Supplementary material

Table 1S – Inclusion and Exclusion Criteria (PICO eligibility criteria**)**

Criteria	**Included**	**Excluded**
Population	Children and adolescents (≤18 years) with ASD diagnosed by DSM or ICD criteria	Adults only or non-human studies
Interventions	Any medical or pharmacological intervention	Behavioral, educational, or psychotherapeutic interventions only
Comparators	Placebo, standard care, or other pharmacological treatments	None
Study Design	RCTs, quasi-experimental, cohort studies, meta-analyses	Case reports, editorials, narrative reviews
Language	English	Non-English publications

Table 2s – Search Strategy conducted in the review

Database	Search Terms	Filters Applied	Date of Search
PubMed	("Autism Spectrum Disorder"[MeSH] OR autism OR ASD) AND ("pharmacological treatment" OR "medical therapy" OR "biomedical therapy" OR "antipsychotics" OR "vitamin B12" OR methylcobalamin OR "vitamin D" OR "omega-3 fatty acids" OR "probiotics" OR "stem cell therapy")	English, Humans, Age ≤18 years	October 2025
Scopus	TITLE-ABS-KEY (autism OR ASD) AND (pharmacological OR biomedical OR "vitamin D" OR risperidone OR melatonin OR "L-carnitine")	English	October 2025
Cochrane Library	autism AND (treatment OR pharmacologic OR biomedical OR vitamin OR probiotic)	No date restriction	October 2025

Table 3S: Risk of Bias Tools

Study Type	Risk of Bias Tool	Domains Evaluated
Randomized Controlled Trials	Cochrane Risk of Bias 2.0	Randomization, blinding, incomplete data, selective reporting
Observational Studies	Newcastle–Ottawa Scale (NOS)	Selection, comparability, outcome
Overall Certainty	GRADE Approach	Risk of bias, inconsistency, indirectness, imprecision, publication bias

Table 4 S: Summary of Key Risperidone Studies in Pediatric ASD

	Study (Author, Year)	**Study Design**	**Population (N, Age Range)**	**Intervention Details**	**Key Outcomes Measured**	**Main Risperidone Findings (vs. Control/Baseline)**
1	McCracken et al., 2002 (RUPP) [9]	8-week, multisite RCT	N=101 (Ris:49, Pla:52), 5–17 years	Risperidone (0.5–3.5 mg/d) vs. Placebo	**ABC-I (Primary)**, CGI-I (Primary), Other ABC subscales, Adverse Events (AEs), Weight	**Significantly reduced ABC-I** (57% vs 14% reduction, P<0.001); Higher CGI-I response (69% vs 12%, P<0.001); Improved ABC Stereotypy & Hyperactivity; Increased weight gain (2.7 vs 0.8 kg, P<0.001), appetite, somnolence, fatigue,

						drooling, dizziness.
2	Shea et al., 2004 [10]	8-week RCT	N=79 (Ris:40, Pla:39), 5-12 years	Risperidone (0.01-0.06 mg/kg/d) vs. Placebo	ABC, CGI, CARS, CBCL, AEs	**Significantly reduced ABC Irritability, Hyperactivity, Stereotypy**; Improved CGI scores; Increased weight gain, somnolence.
3	RUPP Autism Network, 2005 [9,13]	4-month open-label + 2-month discontinuation	N=63 (extension), N=32 (discontinuation)	Continued Risperidone / Placebo substitution	ABC-I, CGI-I, AEs	**Benefit maintained at 6 months** in ~68% of initial responders; Discontinuation led to significant worsening of ABC-I compared to continued treatment.
4	Luby et al., 2006 [11]	8-week RCT	N=24 (Ris:12, Pla:12), 2.5–6 years	Risperidone (0.5-1.5 mg/d) vs. Placebo	ABC, CARS, CGI, Safety measures	**Trend towards improved ABC-I** (not statistically significant);

						Increased somnolence, appetite, weight gain.
5	Nagaraj (Ravishankar) et al., 2006 [12]	8-week RCT	N=39 (Ris:19, Pla:20), 5–12 years	Risperidone (0.5–1 mg/d) vs. Placebo	ABC, CGI, AEs	**Significantly reduced ABC-I** and other subscales; Higher CGI-I response; Increased somnolence and appetite.
6	Aman et al., 2008 [18]	8-week RCT (Cognitive Substudy)	N=38 (Ris:20, Pla:18), 5–17 years	Risperidone (0.5–3.5 mg/d) vs. Placebo	Cognitive tests (attention, learning, coordination, memory)	**No detrimental cognitive effects**; Potential improvement in cancellation task (attention) and verbal learning (recognition).
7	Kent et al., 2013 [19]	6-month Open-Label Extension	N=79 (5–17 years	Risperidone (maximum weight-based	ABC-I, CGI-I, Safety (Weight, Prolactin, AEs)	**Sustained improvement** in ABC-I and CGI-I; Continued weight gain,

				dose of 1.25 mg/ day or 1.75 mg/ day)		elevated prolactin; Somnolence, fatigue common but decreased over time.
8	Aman et al., 2015 [13]	~21-month Naturalistic Follow-up	N=84 (from RUPP RCT), 5–17 years at entry	Risperidone (mean 2.47 mg/d in treated) vs. No Risp	ABC-I, CGI-S, VABS, AEs (Weight, EPS, Enuresis, Lab tests)	**Continued use in 67%**; Lower ABC-I in current users; Improved VABS Social Skills in current users; Associated with weight gain, increased appetite, and **enuresis**; No increase in EPS.
9	Hongkaew et al., 2015 [23]	Cross-sectional	N=147, 3–19 years	Risperidone (0.1–6 mg/d)	Prolactin levels, Clinical factors	**Hyperprolactinemia in 44.9%**; Prolactin levels significantly higher with

						higher doses (P=0.002); No association with duration.
10	Scahill et al., 2016 [20]	24-week RCT (Med vs. Med+Parent Training)	N=124, Mean age 6.9 years	Risperidone monotherapy (data analyzed together)	Weight, Waist, BMI z-score, Fasting labs (Glucose, Insulin, Lipids, HbA1c, ALT, Adiponectin, Leptin)	**Significant weight gain** (mean 5.4 kg), increased waist, increased BMI z-score (faster in <7 yrs, those with early appetite increase); Significant increases in insulin, HOMA-IR, glucose, HbA1c, ALT, leptin; Decreased adiponectin; **Significant increase in Metabolic Syndrome prevalence** (8.4% to 22.6%).

11	Vanwong et al., 2017 [22]	Case-Control	N=127 (Ris) + N=76 (Naïve), 3–20 years	Risperidone (≥12 months) vs. Risperidone-Naïve	Serum Uric Acid, Metabolic parameters	**Higher uric acid** in Ris group (5.70 vs 5.35 mg/dl, P=0.01); 57.5% prevalence of hyperuricemia in Ris group; Associated with dose, duration, age, BMI, TG/HDL-C, insulin, HOMA-IR, leptin, hs-CRP, adiponectin.
12	Vanwong et al., 2020 [21]	Cross-sectional	N=134, Median age 10 years	Risperidone (≥3 months)	BMI (IOTF criteria), HTR2C/ABCB1 genotypes	**Higher prevalence of overweight/obesity** vs. general population (P<0.01); Longer duration significantly associated with overweight/o

						besity; No significant association found for tested HTR2C/ABCB1 polymorphisms after correction.
13	Kloosterboer et al., 2021 [24]	24-week Prospective Observational	N=42, 6–18 years	Risperidone (flexible dose)	PK (Concentrations), BMI z-score, Prolactin, Sedation, ABC-I	**Higher sum trough concentrations predicted higher BMI z-score (P<.001), higher prolactin (P<.001), more sedation (P<.05), and greater ABC-I reduction (P<.01)**.
14	Kouhbanani et al., 2021 [17]	3-month RCT (+ 3-month Follow-up)	N=43 (Ris:15, Ris+VR:15, Ctrl:13), 6–12 yrs	Risperidone vs. Ris + VR vs. Control	Vineland Social Skills, CARS (Behavioral Symptoms)	**Improved Social Skills & Behavioral Symptoms at 3 months** vs Control (P<0.001);

						Benefit relapsed at follow-up (unlike combined Ris+VR group).
15	Al-Huseini et al., 2022 [15]	Retrospective Cohort (2 years)	N=95, 3–18 years	Risperidone (mean ~1 mg/d)	CGI-I, BMI, AEs (Somnolence, EPS, Prolactin)	**79% response rate (CGI-I)**; Better response in those without family history of ASD (P<0.001); Significant BMI increase in males (P=0.001); Somnolence common (43%), higher in females (P=0.003); Hyperprolactinemia associated with dose (P=0.048).

16	Mohamed et al., 2023 [16]	1-year RCT	N=180 (Ris:60, HBOT:60, Pla:60), 5–8 yrs	Risperidone (0.25-0.5 mg/d x 6mo) vs HBOT vs Placebo	CARS, ATEC (Total & Subscales)	**Significant improvement in CARS and ATEC** scores vs baseline (P<0.001); Better improvement than Placebo (P<0.001 for CARS diff); Less improvement than HBOT (P=0.014 for CARS diff).

This table provides a summary of 16 key studies investigating the use of risperidone in children and adolescents with Autism Spectrum Disorder (ASD). The studies range from randomized controlled trials (RCTs) to open-label extensions and observational studies, covering various durations and populations. Overall, the findings consistently demonstrate risperidone's efficacy in reducing irritability, aggression, and other behavioral symptoms associated with ASD. However, significant side effects, including weight gain, metabolic changes, and hormonal alterations, have also been reported across multiple studies.

ABC: Aberrant Behavior Checklist, ABC-I: Aberrant Behavior Checklist-Irritability, ABCB1: ATP Binding Cassette Subfamily B Member 1, ADHD: Attention Deficit Hyperactivity Disorder, AEs: Adverse Events, ALT: Alanine Transaminase, ASD: Autism Spectrum Disorder, ATEC: Autism Treatment Evaluation Checklist, BMI: Body Mass Index, CARS: Childhood Autism Rating Scale, CBCL: Child Behavior Checklist, CGI: Clinical Global Impressions, CGI-I: Clinical Global Impressions-Improvement, CGI-S: Clinical Global Impressions-Severity, Ctrl: Control, EPS:

Extrapyramidal Symptoms, HbA1c: Hemoglobin A1c, HBOT: Hyperbaric Oxygen Therapy, HDL-C: High-Density Lipoprotein Cholesterol, HOMA-IR: Homeostatic Model Assessment of Insulin Resistance, hs-CRP: high-sensitivity C-Reactive Protein, HTR2C: 5-Hydroxytryptamine Receptor 2C, IOTF: International Obesity Task Force, mg/d: milligrams per day, mg/kg/d: milligrams per kilogram per day, N: Number of participants, P: Probability value (p-value), PK: Pharmacokinetics, Pla: Placebo, Ris: Risperidone, RCT: Randomized Controlled Trial, RUPP: Research Units on Pediatric Psychopharmacology, TG: Triglycerides, VABS: Vineland Adaptive Behavior Scales, VR: Virtual Reality.

Table 5S: Different studies of Aripiprazole in Children with ASD with Pooled Meta-analysis.

Feature	**Study Design**	**Population**	**Key Efficacy Findings**	**Key Safety & Tolerability Findings**	**TDM & Pharmacogenetics**
Marcus et al. 2009 [31]	8-week, fixed-dose RCT	N=218, 6-17 yrs	**Significant reduction in ABC-I** vs. placebo at all doses (5, 10, 15 mg). 52% CGI-I response vs 14% placebo.	**Significant weight gain** (mean +1.6kg vs +0.8kg). Most common AEs: sedation, fatigue, vomiting, increased appetite. **Prolactin decreased.**	N/A in this paper.
Owen et al. 2009 [30]	8-week, flexible-dose RCT	N=98, 6-17 yrs	**Significant reduction in ABC-I**	**Significant weight gain** (mean +2.1kg	N/A in this paper.

			vs. placebo (mean change -18.9 vs -12.7).	vs +1.1kg). Most common AEs: somnolence, increased appetite, fatigue. **Prolactin decreased.**	
Ichikawa et al. 2017 [32]	8-week, flexible-dose RCT (Japan)	N=87, 6-17 yrs	**Significant reduction in ABC-I** vs. placebo (mean change -14.1 vs -7.5).	**Significant weight gain** (mean +1.7kg vs +0.6kg). Most common AEs: somnolence (27.9%), increased appetite (18.6%).	N/A in this paper.
Ichikawa et al. 2018 [33]	52-week, Open-Label Extension (OLE)	N=80, 6-17 yrs	**Efficacy (ABC-I reduction) sustained** at 52 weeks.	**Continued weight gain** (mean +4.6kg from baseline). Common AEs: somnolence (42.5%), nasopharyngitis (41.3%).	N/A in this paper.

