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What Can Neonatal Mouse Models of Neurodevelopmental Disorders Tell Us About the Development of Communication and Social Deficits in Children with ASD?

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ABSTRACT

Autism spectrum disorder (ASD) is a neurodevelopmental disorder whose underlying biological changes begin early in development, yet diagnosis typically occurs between ages 3 and 4, after many critical developmental processes have already taken place. This gap limits the ability of human research to investigate the mechanisms driving early communication and social deficits and to identify when intervention may be most effective. Neonatal rodent models provide experimental access to this early period.

This narrative review synthesizes human clinical studies and rodent studies identified through structured searches of PubMed and Google Scholar, supplemented by studies recommended by the faculty mentor. Rodent evidence was classified by the developmental stage at which a manipulation was applied and an outcome was measured, separating neonatal findings (postnatal days 0 to 14) from juvenile and adult findings. Four mechanistic themes shared by the two literatures were compared: excitation/inhibition (E/I) imbalance, serotonin, the developmental shift in gamma-aminobutyric acid (GABA) signaling, and oxytocin.

Neonatal evidence was strongest for oxytocin and GABA polarity, where manipulations in the perinatal period and first postnatal week altered later social behavior in several genetic and environmental models, and for serotonin, where loss of brain serotonin reduced pup ultrasonic vocalizations (USVs). Evidence for E/I imbalance came mainly from adult animals and differed in direction across models. Human data in these areas come largely from older children, and clinical trials of oxytocin and of the NKCC1 inhibitor bumetanide have produced mixed or negative results. Neonatal rodent models are therefore best used to test whether a specific circuit or developmental event is sufficient to alter early communication and social behavior, rather than as complete models of ASD. Differences in developmental timing between species, together with the heterogeneity of ASD, limit direct translation.

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INTRODUCTION

ASD is a neurodevelopmental disorder defined by two core domains: persistent deficits in social communication and interaction, and restricted, repetitive patterns of behavior (APA, 2013). Diagnosis requires that symptoms appear in the early developmental period and cause clinically significant functional impairment (APA, 2013). ASD is estimated to affect roughly 1% of the population and is diagnosed about four times more often in males than in females, although diagnosed females are more likely to also have intellectual disability (APA, 2013; Ey et al., 2011). This prevalence figure conceals substantial variation: individuals with ASD range from minimally verbal to highly verbal and from requiring full-time support to living largely independently (Ey et al., 2011).

The diagnostic category has also changed over time, as reflected in successive editions of the Diagnostic and Statistical Manual of Mental Disorders (DSM). The DSM-IV divided autism into Autistic Disorder, Asperger's Disorder, and pervasive developmental disorder not otherwise specified (PDD-NOS), whereas the DSM-5 consolidated these into a single diagnosis with severity specifiers based on support needs, because the earlier subtype boundaries were applied inconsistently and did not reliably predict outcomes (APA, 2013).

Biologically, ASD is as heterogeneous as its behavioral presentation. Neuropathological changes have been documented across nearly every brain region, from the brainstem and cerebellum to the limbic system and frontal cortex (Frye, 2018). Several neurotransmitter systems, including serotonin, dopamine, glutamate, and GABA, have been implicated (Frye, 2018). No single system appears to account for ASD on its own. In combination, these changes are thought to shift the cortex toward an excitation/inhibition (E/I) imbalance that may contribute to sensory hypersensitivity and, in some cases, seizures (Frye, 2018; Ey et al., 2011). Oxytocin adds a further layer as a modulator of the social circuits affected in ASD (Frye, 2018).

Because so many systems interact, mechanistic questions require a level of control that cannot be achieved in human studies, and animal models can partly fill this gap. Mouse studies allow researchers to isolate a single genetic variable, standardize the environment, and collect tissue at any developmental stage, none of which is possible in human research (Ellegood & Crawley, 2015). Mice are also social animals: they live in groups and communicate through touch, odor, and ultrasonic vocalizations (USVs) during both mother–infant and adult interactions (Ey et al., 2011). Both core ASD domains have established rodent counterparts, although these are behavioral measures rather than reproductions of the human condition. The three-chamber sociability test measures social approach, self-grooming assays measure repetitive behavior, and USV recordings measure vocal communication (Ellegood & Crawley, 2015; Ey et al., 2011). Tools such as structural MRI, diffusion tensor imaging, and immunohistochemistry can then link these behavioral differences to specific brain differences across dozens of mouse lines (Ellegood & Crawley, 2015).

Neonatal USV analysis is a central tool in this review. When separated from the mother, pups emit calls at 30 to 90 kHz that elicit maternal retrieval (Branchi et al., 2001). Because these calls are among the earliest measurable forms of social communication in mice, changes in their number and acoustic structure have been used as an early phenotype in mouse models of neurodevelopmental disorders (Scattoni et al., 2009). Pup calls are not equivalent to infant communication, but they provide a measurable proxy during a period that is otherwise difficult to study. Calling peaks around postnatal day (P) 6 to 7 and disappears by the end of the second postnatal week, providing a bounded window for tracking neonatal communication over time (Branchi et al., 2001). Serotonergic, dopaminergic, GABAergic, and opioid systems all modulate USV rate, so the measure functions as both a behavioral and a neurochemical index (Branchi et al., 2001).

