/A2 subtypes supporting repair. The interplay of these brain cells is summarized below and illustrated in Fig. 1.

i) Microglia: Dual polarization and synaptic phagocytosis. Microglia are the primary immune cells of the central nervous system (CNS) and exhibit dual polarization states, i.e., M1 (classical activation) and M2 (alternative activation). In response to inflammatory stimuli, M1-type microglia secrete proinflammatory factors, such as tumor necrosis factor-α (TNF-α), interleukin (IL)-1β and IL-6 (63-65) and reactive oxygen species (66), driving neuroinflammation and pathological damage. They can also enhance phagocytic activity, causing direct harm to neurons and synapses (67). Conversely, M2-type microglia are induced by anti-inflammatory factors, such as IL-4 and IL-13 (68), and promote the release of neuroprotective mediators, including IL-10 and transforming growth factor-β (TGF-β) (68-70). This balance between M1 and M2 polarization is crucial for regulating neuronal activity, synaptic plasticity, and the clearance of

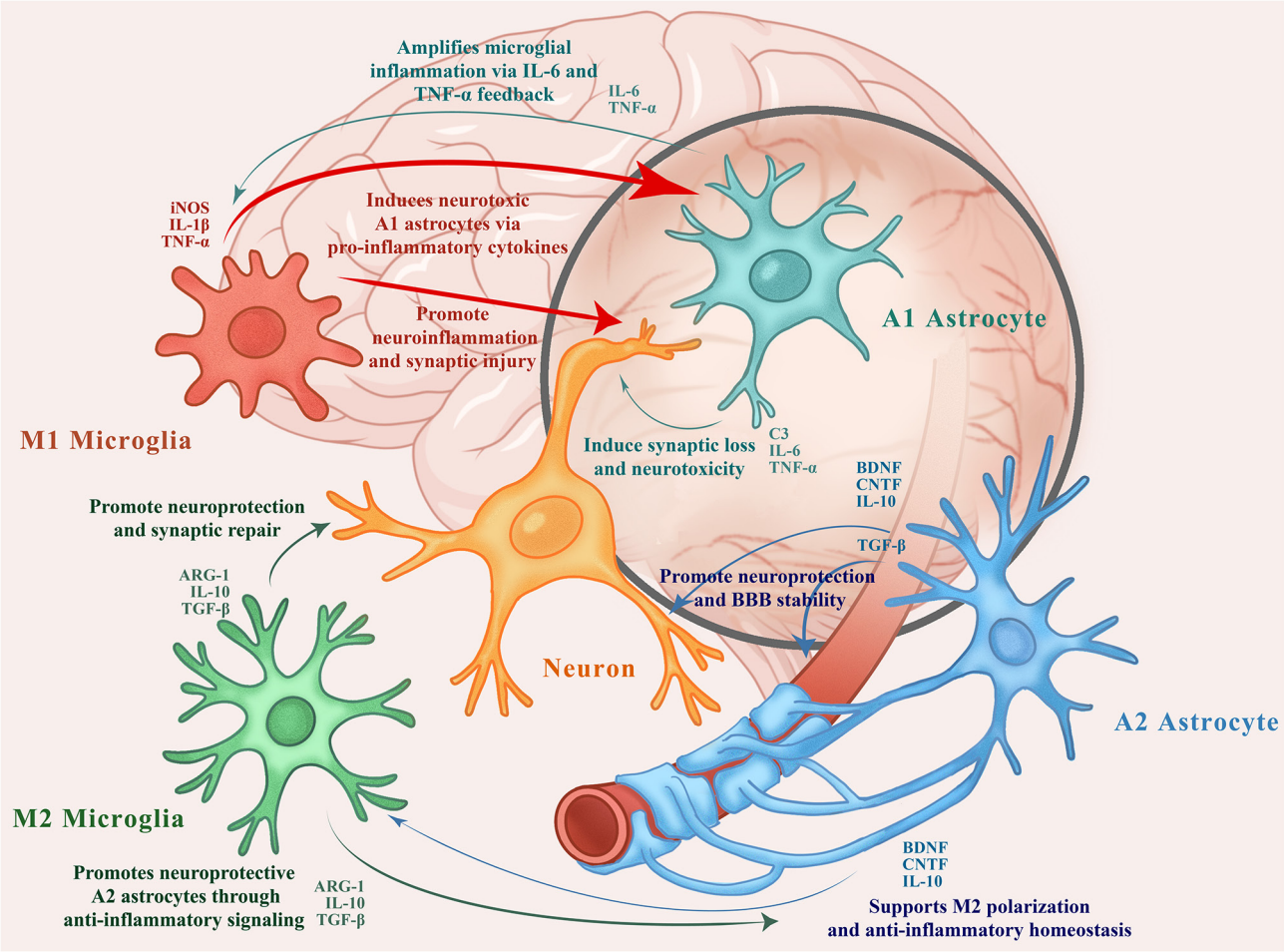

Figure 1. Interactions between microglia, astrocytes, and neurons in the autism spectrum disorder-affected brain. M1 microglia and A1 astrocytes release pro-inflammatory cytokines (e.g., TNF-α, IL-1β and IL-6) that amplify neuroinflammation and induce synaptic damage. In contrast, M2 microglia and A2 astrocytes secrete anti-inflammatory and neurotrophic factors (e.g., IL-10, TGF-β, BDNF and CNTF), promoting synaptic repair, BBB stability and homeostasis. Arrows indicate intercellular regulatory pathways. The authors created this figure referencing multiple sources (40-56,62-68,74-82). TNF-α, tumor necrosis factor-α; IL, interleukin; TGF-β, transforming growth factor β; BMP, bone morphogenetic protein; CNTF, ciliary neurotrophic factor; iNOS, inducible nitric oxide synthase; BBB, blood brain barrier.

apoptotic cells, thus helping maintain CNS homeostasis under normal conditions.