Hermans et al. 2023 [26]	24-week, Observational PK Study	N=24, 6-18 yrs	**No significant association** found between drug concentration and effectiveness (ABC-I).	**Higher sum trough concentrations predicted higher BMI z-scores (P<.001)** and **higher HbA1c (P=.03)**.	TDM may increase *safety* (not efficacy). PK model covariates: Albumin, BMI. Genotypes (CYP2D6, 3A4, 3A5, ABCB1) determined.
Hesapcioglu et al. 2020 [34]	Retrospective Cohort (Ari vs Ris)	N=47, 4–18 yrs	Both drugs reduced ABC scores.	Weight gain was most common for both (50% Ari, 59.3% Ris). Insomnia more common with Aripiprazole.	N/A in this paper.
Sugimoto et al. 2021 [36]	Post-hoc Pooled Analysis (Japan)	N=215 (from 2 Japanese RCTs)	N/A (Methodology paper)	N/A	N/A
POOLED ANALYSIS (3 RCTs)	**Meta-analysis**	**N=403 (Ari: 256, Pla: 147)**	**Pooled MD ABC-I: -5.71** (95% CI: -8.38, -3.04)	**Pooled MD Weight Gain: +0.98 kg** (95% CI: 0.64, 1.33). **Pooled RR Somnolence: 2.89** (95% CI: 1.62, 5.14).	N/A

				Pooled RR Fatigue: 4.17 (95% CI: 1.34, 12.98). **Pooled RR Increased Appetite: 2.88** (95% CI: 1.13, 7.33).	

This table comprehensively summarizes key clinical studies investigating the efficacy and safety of aripiprazole for treating irritability and associated behavioral symptoms in children and adolescents with Autism Spectrum Disorder (ASD). It includes data from pivotal randomized controlled trials (RCTs) such as Marcus et al. (2009), Owen et al. (2009), and Ichikawa et al. (2017), which consistently demonstrate significant reductions in irritability (as measured by ABC-I) compared with placebo. The table also highlights findings from long-term extension studies (Ichikawa et al., 2018) showing sustained efficacy, and observational studies (Hermans et al., 2023; Hesapcioglu et al., 2020) providing insights into real-world effectiveness and safety. Crucially, a pooled meta-analysis of three RCTs confirms the robust efficacy of aripiprazole (pooled MD ABC-I: -5.71) but also underscores the significant risk of adverse events, particularly weight gain, somnolence, fatigue, and increased appetite. The inclusion of pharmacokinetic (PK) and pharmacogenetic data (Hermans et al., 2023) adds depth to precision medicine considerations for aripiprazole.

3A4: Cytochrome P450 3A4, 3A5: Cytochrome P450 3A5, ABC: Aberrant Behavior Checklist, ABC-I: Aberrant Behavior Checklist-Irritability, ABCB1: ATP Binding Cassette Subfamily B Member 1, AEs: Adverse Events, Ari: Aripiprazole, ASD: Autism Spectrum Disorder, BMI: Body Mass Index, CGI-I: Clinical Global Impression-Improvement, CI: Confidence Interval, CYP2D6: Cytochrome P450 2D6, HbA1c: Glycated Hemoglobin, MD: Mean Difference, N: Number of participants, N/A: Not Applicable, OLE: Open-Label Extension, P: Probability value (p-value), PK:

Pharmacokinetics, Pla: Placebo, RCT: Randomized Controlled Trial, Ris: Risperidone, RR: Relative Risk (Risk Ratio), TDM: Therapeutic Drug Monitoring.

Table 6S: Cannabis-Based Medicine Studies in Pediatric ASD

Study (Author, Year)	**Design**	**Population (N, Age Range)**	**Intervention**	**Duration**	**Key Efficacy Findings**	**Key Safety Findings**
Bar-Lev Schleider et al. (2019). [48]	Observational (Prospectively Collected Data)	N=188, (2015-2017)	20:1 CBD: THC oil	6 months	**Positive:** 30.1% "significant improvement," 53.7% "moderate improvement" (global assessment).	**AEs:** Well-tolerated. Most common was restlessness (6.6%).
Barchel et al. (2019) [49]	Observational (Prospective Follow-up)	N=53, 4–22 years	CBD oil	66 days (median)	**Positive:** Parent-reported improvement in rage (67.6%), hyperactivity (68.4%), sleep (71.4%), anxiety (47.1%).	**AEs:** Mild. Mostly somnolence and change in appetite.
Silva Junior et al. (2022) [51]	**RCT (Parallel)**	N=60, 5–11 years	CBD-rich extract vs.	12 weeks	**POSITIVE TRIAL: Significant improvement** vs. placebo in Social Interaction	**AEs:** Few (9.7% in CBD group). Included

			Placebo		(p=0.0002), Anxiety (p=0.016), and Agitation (p=0.003).	dizziness, insomnia, colic, and weight gain.
Mazza et al. 2024 [50]	Retrospective, Observational Cohort	N=30, 5–18 years	33:1 CBD:THC extract	N/A	**Positive:** Significant improvement in communication, attention, eye contact, aggression, and irritability.	**AEs:** "Minimal untoward effects."
Sannar et al. (2025) [52]	**Protocol Paper**	(N=?) 7–14 years	CBD vs. Placebo	27 weeks (crossover)	**(Not Applicable)** - This paper describes the *methods* for a future trial.	**(Not Applicable)**

The table summarizes the emerging clinical evidence for cannabis-based medicine (CBM) in pediatric Autism Spectrum Disorder (ASD). The data, primarily driven by observational studies, consistently suggests a beneficial effect, with parent and clinician reports showing significant or moderate global improvement in behavioral symptoms, including rage, hyperactivity, anxiety, and aggression, across a majority of participants. Crucially, the table includes a positive Randomized Controlled Trial (RCT) by Silva Junior et al. (2022), which provides higher-level evidence supporting the use of CBD-rich extracts to improve social interaction, anxiety, and agitation compared to placebo. Overall, these interventions, typically using high CBD: THC ratios, appear well-tolerated, with Adverse Events (AEs) generally being mild and limited primarily to somnolence and restlessness. The collective evidence suggests CBM is a promising, low-side-effect option for severe behavioral issues in ASD.

However, the field is still in the early stages, with many protocols and long-term safety data yet to be established.

AEs, Adverse Events, **ASD,** Autism Spectrum Disorder, **CBD,** Cannabidiol, **N,** Number of participants, **RCT,** Randomized Controlled Trial, **THC,** Tetrahydrocannabinol, **yrs,** years.

Table 7S: Sertraline Studies in Pediatric ASD

Study (Author, Year)	**Design**	**Population (N, Age Range)**	**Sertraline Dose**	**Duration**	**Key Efficacy Findings**	**Key Safety Findings**
Steingard et al. (1997) [54]	Open-Label Trial (OLT)	N=21, 6–18 years	25–150 mg/day (flexible)	12 weeks	**76% "improved"** on CGI; reductions in aggression, repetitive behaviors.	**AEs:** Motor activation (29%), insomnia (19%), GI upset (14%).
AlOlaby et al. (2017) [55]	**DB-RCT (PGx Efficacy)**	N=57, 24–72 months	Flexible (up to 200mg/d implied)	12 weeks	**No significant difference** vs. placebo on CGI-I, ABC-I, or CY-BOCS-PDD. **PGx: L/L genotype** (SLC6A4) predicted *response* (P=.04); S-carriers did *worse*.	*See Potter 2019 for safety data from this trial.*
Potter et al. (2019) [57]	**DB-RCT (PGx**	N=58, 24–72 months	Flexible (stand	12 weeks	*See AlOlaby 2017 for efficacy data from this trial.*	**Significantly more ADRs** vs.

	Safety)		ard dose)			placebo (82.8% vs 48.3%, P=.004). **AEs:** Hyperactivity, insomnia, stereotypy, decreased appetite. **PGx:** CYP2C19 Poor Metabolizers had more ADRs.
Alolaby et al. (2020) [56]	**DB-RCT**	N=149, 3–6 years	Low-dose (mean 4.6 mg/d)	6 months	**No significant difference** vs. placebo on CGI-I, ABC-I, or Vineland.	**Well-tolerated.** No significant difference in common AEs (GI, sleep, activation) vs. placebo.

ABC-I: Aberrant Behavior Checklist–Irritability subscale; ADRs: Adverse Drug Reactions; AEs: Adverse Events; ASD: Autism Spectrum Disorder; CGI: Clinical Global Impression scale; CGI-I: Clinical Global Impression–Improvement subscale; CY-BOCS-PDD: Children's Yale-Brown Obsessive Compulsive Scale modified for

Pervasive Developmental Disorders; CYP2C19: Cytochrome P450 2C19 (a drug-metabolizing enzyme); DB-RCT: Double-Blind, Randomized Controlled Trial; GI: Gastrointestinal; L/L: Homozygous "long" allele genotype for the SLC6A4 gene; N: Number of participants; OLT: Open-Label Trial; P: P-value (a measure of statistical significance); PDD-NOS: Pervasive Developmental Disorder–Not Otherwise Specified; PGx: Pharmacogenetics; S-carriers: Individuals with at least one "short" allele (L/S or S/S) for the SLC6A4 gene; SLC6A4: Gene encoding the serotonin transporter (SERT); SSRI: Selective Serotonin Reuptake Inhibitor; vs.: Versus.

Table 8S: Fluoxetine Studies in Pediatric ASD

Study (Author, Year)	**Design**	**Population (N, Age Range)**	**Fluoxetine Dose**	**Duration**	**Primary Outcome**	**Efficacy Finding (vs. Placebo)**	**Key Safety Findings**
Fatemi et al. (1998) [60]	Open-Label, Retrospective	N=7, 9–20 years	20–80 mg/day	1.3–32 mos	ABC Subscales	**Positive:** Significant improvement in ABC-Lethargy (P<0.029).	Hyperactivity, vivid dreams, appetite suppression.
DeLong et al. (2002) [61]	Open-Label, Observational	N=129, 2–8 years	0.15–0.5 mg/kg	5–76 mos (mean 32)	Clinical Response	**Positive:** 69% had "excellent" or "good" response. (No placebo). *Response correlated with*	5 children developed bipolar disorder.

						family hx of affective disorder.	
Hollander et al. (2005) [62]	**RCT (Crossover)**	N=45, 5–17 years	Mean 9.9 mg/day	16 weeks (8-wk phases)	CY-BOCS Compulsion Subscale	**POSITIVE:** Fluoxetine superior to placebo (P<0.03).	No significant difference in AEs vs. placebo at this low dose.
Chantiluke et al. (2015) [63]	RCT (fMRI Crossover)	N=18 (ASD), 10.5-17.5 years	Acute Dose	2 sessions	mPFC Activation	**Biologically Active:** Normalized mPFC under-activation seen in ASD.	N/A (fMRI study)
Herscu et al. (2020). (SOFIA Study) [58]	**RCT (Parallel)**	N=158, 5–17 years	Mean 11.8 mg/day	14 weeks	CY-BOCS-PDD Total Score	**NEGATIVE:** No significant difference (P>0.05). Placebo had slightly greater mean improvement.	High rates of activation in *both* groups (42% FLX vs 45% Pla).
Reddihough et al.	**RCT (Parallel)**	N=146, 7.5–18 years	Max 20-30 mg/day	16 weeks	CY-BOCS-PDD	**NEGATIVE/EQUIVOCAL:** (P=.03 in primary model;	High dropout rate (41% in

(2019) [59].					Total Score	P=.21 in adjusted model). Authors conclude null findings.	FLX). AEs included agitation.
POOLED (Herscu & Reddihough)	**Meta-Analysis**	**N=304, 5-18 years**	**N/A**	**14–16 weeks**	CY-BOCS-PDD Total Score	**NEGATIVE** (Pooled MD: -0.18, 95% CI: -1.33 to 0.97; P=0.76)	(See individual trials)

Table 8S summarizes the evidence for the use of **Fluoxetine (FLX)**, a Selective Serotonin Reuptake Inhibitor (**SSRI**), in treating symptoms in pediatric Autism Spectrum Disorder (**ASD**). The evidence is mixed, highlighting a significant shift in understanding. Early open-label and small crossover trials (Fatemi et al., 1998; Hollander et al., 2005) suggested positive effects, primarily on specific repetitive behaviors or lethargy. However, the most extensive and most methodologically rigorous recent trials, including the SOFIA Study (Herscu et al., 2019) and Reddihough et al. (2019), were NEGATIVE, finding no significant difference between fluoxetine and placebo on the primary outcome measure for compulsive symptoms (CY-BOCS-PDD). A pooled meta-analysis of these major RCTs reinforces the null finding. This evidence strongly suggests that SSRIs are not effective for the core restricted and repetitive behaviors (RRBs) of ASD. Safety data indicate that hyperactivity and behavioral activation are common Adverse Events (**AEs**), leading to high dropout rates in some trials.

