doi.org/10.36721/PJPS.2026.39.9.256.1

REVIEW

Pharmacological evaluation frameworks in autism spectrum disorder: Methodological and neurobiological considerations

Michel Bourin
Neurobiology of Anxiety and Mood Disorders, Nantes University, 98 rue Joseph Blanchart, 44100 Nantes, France

Abstract: Autism spectrum disorder (ASD) is a heterogeneous neurodevelopmental condition characterized by persistent deficits in social communication and interaction, alongside restricted and repetitive behaviors. Despite substantial advances in genetics, neurobiology and neuroimaging, no pharmacological treatment currently addresses the core symptoms of ASD. Existing medications primarily target associated behavioral disturbances such as irritability, aggression and hyperactivity. This narrative review synthesizes contemporary neurobiological findings, methodological challenges and evolving clinical trial frameworks relevant to the evaluation of emerging pharmacotherapies in ASD. Particular emphasis is placed on biomarker-informed stratification, rigorous trial design and developmental timing as critical determinants of therapeutic success. Advances in molecular genetics, systems neuroscience and multimodal biomarker integration highlight the need for precision medicine approaches that align pharmacological targets with underlying neurodevelopmental mechanisms. Strengthening methodological rigor while maintaining ethical and neurodiversity-affirming principles will be essential to advancing effective pharmacological interventions for ASD.

Keywords: Autism spectrum disorder; Biomarkers; Clinical trial design; Methodology; Neurodevelopment; Pharmacotherapy

Submitted on 08-11-2025 – Revised on 08-04-2026 – Accepted on 13-04-2026

INTRODUCTION

Autism spectrum disorder (ASD) is a complex and lifelong neurodevelopmental condition that emerges early in childhood and is defined by impairments in social communication and reciprocal interaction, accompanied by restricted, repetitive patterns of behavior, interests, or activities. Current epidemiological estimates indicate a prevalence of approximately 1–2% worldwide, with increasing diagnostic rates largely attributable to improved awareness, screening practices and diagnostic criteria rather than a true rise in incidence (Leisman and Melillo, 2025).While behavioral, educational and psychosocial interventions remain the foundation of ASD management, pharmacological treatments are widely used to address associated symptoms such as irritability, aggression, anxiety and attention-deficit/hyperactivity disorder (ADHD)-like features. Importantly, no medication has yet demonstrated robust efficacy for the core social-communication deficits that define ASD.

Converging neurobiological evidence conceptualizes ASD as a disorder of atypical brain development involving altered synaptic maturation, disrupted neural connectivity, excitation/inhibition (E/I) imbalance and immune dysregulation (Bjorklund et al;, Courchesne *et al*., 2019). Structural and functional neuroimaging studies consistently implicate the amygdala, hippocampus, cerebellum and prefrontal cortex—regions critical for social cognition, emotional regulation and executive function (Courchesne *et al*., 2004; Brambilla *et al*., 2003). Genetic studies have identified hundreds of risk-associated variants, including recurrent mutations in genes such as SHANK3, CHD8, SCN2A, CNTNAP2 and DYRK1A, which converge on synaptic development, chromatin remodeling and neuronal signaling pathways (Satterstrom *et al*., 2020; Sanders *et al*., 2015). Biochemical abnormalities, including elevated whole-blood serotonin and altered glutamatergic signaling, further underscore the biological heterogeneity of ASD and suggest potential pharmacological targets (Muller *et al*., 2016; Uzunova *et al*., 2016).

These advances necessitate the development of methodologically rigorous, biomarker-informed pharmacological evaluation frameworks capable of linking clinical phenotypes with underlying neurobiological mechanisms.

Inclusion criteria for clinical pharmacological studies
Diagnostic heterogeneity

A central methodological challenge in ASD pharmacological research is the marked heterogeneity of the condition. ASD encompasses a wide spectrum of cognitive abilities, language skills, adaptive functioning and comorbid psychiatric or medical conditions. This heterogeneity can obscure treatment effects and contribute to inconsistent trial outcomes.