Microglial synaptic phagocytosis plays a critical role in shaping neural circuits; however, its dysregulation can occur in either direction, either through excessive elimination of synapses or insufficient pruning. This bidirectional imbalance contributes to abnormal neural connectivity, underscoring the complex and multifaceted role of microglia in the pathophysiology of ASD. During normal development, microglia refine neural circuits by pruning superfluous synapses. However, the hyperactivation of microglia in ASD may lead to aberrant over-pruning of synapses, thereby reducing synaptic density and impairing neural circuits, particularly in brain regions linked to social behavior and cognition. This phenomenon may underlie the synaptic deficits observed in specific ASD subtypes (71). By contrast, a dysregulated microglial function can result in insufficient synaptic pruning, leading to excessive synaptic connections and abnormal network synchronization, as well as reduced information-processing efficiency. This effect may be more pronounced in other ASD subgroups. This imbalance in synaptic pruning is influenced by pro-inflammatory cytokines, microglial signaling pathways [e.g., TREM2 (72,73) and STING (74)] and region-specific factors. Consequently, the dual role of microglial phagocytosis highlights the complexity and heterogeneity of the pathogenesis of ASD, supporting the rationale for therapeutic strategies that target microglial modulation.

ii) Astrocytes: Reactive transformation and neuroinflammatory regulation. Astrocytes are instrumental in maintaining CNS homeostasis and supporting neuronal function. However, in ASD, astrocyte activation is likewise closely tied to the progression of neuroinflammation (75). Activated astrocytes can release both pro-inflammatory and anti-inflammatory mediators, such as IL-6 (76) and IL-10 (77), exerting a bidirectional influence on the inflammatory process. Moreover, intermediate filament proteins in astrocytes, including glial fibrillary acidic protein (78) and vimentin (79), exhibit an altered expression and distribution. These proteins regulate the degradation of extracellular matrix components (80). In the brains of patients with ASD, astrocytes often display morphological changes characteristic of reactive astrocytes, i.e., hypertrophic, with thicker and shorter processes, particularly the neurotoxic A1 subtype. These alterations are linked to astrocyte dysfunction, disrupted neurogenesis and neuroinflammation, thereby