ABC, Aberrant Behavior Checklist, **ABC-Lethargy,** Aberrant Behavior Checklist-Lethargy, **AEs,** Adverse Events, **ASD,** Autism Spectrum Disorder, **CY-BOCS,** Children's Yale-Brown Obsessive Compulsive Scale, **CY-BOCS-PDD,** Children's Yale-Brown Obsessive Compulsive Scale-Pervasive Developmental Disorders version, **FLX,** Fluoxetine, **fMRI,** functional Magnetic Resonance Imaging, **hx,** history, **MD,**

Mean Difference, **mg/day,** milligrams per day, **mg/kg,** milligrams per kilogram, **mos,** months, **mPFC,** medial Prefrontal Cortex, **N,** Number of participants, **P,** Probability value (p-value), **Pla,** Placebo, **RCT,** Randomized Controlled Trial, **vs.,** versus, **wk,** week, **yrs,** years.

Table 9S: Valproate (Divalproex) Studies in Pediatric ASD

Study (Author, Year)	Design	Population (N, Age Range)	Valproate Dose	Duration	Primary Outcome	Efficacy Finding (vs. Placebo)	Key Safety Findings
Hollander et al.	DB-RCT (Crosso	N=13, (Mean age ~9)	Flexible	8 weeks	Repetitive Beha	POSITIVE: Significant improvement on	Not detailed

(2006) [79]	ver implied)				viors (C-YBOCS	C-YBOCS (p=0.037).	in the abstract.
Hollander et al. (2010) [78]	DB-RCT (Parallel)	N=27, (Mean age 9.46)	Flexible (blood levels)	12 weeks	Irritability (ABC-I & CGI-I)	POSITIVE: • Significant improvement on ABC-I (p=0.048). • Significant CGI-I response (62.5% vs 9%, p=0.008).	3 discontinuations for side effects/lack of efficacy (2 DVP, 1 Pla).
Aliyev & Aliyev (2018) [80]	DB-RCT (Parallel)	N=100 (50 DVP, 50 Pla)	20–50 mg/kg (syrup)	12 weeks	Global Severity (CGI-I)	POSITIVE: • Significant CGI-I response (80% vs 12%). • RR for response = 1.5.	"Side effects were not observed."
Carta et al. (2024) [81]	Case Report & Lit Review	N=1, (11 years)	IV-VPA (Intravenous)	Acute	Acute Aggression / Agitation	Positive: IV-VPA was "safe and prompt" after first-line therapies failed.	Well-tolerated.

Table 9S summarizes studies investigating the use of Valproate (Divalproex or DVP), a known mood stabilizer and anti-epileptic, in pediatric Autism Spectrum Disorder (ASD). The included studies, though small, suggest a positive trend in DVP's effectiveness in managing specific challenging behaviors. Two double-blind Randomized Controlled Trials (DB-RCTs) by Hollander et al. (2010) and Aliyev & Aliyev (2018) reported significant improvements in primary outcome measures,

specifically irritability (ABC-I) and Global Severity (CGI-I). An earlier small crossover trial (Hollander et al., 2006) also found improvement in Repetitive Behaviors. These findings support the consideration of valproate as a second-line option for severe irritability or aggression, especially in cases where antipsychotics are poorly tolerated or in the presence of associated mood lability. However, the sample sizes are small, and detailed safety reporting is limited, making it essential to remember DVP's known risks (e.g., hepatotoxicity, weight gain, teratogenicity) and the need for therapeutic drug monitoring. The case report also highlights its role in the management of acute agitation (IV-VPA).

ABC-I, Aberrant Behavior Checklist-Irritability, **ASD,** Autism Spectrum Disorder, **C-YBOCS,** Children's Yale-Brown Obsessive Compulsive Scale, **CGI-I,** Clinical Global Impression-Improvement, **DB-RCT,** Double-Blind Randomized Controlled Trial, **DVP,** Divalproex, **IV-VPA,** Intravenous Valproate, **kg,** kilogram, **mg/kg,** milligrams per kilogram, **N,** Number of participants, **P,** Probability value (p-value), **Pla,** Placebo, **RCT,** Randomized Controlled Trial, **RR,** Relative Risk, **vs.,** versus, **yrs,** years.

Table 10S: Methylphenidate RCTs in Pediatric ASD

Study (Author, Year)	**Design**	**Population (N, Age Range)**	**MPH Dose**	**Duration**	**Primary Outcomes**	**Efficacy Findings (vs. Placebo)**	**Key Safety Findings**
Quintana et al. (1995) [90]	DB, Crossover	N=10, 7–11 years	10 mg or 20 mg bid	Short-term	Hyperactivity	POSITIVE: Modest but statistically significant improvement in hyperactivity.	No significant side effects or worsening of stereotypies.

Handen et al. (2000) [91]	DB, Crossover	N=13, 5.6–11.2 years	0.3 & 0.6 mg/kg	Short-term	Conners Hyperactivity Index, CARS, ABC	POSITIVE (for 8/13): 50% decrease in Conners. Decreased stereotypy/speech. NEGATIVE on CARS.	Tolerability issues: Significant AEs (social withdrawal, irritability) at 0.6 mg/kg dose.
Posey et al. (2007). (RUPP) [92]	DB, Crossover	N=66, Mean age 7.5	.125, .25, .5 mg/kg (TID)	4 weeks (1 wk/dose)	SNAP-IV (ADHD/ODD), CY-BOCS (Repetitive)	POSITIVE: Significant improvement in Hyperactivity & Impulsivity. NEGATIVE: No significant effects on ODD or repetitive behaviors.	Abstract does not detail AEs.
Jahromi et al. (2009). (RUPP Substudy) [94]	DB, Crossover	N=33, 5–13 years	(Same as Posey 2007)	4 weeks	Observational measures (Social communication, self-regulation)	POSITIVE: Significant improvement in joint attention, response to bids, and self-regulation.	(See Posey 2007 / Sturman 2017)

Pearson et al. (2013) [93]	DB, Crossover	N=24, Mean age 8.8	4 dose levels (ER + IR)	Short-term	Parent/ Teacher ratings (ADHD)	POSITIVE: Significant declines in hyperactive, impulsive, inattentive, and oppositional behavior. Improved social skills.	Well-tolerated: Side effects similar to typical ADHD. No exacerbation of stereotypies.
Pooled analysis	Meta-Analysis of 5RCTs	N=146	N/A	1 week (per arm)	ADHD & ASD symptoms	POOLED POSITIVE: for Hyperactivity (Teacher SMD -0.78, P<0.001). POOLED NEGATIVE: for Social Interaction (P=0.07) & Stereotypy (P=0.19).	POOLED AE: Increased risk of Reduced Appetite (RR 8.28, P<0.001).

Table 10S summarizes the randomized controlled trials (RCTs) for Methylphenidate (MPH) in children with Autism Spectrum Disorder (ASD) and co-occurring ADHD symptoms. The consistent finding across these crossover trials (Quintana et al., 1995; Handen et al., 2000; Posey et al., 2007) is that MPH is effective in reducing core ADHD symptoms, specifically hyperactivity and impulsivity, as confirmed by the pooled meta-analysis (Teacher SMD -0.78, P<0.001). However, a critical finding is the poor tolerability compared to its use in neurotypical children. High-dose levels often lead

to significant Adverse Events (AEs) like social withdrawal and irritability (Handen et al., 2000), which are key dose-limiting factors in this population. Furthermore, the pooled analysis showed no significant benefit on core ASD symptoms (Social Interaction or Stereotypy), confirming its role as a treatment for comorbidity, not the core disorder. The most common pooled adverse event was Reduced Appetite (RR 8.28).

ABC, Aberrant Behavior Checklist, **ADHD,** Attention-Deficit/Hyperactivity Disorder, **AEs,** Adverse Events, **ASD,** Autism Spectrum Disorder, **bid,** twice daily, **CARS,** Childhood Autism Rating Scale, **Crossover,** a study design where participants receive both drug and placebo at different times, **CY-BOCS,** Children's Yale-Brown Obsessive Compulsive Scale, **DB,** Double-Blind, **ER,** Extended Release, **IR,** Immediate Release, **mg/kg,** milligrams per kilogram, **MPH,** Methylphenidate, **N,** Number of participants, **N/A,** Not Applicable, **ODD,** Oppositional Defiant Disorder, **P,** Probability value (p-value), **RCT,** Randomized Controlled Trial, **RR,** Relative Risk (Risk Ratio), **RUPP,** Research Units on Pediatric Psychopharmacology, **SMD,** Standardized Mean Difference, **SNAP-IV,** Swanson, Nolan, and Pelham Rating Scale IV, **TID,** three times daily, **wk,** week, **yrs,** years.

Table 11S: Atomoxetine Studies in Pediatric ASD:

Study (Author, Year)	Design	Population (N, Age Range)	Atomoxetine Dose	Duration	Primary Outcome	Efficacy Finding (vs. Placebo)	Key Safety Findings
Arnold et al. (2006) [96]	**DB-RCT (Crossover)**	N=16, 5–15 years	Clinically titrated	6 weeks (per arm)	**ABC-Hyperactivity**	**POSITIVE:** Significant improvement on ABC-Hyperactivity (P=.043).	Tolerable; no increase in stereotypy. 1 rehospitalization for violence.
Zeiner et al. (2011) [98]	Open-Label Trial (OLT)	N=14, 7–17 years	1.2–1.4 mg/kg/day	10 weeks	ADHD-RS, CGI	**Positive:** 7/14 were responders. Significant reduction in parent/teacher ADHD-RS. (No placebo).	Well-tolerated. Most common AEs: nausea, headache. 2 dropouts.
Harfterkamp et al. (2012) [95]	**DB-RCT (Parallel)**	N=97, 6–17 years	1.2 mg/kg/day	8 weeks	**ADHD-RS**	**POSITIVE:** Significant improvement on ADHD-RS	Generally well-tolerated. Most common

						(MD -6.7, P<.001). **NEGATIVE** on CGI-I (P=0.14).	AEs: nausea, decreased appetite, fatigue.
Eslamzadeh et al. (2018) [97]	**DB-RCT (Adjunct)**	N=44, 6–17 years	0.5–1.2 mg/kg/day	8 weeks	**CARS, CGI**	**POSITIVE (Adjunct):** Significant improvement in CGI and CARS total score (P≤0.05) vs. Placebo+Risperidone.	AEs: Mood change, irritability, GI disturbance; "tend to subside."
POOLED (Arnold & Harfterkamp)	**Meta-Analysis**	**N=113**	**N/A**	**6–8 weeks**	**ADHD/Hyperactivity**	**POSITIVE (Pooled SMD: -0.68, 95% CI: -1.06 to -0.31; P=0.0004)**	(See individual trials)

Table 11S reviews the key clinical trials for **Atomoxetine**, a selective norepinephrine reuptake inhibitor, used to treat ADHD symptoms in pediatric Autism Spectrum Disorder (**ASD**). The evidence from these studies is consistently **POSITIVE**, establishing atomoxetine as an effective non-stimulant option. Large randomized controlled trials (**RCTs**), such as Harfterkamp et al. (2012), reported significant improvements in the ADHD-RS score, findings strongly supported by the pooled meta-analysis (SMD: -0.68, P=0.0004). The efficacy is primarily targeted at hyperactivity and inattention. Furthermore, atomoxetine appears to be generally well-tolerated, with common Adverse Events (**AEs**) like nausea, decreased appetite, and

fatigue, but without the high rates of irritability and social withdrawal often seen with stimulants. Eslamzadeh et al. (2018) also suggest its potential as an adjunct therapy to risperidone, enhancing global and ASD symptom improvement. Overall, atomoxetine provides a valuable alternative for children with ASD who cannot tolerate or do not respond adequately to traditional stimulants.

ABC, Aberrant Behavior Checklist, **ABC-Hyperactivity,** Aberrant Behavior Checklist-Hyperactivity, **ADHD,** Attention-Deficit/Hyperactivity Disorder, **ADHD-RS,** Attention-Deficit/Hyperactivity Disorder Rating Scale, **AEs,** Adverse Events, **ASD,** Autism Spectrum Disorder, **CARS,** Childhood Autism Rating Scale, **CGI,** Clinical Global Impressions, **CGI-I,** Clinical Global Impressions-Improvement, **CI,** Confidence Interval, **DB-RCT,** Double-Blind Randomized Controlled Trial, **GI,** Gastrointestinal, **MD,** Mean Difference, **mg/kg/day,** milligrams per kilogram per day, **N,** Number of participants, **N/A,** Not Applicable, **OLT,** Open-Label Trial, **P,** Probability value (p-value), **RCT,** Randomized Controlled Trial, **SMD,** Standardized Mean Difference, **vs.,** versus, **wk,** week, **yrs,** years.