Core diagnostic domains include persistent deficits in social communication and interaction, restricted or repetitive behaviors, early developmental onset and clinically significant functional impairment (APA, 2013). In pharmacological trials, failure to adequately

*Corresponding author: e-mail: Michel.Bourin@univ-nantes.fr

characterize participant subgroups may dilute efficacy signals and limit reproducibility. Stratification based on age, intellectual functioning, language ability and comorbidities is therefore essential to enhance both internal validity and translational relevance (Hodges *et al*., 2020).

Diagnostic tools and biomarker support

Reliable and standardized diagnostic instruments are critical for participant selection. The Autism Diagnostic Interview–Revised (ADI-R) and Autism Diagnostic Observation Schedule, Second Edition (ADOS-2), remain the gold standard diagnostic tools, ensuring cross-study consistency (Lord *et al*., 1994). Dimensional measures such as the Social Responsiveness Scale (SRS-2) and Vineland Adaptive Behavior Scales (VABS-3) provide quantitative assessments of social and adaptive functioning.

Beyond behavioral diagnostics, neurobiological markers offer an opportunity to refine inclusion criteria. Neuroimaging studies have demonstrated early brain overgrowth followed by atypical synaptic pruning, with consistent abnormalities in the amygdala, corpus callosum and cerebellum (Brambilla *et al*., 2003). Incorporating genetic profiles, serotonergic markers, or EEG-derived indices of cortical E/I balance may facilitate the identification of biologically meaningful subgroups with differential pharmacological responsiveness (Hudac and Webb, 2024; Beversdorf *et al*., 2023).

Intervention models

Pharmacological interventions in ASD generally follow two conceptual models. The symptom-based model targets secondary behavioral manifestations such as irritability, aggression, anxiety, or hyperactivity and underlies the clinical use of antipsychotics, mood stabilizers and adrenergic agents. In contrast, the disease-modifying model aims to influence core neurodevelopmental processes by targeting synaptic plasticity, neurotransmitter balance, neuroimmune pathways, or social neuropeptide systems (Josselsohn *et al*., 2024).

To date, most clinical trials have focused on symptom reduction, while mechanistically driven, disease-modifying approaches remain largely experimental. Strengthening translational links between animal models, human biomarkers and clinical endpoints is critical for progress beyond symptomatic management.

Evaluation criteria

Behavioral outcome measures

Outcome measure selection is central to evaluating pharmacological efficacy in ASD. While ADI-R and ADOS-2 are diagnostic tools, they have limited sensitivity to short-term change. The Aberrant Behavior Checklist (ABC), particularly the irritability subscale, is widely used in pharmacological trials, with a 25% reduction commonly defined as a clinically meaningful response (Kildahl *et al*., 2025).

The Clinical Global Impression (CGI) scale provides a broad clinician-rated assessment of improvement but lacks ASD specificity. Instruments such as the Children's Yale–Brown Obsessive Compulsive Scale (CY-BOCS), SRS-2 and VABS-3 capture repetitive behaviors, social responsiveness and adaptive functioning, respectively and are increasingly incorporated as secondary or exploratory outcomes.

Given the limited verbal abilities of many individuals with ASD, direct behavioral observation by trained clinicians remains an essential and objective assessment method. Composite outcome measures integrating caregiver reports, clinician ratings and objective biomarkers may enhance reliability and ecological validity (Gabriels *et al*., 2001).

Objective biomarkers

Objective biomarkers are increasingly incorporated into early-phase trials to assess target engagement and neurobiological effects. Functional MRI studies demonstrate that intranasal oxytocin modulates activity in amygdala–prefrontal circuits involved in social processing (Yamasue *et al*., 2020; Gordon *et al*., 2013). EEG markers, including gamma-band synchrony and mismatch negativity, offer scalable indicators of cortical E/I balance and sensory processing (Hudac and Webb, 2024).