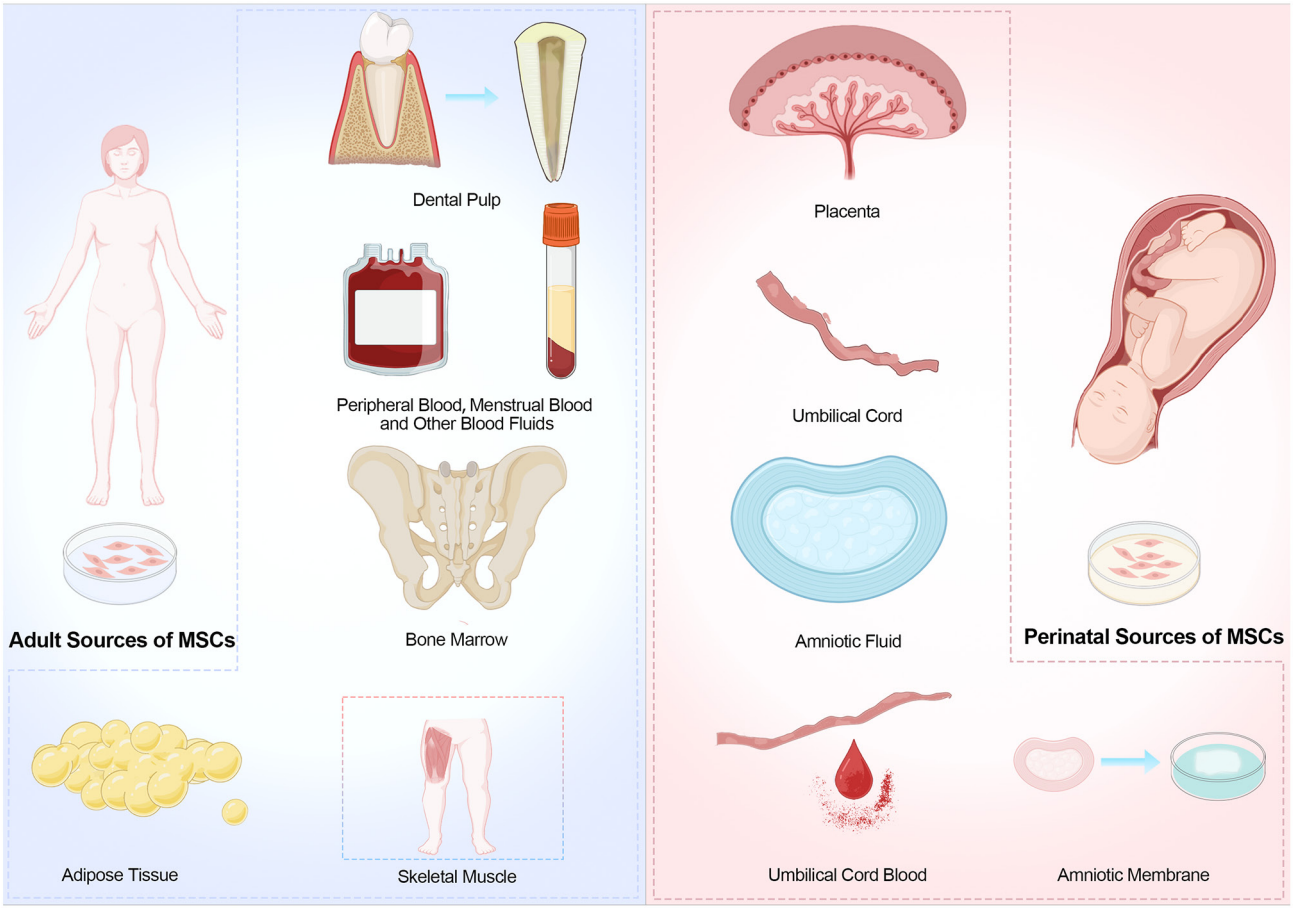

Figure 2. Sources of MSCs. Adult MSCs can be derived from bone marrow, adipose tissue, peripheral blood, skeletal muscle, and dental pulp. Perinatal MSCs are typically harvested from the placenta, umbilical cord, cord blood, amniotic fluid, and amniotic membrane. This figure was created based on data or concepts adapted from the study by Brown *et al* (88), but was drawn and designed by the authors. MSCs, mesenchymal stem cells.

exacerbating neurological abnormalities in individuals with ASD (81-83).

iii) Pro-inflammatory cytokines and neuroinflammatory signaling. Pro-inflammatory cytokines are key factors driving neuroinflammation in ASD. Their dysregulated expression signifies aberrant immune activation and may directly or indirectly inflict neuronal damage. Elevated levels of IL-1β and TNF-α have been strongly associated with compromised synaptic function and reduced neuronal viability (84). Furthermore, cytokines can modify neurotransmitter metabolism and release, further destabilizing neural networks. For example, IL-6 has been shown to regulate glutamate transporter expression, thereby exacerbating excitotoxicity (85). These inflammatory mediators also function through signaling cascades, such as nuclear factor κ-light-chain-enhancer of activated B cells (NF-κB) and JAK-STAT to regulate the intensity and duration of the immune response, thereby exacerbating neuronal dysfunction and cell death (86).

In summary, the activation of microglia and astrocytes significantly modulates neuroinflammation and immune responses in ASD, profoundly influencing pathological progression. By releasing pro-inflammatory cytokines and altering synaptic function, aberrant glial activation not only promotes neuronal damage, but also undermines the stability of neuronal networks. A more precise understanding of the atypical inflammatory and immune mechanisms in ASD is therefore pivotal for illuminating its pathophysiology and developing effective therapeutic strategies.

3. Mesenchymal stem cells and their derived exosomes in ASD: Research and applications

Biological characteristics and immunoregulatory mechanisms of action of MSCs. MSCs are adult stem cells characterized by multipotent differentiation and self-renewal capacities, initially identified in bone marrow by Friedenstein *et al* (87). These cells adhere to plastic culture flasks and exhibit robust biological functions. They are widely distributed in various tissues, such as bone marrow, adipose tissue, umbilical cord, placenta, and dental pulp (88). A schematic overview of these MSC sources is illustrated in Fig. 2. Owing to their diverse sources and distinct functional properties, have emerged as a significant research focus in regenerative medicine and cell therapy.

Firstly, MSCs possess notable multipotent differentiation potential under specific conditions; they can differentiate into various cell types, including osteoblasts, chondrocytes, myocytes, adipocytes, and neuron-like cells. This property is under precise genetic regulation (89). For instance, RUNX family transcription factor 2 (*RUNX2*) plays a pivotal role in osteogenic differentiation (90); peroxisome proliferator-activated receptor γ (*PPAR-γ*) controls adipogenic differentiation (91); SRY-Box transcription factor 9 (*SOX9*)

is crucial for chondrogenesis (92); and the differentiation of neuron-like cells relies on the expression of proteins, such as Nestin and neuronal differentiation 1 (NeuroD1) under the induction of neurotrophic factors (93). These genes, which fine-tune the Wnt/β-catenin TGF-β/bone morphogenetic protein (BMP) and Notch 1 signaling pathways, provide the molecular underpinnings for the applications of MSCs in regenerative medicine.

Secondly, MSCs play a prominent role in immunomodulation. Research suggests that MSCs can secrete multiple immunoregulatory factors and interact with immune cells, thus exerting immunosuppressive and immunobalancing effects. For example, PGE2, TGF-β, IL-10 and HLA-G secreted by MSCs can effectively inhibit T-cell proliferation and induce the generation of regulatory T-cells (Tregs), thereby promoting immune tolerance (94). Furthermore, by causing the polarization of macrophages toward the M2 phenotype, MSCs can significantly reduce the release of pro-inflammatory cytokines, such as IL-6 and TNF-α (95), thereby improving the pathological environment associated with inflammation. These properties render MSCs promising therapeutic candidates for various immune-related diseases (96).

Additionally, the low immunogenicity of MSCs aids in their long-term survival in the host. This advantage arises from their low expression of Major Histocompatibility Complex (MHC) class I molecules and the lack of MHC class II and costimulatory molecules (e.g., CD80 and CD86) (97). Such features markedly reduce T-cell activation and diminish strong host immune rejection. Moreover, MSCs secrete multiple immunosuppressive factors (e.g., PGE2, IDO1 and HLA-G) to attenuate immune responses further (98). These combined mechanisms enable MSCs to survive and exert therapeutic effects in allogeneic transplantation settings, with minimal reliance on immunosuppressants.