Mouse models also have important limitations. Most studies to date have used only male animals because ASD is diagnosed more often in males; this should be regarded as a gap in the research rather than as evidence that female biology is the same (Ey et al., 2011). Genetic background also matters: the same mutation can produce different phenotypes depending on the inbred strain, as reported for Fmr1 and Nlgn3 lines (Ey et al., 2011; Ellegood & Crawley, 2015). Age is the most consequential limitation for the question addressed here. ASD symptoms in children are typically recognized between 12 and 24 months of age (APA, 2013), whereas most mouse studies characterize adult animals (Ey et al., 2011; Ellegood & Crawley, 2015). Consequently, most of what is known about ASD-relevant behavior in mice describes developmental end points rather than the processes that produced them, and findings in the adult mouse brain cannot be assumed to hold in the neonatal brain. The same caution applies across sex and strain (Ey et al., 2011).

Few reviews have examined human clinical research and neonatal rodent research side by side to identify where they agree, where they diverge, and which areas remain largely unexplored. This review addresses that gap by comparing four mechanistic themes that appear in both literatures: E/I imbalance, serotonin, the GABA developmental shift, and oxytocin. For each theme, neonatal rodent evidence is distinguished from evidence obtained in juvenile or adult animals. The review is organized around the following question: what can neonatal mouse models of neurodevelopmental disorders reveal about the development of communication and social deficits in children with ASD, and where are the limits of that translation?

METHODS

Search Strategy

This review was conducted as a narrative review with a structured literature search. It was completed by a single author and was not pre-registered. Literature was identified through searches of PubMed and Google Scholar conducted between June 2026 and August 2026, followed by an additional targeted search in October 2026, during revision, to locate original human studies and neonatal rodent studies. Rodent searches combined a population term (“autism” OR “autism spectrum disorder” OR “ASD”), a

species and age term ("mouse" OR "mice" OR "rodent") AND ("neonatal" OR "pup" OR "postnatal"), and a mechanism or outcome term ("ultrasonic vocalization" OR "oxytocin" OR "GABA" OR "NKCC1" OR "KCC2" OR "serotonin" OR "excitation/inhibition" OR "sociability"). Human searches combined "autism" OR "ASD" with "oxytocin levels," "intranasal oxytocin," "bumetanide," "folinic acid," "early markers," "motor skills," or "bilingual." Additional studies were identified through recommendations from the faculty mentor.

Inclusion and Exclusion Criteria

Titles and abstracts were screened for relevance, followed by full-text review of articles that appeared to meet the criteria in Table 1. No restriction on publication date was applied; included studies were published between 2001 and 2025, and the DSM-5 (APA, 2013) was used as the diagnostic reference.

Inclusion criteria	Exclusion criteria
Peer-reviewed original studies, meta-analyses, or reviews published in English	Publications not written in English
Rodent studies using a genetic or environmental model relevant to ASD (e.g., Fmr1, Magel2, Shank3, Tph2, VPA)	Conference abstracts without a full paper, preprints, and other non-peer-reviewed sources
Rodent studies reporting social behavior, repetitive behavior, USVs, or a mechanism within the four themes (E/I balance, serotonin, GABA polarity, oxytocin)	Non-rodent animal models (e.g., zebrafish, non-human primates)
Human studies of individuals with ASD reporting communication or social outcomes, biomarkers, or treatment response	Rodent studies without a social, communicative, repetitive-behavior, or mechanistic outcome relevant to the four themes
Diagnostic manuals and narrative reviews, used for background and definitions only	Case reports

Table 1: Inclusion and exclusion criteria for studies in this review.

Classification by Developmental Stage

Because the central question concerns early development, each rodent study was classified by two ages: the age at which a manipulation (genetic, pharmacological, or chemogenetic) was applied, and the age at which outcomes were measured. Three stages were defined: neonatal (P0 to P14, the period during which pups emit isolation calls), juvenile/adolescent (P15 to P60), and adult (older than P60). For genetic models, in which the mutation is present throughout life, the age of the measured outcome determined the classification. Findings were treated as direct evidence for neonatal mechanisms only when the manipulation or the outcome fell within the neonatal period. Findings from adult animals are reported separately and are used to describe how a system functions after development, not how it develops. Table

2 lists each rodent study with its classification. One study using rats (Tyzio et al., 2014) was included because it tested the same mechanism in a mouse model within the same paper.

Data Extraction and Synthesis

From each study, the following data were extracted: species and model, developmental stage of manipulation and measurement, outcome measures (for example, sociability, self-grooming, USV number or acoustic structure, and electrophysiological or molecular measures), and main findings. Studies were then grouped under the four mechanistic themes, and human and rodent evidence within each theme were compared. Because study designs and outcomes were heterogeneous, no meta-analysis or formal risk-of-bias scoring was performed. Throughout this review, rodent phenotypes are described in terms of the specific behavior measured, such as reduced sociability, increased self-grooming, or altered USV emission, rather than as "autistic-like" behavior, because mice do not reproduce ASD itself.