Multimodal biomarker integration—combining neuroimaging, electrophysiology and biochemical measures—may provide a more comprehensive understanding of treatment mechanisms and individual variability in response.

Statistical and methodological design

Randomized, double-blind, placebo-controlled trials remain the gold standard for evaluating pharmacological efficacy in ASD (Jacob *et al*., 2022). However, small sample sizes and heterogeneity necessitate innovative designs, including adaptive and Bayesian approaches that allow interim analyses and flexible dosing strategies.

Stratified analyses based on age, sex, genetic background and biomarker-defined subgroups improve interpretability and translational relevance. Longitudinal follow-up is particularly important, as ASD is a developmental condition and treatment effects may evolve over time.

Pharmacological evaluation framework

Current therapeutic strategies

Current pharmacological management in ASD is predominantly symptomatic. Atypical antipsychotics such

as risperidone and aripiprazole are effective in reducing irritability and aggression through dopaminergic and serotonergic modulation (McCracken *et al*., 2002; Nagaraj *et al*., 2006). Mood stabilizers and adrenergic agents are used to manage impulsivity, mood lability and hyperarousal (McDougle *et al*., 2003).

Selective serotonin reuptake inhibitors have been evaluated for repetitive behaviors, but results remain inconsistent, with frequent reports of adverse effects (Kolevzon *et al*., 2006). Trials involving secretin, fenfluramine and several mood stabilizers have largely yielded negative or inconclusive findings (McQueen and Heck, 2002).

Overall, existing treatments provide symptomatic relief without modifying core neurodevelopmental dysfunctions.

Emerging and experimental therapies

Recent research emphasizes mechanism-based interventions. Oxytocin and vasopressin analogues show modest but variable effects on social cognition and reciprocity (Josselsohn *et al*., 2024). Glutamatergic modulators targeting NMDA or mGluR5 receptors aim to restore E/I balance, though clinical efficacy remains under investigation (Canitano and Palumbi, 2021).

Microbiome-based interventions represent an emerging frontier, with pilot studies suggesting improvements in gastrointestinal and behavioral symptoms via modulation of the gut–brain axis (Kang *et al*., 2017). Anti-inflammatory and microglial-modulating agents, including minocycline, are also under exploration due to evidence of neuroinflammation in ASD (Fan *et al*., 2023; Vargas *et al*., 2005).

Methodological and developmental considerations

Robust ASD pharmacological trials require careful participant stratification, multimodal outcome measures and sufficient duration (≥12 weeks) to assess both acute and sustained effects. Ethical considerations are paramount, particularly in pediatric populations and protocols must respect neurodiversity, ensure informed assent and minimize harm.

Developmental timing is a critical variable. Early childhood represents a window of heightened neuroplasticity during which pharmacological interventions may exert disproportionate long-term effects when combined with behavioral therapies (Dawson, 2008).

Future directions

Future ASD pharmacotherapy research should adopt a precision medicine framework, integrating genetic, neuroimaging and electrophysiological markers to identify responders and clarify mechanisms of action (Beversdorf *et al*., 2023). Large-scale open datasets, such as ABIDE, facilitate replication and meta-analysis, thereby strengthening the reliability of evidence.

Synergistic combinations of pharmacological and behavioral interventions may enhance learning and social engagement. Longitudinal studies should prioritize functional outcomes, including adaptive behavior, quality of life and community participation (Sterrett *et al*., 2023). Active involvement of autistic individuals and families in study design is essential to ensure ethical, relevant and respectful research (Lerner *et al*., 2023).

CONCLUSION

Autism spectrum disorder remains a profoundly heterogeneous neurodevelopmental condition for which effective pharmacological treatments targeting core symptoms are still lacking. Advances in genetics, neuroimaging and biomarker science now enable the development of stratified, mechanism-based trials. Progress will depend on methodological rigor, biomarker integration, developmental sensitivity and ethical commitment to neurodiversity-affirming research. Aligning pharmacological targets with developmental neurobiology offers the most promising path toward meaningful therapeutic innovation in ASD.