Table 12S: Clonidine Studies in Pediatric ASD

Study (Author, Year)	Design	Population (N, Age Range)	Clonidine Dose	Duration	Primary Outcome	Efficacy Finding (vs. Placebo)	Key Safety Findings
Fankhauser et al. (1992) [100]	DB, Crossover RCT	N=9, 5–33 years	~0.005 mg/kg/day (Transdermal)	4 weeks (per arm)	Ritvo-Freeman Scale, CGI	**POSITIVE:** Significant improvement on social, affectual, and sensory subscales, and on CGI.	**AEs:** Sedation and fatigue (most common, esp. first 2 weeks).
Jaselskis et al. (1992) [101]	DB, Crossover RCT	N=8, (Mean age 8.1)	10 mg or 20 mg bid (oral)	Short-term	ABC, Conners, ADD-CTRS, CGI	**MODEST/ MIXED:** • **Positive:** Parent (Conners) & Teacher (ABC) ratings improved. • **Negative:** Clinician	**AEs:** Drowsiness and decreased activity.

						(CGI) & other Teacher (ADD-CTRS) ratings did not.	
Ming et al. (2008) [102]	Open-Label, Retrospective	N=19, (Children)	Not specified	Not specified	Sleep, Behavior (Parent Report)	**POSITIVE (Open-label):** • Effective for sleep initiation & maintenance. • Less effective for hyperactivity, mood, and aggression.	Side effects were "largely tolerable."

Table 12S summarizes early clinical research on Clonidine, an alpha-2 adrenergic agonist, in children with Autism Spectrum Disorder (ASD). The evidence, primarily from small, double-blind, crossover Randomized Controlled Trials (DB, Crossover RCTs) conducted in the early 1990s, suggests a modest positive effect. Clonidine showed significant improvement in areas such as social interaction and sensory symptoms (Fankhauser et al., 1992) and demonstrated benefit on parent/teacher ratings of hyperactivity (ABC and Conners). However, the therapeutic effect appears inconsistent, as clinician ratings (CGI) did not constantly improve (Jaselskis et al., 1992). An important finding is its efficacy as a sleep aid (Ming et al., 2008). The primary Adverse Event (AE) across all studies is sedation and drowsiness, which can be a therapeutic benefit for sleep or a limiting side effect during the day. Overall, clonidine

is supported as a second-line option for co-occurring hyperactivity, hyperarousal, or sleep initiation difficulties in ASD.

ABC, Aberrant Behavior Checklist, **ADD-CTRS,** Attention Deficit Disorder-Clinical Teacher Rating Scale, **AEs,** Adverse Events, **ASD,** Autism Spectrum Disorder, **bid,** twice daily, **CGI,** Clinical Global Impressions, **Crossover,** a study design where participants receive both drug and placebo at different times, **DB,** Double-Blind, **mg/kg/day,** milligrams per kilogram per day, **N,** Number of participants, **RCT,** Randomized Controlled Trial, **yrs,** years.

Table 13S: Guanfacine Studies in Pediatric ASD

Study (Author, Year)	**Design**	**Population (N, Age Range)**	**Dose**	**Duration**	**Primary Outcome**	**Efficacy Finding (vs. Placebo)**	**Key Safety Findings**
Handen et al. (2008) [104]	DB, Crossover RCT	N=11, 5–9 years	Max 3 mg/day	6 weeks	**ABC-Hyperactivity**	**POSITIVE:** Significant benefit on ABC-H (45% responders). No gains on other ABC subscales.	**AEs:** Drowsiness and irritability reported.
Scahill et al. (2015) [103]	DB, Parallel RCT	N=62 (30 GXR, 32 Pla), 5–14 years (Mean 8.5)	1–4 mg/day (Modal 3 mg/d)	8 weeks	**ABC-Hyperactivity**	**POSITIVE:** Significant 43.6% decline in ABC-H (vs 13.2% placebo; effect size=1.67).	**AEs:** Drowsiness, fatigue, decreased appetite. • Temporary decline in blood

						• **CGI-I Response:** 50% vs 9.4% placebo.	pressure/ pulse, returning near baseline by week 8.
Politte et al. (2018) [105] *(Secondary analysis of Scahill 2015)*	DB, Parallel RCT	N=62, 5–14 years	1–4 mg/ day	8 weeks	Oppositional Behavior (HSQ), Repetitive Behavior (CYBOCS-ASD), Anxiety (CASI), Sleep (CSHQ)	**POSITIVE:** • **Oppositional Behavior:** 44% decline vs 12% placebo (p=0.004). • **Repetitive Behavior:** 24% decline vs <1% placebo (p=0.01). **NEGATIVE:** • No difference for Anxiety or Sleep.	(See Scahill et al. 2015)

ABC-H: Aberrant Behavior Checklist - Hyperactivity subscale, **AEs:** Adverse Events, **CASI:** Child and Adolescent Symptom Inventory, **CGI-I:** Clinical Global Impression - Improvement scale, **CSHQ:** Children's Sleep Habits Questionnaire, **CYBOCS-ASD:** Children's Yale-Brown Obsessive Compulsive Scale - Modified for ASD, **DB:** Double-Blind, **GXR:** Guanfacine Extended-Release, **HSQ:** Home Situation Questionnaire, **N:** Number of participants, **Pla:** Placebo, **RCT:** Randomized Controlled Trial

Table 14S: Memantine Studies in Pediatric ASD

Study (Author, Year)	Design	Population (N, Age Range)	Memantine Dose	Duration	Primary Outcome(s)	Efficacy Finding (vs. Baseline or Placebo)	Key Safety Findings
Owley et al. (2006) [107]	Open-Label Trial	N=14, 3–12 years	0.4 mg/kg	8 weeks	Memory (Dot Learning), ABC, CGI	**MIXED:** • **Positive:** Improved Memory & ABC (Hyperactivity, Lethargy, Irritability). • **Negative:** No significant change in CGI-S or CGI-I response.	(Not detailed in abstract).
Chez et al. (2007) [108]	Open-Label, Observational	N=151, (Children)	2.5–30 mg/day (per summary)	21 months (recruitment)	CGI-I (Language, Behavior, Self-stim)	**POSITIVE (Open-label):** • Significant improvement in language and social behavior.	"No serious side effects." Agitation led to discontinuation in ~10%.
Ghaleiha et al. (2013) [109]	**Adjunctive DB-RCT**	(N=44 in abstract),	Up to 20 mg/day (adju	10 weeks	**ABC-C Irritability**	**POSITIVE (Adjunct):** • Memantine + Risperidone was significantly	"Generally well tolerated"; no significan

		(Children)	nct to Risperidone)			superior to Placebo + Risperidone for Irritability, Stereotypy, and Hyperactivity.	t difference in AEs vs. placebo group.

Table 14S summarizes the findings from clinical trials evaluating **Memantine**, an NMDA-receptor antagonist, for use in pediatric Autism Spectrum Disorder (**ASD**). The evidence, primarily from open-label and adjunctive double-blind randomized controlled trials (**DB-RCTs**), suggests a **positive and synergistic effect**. The primary mechanism of memantine is to restore the glutamate system balance, which is often implicated in ASD pathophysiology. Studies show that when used as an **adjunct** to risperidone (Ghaleiha et al. 2013), memantine significantly enhances the improvement in core symptoms like **Irritability**, **Stereotypy**, and **Hyperactivity** compared to risperidone alone. Open-label data also reported improvement in **language** and **social behavior**. While generally well-tolerated, with "no serious side effects," some studies reported **agitation** leading to discontinuation in a small percentage of patients. Overall, memantine shows promise as an **augmentation strategy** for challenging behaviors that have not fully responded to atypical antipsychotics.

ABC, Aberrant Behavior Checklist, **ABC-C,** Aberrant Behavior Checklist-Community, **AEs,** Adverse Events, **ASD,** Autism Spectrum Disorder, **CGI,** Clinical Global Impressions, **CGI-I,** Clinical Global Impressions-Improvement, **CGI-S,** Clinical Global Impressions-Severity, **DB-RCT,** Double-Blind Randomized Controlled Trial, **mg/day,** milligrams per day, **mg/kg,** milligrams per kilogram, **N,** Number of participants, **RCT,** Randomized Controlled Trial, **vs.,** versus, **yrs,** years.

Table 15S: D-cycloserine (DCS) Studies in Pediatric ASD

Study (Author, Year)	Design	Population (N, Age Range)	Intervention	Duration	Primary Outcome	Efficacy Finding (vs. Placebo or Baseline)	Key Safety Findings
Posey et al. (2004) [111]	Pilot Study (Open-label after placebo lead-in)	N=14 (completers=10), (ages 5.9-10.7 yrs)	DCS (Dose-ranging)	6 weeks (total)	ABC-Social Withdrawal, CGI	**POSITIVE (Pilot):** Significant improvement in ABC-Social Withdrawal and CGI.	"Well tolerated".
Minshawi et al. (2016) [112]	**DB-RCT (Parallel)**	N=67 (randomized), 5–11 years	50 mg DCS (weekly) + SST vs. Placebo + SST	10 weeks	**Social Responsiveness Scale (SRS)**	**NEGATIVE (Short-Term):** No significant difference vs. placebo on SRS (p=0.45) or any secondary outcomes.	Well tolerated.
Wink et al. (2017) [114]	**RCT (Long-Term Follow-up)**	N=60 (completers), 5–11 years	(Analysis of Minshawi 2016 cohort)	22 weeks (11 weeks post-treatment)	**SRS Total Score** (at 22 weeks)	**POSITIVE (Durability):** DCS group showed significantly greater *maintenance* of skills (lower SRS score) vs. placebo (p=0.042).	Well tolerated. Irritability was the most common AE, but equal in both groups.

Aye et al. (2021) [113]	Systematic Review	(Includes Minshawi 2016)	N/A	N/A	Social & Communication Skills	**NEGATIVE (Short-Term):** Confirms "low certainty evidence of little to no difference" at 1-week post-treatment.	

Table 15S outlines the research on D-cycloserine (DCS), an NMDA partial agonist, as a potential treatment for improving social and communication deficits in pediatric Autism Spectrum Disorder (ASD). Early pilot studies suggested positive effects on social withdrawal (Posey et al., 2004). However, the critical double-blind Randomized Controlled Trial (DB-RCT) (Minshawi et al., 2016) was NEGATIVE for the primary outcome measure, the Social Responsiveness Scale (SRS), when assessed immediately post-treatment. This suggests DCS is ineffective as a standalone agent for symptom reduction. A subsequent long-term follow-up of that cohort (Wink et al., 2017) yielded a more nuanced, positive result: the DCS group demonstrated significantly greater durability and maintenance of social skills learned during Social Skills Training (SST) up to 22 weeks post-treatment compared to the placebo group. This suggests DCS's therapeutic role may be as a cognitive enhancer or learning facilitator when paired with behavioral intervention, rather than a direct symptom reducer. DCS was consistently reported as well-tolerated.

ABC, Aberrant Behavior Checklist, **ABC-Social Withdrawal,** Aberrant Behavior Checklist-Social Withdrawal, **AE,** Adverse Event, **ASD,** Autism Spectrum Disorder, **CGI,** Clinical Global Impressions, **DB-RCT,** Double-Blind Randomized Controlled Trial, **DCS,** D-cycloserine, **N,** Number of participants, **N/A,** Not Applicable, **P,** Probability value (p-value), **Pilot,** Preliminary, small-scale study, **RCT,** Randomized Controlled Trial, **SRS,** Social Responsiveness Scale, **SST,** Social Skills Training, **vs.,** versus, **yrs,** years.

Table 16S: Baclofen (Arbaclofen) Studies in Pediatric ASD

Study (Author, Year)	Design	Population (N, Age Range)	Intervention	Duration	Primary Outcome	Efficacy Finding (vs. Placebo or Baseline)	Key Safety Findings
Erickson et al. (2014) [116]	Open-Label Trial	N=32, 5–17 years	**Arbaclofen** (monotherapy)	8 weeks	**ABC-Irritability**	**POSITIVE (Open-label):** Significant improvement in ABC-Irritability, Lethargy/Social Withdrawal, and SRS.	**AEs:** Agitation and irritability were most common.
Veenstra-VanderWeele et al. (2017) [117]	**DB-RCT (Parallel)**	N=150, 5–21 years	**Arbaclofen** (monotherapy) vs. Placebo	8 weeks	**ABC-Social Withdrawal/Lethargy**	**NEGATIVE:** No significant difference vs. placebo on primary outcome. *Positive signal on secondary (CGI-S) & post-hoc (Vineland) measures.*	**AEs:** Affect lability (11%) and sedation (9%) were most common.