Research advances of MSCs in ASD. MSCs promote neuroprotection and functional recovery in various neurological disorders through multiple pathways. For instance, MSCs secrete brain-derived neurotrophic factor (BDNF), vascular endothelial growth factor (VEGF) and neurotrophin-3 (NT-3), which can enhance neuronal survival, axonal regeneration, and synaptic function by activating the phosphatidylinositol 3 kinase (PI3K)/protein kinase B (Akt) and MAPK/ERK signaling pathways (99,100). Through reducing neuroinflammation and oxidative stress, MSCs have been shown to attenuate disease progression in Alzheimer's disease and Parkinson's disease, while improving neurological and cognitive functions (101,102). In spinal cord injury models, MSCs facilitate axonal regeneration and functional recovery (103).

Notably, relevant studies on ASD also demonstrate promising results. In a rat model of VPA-induced ASD, MSC transplantation was shown to markedly improve social interaction, mitigate repetitive behaviors and alleviate anxiety-like behaviors (104). These effects may be attributed to the immunomodulatory and neuroprotective functions of MSCs which reduce neuroinflammation and enhance synaptic plasticity, ultimately improving core behavioral symptoms.

Despite the considerable therapeutic potential of MSCs in ASD, challenges remain regarding clinical translation, including immunorejection, diminished proliferative capacity over time, and concerns over transplantation safety. In this regard, exosomes secreted by MSCs have emerged as a research hotspot in recent years due to their critical roles in immune regulation and tissue repair (105). Compared with conventional cell transplantation, MSC-derived exosomes offer a 'cell-free therapy' strategy characterized by small size, ease of storage, low immunogenicity, and suitability for standardized production and quality control. These features not only provide new perspectives and directions for the precision treatment of neurodevelopmental disorders such as ASD, but may also enhance the feasibility of clinical translation (106).

Structure and biological functions of exosomes. Exosomes are extracellular vesicles measuring ~30-150 nm in diameter, originating from endosomal pathways and released into bodily fluids through the fusion of multivesicular bodies with the cell membrane (107,108). A lipid bilayer encases exosomes enriched with various biologically active components, including proteins, nucleic acids (mRNAs, miRNAs and lncRNAs, etc.) and lipids. Their stable structure enables exosomes to serve as unique mediators of intercellular communication and regulation, thus providing a theoretical basis for potential therapeutic applications (109). The composition and biogenesis pathways of MSC-Exos, as well as their downstream effects on neuronal and glial cells in ASD models, are summarized in Fig. 3. Exosomes exert their therapeutic effects primarily through interacting with their cargo in recipient cells. Exosomes deliver their contents directly into target cells, either by fusing with the plasma membrane or undergoing receptor-mediated endocytosis, and thereby modulate gene expression and signaling pathways.

i) Nucleic acid cargo and gene regulation. The miRNAs in exosomes play vital roles in regulating gene expression, as they can modulate inflammation, neuronal survival, and synaptic plasticity by targeting specific signaling cascades (110,111). Previous studies have demonstrated that miRNAs encapsulated in exosomes, such as miR-124 packaged within exosomes, have been demonstrated to enhance neuronal regeneration and reduce neuroinflammation (112-115). Other miRNAs, such as miR-21, miR-155 and miR-146a, can suppress the NF-κB pathway, thereby reducing the secretion of pro-inflammatory factors (e.g., IL-6 and TNF-α) and increasing the levels of anti-inflammatory IL-10, ultimately mitigating neuroinflammation. These miRNAs may also inhibit excessive activation of the mTOR pathway, reducing aberrant neuronal activity (115-118). Additionally, miR-873a-5p within exosomes can promote microglial polarization toward the anti-inflammatory M2 phenotype (120). Apart from miRNAs, mRNAs and lncRNAs, exosomes can also contain these molecules, which can affect recipient cell functions through translation or transcriptional regulation, influencing tissue repair and immune modulation (121-123).

ii) Protein cargo and neural repair. Exosomes also harbor proteins, such as heat shock proteins (HSPs) (124), neuronal adhesion molecules [e.g., intercellular adhesion molecule 1 (125)] and cytokines [e.g., TGF-β (126)] that can directly modulate axonal growth, promote synaptic remodeling, and strengthen neural network connectivity, are considered critical processes in immune regulation, neuroprotection, and tissue repair. Furthermore, exosomal membrane