Acknowledgments

None.

Author's contributions

MB conceived the manuscript and performed the literature review.

Funding

There was no funding.

Data availability statement

All data analyzed during this study are included in this published article and its reference list.

Ethical approval

Not applicable.

Conflict of interest statement

The author declares no conflict of interest.

REFERENCES

American Psychiatric Association. Diagnostic and Statistical Manual of Mental Disorders. 5th ed. Washington DC: APA; **2013**.

Beversdorf DQ, Carpenter RL and Miller RF (2023). Precision medicine approaches in autism spectrum disorder. *Neurotherapeutics*, **20**(1): 123–136.

Bjorklund G, Saad K, Chirumbolo S, Kern JK, Geier DA, Geier MR and Urbina MA (2016). Immune dysfunction and neuroinflammation in autism spectrum disorder. *Acta Neurobiol Exp (Wars)*, **76**(4): 257-268.

Brambilla P, Hardan A, Di Nemi S U, Perez J, Soares J C and Barale F (2003). Brain anatomy and development in autism: Review of structural MRI studies. *Brain Res Bull*., **61**: 57–569.

Canitano R and Palumbi R (2021). Excitation/inhibition imbalance in autism spectrum disorders: Implications for pharmacological treatment. *Neuropsychiatr Dis Treat*, **17**: 357–366.

Courchesne E, Campbell K and Solso S (2019). Brain growth across the life span in autism: Age-specific changes in anatomical pathology. *Brain Res*., **1380**: 138–145.

Courchesne E, Redcay E and Kennedy DP (2004). The autistic brain: Birth through adulthood. *Curr Opin Neurol*., **17**(4): 489–496.

Dawson G (2008). Early behavioral intervention, brain plasticity and the prevention of autism spectrum disorder. *Dev Psychopathol*., **20**(3): 775–803.

Gabriels RL, Hill DE, Pierce RA, Rogers SJ and Wehner B (2021). Predictors of treatment outcome in young children with autism. *J Autism Dev Disord*., **31**: 205–216.

Gordon I, Vander Wyk BC, Bennett RH, Cordeaux C, Lucas MV, Eilbott JA, Zagoory-Sharon O, Leckman JF, Feldman R and Pelphrey KA (2013). Oxytocin enhances brain function in children with autism. *Biol Psychiatry*, **73**: 66–72.

Hodges H, Fealko C and Soares N (2020). Autism spectrum disorder: Definition, epidemiology, causes and clinical evaluation. *Transl Pediatr*., **9**(Suppl 1): S55–S65.

Hudac CM and Webb SJ (2024). Biomarkers of autism spectrum disorder: EEG and eye-tracking approaches. *Curr Psychiatry Rep*., **26**(2): 45–57.

Jacob S, Wolff JJ, Steinbach MS, Doyle CB, Kumar V and Elison JT (2019). Neurodevelopmental heterogeneity and computational approaches for understanding autism. *Transl Psychiatry*, **9**(1):63.

Josselsohn RH, Modabbernia A and Kolevzon A (2024). Emerging pharmacological treatments for autism spectrum disorder. *Curr Opin Psychiatry*, **37**(2): 89–97.

Kang DW, Adams JB, Gregory AC, Borody T, Chittick L, Fasano A, Khoruts A, Geis E and Maldonado J (2017). McDonough-Means S, Pollard EL, Roux S, Sadowsky MJ, Lipson KS, Sullivan MB, Caporaso JG, Krajmalnik-Brown R (2017). Microbiota transfer therapy alters gut ecosystem and improves strointestinal and autism symptoms: An open-label study. *Microbiome*, **5**(1): 10.