Mahdavinasab et al. (2019) [118]	**DB-RCT (Adjunctive)**	N=64, 3–12 years	**Baclofen** + Risperidone vs. Placebo + Risperidone	10 weeks	ABC Subscales	**POSITIVE (for Hyperactivity):** Significant improvement in ABC-Hyperactivity (P<0.001) vs. control. **NEGATIVE:** No significant difference for other ABC subscales.	Reported as safe and efficacious.
Gao et al. (2021) [115]	Computational (In Silico)	N/A (Brain tissue data)	N/A	N/A	Drug Repositioning	**(Rationale):** Identified baclofen as a high-potential candidate drug for ASD based on gene network analysis.	N/A

Table 16S summarizes the clinical evidence for the use of **Arbaclofen** (an R-enantiomer of Baclofen and a potent GABA_{B} receptor agonist) in pediatric Autism Spectrum Disorder (**ASD**). The results are **mixed**, reflecting the ongoing challenge of targeting core symptoms in this heterogeneous population. An initial open-label trial (Erickson et al., 2014) showed broad **POSITIVE** effects on irritability, lethargy, and social withdrawal. However, the definitive large-scale DB-RCT (Veenstra-VanderWeele et al., 2017) was NEGATIVE for its primary outcome (ABC-Social Withdrawal/Lethargy), failing to show a significant difference relative to placebo. Notably, post-hoc analysis in that study suggested a potential positive signal on secondary measures, such as the **CGI-S**. Furthermore, Baclofen demonstrated

benefit as an **adjunctive** therapy, significantly improving **Hyperactivity** when combined with risperidone (Mahdavinasab et al., 2019). Safety concerns include **affect's lability** and **sedation**. The computational analysis provides a mechanistic rationale, positioning baclofen as a strong candidate for further investigation based on its role in normalizing E/I (Excitatory/Inhibitory) balance in ASD.

ABC, Aberrant Behavior Checklist, **AEs,** Adverse Events, **ASD,** Autism Spectrum Disorder, **CGI-S,** Clinical Global Impressions-Severity, **DB-RCT,** Double-Blind Randomized Controlled Trial, **GABA,** Gamma-Aminobutyric Acid, **N,** Number of participants, **N/A,** Not Applicable, **P,** Probability value (p-value), **RCT,** Randomized Controlled Trial, **SRS,** Social Responsiveness Scale, **vs.,** versus, **yrs,** years.

Table 17S: N-acetylcysteine (NAC) Studies in Pediatric ASD

Study (Author, Year)	Design	Population (N, Age Range)	NAC Dose	Duration	Primary Outcome(s)	Efficacy Finding (vs. Placebo or Baseline)	Key Safety Findings
Hardan et al. (2012) [120]	**DB-RCT (Monotherapy)**	N=33, 3.2–10.7 years	900–2700 mg/day (titrated)	12 weeks	**ABC-Irritability**	**POSITIVE:** Significant improvement in ABC-Irritability ($p < .001$) vs. placebo.	"Well tolerated with limited side effects".
Ghanizadeh et al. (2012) [123]	**DB-RCT (Adjunct)**	N=40, (Children)	1200 mg/day (adjunct to Risperidone)	8 weeks	**ABC-Irritability**	**POSITIVE (Adjunct):** Significant reduction in irritability ($p < 0.05$) vs. placebo. No change in core symptoms.	"Generally tolerated well." Most common AEs: constipation, increased appetite, fatigue.
Nikoo M et al.	**DB-RCT**	N=40, 4–12 years	600–900 mg/	10 weeks	**ABC-Irritability**	**POSITIVE (Adjunct):** Significant	"Generally well tolerate

(2015) [124]	**(Adjunct)**		day (adjunct to Risperidone)			reduction in Irritability (p=0.02) and Hyperactivity (p=0.01) vs. placebo.	d." No significant difference in AEs vs. placebo.
Wink et al. (2016) [121]	**DB-RCT (Monotherapy)**	N=31, 4–12 years	60 mg/kg/day	12 weeks	**CGI-I (Social Impairment)**	**NEGATIVE:** No significant difference vs. placebo ($p > 0.69$). Boosted glutathione (GSH) levels ($p < 0.05$).	"Well tolerated." AE frequency was too low to compare groups.
Dean et al. (2017) [122]	**DB-RCT (Monotherapy)**	N=102, 3.1–9.9 years	500 mg/day	6 months	SRS, CCC-2, RBS-R	**NEGATIVE:** No differences between NAC and placebo on any primary or secondary outcome measures.	No significant difference in AEs vs. placebo.
Lee et al. (2021) [119]	**Meta-Analysis**	5 RCTs	N/A	8–12 weeks	ABC, SRS, RBS-R	**POSITIVE (Pooled):** Significant improvement in **ABC-Total,**	"Safe and tolerable".

						Irritability, Hyperactivity, and **SRS-Social Awareness**. **NEGATIVE (Pooled):** No difference on RBS-R.	
Nalbant K, Erden S (2023) [125]	Retrospective Cohort	N=58 (37 NAC, 21 Ctl)	400–600 mg/day	8 weeks	CARS, ABC, CEBQ, CSHQ	**POSITIVE (Observational):** NAC group improved significantly on ABC-Irritability, Stereotypy, Hyperactivity, and CARS scores (social, verbal) vs. control.	No difference in eating or sleeping habits.
Dean et al., 2018) [127]	Qualitative	Small N	Various	N/A	Behavioral Symptoms	Generally **Positive (Low Evidence):** Reports of reduced irritability, stereotypy, and aggression; improved calmness and	Well tolerated.

						verbal communication.	

Table 17S presents the evidence for N-acetylcysteine (NAC), a precursor to the antioxidant glutathione (GSH), in pediatric Autism Spectrum Disorder (ASD). The hypothesis for NAC use centers on its ability to modulate the glutamate/glutamine cycle and restore redox balance, which is often impaired in ASD. The evidence is highly MIXED, reflecting conflicting results from the clinical trials. Studies investigating NAC as an adjunct to risperidone (Ghanizadeh et al., 2013; Nikoo et al., 2015) and an early monotherapy trial (Hardan AY et al., 2012) all reported POSITIVE findings, specifically showing significant reductions in Irritability (ABC-I) and Hyperactivity. This suggests a potential role for NAC in managing challenging behaviors. However, two subsequent, larger DB-RCTs of NAC as monotherapy (Wink LK et al., 2016; Dean OM et al., 2017) were NEGATIVE for their primary outcomes, finding no significant difference from placebo in social impairment (CGI-I, SRS) or repetitive behaviors (RBS-R). Despite these null findings, a recent meta-analysis of five RCTs reported a POOLED POSITIVE effect on global symptoms and specific behavioral subscales (ABC-Total, Irritability, Hyperactivity). In terms of safety, NAC is consistently reported as safe and well-tolerated, with minimal Adverse Events (AEs). The clinical conclusion is that NAC may serve as a safe, moderate adjunct for irritability and hyperactivity, but it is ineffective for core social or repetitive symptoms when used alone.

ABC, Aberrant Behavior Checklist, **ABC-I,** Aberrant Behavior Checklist-Irritability, **AEs,** Adverse Events, **ASD,** Autism Spectrum Disorder, **CARS,** Childhood Autism Rating Scale, **CCC-2,** Children's Communication Checklist-Second Edition, **CEBQ,** Child Eating Behavior Questionnaire, **CGI-I,** Clinical Global Impressions-Improvement, **CSHQ,** Children's Sleep Habits Questionnaire, **Ctl,** Control, **DB-RCT,** Double-Blind Randomized Controlled Trial, **GSH,** Glutathione, **mg/day,** milligrams per day, **mg/kg/day,** milligrams per kilogram per day, **N,** Number of participants, **NAC,** N-acetylcysteine, **N/A,** Not Applicable, **P,** Probability value (p-value), **RBS-R,** Repetitive Behavior Scale-Revised, **RCT,** Randomized Controlled Trial, **SRS,** Social Responsiveness Scale, **vs.,** versus, **wk,** week, **yrs,** years.

Table 18S: Prednisolone Studies in Pediatric ASD

Study (Author, Year)	Design	Population (N, Age Range)	Intervention	Duration	Primary Outcome	Efficacy Finding (vs. Placebo/Control)	Key Safety Findings
Duffy et al. (2014) [132]	Retrospective Cohort	N=44 (20 Steroid, 24 Ctl), 3–5 years	Corticosteroids	N/A	FMAER (EEG), Language, Behavior	**POSITIVE (Retrospective):** Significant improvement in auditory evoked response, language, and behavior.	"No lasting morbidity" reported.
Malek et al. (2020) [131]	**SB-RCT (Adjunctive)**	N=37, (Children)	Prednisolone (1 mg/kg) + Risperidone	12 weeks	**ABC-Irritability**	**POSITIVE (Adjunct):** Significant improvement in CARS, Irritability, Hyperactivity, Lethargy, Stereotypy.	No significant adverse events detected. Decreased inflammatory markers.
Brito et al. (2021) [129]	**DB-RCT (Monotherapy)**	N=38, 3–7 years	Prednisolone (1 mg/kg tapered)	24 weeks	**Language (ADL, ABFW)**	**MIXED/SPECIFIC:** Significant language improvement **only** in children <5 years with **regression**.	**AEs:** Hyperglycemia (n=5), Hypertension (n=2), Varicella

							(n=2). Considered "mild."
Figueiredo (2021) [133]	Case Report	N=1, 7 years	Acute Prednisolone	Acute	Motor Tics	**(Adverse Event):** Recurrence/worsening of motor tics.	Worsening of tics.

Table 18S summarizes the limited, yet mechanistically important, research on Prednisolone and other corticosteroids in pediatric Autism Spectrum Disorder (ASD). These interventions are typically used to modulate inflammation and immune dysregulation, which is hypothesized to contribute to ASD in a subset of children. The findings are MIXED and highly specific. A retrospective cohort (Duffy et al., 2014) showed a POSITIVE signal, with corticosteroids improving auditory evoked response (FMAER), language, and behavior. An adjunctive DB-RCT (Malek et al., 2020) demonstrated that adding prednisolone to risperidone significantly improved a range of behavioral symptoms (CARS, Irritability, Hyperactivity), suggesting a synergistic anti-inflammatory effect. However, a subsequent DB-RCT of prednisolone monotherapy (Brito et al., 2021) showed a very SPECIFIC positive finding: only children under 5 years old with a history of developmental regression benefited significantly in language outcomes. This suggests corticosteroids are not a broad treatment for ASD but may benefit a highly immunologically defined subgroup. The key safety concerns, including hyperglycemia and hypertension, highlight the need for cautious use and monitoring, which is typical for systemic steroids.

ABC, Aberrant Behavior Checklist, **ABC-I,** Aberrant Behavior Checklist-Irritability, **ABFW,** ABFW Language Test, **ADL,** Assessment of Developmental Level, **AEs,** Adverse Events, **ASD,** Autism Spectrum Disorder, **CARS,** Childhood Autism Rating Scale, **CGI-I,** Clinical Global Impressions-Improvement, **Ctl,** Control, **DB-RCT,** Double-Blind Randomized Controlled Trial, **EEG,** Electroencephalogram, **FMAER,** Frequency-Modulated Auditory Evoked Response, **mg/kg,** milligrams per kilogram, **N,** Number of participants, **N/A,** Not Applicable, **P,** Probability value (p-value), **RCT,**

Randomized Controlled Trial, **SB-RCT,** Single-Blind Randomized Controlled Trial, **vs.,** versus, **yrs,** years.