EARLY DEVELOPMENT AND DIAGNOSIS OF ASD IN CHILDREN

Early Neurobiological Changes

ASD is classified as a neurodevelopmental disorder, so the brain changes associated with it begin in early development, well before diagnosis (APA, 2013). Frye (2018) reviews evidence that a subset of individuals with ASD show accelerated brain growth in the first two years of life followed by a premature slowdown, a trajectory associated with excess white matter and altered connectivity between cortical regions. In frontotemporal areas, which support both language processing and social cognition, the balance between local and long-range connections is altered, and cortical minicolumns are smaller, more numerous, and receive less lateral inhibition from interneurons (Frye, 2018). Regions involved in communication and social perception are therefore structurally atypical early in life.

The first three years of life appear to be especially important. Ayoub (2024) argues that disruptions during this window, whether from genetic variants, maternal immune activity, or nutritional deficiencies, shift the developmental trajectory in ways that become harder to correct over time. Folate metabolism is one example. Folate is required for neurogenesis and neural pathway formation, and folate receptor alpha autoantibodies (FRAAs), which can impair folate transport into the brain, were present in 71% of individuals with ASD in a meta-analysis (Rossignol & Frye, 2021). In a randomized, double-blind, placebo-controlled trial of 48 children with ASD and language impairment aged 3 to 14 years, 12 weeks of high-dose folinic acid improved verbal communication more than placebo, with a larger effect in FRAA-positive children (Frye et al., 2018). Ayoub (2024) also describes an age gradient in which benefit is greatest at ages 2 to 3 and declines by age 5; however, this gradient is drawn from data that had not been published at the time of that review and should be regarded as preliminary.

The Gap Between Biology and Diagnosis

Although neurobiological changes associated with ASD begin before birth, the disorder is identified from behavior, which creates a lag between biology and diagnosis. The DSM-5 requires symptoms to appear in

the early developmental period while acknowledging that they may not become fully apparent until social demands exceed the child's capacities (APA, 2013). In practice, children with lower support needs and female children tend to be diagnosed considerably later than their peers (Ey et al., 2011; Frye, 2018). Parents typically report initial concerns at 12 to 18 months, but formal diagnosis usually does not occur until age 3 to 4 (APA, 2013; Frye, 2018). This interval represents a period during which intervention could, in principle, begin earlier.

Recent research has sought to narrow this gap by identifying earlier biological and behavioral markers. FRAAs are one candidate: they are common in children with ASD (Rossignol & Frye, 2021) and can be detected in parental blood samples before or during pregnancy (Ayoub, 2024). If folinic acid is more effective in FRAA-positive children, as reported by Frye et al. (2018), earlier identification of FRAA status could allow earlier treatment.

Motor difficulties are a second early marker. Peyre et al. (2024) followed 162 children with ASD from the ELENA cohort over three years and found that about 70% had motor skill difficulties at baseline. These children had significantly worse social skill trajectories over the follow-up period than children without motor difficulties ($\beta = -0.42$, $p < 0.01$), a pattern that remained after controlling for age, IQ, and sex and whether the difficulties were in fine motor skills, gross motor skills, or both (Peyre et al., 2024). Motor incoordination is not a diagnostic criterion for ASD; however, its frequency and its early appearance relative to social deficits suggest that it warrants further study as an early marker.

RODENT MODELS OF ASD ACROSS DEVELOPMENTAL STAGES

Rodent models relevant to ASD generally fall into two types: genetic and environmental (Ey et al., 2011). Genetic models delete or mutate genes associated with ASD risk in humans; Fmr1, Nlgn3, Shank3, and Magel2 are among the most common (Ey et al., 2011; Ellegood & Crawley, 2015). In the environmental valproic acid (VPA) model, pregnant animals are exposed to valproic acid, a drug associated with elevated ASD risk in humans, and the offspring are then studied behaviorally and neurologically (Ey et al., 2011; Wang et al., 2025). Both model types are assessed with a consistent set of behavioral tests, including the three-chamber sociability test, measurement of self-grooming, marble burying, and USV recording (Ellegood & Crawley, 2015). Table 2 summarizes the rodent studies included in this review by the developmental stage of manipulation and outcome measurement.

Study	**Species and model**	**Manipulation (age)**	**Outcome measured (age)**	**Stage of evidence**
Branchi et al. (2001)	Mouse; review of pup USV methods	Genetic and environmental manipulations reviewed	Isolation-induced USVs (P0–P14)	Neonatal

Mosienko et al. (2015)	Mouse; Tph2−/− (no brain serotonin)	Genetic (lifelong)	Pup USVs at P3, P6, and P9; deficits at P3 and P6	Neonatal
Tyzio et al. (2014)	Rat VPA; mouse Fmr1-KO	Maternal bumetanide or OXTR antagonist one day before delivery	GABA driving force from E20 to P30 (treatment effect at P15); pup USVs at P4 (rats) and P8 (mice)	Perinatal, with neonatal and juvenile outcomes
Bertoni et al. (2021)	Mouse; Magel2-KO	OXT at P0, P2, P4, P6	GABA driving force at P7; hippocampal activity and social memory in adults	Neonatal manipulation; neonatal and adult outcomes
Wang et al. (2025)	Mouse; VPA and Fmr1-KO; wild-type with chemogenetic suppression	Suppression of oxytocin neurons at P1–P7 (compared with P21–P27)	Oxytocin levels at P1 and P7; GABA reversal potential; self-grooming and social index at P30	Neonatal manipulation; juvenile outcome
Gigliucci et al. (2023)	Mouse; Magel2-KO	OXT at P0, P2, P4, P6	OXTR levels and USVs at P8; OXTR at P90	Neonatal manipulation; neonatal and adult outcomes
Lee et al. (2017)	Mouse; Shank3, Syngap1, and other lines (review)	Genetic (lifelong)	Synaptic transmission and circuit activity, mainly juvenile and adult	Juvenile/adult
Schoch et al. (2017)	Mouse; Pcdh10+/−	Genetic (lifelong); acute d-cycloserine before juvenile testing	Pup USVs at P6; social approach and amygdala gamma power at P28–P32; spines and NMDA receptor subunits in adults	Neonatal, juvenile, and adult outcomes
Gonçalves et al. (2017)	Mouse; Nf1+/−	Genetic (lifelong)	MRS GABA/glutamate and GABA(A) receptors at 7–8 months	Adult