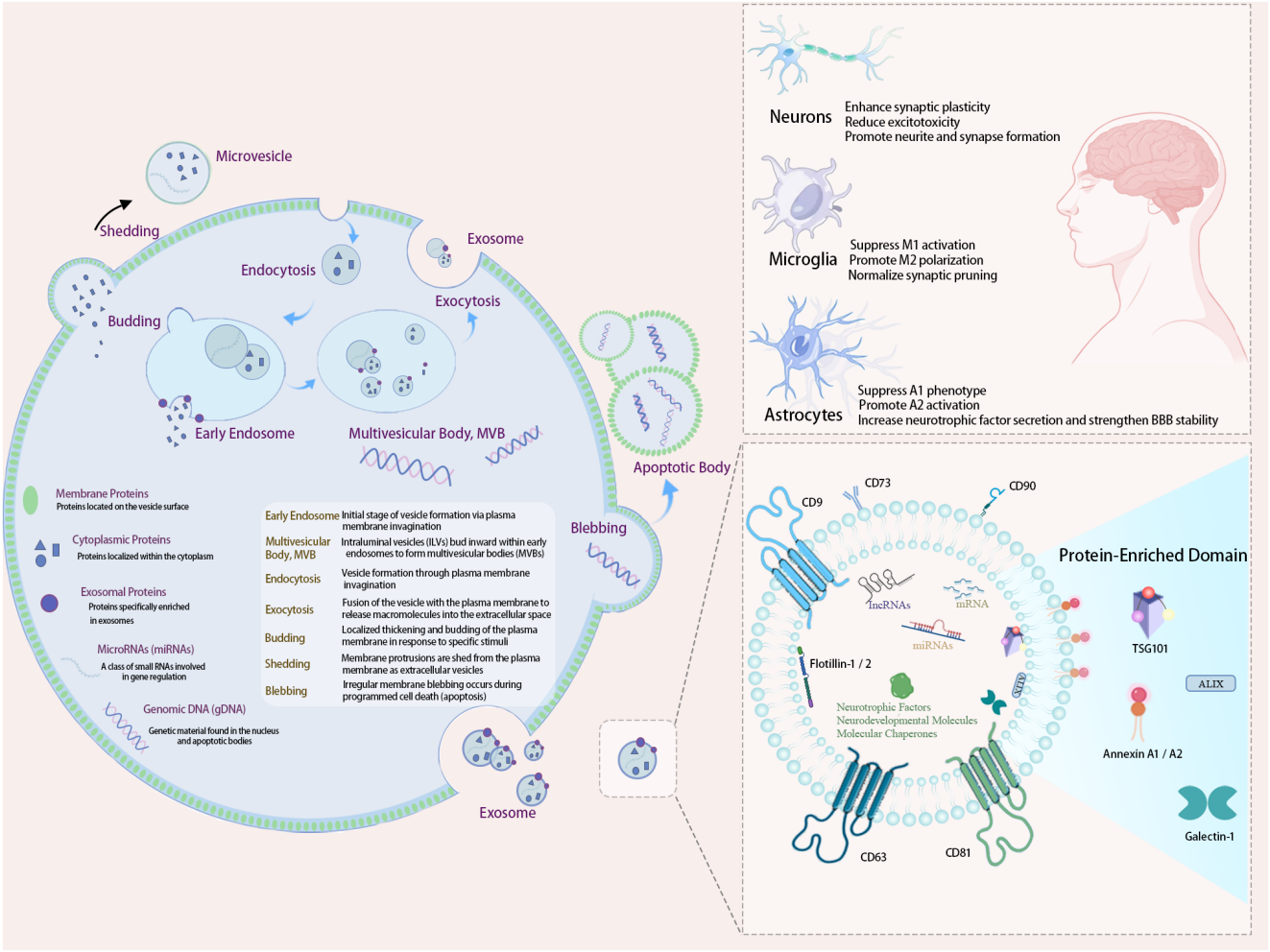

Figure 3. Biogenesis, molecular contents and mechanisms of MSC-derived exosomes in ASD therapy. (Left panel) MSC-derived exosomes originate from endosomal pathways. They encapsulate proteins, mRNAs and miRNAs. (Top right panel) These exosomes interact with neurons, microglia and astrocytes, modulating synaptic plasticity, microglial polarization, and astrocyte phenotype to alleviate neuroinflammation associated with ASD. (Bottom right panel) Exosomal surface and cargo proteins include CD63, CD81, CD9, CD73, flotillin-1/2, TSG101, Annexin A1/A2, galectin-1, neurotrophic factors and molecular chaperones. The authors created a figure referencing multiple sources (119-123,133-139,141-143). ASD, autism spectrum disorder; MSCs, mesenchymal stem cells.

lipids (e.g., sphingomyelin and phosphatidylserine) can enhance synapse formation and stability, thereby influencing the functional properties of recipient cells (127). These molecular mechanisms provide a basis for improving cognitive and behavioral deficits in ASD.

iii) Neurotrophic factors and signaling pathways. Exosomes are often enriched with neurotrophic factors and neurodevelopmental molecules, such as Nestin, NeuroD1, BDNF and VEGF. These factors act through signaling pathways, including the Wnt/β-catenin, TGF-β/BMP and PI3K/Akt pathways, thereby supporting neuronal proliferation, migration and differentiation (128-130).

iv) Gut microbiota and the gut-brain axis. ASD has been closely linked to gut microbiome dysbiosis. Exosomes can modulate gut microbiota diversity by increasing the abundance of bifidobacteria and lactobacilli and improving gut barrier function (131). Exosomes containing miR-155, miR-181a and TGF-β can reduce intestinal inflammation and permeability. Exosomes containing miR-155, miR-181a and TGF-β can reduce intestinal inflammation and permeability, ultimately influencing the gut-brain axis and indirectly affecting the central nervous system function (132-134).

Exosomes carry a versatile repertoire of bioactive molecules that can modulate inflammatory responses, promote neural repair and regulate the gut-brain axis. These properties underscore the potential of exosome-based, cell-free therapy for ASD and other neurodevelopmental disorders.

Potential value of exosomes in the treatment of ASD. In recent years, MSC-Exos have exhibited immense promise in ASD research and interventions due to their unique composition and diverse functionalities (29,129). Compared with direct cell transplantation, exosomes provide greater safety, controllability and ease of storage. By carrying various miRNAs, proteins and bioactive molecules, MSC-Exos can exert immunomodulatory, neuroprotective and synaptic repair effects in the CNS (135,136). Their ability to regulate neuroinflammation, restore synaptic plasticity, and rebalance the gut-brain axis underscores the potential of exosome-based 'cell-free therapy' (129), paving the way for new avenues of research and clinical applications in ASD.