Table 19S: Pregnenolone Studies in ASD

Study (Author, Year)	**Design**	**Population (N, Age Range)**	**Intervention**	**Primary Outcome**	**Efficacy Finding**	**Key Safety Findings**
Ayatollahi et al. (2020) [135]	**DB-RCT (Adjunctive)**	N=64, Adolescents	Pregnenolone + Risperidone	**ABC-Irritability**	**POSITIVE:** Significant improvement in Irritability, Stereotypy, and Hyperactivity vs. Placebo.	No significant difference in AEs vs. Placebo.
Fung et al. (2014) [136]	Open-Label Pilot	N=12, Adults (Mean 22.5y)	Pregnenolone (Monotherapy)	ABC-Irritability	**POSITIVE (Open Label):** Significant improvement in Irritability, Lethargy, and Sensory Profile.	Well-tolerated. Mild fatigue/ GI reported.
McGrath et al. (2025) [137]	Pilot / Methodology	N=25, 14–25 years	Placebo Lead-in	Placebo Effect	**(Methodological Finding):** 30.2% reduction in irritability on Placebo alone.	N/A

Table 19S summarizes the initial clinical findings for Pregnenolone, a neurosteroid and precursor to various neuroactive hormones, in the treatment of Autism Spectrum Disorder (ASD). The evidence is currently preliminary but highly promising. An early open-label pilot (Fung et al., 2014) reported POSITIVE effects across multiple domains, including Irritability, Lethargy, and Sensory Profile. A subsequent Double-Blind

Randomized Controlled Trial (DB-RCT) (Ayatollahi et al., 2020) showed that Pregnenolone, when used as an adjunct to risperidone, produced significant improvement in challenging behaviors, including Irritability, Stereotypy, and Hyperactivity, compared to placebo. This suggests Pregnenolone may modulate neurochemical pathways to enhance the efficacy of existing medications. Importantly, the neurosteroid was consistently well-tolerated, with no significant difference in Adverse Events (AEs) compared with placebo. The inclusion of a methodology paper (McGrath et al., 2025) serves as an important caution, highlighting the substantial placebo effect (over 30% reduction in irritability) common in ASD trials, which underscores the necessity of the rigorous DB-RCT design.

ABC, Aberrant Behavior Checklist, **AEs,** Adverse Events, **ASD,** Autism Spectrum Disorder, **DB-RCT,** Double-Blind Randomized Controlled Trial, **GI,** Gastrointestinal, **N,** Number of participants, **N/A,** Not Applicable, **RCT,** Randomized Controlled Trial, **vs.,** versus, **y,** years.

Table 20S: Oxytocin Studies in Pediatric ASD

Study (Author, Year)	**Design**	**Population (N, Age Range)**	**Duration**	**Primary Outcome**	**Efficacy Finding (vs. Placebo)**	**Key Safety Findings**
Guastella et al.	DB Crossover	N=16, 12–19 years	Acute (1	Emotion Recogniti	POSITIVE: Improved emotion recognition performance.	(Not detailed)

(2010) [146]			dose)	on (RMET)		
Gordon et al. 2013 [137]	fMRI	N=17, (Children)	Acute	Brain Activity (fMRI)	POSITIVE (Neural): Increased activity in social brain regions (striatum, mPFC).	(Not detailed)
Dadds et al. (2014) [145]	DB RCT	N=38, 7–16 years	4 days	Social Interaction, Emotion Rec.	NEGATIVE: No significant improvement in social skills or emotion recognition.	(Not detailed)
Yatawara et al. (2016) [143]	DB Crossover	N=31, (Young children)	5 weeks	Social Responsiveness (SRS)	POSITIVE: Significant improvement in social responsiveness.	Well-tolerated. AEs: Thirst, urination, constipation.
Parker et al. (2017) [142]	DB Parallel	N=32, 6–12 years	4 weeks	Social Responsiveness (SRS)	POSITIVE: Significant improvement in SRS. Benefit predicted by low baseline oxytocin.	Well-tolerated.
Mayer et al. (2021) [148]	fMRI Crossover	N=25, (Male)	Acute	Empathy Neural Activation	MIXED/NEURAL: Increased amygdala response to pain. No other substantial modulation.	(Not detailed)
Sikich et al. (2021) [144]	DB Parallel	N=290, 3–17 years	24 weeks	ABC-mSocial Withdrawal	NEGATIVE: No significant difference vs. placebo on social or cognitive functioning.	Similar AE incidence

						to placebo.

Table 20S summarizes the research on Oxytocin, often called the "social neuropeptide," in pediatric Autism Spectrum Disorder (ASD). The evidence presents a pattern of highly promising acute and neurobiological findings that fail to translate into sustained clinical benefit in large-scale trials. Early and small-scale studies, including double-blind crossover trials (DB Crossover) and functional Magnetic Resonance Imaging (fMRI) studies, reported POSITIVE acute effects, such as improved emotion recognition (RMET) and increased activity in social brain regions like the mPFC. However, the definitive, largest, and longest duration randomized controlled trial (RCT) (Sikich et al. 2021, N=290) was NEGATIVE, finding no significant difference between Oxytocin and placebo on social or cognitive functioning over 24 weeks. This divergence suggests that while Oxytocin may acutely modulate neural pathways related to social processing, the effect is not robust or durable enough to clinically impact the core social withdrawal symptoms of ASD. Safety data across the studies generally indicates that Oxytocin is well-tolerated, with Adverse Events (AEs) being mild and similar in incidence to placebo.

ABC-mSW: Aberrant Behavior Checklist-modified Social Withdrawal, **AEs:** Adverse Events, **DB:** Double-Blind. **fMRI:** Functional Magnetic Resonance Imaging, **IU:** International Units, **mPFC:** Medial Prefrontal Cortex, **RCT:** Randomized Controlled Trial, **RMET:** Reading the Mind in the Eyes Task, **SRS:** Social Responsiveness Scale

Table 21: Vasopressin Agonists/Antagonist Studies in Patients with ASD

Study (Author, Year)	**Design**	**Population (N, Age Range)**	**Intervention**	**Primary Outcome**	**Efficacy Finding (vs. Placebo)**
Parker et al. (2019) [152]	DB-RCT (Parallel)	N=30, 6–12.9 years	Intranasal AVP	SRS-2 Total Score	POSITIVE: Significant improvement in social abilities (P=0.0052,

					d=1.40) and anxiety.
Hollander et al. (2022) [155]	DB-RCT (Phase 2)	N=339, 5–17 years	Balovaptan (Antagonist)	Vineland-II 2DC	NEGATIVE: No significant difference vs. placebo (P=0.91).
Jacob et al. (2022) [155]	DB-RCT (Phase 3)	N=322, Adults	Balovaptan (Antagonist)	Vineland-II 2DC	NEGATIVE: Terminated for futility. No improvement vs. placebo.
Bolognani et al. (2019) [156]	DB-RCT (Phase 2)	N=223, Men (Adults)	Balovaptan (Antagonist)	SRS-2	Mixed: Negative on primary (SRS-2), but positive on secondary Vineland-II scores.
Umbricht et al. (2017) [157]	DB-RCT (Crossover)	N=19, Men (Adults)	RG7713 (Antagonist)	Eye tracking / ASR	Mixed: Improved eye tracking; reduced emotion recognition.

Table 21S summarizes the key studies investigating the therapeutic potential of modulating the Vasopressin (AVP) system in patients with Autism Spectrum Disorder (ASD). The results highlight a critical example of the difficulty in translating biological rationale into clinical efficacy. While a small early trial (Parker et al. 2019) suggested that the AVP agonist (Intranasal AVP) could significantly improve social abilities, the large-scale clinical development program for the V1a receptor antagonist,

Balovaptan, was overwhelmingly NEGATIVE. Pivotal Phase 2 and Phase 3 double-blind randomized controlled trials (DB-RCTs) in both children and adults failed to show a significant difference from placebo on the primary outcome measure, the Vineland-II 2DC, leading to the termination of the Phase 3 trial due to futility. This evidence establishes that V1a receptor antagonism is not an effective treatment strategy for improving the core social deficits of ASD.

ASD, Autism Spectrum Disorder, **ASR,** Auditory Startle Response, **AVP,** Arginine Vasopressin, **DB-RCT,** Double-Blind Randomized Controlled Trial, **N,** Number of participants, **P,** Probability value (p-value), **RCT,** Randomized Controlled Trial, **SRS-2,** Social Responsiveness Scale-2, **Vineland-II 2DC,** Vineland Adaptive Behavior Scales, Second Edition, Two-Domain Composite, **yrs,** years.

Table 22S: Intravenous Immunoglobulin (IVIG) Studies in ASD

Study (Author, Year)	**Design**	**Population (N)**	**Intervention**	**Primary Outcomes**	**Efficacy Finding**	**Key Safety Findings**
Plioplys (1998) [166]	Open-Label	N=10	IVIG (monthly)	Behavior	Limited: 10% significant response; 40% mild; 50% no response.	(Not detailed)

Niederhofer et al. (2003) [165]	DB Crossover	N=12	IVIG vs. Placebo	ABC, Symptom Checklist	POSITIVE: Significant improvement in Irritability, Hyperactivity, Eye Contact, Speech ($p<0.05$).	AEs: Significantly more drowsiness and decreased activity vs. placebo.
Boris et al. 2005 [162]	Open-Label	N=26	IVIG (monthly)	ABC	POSITIVE: Reductions in hyperactivity, speech, and irritability. Relapse is common after stopping.	(Not detailed)
Connery et al. (2018) [163]	Case Series	N=31	IVIG	SRS, ABC	POSITIVE: Significant improvement in SRS and ABC. Bio-markers predicted the response.	AEs are common (62%) but transient. 6% discontinued.
Melamed et al. (2018) [167]	Pilot Study	N=14	High-dose IVIG	CCC-2, SRS, ABC	POSITIVE: Significant improvements in communication, social interaction, and stereotypy.	Well tolerated; no withdrawals due to AEs.
Rossignol & Frye (2021) [161]	Meta-Analysis	27 studies	IVIG	ABC	POSITIVE (Pooled): Significant improvement in Aberrant Behavior & Irritability (large effect).	AEs: Headache, fever, nausea,

						fatigue, rash.
Maltsev & Yevtushenko (2022) [155]	Controlled Cohort	N=78	High-dose IVIG	ABC	POSITIVE: "Complete elimination" of ASD phenotype in 21/78. Improvement in comorbidities.	(Not detailed)

Table 22S summarizes the clinical evidence for Intravenous Immunoglobulin (IVIG) in pediatric ASD, an intervention based on the hypothesis of underlying immune or inflammatory dysregulation. The data is consistently POSITIVE, with multiple open-label, case series, and a meta-analysis reporting significant improvements in challenging behaviors, including Irritability, Hyperactivity, and Communication (as measured by ABC, SRS, and CCC-2). One small double-blind crossover trial also reported significant benefit. The pooled meta-analysis indicates a large effect size for the reduction of aberrant behavior. However, the overall level of evidence is generally low certainty, as most positive reports stem from small, uncontrolled cohorts. Safety remains a significant consideration: Adverse Events (AEs) are reported as common (up to 62%), though often transient (e.g., headache, fever), and may lead to discontinuation in a minority of patients. The findings suggest that IVIG may be a viable treatment reserved for a specific, immunologically defined subgroup of children with ASD.

ABC, Aberrant Behavior Checklist, **AEs,** Adverse Events, **ASD,** Autism Spectrum Disorder, **CARS,** Childhood Autism Rating Scale, **CCC-2,** Children's Communication Checklist-Second Edition, **Crossover,** Crossover study design, **Ctl,** Control, **DB,** Double-Blind, **IVIG,** Intravenous Immunoglobulin, **N,** Number of participants, **SRS,** Social Responsiveness Scale.

Table 23S: Methylcobalamin & Folinic Acid Studies in ASD

Study (Author, Year)	Design	Population (N)	Intervention	Primary Outcome	Efficacy Finding (vs. Placebo or Baseline)	Key Safety/Biomarker Findings
Bertoglio et al. (2010) [177]	DB-RCT (Crossover)	N=30	Methyl B12 (SC)	Behavior, GSH	NEGATIVE: No significant difference in overall means. 30% were responders.	Responders had increased Glutathione (GSH).
Hendren et al. (2016) [175]	DB-RCT (Parallel)	N=57	Methyl B12 (SC)	CGI-I, ABC	MIXED: Positive on clinician-rated CGI-I (p=0.005). Negative on parent-rated ABC/SRS.	Improvement correlated with increased methylation capacity.
Frye et al. (2018) [181]	DB-RCT (Parallel)	N=48	High-Dose Folinic Acid	Verbal Communication	POSITIVE: Significant improvement in verbal communication.	Response predicted by Folate Receptor-α Autoantibodies.
James et al. 2009 [182]	Open-Label	N=40	Methyl B12 + Folinic Acid	Metabolites	POSITIVE (Metabolic): Normalized glutathione redox status and methylation markers.	(Biomarker study)
Frye et al.	Open-Label	N=37	Methyl B12 +	Adaptive Behavio	POSITIVE (Behavior): Significant gains in	(See James et al. 2009)

(2013) [183]			Folinic Acid	r (VABS)	adaptive behavior, correlated with metabolic improvement.	
Geier & Geier (2010) [180]	Cohort Study	N=72	Methyl B12 (SC)	Cobalt Levels	(Toxicity Concern): Injections significantly increased plasma/urinary cobalt levels.	Raised concern for cobalt toxicity.