Pantouli et al. (2024)	Mouse; Oprm1−/−	Acute and chronic intranasal OXT in adults	Social interaction at 8–10 weeks	Adult
Ey et al. (2011); Ellegood & Crawley (2015)	Mouse; multiple models (reviews)	Genetic and environmental	Behavioral and neuroanatomical phenotypes, mostly adult	Mostly adult

Table 2: Rodent studies included in this review, classified by the developmental stage of manipulation and outcome measurement.

Evidence From Juvenile and Adult Rodents

Most rodent work relevant to ASD has been conducted in adolescent or adult animals, usually after P30. The reviews by Ey et al. (2011) and Ellegood and Crawley (2015) describe mostly adult phenotypes, so the field's picture of rodent ASD-relevant behavior comes largely from animals that have completed development. Since ASD symptoms in children emerge before age 3 (APA, 2013), this creates a mismatch: most rodent research describes where neural development ends rather than how it gets there. The E/I studies discussed below (Lee et al., 2017; Schoch et al., 2017; Gonçalves et al., 2017) were likewise conducted mainly in juvenile or adult animals.

Pantouli et al. (2024) illustrate both the value and the limits of adult evidence. They tested intranasal oxytocin (OXT) in adult (8- to 10-week-old) Oprm1−/− mice, a model with reduced social interaction and altered oxytocin signaling. A 0.3 IU dose given five minutes before testing restored social interaction, whereas 0.6 IU had no effect, and the effect of 0.3 IU had faded by 15 to 30 minutes after dosing. Chronic OXT improved social interaction in Oprm1−/− mice but reduced it in wild-type controls, showing that the same treatment can produce opposite effects depending on genotype. Because these experiments were performed in adults, they describe how the mature oxytocin system responds to treatment and do not provide evidence about neonatal mechanisms. They remain relevant to human trials, which cannot stratify participants by genotype in the same way.

Neonatal Evidence

The neonatal period is studied mainly through USVs. Branchi et al. (2001) documented pups emitting calls in the 30 to 90 kHz range after maternal separation, peaking around P6 to P7 and ceasing by the end of the second postnatal week. Because USV rate is modulated by serotonin, dopamine, GABA, and opioid signaling, all systems implicated in ASD, the measure serves as a behavioral and neurochemical readout (Branchi et al., 2001). Mosienko et al. (2015) used this approach to show that pups lacking brain serotonin emit fewer isolation calls in the first postnatal week (see Serotonin). USV changes are not always reductions: Schoch et al. (2017) found that Pcdh10+/− pups of both sexes emitted more isolation calls than wild-type littermates at P6.

A small number of studies have manipulated the perinatal period or the first postnatal week directly. Tyzio et al. (2014) intervened at birth: in VPA-exposed rats and Fmr1-knockout (KO) mice, the transient reduction in neuronal chloride that normally occurs during delivery was absent, and treating the mother with the NKCC1 inhibitor bumetanide one day before delivery restored control-like electrophysiological and behavioral phenotypes in the offspring. These included pup USVs, which were reduced in number in VPA-exposed rats at P4 and altered in call type in Fmr1-KO mice at P8. Wang et al. (2025) chemogenetically suppressed oxytocin neurons from P1 to P7 and found that by P30 the treated mice self-groomed significantly more and had lower social index scores, whereas the same suppression from P21 to P27 had no comparable effect.

Two studies gave OXT to Magel2-KO pups at P0, P2, P4, and P6. Bertoni et al. (2021) found that this treatment reduced an abnormally high GABA driving force in hippocampal neurons at P7, normalized GABAergic activity in adult hippocampal neurons, and restored social memory in adulthood. Gigliucci et al. (2023) found that Magel2-KO pups of both sexes had reduced oxytocin receptor (OXTR) levels and fewer separation-induced USVs at P8, and that neonatal OXT changed neither measure at that age. In adulthood (P90), the same treatment normalized OXTR levels in the central amygdala, hippocampus, and piriform cortex of males, regions in which OXTR was upregulated in the knockout. Magel2-KO females, which do not show the social deficits observed in males, showed a different pattern of receptor expression (Gigliucci et al., 2023). Taken together, the Magel2 findings show that a neonatal intervention can change adult social behavior without changing neonatal vocal communication, which cautions against treating pup USVs as a direct readout of later social outcomes.