Studies using animal models of ASD have demonstrated that the intranasal administration of MSC-Exos markedly improves social interaction, cognition and

repetitive behaviors (22,29,135,136). These effects may be attributed to miRNAs and anti-inflammatory agents delivered by exosomes, which modulate immune responses and neuronal network functionality. For example, exosomal miRNAs [such as miR-21-5p (137), miR-17-92 (138) and miR-146a (139)] and multiple bioactive proteins [including HSP70 and TGF-β (140)] help regulate neuroinflammation, synaptic plasticity, and neurogenesis via intercellular signaling. Additionally, research suggests that neurotrophic factors present in exosomes can enhance neuronal survival and differentiation, thereby improving behavioral outcomes. These factors also enhance synaptic plasticity, promote network-level functional integration, and reduce CNS inflammation by increasing anti-inflammatory cytokines (22,29,135,141).

In summary, MSC-Exos, owing to their diverse bioactive molecules and stable delivery system repertoire, have shown comprehensive effects on immunoregulation, neural repair and synaptic regeneration in ASD. They are thus emerging as a promising therapeutic alternative to conventional cell-based treatments. Ongoing research will further elucidate the underlying mechanisms of exosomes in ASD, optimize exosome preparation and administration strategies, and, through multidisciplinary collaboration, develop safe, efficacious and scalable exosome-based therapeutics. These efforts hold the potential to propel the field toward precision interventions and clinical translation for ASD.

4. Clinical progress of stem cells and exosomes in ASD

In recent years, stem cell-based therapy has emerged as one of the key areas of interest for the treatment of ASD. By employing stem cells or their exosomes, researchers aim to modulate immune responses, improve cerebral blood flow, and enhance neuronal repair in individuals with ASD (21). Early clinical trials have documented varying degrees of improvement in behavior, language skills and social abilities among ASD patients receiving MSC therapy (142) with minimal adverse events (143). Additionally, these trials noted decreases in inflammatory markers and improvements in brain metabolic activity, further supporting the potential role of MSCs in the treatment of ASD (142).

Current status of clinical research and key findings. Several completed clinical trials worldwide have investigated the use of stem cell therapy for the treatment of ASD. The following paragrpahs summarizes the primary clinical studies and their principal outcomes:

i) Use of umbilical cord blood-derived stem cells. Lv *et al* (144) conducted a study combining cord blood mononuclear cells (CBMNCs) with umbilical cord-derived mesenchymal stem cells, enrolling 37 patients with ASD. Their results revealed that the combination treatment group had significantly greater improvements in the Childhood Autism Rating Scale (CARS) and Aberrant Behavior Checklist (ABC) scores compared to either the CBMNC-only group or the control group (which received only rehabilitative interventions). No severe adverse events were reported, underscoring both the safety and the superior efficacy of combination therapy (144).

ii) Preliminary attempts with human embryonic stem cells. Shroff (145) reported outcomes for 3 patients with ASD treated with human embryonic stem cells. Improvements were observed in motor skills, cognition, social interaction and sensory sensitivities, with no adverse events noted. Although the sample size was small, this study highlights the potential of stem cell therapies for alleviating core symptoms of ASD (145).

iii) Potential of autologous umbilical cord blood therapy. Chez *et al* (146) conducted a randomized, double-masked, placebo-controlled crossover study involving 29 children with ASD aged 2 to 6 years, evaluating the safety and efficacy of autologous umbilical cord blood transfusions. Although treatment did not significantly improve its primary endpoints, some participants exhibited trends toward improved socialization and language skills. No severe adverse events occurred, suggesting that stem cell therapy may hold promise for enhancing social functioning (146).

iv) Changes in brain network connectivity. Simhal *et al* (147) used diffusion tensor imaging to assess the effect of umbilical cord blood stem cell therapy on white matter networks in patients with ASD. Their study revealed that post-treatment, the robustness of the white matter network was markedly increased, especially in regions involved in social and communication functions. These findings may be related to the ability of stem cells to regulate neuroinflammation and promote neuroplasticity (147).

v) Stem cells combined with behavioral interventions. Thanh *et al* (148) conducted an open-label clinical trial in Vietnam, enrolling 30 children with ASD aged 3 to 7 years. The intervention combined autologous bone marrow mononuclear cell transplantation with the ESDM, an early behavioral intervention. The results revealed a reduction in median CARS scores from 50 to 46.5 ($P<0.05$) and an increase in Vineland Adaptive Behavior Scale scores from 53.5 to 60.5, indicating substantial improvements in social interaction, language abilities, and daily living skills, with no severe adverse events reported (148).

Collectively, the aforementioned clinical studies suggest that MSC-based and MSC exosome therapies hold promise for improving social communication, language skills, behavioral challenges and cognition in individuals with ASD. Notably, combined treatments, including the incorporation of behavioral interventions or personalized therapeutic regimens that appear effective, may be due to their multi-target mechanisms of action. The majority of studies did not report severe adverse events, reflecting an overall favorable safety profile. However, a number of existing studies involve small sample sizes and open-label designs, lacking the robust evidence from large-scale randomized controlled trials (RCTs). Additionally, the precise mechanisms underlying the observed therapeutic effects remain incompletely understood. Future investigations should further refine treatment protocols, clarify the role of exosomes in cell therapy, and establish the long-term safety and efficacy of these interventions. Expanding multicenter, large-sample RCTs is crucial for evaluating the performance of different stem cell types and administration strategies. Moreover, exploring synergistic effects between stem cells and behavioral or pharmacological interventions will help optimize individualized treatment plans. Finally, integrating

gene-editing technologies and engineered exosomes could enhance their clinical utility. Such efforts will provide a more robust theoretical and practical foundation for applying stem cell and exosome-based therapies in ASD.