Table 23S summarizes the research on supplementing with Methylcobalamin (Methyl B12) and High-Dose Folinic Acid, interventions designed to correct hypothesized deficits in methylation and folate metabolism in children with Autism Spectrum Disorder (ASD). The evidence is highly MIXED but strongly supports a precision medicine approach. Early open-label trials suggested broad metabolic and behavioral benefits. However, subsequent double-blind randomized controlled trials (DB-RCTs) for Methyl B12 monotherapy were inconsistent, showing either no difference in overall means or mixed results between clinician- and parent-rated outcomes. The most significant finding comes from the Folinic Acid RCT (Frye et al. 2018), which reported a significant POSITIVE effect on verbal communication, a benefit that was highly correlated with the presence of Folate Receptor-α Autoantibodies in the children. This suggests these therapies are not a blanket treatment but are effective for a specific, biologically defined subgroup with documented methylation/folate pathway abnormalities. Safety concerns were raised in one cohort study regarding the potential for elevated Cobalt levels following subcutaneous Methyl B12 injections.

ABC, Aberrant Behavior Checklist, **ASD,** Autism Spectrum Disorder, **CGI-I,** Clinical Global Impressions-Improvement, **Crossover,** Crossover study design, **DB-RCT,** Double-Blind Randomized Controlled Trial, **GSH,** Glutathione, **Methyl B12,** Methylcobalamin, **N,** Number of participants, **SC,** Subcutaneous, **SRS,** Social Responsiveness Scale, **VABS,** Vineland Adaptive Behavior Scales.

Table 24S: L-Carnitine Studies in Pediatric ASD

Study (Author, Year)	Design	Population (N)	Intervention	Primary Outcome	Efficacy Finding (vs. Placebo/Baseline)	Key Safety Findings
Shakibaei & Jelvani (2023) [187]	DB-RCT (Adjunctive)	N=50	L-Carnitine + Risperidone	ABC	POSITIVE: Significant reduction in ABC-Total and Lethargy/Social Isolation.	(Not detailed)
Nasiri et al. (2024) [188]	DB-RCT (Adjunctive)	N=68	L-Carnitine (150 mg) + Risperidone	ABC-Irritability	POSITIVE: Significant reduction in Irritability (P=0.033) and Hyperactivity (P<0.001).	Safe; no difference in AEs vs. placebo.
Hajizadeh-Zaker et al. (2018) [189]	DB-RCT (Adjunctive)	N=70	*L-Carnosine* + Risperidone	ABC-Irritability	MIXED: Positive for Hyperactivity (P=0.044). Negative for Irritability.	No significant difference in AEs vs. placebo.
Goin-Kochel et al.	Open-Label Pilot	N=10	High-Dose L-Carni	Safety/Behavior	POSITIVE (Trend): Improvements in Hyperactivity & Social	AEs: Heavy odor, diarrhe

(2019) [190]			tine (up to 400 mg/kg)		Communication (not sig. after correction).	a, vomiting. led to some dropouts.
Lv QQ et al. (2018) [186]	Case-Control	N=90	Biomarker Analysis	Acyl-carnitines	(Biomarker): ASD children had significantly lower levels of free and acyl-carnitines.	N/A

Table 24S summarizes the clinical evidence for L-Carnitine (and related L-Carnosine), a supplement critical for mitochondrial function and fatty acid transport, in pediatric Autism Spectrum Disorder (ASD). The evidence is largely POSITIVE when L-Carnitine is used as an adjunct to risperidone in double-blind randomized controlled trials (DB-RCTs). Two recent adjunctive RCTs reported significant reductions in challenging behaviors, including Irritability, Hyperactivity, and Lethargy/Social Isolation (ABC). These efficacy findings are supported by biomarker data showing that children with ASD have significantly lower free and acyl-carnitine levels compared to controls. This suggests a potential therapeutic role for L-Carnitine as an augmentation strategy for children with ASD, particularly those on atypical antipsychotics or with suspected mitochondrial dysfunction. L-Carnitine is reported to be safe and well-tolerated, though very high doses may cause temporary gastrointestinal (GI) side effects.

ABC, Aberrant Behavior Checklist, **ABC-I,** Aberrant Behavior Checklist-Irritability, **AEs,** Adverse Events, **ASD,** Autism Spectrum Disorder, **DB-RCT,** Double-Blind Randomized Controlled Trial, **GI,** Gastrointestinal, **mg,** milligrams, **mg/kg,** milligrams per kilogram, **N,** Number of participants, **N/A,** Not Applicable, **P,** Probability value (p-value), **RCT,** Randomized Controlled Trial, **vs.,** versus.

Table 25S: Coenzyme Q10 (Ubiquinol) Studies in Pediatric ASD

Study (Author, Year)	Design	Population (N)	Intervention	Primary Outcomes	Efficacy Finding (vs. Baseline)	Key Safety Findings
Mousavinejad et al. (2018) [196]	Open-Label	N=90	CoQ10 (30-60 mg)	Oxidative Stress, Symptoms	POSITIVE: Improved sleep (P=0.005) and GI problems (P=0.004). Reduced oxidative stress.	(Not detailed)
Gvozdjáková et al. (2014) [197]	Open-Label	N=24	Ubiquinol (100 mg)	Parent Report	POSITIVE: Improvements in communication, sleep, and food refusal.	*Expression of Concern issued regarding study ethics/validity.*
Legido et al. (2018) [199]	Open-Label Pilot	N=11	MitoCocktail (CoQ10 + Carnitine + ALA)	Mitochondrial function, ABC	POSITIVE: Significant improvement in Lethargy and Speech. Improvement waned post-treatment.	(Not detailed)
Cucinotta et al. (201) [201]	Retrospective Chart Review	N=59 (Mixed ND)	Ubiquinol + Vit E + B-Complex	CGI-I	POSITIVE: 76% responders. Improvements in cognition, adaptive function, social motivation.	Mild AEs: Increased hyperactivity (15%). No discontinuations.

Chen et al. (2021) [194]	Meta-Analysis	N=9109	Biomarkers	Oxidative Stress Markers	(Rationale): Confirmed oxidative stress (high GSSG, low GSH) in ASD, supporting antioxidant use.	N/A

Table 25S summarizes the studies exploring the use of Coenzyme Q10 (CoQ10) and its reduced form, Ubiquinol, in pediatric Autism Spectrum Disorder (ASD). These agents are central to mitochondrial function and act as powerful antioxidants, targeting the oxidative stress confirmed to be present in children with ASD by a meta-analysis. The evidence is derived solely from open-label trials and retrospective reviews and is therefore of low certainty. However, the findings are consistently POSITIVE, suggesting benefits in several associated symptoms, including improved sleep, gastrointestinal (GI) problems, communication, and social motivation. The observed improvement in behavior and language is often attributed to the intervention's ability to boost mitochondrial function and reduce oxidative damage. One open-label pilot study involving a MitoCocktail (CoQ10 + Carnitine + Alpha-Lipoic Acid) showed significant improvement in Lethargy and Speech, though the effect was not durable. Overall, while CoQ10/Ubiquinol appears safe and well-tolerated, rigorous double-blind randomized controlled trials (DB-RCTs) are necessary to confirm these preliminary efficacy signals.

ABC, Aberrant Behavior Checklist, **AEs,** Adverse Events, **ALA,** Alpha-Lipoic Acid, **ASD,** Autism Spectrum Disorder, **CGI-I,** Clinical Global Impressions-Improvement, **CoQ10,** Coenzyme Q10, **DB-RCTs,** Double-Blind Randomized Controlled Trials, **GI,** Gastrointestinal, **GSH,** Glutathione (Reduced form), **GSSG,** Glutathione disulfide (Oxidized form), **mg,** milligrams, **N,** Number of participants, **N/A,** Not Applicable, **ND,** Neurodevelopmental, **P,** Probability value (p-value), **Vit E,** Vitamin E.

Table 26S: Sulforaphane Studies in ASD

Study (Author, Year)	Design	Population (N)	Intervention	Primary Outcomes	Efficacy Finding (vs. Placebo)	Key Safety Findings
Singh et al. (2014) [202]	DB-RCT	N=44, 13–27 years	SFN (Monotherapy)	ABC, SRS, CGI-I	POSITIVE: Significant improvement in ABC (-34%), SRS (-17%), and Social Interaction.	Well-tolerated. Symptoms returned after stopping.
Momtazmanesh et al. (2020) [206]	DB-RCT (Adjunctive)	N=60, 4–12 years	SFN + Risperidone	ABC-Irritability	POSITIVE (Adjunct): Significant improvement in Irritability and Hyperactivity.	Safe; no difference in AEs vs. placebo.
Zimmerman et al. (2021) [204]	DB-RCT	N=57, 3–12 years	SFN (Monotherapy)	OACIS, ABC, SRS	MIXED: Primary (OACIS) not significant. Secondary (ABC) significantly improved. Biomarker correlations found.	Well-tolerated. Rare insomnia/irritability.
Ou et al. (2024) [203]	DB-RCT	N=108, 4–12 years	SFN (Monotherapy)	Clinician & Caregiver Scales	MIXED: Clinician ratings positive (significant improvement). Caregiver ratings negative (no change). Effect greater in age >10.	Safe and well-tolerated.
Magner et al. (2023) [205]	DB-RCT	N=40, 3–7 years	SFN (Month	ADOS-2, SRS-	NEGATIVE: No significant difference in behavioral outcomes for this young cohort.	(Not detailed)

			erapy)	2, ABC		
Lynch et al. (2017) [207]	Case Series	N=16 (Follow-up)	SFN Supplements	Qualitative Report	Positive (Long-term): Caregivers reported sustained benefits over 3 years.	N/A

Table 26S summarizes the clinical evidence for Sulforaphane (SFN), a compound derived from broccoli sprouts that acts as a potent Nrf2 pathway activator, in individuals with Autism Spectrum Disorder (ASD). The evidence is MIXED but consistently suggests a signal of therapeutic efficacy for core and associated symptoms. The initial definitive double-blind randomized controlled trial (DB-RCT) reported significant POSITIVE results on global assessment (CGI-I) and challenging behaviors (ABC, SRS). Subsequent large RCTs have been more nuanced, showing mixed outcomes: clinician ratings were often positive, but caregiver ratings were not (Ou et al., 2024), or secondary outcomes (ABC) improved, but the primary outcome (OACIS) did not (Zimmerman et al., 2021). SFN appears most beneficial in older children and adolescents, as one RCT in a young cohort (3–7 years) was NEGATIVE. As an adjunct to risperidone, SFN showed positive synergy in reducing irritability and hyperactivity. Crucially, SFN is consistently reported as safe and well-tolerated, with symptoms often returning when the treatment is stopped, indicating a need for continuous dosing.

ABC, Aberrant Behavior Checklist, **ABC-Irritability,** Aberrant Behavior Checklist-Irritability, **ADOS-2,** Autism Diagnostic Observation Schedule-2, **AEs,** Adverse Events, **ASD,** Autism Spectrum Disorder, **CGI-I,** Clinical Global Impressions-Improvement, **DB-RCT,** Double-Blind Randomized Controlled Trial, **N,** Number of participants, **OACIS,** Observer-rated Autism Clinical Impairment Scale, **RCT,** Randomized Controlled Trial, **SFN,** Sulforaphane, **SRS,** Social Responsiveness Scale, **SRS-2,** Social Responsiveness Scale-2, **vs.,** versus, **yrs,** years.

Table 27S: Bumetanide Studies in Pediatric ASD

Study (Author, Year)	Design	Population (N)	Intervention	Primary Outcome	Efficacy Finding (vs. Placebo)	Key Safety Findings
Lemonnier et al. (2012) [210]	DB-RCT	N=60	Bumetanide (1 mg)	CARS	POSITIVE: Significant reduction in CARS (P<0.004) and CGI.	Occasional mild hypokalemia.

Lemonnier et al. (2017) [211]	DB-RCT	N=88	Bumetanide (various doses)	CARS, SRS	POSITIVE: Significant improvement in CARS, SRS, and CGI. 1.0 mg BID optimal.	Hypokalemia, dehydration, and diuresis.
Sprengers et al. (2021) (BAMBI) [2018]	DB-RCT	N=92	Bumetanide (1 mg BID)	SRS-2	NEGATIVE: No difference on primary SRS-2. Positive for Repetitive Behaviors (RBSR).	Hypokalemia (51%), Orthostatic hypotension.
Dai et al. (2021) [212]	DB-RCT	N=120 (3-6 yrs)	Bumetanide (0.5 mg)	CARS	POSITIVE: Significant reduction in CARS and insular GABA levels.	Safe; no withdrawals due to AEs.
Fuentes et al. (2023) (SIGN 1 & 2) [217]	2 Phase 3 RCTs	N=422 (Total)	Bumetanide	CARS2	NEGATIVE: Trials were terminated early due to futility—no benefit vs. placebo.	Thirst, polyuria, and hypokalemia are more common.
Shaker et al. (2024) [214]	DB-RCT	N=80	Bumetanide	CARS	POSITIVE: Significant decrease in CARS scores ($p<0.001$).	Minimal and tolerable AEs.
Fernell et al. (2025) [216]	Wait list-RCT	N=15	Bumetanide	Parent Ratings	POSITIVE (Small): 4/9 completers showed significant improvement.	2 were excluded for behavioral problems.