COMPARING HUMAN AND RODENT LITERATURE

Excitation/Inhibition Imbalance

E/I imbalance has been implicated in ASD in both literatures. Frye (2018) reviews postmortem and imaging work showing reduced inhibitory interneuron density in frontotemporal minicolumns and a seizure co-occurrence rate near 30%, both consistent with cortical hyperexcitability in ASD. In rodents, however, E/I imbalance does not take the same form across genetic models. Lee et al. (2017) describe reduced AMPA receptor-mediated transmission and suppressed long-term potentiation in Shank3 heterozygous mice, whereas Syngap1 heterozygous mice show elevated AMPA receptor levels and hyperactive hippocampal circuits. The same label therefore describes effects in opposite directions, which suggests that E/I imbalance is a broad category reached through distinct mechanisms rather than a single mechanism shared across models.

The Pcdh10 model provides a region-specific example. Schoch et al. (2017) studied Pcdh10+/− mice, which lack one copy of an ASD-associated protocadherin gene whose expression is enriched in the basolateral amygdala. Juvenile males (P28 to P32) showed reduced social approach, and brain slices from juvenile mice showed reduced gamma-frequency power in the basolateral amygdala but not in the striatum. In adult males, neurons of the lateral and basolateral amygdala had an increased density of

immature, filopodia-like dendritic spines, and amygdala synaptic fractions contained lower levels of the NMDA receptor subunits GluN1 and GluN2A. Acute administration of d-cycloserine, an NMDA receptor co-agonist, restored social approach in juvenile males without affecting nonsocial exploration or locomotor activity, although it did not change gamma power in amygdala slices (Schoch et al., 2017). This result links a social behavioral deficit to glutamatergic signaling in a defined circuit, a degree of causal specificity that human neuroimaging studies of the amygdala in ASD cannot currently provide. It was, however, obtained after the neonatal period, in juvenile and adult animals, and does not indicate when in development the circuit alteration arises.

Mouse studies also offer regional resolution that is difficult to obtain in humans. Using in vivo magnetic resonance spectroscopy in Nf1+/− mice aged seven to eight months, Gonçalves et al. (2017) found increased GABA/glutamate ratios in the prefrontal cortex and striatum, whereas in the hippocampus a small reduction in GABA was accompanied by a large increase in GABA(A) receptor expression. The authors concluded that inhibitory tone was enhanced in all three regions, but through different mechanisms. None of the E/I studies reviewed here measured E/I balance during the neonatal period. How E/I imbalance develops, and how it varies across a heterogeneous clinical population rather than a single inbred strain, therefore remains unknown.

The GABA Developmental Shift

GABA, the main inhibitory neurotransmitter in the mature brain, is depolarizing in immature neurons. Early in development, neurons express the chloride importer NKCC1, which keeps intracellular chloride high enough that GABA(A) receptor activation depolarizes the membrane; as expression of the chloride exporter KCC2 increases, intracellular chloride falls and GABA becomes hyperpolarizing (Ben-Ari, 2002). In rodents, this shift takes place mainly during the first two postnatal weeks, but its timing differs between brain structures and between cell types, with interneurons completing the transition earlier than pyramidal cells (Ben-Ari, 2002). It is better described as a sequence of region- and cell-specific transitions than as a single event at a fixed postnatal day. Tyzio et al. (2014) additionally described a brief, oxytocin-mediated reduction in neuronal chloride during delivery.

Rodent evidence links this shift to ASD-relevant models, but the reported direction of the alteration differs between studies. In VPA-exposed rats and Fmr1-KO mice, Tyzio et al. (2014) found that the GABA driving force remained elevated in fetal and early postnatal neurons and at P15 to P30, consistent with GABA remaining depolarizing for longer than normal. Maternal bumetanide one day before delivery restored control-like electrophysiology at P15 and behavior in the offspring, and blocking oxytocin receptors in typical mothers at the same time point produced offspring with similar alterations. Bertoni et al. (2021) reported a comparable delay in Magel2-KO mice, with an elevated GABA driving force at P7 that neonatal OXT reduced. Wang et al. (2025), by contrast, reported that reduced oxytocin at P1 and P7 in VPA-exposed and Fmr1-KO mice was associated with reduced NKCC1 expression and a more negative GABA reversal potential in CA3 neurons, and that OXT acted by shifting the reversal potential in the positive direction through NKCC1. The three studies agree that oxytocin regulates GABA polarity in the perinatal and neonatal period, but they disagree on whether ASD-relevant models show too much or

too little depolarizing GABA. Differences in timing (delivery versus the first postnatal week), model, and method may account for this discrepancy, which argues against a single "delayed switch" explanation.

In the human literature, the evidence is indirect. Postmortem studies show that KCC2 expression begins prenatally, earlier in subcortical structures (from about 10 weeks after conception in the amygdala, cerebellum, and thalamus) than in most of the neocortex (about 19 to 24 weeks), and reaches adult-like levels between birth and six months of age (Sedmak et al., 2016). The human counterpart of the rodent shift therefore appears to begin before birth and to proceed region by region. Clinical trials of bumetanide, which was tested on the premise that GABA remains excessively depolarizing in ASD, have produced conflicting results. A phase IIb trial in 88 children and adolescents aged 2 to 18 years reported improvement on the Childhood Autism Rating Scale among participants who completed treatment (Lemonnier et al., 2017), but two phase III trials in a total of 422 children aged 2 to 6 and 7 to 17 years were terminated early because bumetanide did not differ from placebo (Fuentes et al., 2023). No human study has characterized the timing or regional specificity of this process in infants later diagnosed with ASD. The rodent literature identifies a candidate window and mechanism, but the conflicting directions reported in rodents and the negative phase III results in children indicate that this mechanism is not yet established enough to guide early intervention.