Potential for clinical translation and existing barriers. Due to pronounced immunomodulatory properties, tissue repair potential and favorable safety profile, MSCs have attracted widespread attention in recent years, gradually becoming an essential focus of stem cell-based therapies. As MSC research advances, the exosomes they secrete (MSC-Exos) have been identified as a key functional mediator and are expected to play a potential role in ASD interventions (149). The majority of clinical studies on MSCs and MSC-Exos remain in phase I/II trials (150). Early data suggest that patients with ASD treated with these therapies experience improved social interaction and language skills, as well as decreased inflammatory markers, such as TNF-α and IL-6 (151,152). Nonetheless, the clinical translation of MSCs and their exosomes depends on the efficacy of preclinical studies, as well as substantial practical and regulatory challenges. Although preliminary findings are encouraging, limitations in sample size, follow-up duration, and study design mean that large-scale, multicenter clinical data are still required to confirm their long-term efficacy and safety. A discussion of the advantages and challenges inherent in clinical translation is presented below:

a) Core advantages and multifaceted mechanisms:

i) Multifunctionality: Immunomodulation and neuroprotection. The immunomodulatory and neuroprotective properties of MSCs render them attractive for addressing multiple pathological mechanisms underlying ASD. Research indicates that MSC-Exos can regulate T- and B-cell activation, induce immune tolerance, and suppress the secretion of inflammatory cytokines, thus improving neuroinflammatory conditions (22). Growth factors, miRNAs, and other bioactive substances in MSC-Exos also promote neuronal survival and synaptic plasticity (111). Owing to their differentiation potential, MSCs and their exosomes may also serve as tools for neural repair, providing potential therapeutic value for ASD-related neurodevelopmental anomalies.

ii) Low immunogenicity and favorable safety profile. MSCs exhibit relatively low immunogenicity in allogeneic transplantation (153). Their exosomes, which lack complete cell-surface antigen expression (154) and do not carry a nucleus or full genomic content (155), further minimize risks of immune rejection and tumorigenesis. Consequently, MSC-Exos do not require stringent donor-recipient matching to the same extent as allogeneic MSCs, reducing potential complications and ethical concerns.

iii) Targeted action and personalized therapy. Exosomes can enter target cells through receptor-mediated endocytosis or membrane fusion, delivering a diverse array of active molecules that influence cell proliferation, differentiation, apoptosis, and immune responses. Owing to their nanoscale size and lipid bilayer structure, exosomes demonstrate relatively high transmembrane delivery efficiency; research suggests that exosomes can traverse the blood-brain barrier (156). As the pathogenesis of ASD involves abnormal neural networks and central neuroinflammation, MSC-Exos capable of penetrating the CNS may help restructure the brain microenvironment and enhance therapeutic outcomes. Moreover, engineering MSCs or modifying exosome cargo based on individual patient pathology (e.g., through gene editing) may enable more precise, personalized treatments.

iv) Large-scale production, transport and storage. Under specific culture conditions, exosomes can be produced and purified in large quantities. Compared to cell transplantation, which requires highly viable, well-characterized cells in sufficient numbers, exosomes are easier to standardize and mass-produce, rendering clinical deployment more feasible. Additionally, exosomes can be stored for extended periods under appropriate conditions while retaining their bioactivity, thereby circumventing many of the stability challenges encountered with live MSCs. This facilitates large-scale manufacturing, clinical stockpiling, and broader geographic distribution.

b) Real-world challenges and bottlenecks:

i) Production techniques and quality control. Variations in MSC culture conditions and tissue sources can produce substantial differences in exosome yield and quality. Standard protocols for extracting, purifying and characterizing exosomes have yet to be universally adopted. Reagents, equipment and technical approaches vary greatly among laboratories, which can potentially affect the reproducibility of results. To achieve industrial-scale production and clinical adoption of MSC-Exos, international or national-level standardization is needed, encompassing MSC culture conditions, exosome extraction, purification, and the establishment of quality metrics. Key parameters include identifying and quantifying cargo molecules, particle size distribution and surface markers. Without such standards, ensuring the stability and consistency of MSC-Exos across different batches remains difficult.

ii) Heterogeneity in sources and cell states. MSCs can be derived from various sources, including bone marrow, adipose tissue, umbilical cord, and other tissues, resulting in variability based on tissue origin, donor characteristics and passage number. For instance, adipose-derived MSCs generally have more substantial adipogenic potential, whereas bone marrow-derived MSCs may be more adept at osteogenic differentiation (157). In the context of ASD therapy, MSC-Exos from different sources or differing cell expansion histories may yield divergent immunomodulatory and neuroprotective effects. A systematic investigation is required to determine an optimal source or a combination strategy for clinical use.

iii) Complexity of mechanisms. Although a number of studies have documented the neuroprotective, immunoregulatory and neural network repair functions of MSC-Exos, the precise molecular mechanisms underlying these functions remain incompletely understood (22,136,149). It remains to be determined which miRNAs or proteins in MSC-Exos are essential for regulating neuronal repair and immune balance in ASD. In addition, the mechanisms through which exosomes interact with target cells and influence downstream signaling pathways remain to be elucidated. Further research using *in vitro* and *in vivo* models is required to clarify these mechanisms, improving the specificity and reliability of future clinical applications.

iv) Safety and ethical oversight. While MSC-Exos are associated with a lower immunogenicity and tumorigenic risk than direct cell transplantation, comprehensive preclinical safety

evaluations and multicenter clinical trials are necessary to assess long-term risks. The clinical use of stem cells and their derivatives is strictly regulated in many countries, requiring compliance with guidelines for pharmaceutical and biological products, as well as adherence to ethical considerations, including donor sourcing and traceability during preparation and application (158).

v) Large-scale manufacturing and cost-effectiveness. Generating MSC-Exos on a large scale requires efficient isolation and concentration protocols for MSC culture. Once translated to clinical practice, cost and patient accessibility become critical factors. In the event that production remains prohibitively costly without relevant policy support, the widespread adoption of MSC-Exos in the treatment of ASD may be limited.