Zhang (2020) [213]	RCT	N=83;	bumetanide treatment, 0.5 mg twice daily	CARS	POSITIVE: Clinical improvement correlated with reduced GABA/Glx ratio.	Mechanism Validated: Restored E/I balance.
Mollajani (2025) [221]	Pre-Post	N=15	1 mg daily (0.5 mg twice a day) for 3 months	ERP, CARS	POSITIVE: Improved emotion recognition and N170/N250 ERP components.	Mechanism Validated: Improved neural processing.
Juarez-Martinez (2023) [222]	EEG Analysis	N=82	1 mg daily (0.5 mg twice a day) for 91 days	EEG	(Prediction): EEG profiles predicted responders with 92% accuracy.	Efficacy is likely subgroup-specific.

Table 27S summarizes the pivotal, though highly MIXED, clinical research on Bumetanide, a diuretic that acts as an inhibitor to restore the theorized Excitatory/Inhibitory (E/I) imbalance in Autism Spectrum Disorder (ASD). Early small double-blind randomized controlled trials (DB-RCTs) in younger cohorts consistently reported POSITIVE findings, showing significant improvements in core symptoms (CARS, SRS, CGI) and supporting the mechanism by demonstrating a corresponding reduction in levels. However, the definitive, large-scale Phase 3 trials (SIGN 1 & 2) were subsequently terminated early due to futility (NEGATIVE), finding no benefit compared to placebo. This divergence suggests Bumetanide is not broadly effective across the ASD population. Crucially, recent studies are shifting focus to predictive biomarkers, showing that improvement correlates with changes in neural

activity (ERP, EEG) and metabolite levels (GABA/Glx ratio), indicating that efficacy is likely limited to a specific, biologically defined subgroup of children. Safety concerns are common, including Hypokalemia, thirst, and diuresis, necessitating cautious use and monitoring.

AEs, Adverse Events, **ASD,** Autism Spectrum Disorder, **BID,** twice daily, **BAMBI,** Bumetanide in AutisM British/International trial, **CARS,** Childhood Autism Rating Scale, **CARS2,** Childhood Autism Rating Scale-Second Edition, **CGI,** Clinical Global Impressions, **DB-RCT,** Double-Blind Randomized Controlled Trial, **EEG,** Electroencephalogram, **E/I,** Excitatory/Inhibitory, **ERP,** Event-Related Potential, **GABA,** Gamma-Aminobutyric Acid, **Glx,** Combined Glutamate, Glutamine, and GABA measurement (in magnetic resonance spectroscopy), **Hypokalemia,** Low potassium in the blood, **mg,** milligrams, **N,** Number of participants, **N170/N250,** Specific components of the Event-Related Potential waveform, **RBSR,** Repetitive Behavior Scale-Revised, **RCT,** Randomized Controlled Trial, **SIGN 1 & 2,** Names of specific Phase 3 clinical trials, **SRS,** Social Responsiveness Scale, **SRS-2,** Social Responsiveness Scale-2, **Waitlist-RCT,** Waitlist-Controlled Randomized Controlled Trial, **yrs,** years.

Table 28S: Spironolactone Studies in ASD

Study (Author, Year)	Design	Population	Intervention	Key Findings
Bradstreet et al. (2007) [227]	Review / Hypothesis	N=1 (Case Report)	Spironolactone	POSITIVE (Case): Clinical improvement in one child. Proposed utility for neuroinflammation & hyperandrogenism.
Zarate-Lopez et al. (2025) [225]	Preclinical (Mice)	VPA Model	Spironolactone (50 mg/kg)	POSITIVE (Animal): Reduced repetitive behaviors. Mechanism: Antagonism of ErbB4/mTOR pathway.
Mirza et al. (2023) [226]	Preclinical (Rats)	PPA Model	Spironolactone (25-50 mg/kg)	POSITIVE (Animal): Improved social behavior, anxiety, repetitive behavior, and restored BDNF & Synapsin II.
Majewska et al.	Case-Control	N=Prepubertal Children	Biomarker Analysis	(Rationale): Found elevated salivary

(2014) [228]				androgens in ASD, supporting the use of anti-androgens like spironolactone.

Table 28S summarizes the preliminary and mechanistic evidence for Spironolactone, an aldosterone and androgen antagonist, in Autism Spectrum Disorder (ASD). Currently, there are no completed large-scale randomized controlled trials (RCTs) in human subjects. The rationale for its use stems from two key biological hypotheses: 1) the role of neuroinflammation, and 2) the finding of elevated salivary androgens in prepubertal children with ASD. The early evidence is highly supportive at the preclinical (animal) level, where Spironolactone improved core symptoms such as repetitive behaviors and social behavior across multiple rodent models of ASD. Mechanistically, these effects were linked to antagonizing the ErbB4/mTOR pathway and restoring key neuroplasticity markers BDNF and Synapsin II. While an initial human case report suggested clinical improvement, the field requires rigorous double-blind RCTs to validate the promising mechanistic and animal findings before Spironolactone can be considered a therapeutic option in ASD.

ASD, Autism Spectrum Disorder, **BDNF,** Brain-Derived Neurotrophic Factor, **ErbB4,** Epidermal Growth Factor Receptor 4 (a receptor implicated in synaptic function), **mTOR,** mechanistic Target of Rapamycin (a pathway involved in cell growth and metabolism), **N,** Number of participants, **N/A,** Not Applicable, **PPA,** Propionic Acid (a rat model of ASD), **RCTs,** Randomized Controlled Trials, **VPA,** Valproic Acid (a mouse model of ASD).

Table 29S: Microbiota-Gut-Brain Axis Interventions: Probiotics & Prebiotics

Probiotics, Prebiotics, and Microbiota-Targeted Interventions in Children with ASD

Study (Author, Year)	Intervention / Comparator	Study Design	Population (N)	Primary Outcome / Target Symptom	Key Findings
Kang DW et al. (2019) [250]	Microbiota Transfer Therapy (FMT/MTT)	Open-label trial with 2-year follow-up	18 children with ASD (7–16 y)	GI symptoms; ASD severity (ATEC, CGI-I)	Significant and sustained improvement in GI symptoms and ASD-related behaviors; benefits persisted or further improved at 2 years; increased microbial diversity (↑ *Bifidobacteria*, *Prevotella*).
Arnold LE et al. (2019) [239]	VISBIOME® (8-strain probiotic) vs placebo	Randomized, placebo-controlled crossover pilot	13 children with ASD + GI symptoms (3–12 y)	GI symptoms (PedsQL-GI); QoL	Safe and feasible; significant improvement in parent-selected GI target symptoms; moderate effect sizes on QoL; microbiota

					changes correlated with symptom improvement.
Liu YW et al. (2019) [238]	*Lactobacillus plantarum* PS128 vs placebo	Randomized, double-blind, placebo-controlled trial	Boys with ASD (7–15 y)	Behavioral symptoms (ABC, SNAP-IV, SRS)	Improved opposition/defiance behaviors; significant improvement in SNAP-IV total score in younger children (7–12 y).
Li YQ et al. (2021) [236]	Probiotics + ABA vs ABA alone	Randomized controlled trial	41 children with ASD	Behavioral symptoms (ATEC); gut microbiota	Combination therapy significantly reduced ATEC scores vs ABA alone; increased beneficial bacteria (*Bifidobacterium*, *Lactobacillus*).
Zeng P et al. (2024) [230]	Probiotics vs placebo	Meta-analysis of 6 RCTs	302 children with ASD	ASD symptoms; GI symptoms	Significant reduction in GI symptom severity; no significant improvement in core ASD behavioral symptoms.

Liu Y et al. (2022) [253]	Probiotics / Prebiotics	Systematic review & meta-analysis	15 RCTs (N = 833)	Core ASD symptoms; GI symptoms	Overall limited and inconsistent efficacy for core ASD symptoms; modest benefits for GI symptoms and secondary behavioral issues (e.g., irritability, anxiety) in selected trials.
Rahim F et al. (2023) [231]	Probiotics / Prebiotics / Synbiotics	Meta-analysis of RCTs	720 children with ASD	Behavioral symptoms; neurophysiology	No significant effect on behavioral outcomes; significant modulation of brain connectivity correlated with inflammatory markers.

Table 29S summarizes clinical studies evaluating probiotics, prebiotics, and microbiota-targeted interventions in children with Autism Spectrum Disorder (ASD). The table includes randomized controlled trials, pilot studies, open-label trials, and systematic reviews/meta-analyses assessing the effects of microbiome-modulating therapies on gastrointestinal (GI) symptoms, core and associated ASD behavioral symptoms, quality of life, and inflammatory or neurophysiological outcomes. Only human studies included in the reviewed dataset are presented.

ASD: Autism Spectrum Disorder; ABA: Applied Behavior Analysis; ABC: Aberrant Behavior Checklist; ATEC: Autism Treatment Evaluation Checklist; CGI-I: Clinical

Global Impression–Improvement; EEG: Electroencephalography; FMT: Fecal Microbiota Transplantation; GI: Gastrointestinal; MTT: Microbiota Transfer Therapy; PedsQL: Pediatric Quality of Life Inventory; QoL: Quality of Life; RCT: Randomized Controlled Trial; SNAP-IV: Swanson, Nolan and Pelham Rating Scale–IV; SRS: Social Responsiveness Scale; TNF-α: Tumor Necrosis Factor-alpha; VPA: Valproic Acid.

Table 30S: The various studies concerned with using Fecal Microbiota Transplantation (FMT) in ASD

Study (Author, Year)	**Design**	**Population**	**Intervention**	**Efficacy Finding**	**Key Safety/Notes**
Wan L et al. 2024 [248]	DB-RCT	N=103	Oral FMT Capsules	NEGATIVE Primary: No diff on SRS-2. POSITIVE Secondary: Improved Vineland-3 Socialization.	Safe. Fever is the most common AE.
Wang L et al. 2024 [248]	DB-RCT	N= Not listed	FMT	POSITIVE: Significant improvement in GSRS, CARS, SRS, and ABC vs Placebo.	Reduced urinary 5-HIAA.
Kang et al. 2017,20	Open Label	N=18	MTT (Antibiotic +FMT)	POSITIVE (Long-term): 80% GI reduction. ASD symptoms improved further at the 2-year follow-up.	Increased *Bifidobacteria* & *Prevotella*.

19 [249, 250]					
Li Y et al. 2024 [253]	Prospective	N=98	Capsules vs. Tube	POSITIVE: Upper GI routes (Capsules/NJT) are superior to Lower GI (TET) for behavior.	Capsules had the fewest AEs (8.2%).
Ye C et al. 2022 [251]	Longitudinal	N=328	FMT	POSITIVE (Waning): Improvements sustained for 3-4 years, but returned to baseline by year 5.	No serious AEs.
Pan ZY et al. 2022 [242]	Retrospective	N= Various	Washed MT (WMT)	POSITIVE: Repeated courses yielded better ABC/Sleep scores than a single course.	Reduced systemic inflammation.

Table 30S examines the linical studies evaluating fecal microbiota transplantation (FMT) and microbiota transfer–based interventions in children with Autism Spectrum Disorder (ASD). The table includes randomized controlled trials and observational studies assessing different FMT delivery methods, efficacy on gastrointestinal and behavioral outcomes, durability of response, and safety profiles.

ABC, Aberrant Behavior Checklist, **AEs,** Adverse Events, **ASD,** Autism Spectrum Disorder, **CARS,** Childhood Autism Rating Scale, **DB-RCT,** Double-Blind Randomized Controlled Trial, **FMT,** Fecal Microbiota Transplantation, **GI,** Gastrointestinal, **GSRS,** Gastrointestinal Symptom Rating Scale, **MTT,** Microbiota Transfer Therapy, **N,** Number of participants, **NEGATIVE Primary,** The main outcome measure did not show a statistically significant difference from placebo, **NJT,** Nasojejunal Tube (an upper GI route of delivery), **POSITIVE Secondary,** A secondary outcome measure showed a statistically significant improvement, **SRS,** Social Responsiveness Scale, **SRS-2,** Social Responsiveness Scale-2, **TET,** Transendoscopic Tube (a lower GI route of delivery), **Vineland-3,** Vineland Adaptive Behavior Scales, Third Edition, **WMT,** Washed Microbiota Transplantation, **5-HIAA,** 5-Hydroxyindoleacetic acid