Serotonin

Elevated blood serotonin was one of the first ASD biomarkers to be identified, first reported in the 1960s; a meta-analysis found elevated whole-blood serotonin in 28.3% of individuals with ASD (Gabriele et al., 2014). The serotonergic circuits involved span the amygdala, striatum, and prefrontal cortex, regions associated with both social and repetitive behaviors (Frye, 2018). The neonatal rodent evidence is more detailed. Mosienko et al. (2015) found that pups of Tph2−/− mice, which lack brain serotonin, emitted fewer isolation-induced USVs at P3 and P6, but not at P9, with lower peak frequencies and amplitudes and a disrupted temporal sequence of calls. Branchi et al. (2001) reported that manipulating serotonin in either direction, by depleting it with 5,7-dihydroxytryptamine or raising it acutely with selective serotonin reuptake inhibitors, reduced neonatal USVs. This suggests that early vocal behavior depends on serotonin remaining within a functional range rather than on its absolute level.

These neonatal findings have no direct counterpart in the human literature. Most human serotonin data come from older children and adults, as does most rodent serotonin research outside these USV studies. The neonatal window is therefore understudied in both literatures, even though the rodent methods needed to study it are already established.

Oxytocin

Oxytocin shows the clearest agreement between the two literatures. A meta-analysis of 31 studies found lower endogenous oxytocin in autistic individuals than in neurotypical controls; the difference was present in children ($d = -0.44$) but not in adults ($d = 0.03$) (John & Jaeggi, 2021). In rodents, VPA-exposed, Fmr1-KO, and Magel2-KO mice all show reduced oxytocin or altered OXTR expression, with alterations detectable in the first postnatal week (Wang et al., 2025; Gigliucci et al., 2023).

The rodent literature adds mechanism and timing. As described above, neonatal OXT altered GABA polarity in hippocampal neurons (Bertoni et al., 2021; Wang et al., 2025) and changed social behavior measured at P30 or in adulthood, whereas suppressing oxytocin neurons at P21 to P27 had no comparable effect (Wang et al., 2025). Neonatal OXT did not, however, restore reduced USVs at P8 in Magel2-KO pups (Gigliucci et al., 2023), so the neonatal effects of oxytocin on later social behavior are better supported than its effects on early vocal communication.

Human oxytocin trials, by contrast, have mostly tested intranasal OXT in children and adults who are already well past this period. The largest trial, in 290 children and adolescents aged 3 to 17 years, found no effect on social withdrawal after 24 weeks (Sikich et al., 2021). A trial in 87 children aged 3 to 12 years found no overall effect, but children aged 3 to 5 years improved on caregiver-rated social responsiveness, whereas children aged 6 to 12 years did not (Guastella et al., 2023). Adult mouse data from Pantouli et al. (2024) suggest one reason for this variability: oxytocin's effects depend simultaneously on dose, context, and genotype, so inconsistent trial results might reflect trial design as much as an absence of effect. The age-dependent response reported by Guastella et al. (2023) is consistent with the rodent finding that earlier treatment is more effective, but the two are not directly comparable, since ages 3 to 5 in a child fall far beyond the first postnatal week in a mouse. Whether OXT given to high-risk human infants could reproduce the rodent findings is a question that no existing human study is positioned to answer.

DISCUSSION

What Neonatal Models Add

The main contribution of neonatal rodent models is experimental access to mechanism during a period that cannot be studied directly in children. They allow researchers to establish causal links between genetic background, circuit function, and behavior and to intervene at defined postnatal time points. Wang et al. (2025) provide an example: oxytocin signaling during the first postnatal week altered social behavior measured weeks later, through a mechanism involving NKCC1 and GABA polarity that was specific to this early window.

USVs are the main behavioral readout available during this period. Branchi et al. (2001) showed that USV parameters in neonates track neurobehavioral development closely enough to respond to both genetic and environmental manipulation, and Mosienko et al. (2015) and Tyzio et al. (2014) used them to detect early effects of serotonin loss and altered GABA polarity. The relationship between pup USVs and later social behavior is not straightforward, however. In the Magel2 model, neonatal OXT changed adult social behavior and OXTR expression without changing USVs at P8 (Gigliucci et al., 2023). In Pcdh10+/− mice, increased USVs at P6 were found in both sexes, whereas reduced social approach at P28 to P32 was found only in males (Schoch et al., 2017). The direct link between the GABA shift and neonatal USV phenotypes has not been systematically tested, and most E/I imbalance work measures adult social behavior rather than early vocalization.