vi) Delivery efficiency and long-term safety. Achieving the efficient, targeted delivery of exosomes to the CNS and specific lesion sites to enhance therapeutic specificity poses a significant challenge. Moreover, current exosome-related clinical trials typically feature short follow-up periods (144,146,147). Exosomes hold potential as a long-term intervention strategy; however, their extended use may carry unknown risks, including effects on the immune system and neural function. Rigorous long-term monitoring and evaluation are therefore necessary.

vii) Individual differences and clinical applicability. ASD is highly heterogeneous, with variability in age, subtype and comorbid conditions affecting treatment responses to MSCs or MSC-Exos. Personalized therapies tailored to individual disease stages and pathological profiles remain a significant challenge. Biomarker analysis and molecular subtyping may enhance individualized treatment strategies. Systematic clinical trials are necessary to establish optimal dosing and administration protocols for diverse patient populations.

5. Conclusion

In conclusion, MSCs and MSC-Exos have demonstrated considerable therapeutic potential in the treatment of ASD. MSCs and MSC-Exos can alleviate neuroinflammation, enhance synaptic plasticity, and promote neural network repair through their immunomodulatory and neuroprotective effects. MSC-Exos offers substantial advantages for ASD treatment, including multifaceted therapeutic mechanisms, low immunogenicity and potential for large-scale production. Nevertheless, critical hurdles, such as manufacturing protocols, quality control, elucidation of the mechanism, long-term safety, and personalized application, need to be overcome before MSC-Exos can be widely integrated into ASD therapies. Addressing these challenges through multidisciplinary collaboration and further research will pave the way for more precise, safe, and effective interventions. With their low immunogenicity, ability to cross the blood-brain barrier, and efficient delivery of bioactive molecules, exosomes are promising candidates for treating ASD and other neurodevelopmental disorders. However, several critical challenges remain, including issues related to large-scale production, quality control, and elucidation of underlying mechanisms. Moreover, rigorous clinical trials under strict regulatory and ethical frameworks are required to establish the safety and efficacy of MSC-Exos. Therefore, it is urgent to clarify their mechanisms of action, develop standardized manufacturing and quality control protocols, and optimize long-term safety and personalized treatment strategies. With the integration of high-throughput omics technologies, interdisciplinary collaboration, and continuous innovation, MSC-Exos are expected to advance in basic research and clinical application, offering greater precision and broader therapeutic potential in ASD. Ultimately, only by overcoming key scientific and technical hurdles can MSC-Exos become a viable strategy for individualized interventions.

6. Future perspectives and application directions

Advancing the therapeutic efficacy and clinical translation of MSC-derived exosomes in ASD will require sustained innovation across multiple dimensions. To improve *in vivo* delivery efficiency, various delivery platforms, such as magnetic nanoparticles (159) and hydrogels (160), have been developed to enhance targeting and stability in the central nervous system. Additionally, chemical modifications (161) and genetic engineering techniques (162) have been employed to increase the therapeutic specificity and prolong the half-life of exosomes. Engineered MSCs that overexpress functional proteins and miRNAs, such as neurotrophic factors or anti-inflammatory molecules (163), have shown promise in enhancing the neuroprotective and immunoregulatory effects of their exosomes, thereby broadening their application potential in ASD and other neurological disorders. To meet the demands of personalized medicine, customized exosome formulations and treatment protocols tailored to individual patient profiles may improve therapeutic outcomes and reduce adverse effects. The application of high-throughput omics technologies, such as genomics, proteomics and metabolomics continues to unveil the key molecular pathways and networks through which MSCs and their exosomes modulate neurorepair and immune responses. In particular, in-depth investigations into the roles and interactions of miRNAs, proteins, and other bioactive cargos will provide a robust foundation for evaluating therapeutic efficacy and guiding the engineering of exosomes. For clinical translation, it is essential to establish standardized and efficient protocols for MSC cultivation and exosome purification, along with internationally recognized quality control standards to ensure batch-to-batch and inter-institutional consistency. Large-scale, multicenter randomized controlled trials will be crucial for validating the safety and efficacy of MSC-Exos, necessitating refined trial designs that address patient selection, dosage regimens, outcome measures, and long-term follow-up. Given the complexity of ASD pathogenesis and its clinical heterogeneity, future breakthroughs in MSC-Exos therapy will depend on interdisciplinary collaboration across neuroscience, molecular biology, immunology, and materials science, combined with pharmacological and behavioral interventions. Continuous technological innovation and integrative strategies will ultimately pave the way for more effective and safer treatment options for individuals with ASD.

Acknowledgements

Not applicable.

Funding

The present study was supported by a grant from the Enterprise Joint Fund Project of Hunan Provincial Natural Science Foundation (grant no. 2024JJ9097).

Availability of data and materials

Not applicable.

Authors' contributions

ZS was involved in the conceptualization of the study. ZS and NA were involved in the collection and curation data from the literature, and in the writing and preparation of the original draft of the manuscript. MM was involved in visualization and literature analysis. ZL supervised the study. ZS, NA and ZL reviewed and edited the final manuscript. All authors have read and approved the final manuscript. Data authentications is not applicable.

Ethics approval and consent to participate

Not applicable.

Patient consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

Use of artificial intelligence tools

During the preparation of this work, AI tools were used to improve the readability and language of the manuscript or to generate images, and subsequently, the authors revised and edited the content produced by the AI tools as necessary, taking full responsibility for the ultimate content of the present manuscript.

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