Kildahl AN, Bakken TL and Helverschou SB (2025). Pharmacological treatment of irritability and aggression in autism spectrum disorder: A systematic review. *J Child Adolesc Psychopharmacol*., **35**(1): 12–24.

Kolevzon A, Mathewson KA and Hollander E (2006). Selective serotonin reuptake inhibitors in autism: A review of efficacy and tolerability. *J Clin Psychiatry*, **67**(3): 407–414.

Leisman G and Melillo R (2025). Autism spectrum disorders: A review of prevalence, etiology and epidemiology. *Neurol Int*., **17**(1): 1–18.

Lerner MD, Botha M and Morris SL (2023). Collaborative and neurodiversity-affirming autism research. *Autism*, **27**(4):903–910.

Lord C, Rutter M and Le Couteur A (1994). Autism Diagnostic Interview–Revised: A revised version of a diagnostic interview for caregivers of individuals with possible pervasive developmental disorders. *J Autism Dev Disord*., **24**(5): 659–685.

McCracken JT, McGough J, Shah B, Cronin P, Hong D, Aman MG, Arnold LE, Lindsay R, Nash P, Hollway J and McDougle CJ (2002). Risperidone in children with autism and serious behavioral problems. *N Engl J Med*., **347**: 314–321.

McDougle CJ, Scahill L, McCracken JT, Tierney E, Davies M, Arnold LE, Posey DJ, Martin A, Ghuman JK, Shah B, Chuang SZ, Swiezy NB, Gonzalez NM, Hollway J, Koenig K, McGough JJ, Ritz L and Vitiello B (2003). Research units on pediatric psychopharmacology autism network: Risperidone for the core symptom domains of autism. *Am J Psychiatry*, **160**(6): 1142–1148.

McQueen JM and Heck AM (2002). Secretin for the treatment of autism. *Ann Pharmacother*., **36**: 305-311.

Muller CL, Anacker AMJ and Veenstra-VanderWeele J (2016) The serotonin system in autism spectrum disorder: From biomarker to animal models. *Neurosci*., **321**: 24-41.

Nagaraj R, Singhi P and Malhi P (2006). Risperidone in children with autism: Randomized, placebo-controlled, double-blind study. *J Child Neurol*., **21**: 450-445.

Sanders SJ, He X, Willsey AJ, Ercan-Sencicek AG, Samocha KE, Cicek AE, Murtha MT, Bal VH, Bishop SL, Dong S and Goldberg AP (2015). Insights into autism spectrum disorder genomic architecture and biology from 71 risk loci. Neuron., **87**(6): 1215-33.

Satterstrom FK, Kosmicki JA, Wang J, Breen MS, De Rubeis S, An JY, Peng M, Collins R, Grove J, Klei L and Stevens C (2020). Large-scale exome sequencing study implicates both developmental and functional changes in the neurobiology of autism. *Cell*, **180**(3): 568–584.

Sterrett K, Lerner MD and Shattuck PT (2023). Long-term outcomes in autism spectrum disorder: Quality of life and community participation. *Autism*, **27**(1): 15–28.

Uzunova G, Hollander E and Shepherd J (2016). The role of ionotropic glutamate receptors in childhood neurodevelopmental disorders: Autism spectrum

disorders and fragile X syndrome. *Curr Neuropharmacol*., **14**(3): 206–226.

Vargas DL, Nascimbene C, Krishnan C, Zimmerman AW and Pardo CA. (2005). Neuroglial activation and neuroinflammation in the brain of patients with autism. *Ann Neurol. J*, **57**: 67-81.

Yamasue H, Okada T, Munesue T, Kuroda M, Fujioka T, Uno Y, Matsumoto K, Kuwabara H, Mori D, Okamoto Y and Yoshimura Y (2020). Effect of intranasal oxytocin on the core social symptoms of autism spectrum disorder: A randomized clinical trial. *Mol psychiatry*, **25**(8): 1849-58.