Gaps and Future Directions

A timing mismatch runs through the rodent literature on ASD. The developmental stage most relevant for translation is the early communicative and social period, yet most published rodent work uses adolescent or adult animals (Ellegood & Crawley, 2015). The neonatal studies covered in this review (Branchi et al., 2001; Mosienko et al., 2015; Tyzio et al., 2014; Bertoni et al., 2021; Gigliucci et al., 2023; Wang et al., 2025) represent a small body of work. Systematic neonatal phenotyping across genetic models and both sexes remains uncommon, and expanding it would align rodent findings more closely with the clinically relevant developmental window.

Sex comparisons are another significant gap. ASD is diagnosed roughly four times more often in males, and most rodent studies still rely almost entirely on male subjects (APA, 2013; Ey et al., 2011). The age-, sex-, and region-specific OXTR findings of Gigliucci et al. (2023) show that the oxytocin system does not develop identically in males and females. Without matching female data at the neonatal stage under the same intervention conditions, conclusions about early treatment windows remain incomplete. This is relevant because diagnosed females differ from males in clinical profile, including a higher likelihood of intellectual disability, and are often diagnosed later (APA, 2013; Ey et al., 2011; Frye, 2018).

Whether rodent and human developmental timelines correspond is a separate question. Rodents at P7 to P10 have historically been treated as roughly equivalent to a term human infant, based mainly on comparisons of brain weight. The sequence of major maturational events is largely conserved between species, but the correspondence differs by process and brain region (Semple et al., 2013). The P1 to P7 window studied by Wang et al. (2025) therefore should not be treated as a precise equivalent of a single human developmental period. For GABA signaling specifically, KCC2 expression in humans begins in mid-gestation and reaches adult-like levels between birth and six months (Sedmak et al., 2016), so the human processes that correspond to the rodent first postnatal week may occur partly before birth and at different times in different regions. Few, if any, human studies have measured neonatal markers such as oxytocin signaling, GABA maturation, or E/I balance in children who later receive an ASD diagnosis, although prospective studies of infants at elevated familial likelihood offer one route to such data. For now, rodent models provide the main experimental access to this period of development. Figure 1 shows the timing of the rodent studies reviewed here and, on a separate axis, the corresponding human developmental processes, clinical milestones, and treatment trials.

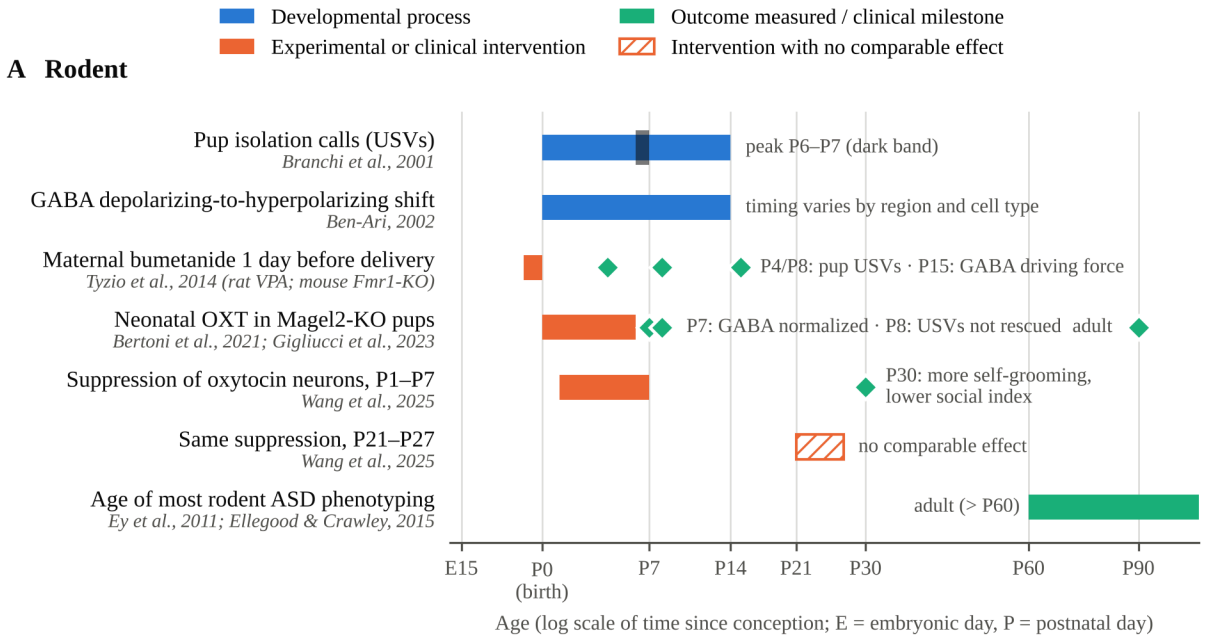

Figure 1: Timing of the developmental processes, experimental manipulations, and clinical milestones discussed in this review. (A) Rodent studies, with ages in embryonic (E) and postnatal (P) days. (B) Human development, diagnosis, and clinical trials. Each panel uses its own logarithmic scale of time since conception; the panels are not aligned, and no direct age correspondence between species is implied. Sources are listed beside each row.

Therapy Testing and Translational Limits

Oxytocin's record as a therapeutic candidate shows both the promise and the difficulty of translating rodent rescue into human benefit. Large pediatric trials have been negative overall (Sikich et al., 2021), with possible benefit limited to younger children (Guastella et al., 2023). Rodent data offer a possible explanation. Pantouli et al. (2024) found that dose, timing, and social context all shift the oxytocin response in adult mice, and that the behavioral effect depends on the model's opioid–oxytocin circuit configuration. If similar interactions exist in humans, trials in heterogeneous populations without genotypic or contextual stratification would be expected to yield null or mixed results, as observed so far. This points toward stratified trials as a more promising next step.

Folinic acid is a second candidate, with randomized evidence in children. Frye et al. (2018) reported improved verbal communication after 12 weeks of treatment, with a larger effect in FRAA-positive children. Ayoub (2024) interprets nutritional and metabolic interventions in ASD as following an age-dependent pattern consistent with critical-period biology, although the specific age gradient described in that review has not yet been published as original data. Most enrollment in these trials occurs after age 2. If the critical-period logic from rodent models applies in humans, the main limitation is diagnostic: at-risk infants would need to be identified earlier for treatment to fall within the effective window.

Bumetanide illustrates a different outcome. A mechanism supported by perinatal rodent rescue (Tyzio et al., 2014) produced encouraging results in an earlier trial (Lemonnier et al., 2017) but no benefit over placebo in phase III trials (Fuentes et al., 2023). The rodent rescue was achieved by treating the mother before delivery, whereas the human trials treated children aged 2 to 17 years; this difference in timing may partly explain the gap between the two results.

Specificity is a limitation shared by all rodent models of ASD. Each genetic line captures a single entry point into a heterogeneous disorder, and neuroanatomical and behavioral profiles vary substantially across models (Ellegood & Crawley, 2015; Ey et al., 2011). A rescue in the Magel2 model (Bertoni et al., 2021; Gigliucci et al., 2023) or the mu-opioid receptor knockout model (Pantouli et al., 2024) does not generalize to ASD as a whole, and whether the early-postnatal oxytocin–GABA mechanism identified by Wang et al. (2025) applies to other genetic subtypes remains open. With current tools, rodent models are best treated as hypothesis-testing platforms: they can show whether a given circuit, molecule, or developmental event is sufficient to alter a defined behavior, but they cannot by themselves establish the mechanism of ASD.

Bilingual Exposure as a Translational Case Study

Human studies of bilingual exposure illustrate the type of question that neonatal rodent models could help address. In a study of 412 toddlers aged 15 to 35 months, bilingual exposure showed no negative interaction with ASD on language, cognitive, or social-emotional outcomes (Phillips et al., 2024). At school age, bilingual children with ASD have been reported to outperform monolingual peers with ASD on executive function measures such as cognitive flexibility and inhibitory control (Baldimtsi et al., 2020) and on reading and listening comprehension (Peristeri & Tsimpli, 2020), and bilingual exposure has been associated with stronger first-language production (Ge et al., 2024). These studies describe outcomes but

not mechanism or developmental timing. Bilingualism itself cannot be modeled in mice; the testable question is the more general one of whether the complexity of early social and acoustic experience during the neonatal USV period alters later social and executive phenotypes. Executive function phenotypes, including attentional flexibility and working memory, have been documented in several genetic models (Ey et al., 2011), so the relevant behaviors are accessible to rodent testing. This case study is included to indicate a direction for future work, not as evidence of a shared mechanism.

Limitations of This Review

This review has several limitations. It was conducted by a single author and was not pre-registered, which may introduce bias in study selection and interpretation. Although the search was structured, this was not a systematic review: screening counts were not recorded in a PRISMA flow diagram, and no formal risk-of-bias assessment was performed. The number of neonatal rodent studies that met the inclusion criteria was small, and several themes rely on one or two studies, often in a single model. Finally, some background statements rely on narrative reviews (Frye, 2018; Ayoub, 2024); where possible, these have been supplemented with original human studies and meta-analyses.

CONCLUSION

This review compared human clinical and neonatal rodent evidence on four mechanisms relevant to ASD, separating neonatal findings from those obtained in juvenile and adult animals. The oxytocin–GABA axis has the most consistent neonatal support: manipulations of oxytocin in the perinatal period and first postnatal week altered GABA polarity and later social behavior across several models (Tyzio et al., 2014; Bertoni et al., 2021; Wang et al., 2025). The studies disagree, however, on the direction of the GABA alteration, and neonatal OXT did not restore reduced pup USVs in the Magel2 model (Gigliucci et al., 2023). Human data agree that endogenous oxytocin is lower in autistic children (John & Jaeggi, 2021), but clinical trials of oxytocin and bumetanide in children have produced mixed or negative results (Sikich et al., 2021; Guastella et al., 2023; Fuentes et al., 2023). Serotonin's role in early communication is supported by neonatal USV studies but remains poorly characterized in humans at comparable ages. Evidence for E/I imbalance comes mainly from adult rodents and differs in direction across models.

Underlying each theme is the same translational problem: neurodevelopmental disruption begins prenatally and in early infancy, but ASD symptoms are usually recognized only in the second year of life, and formal diagnosis often occurs at age 3 to 4 (APA, 2013; Ayoub, 2024; Frye, 2018). Because developmental timing differs between species and across brain regions (Semple et al., 2013; Sedmak et al., 2016), neonatal rodent models can identify candidate mechanisms and windows but cannot by themselves define the corresponding period in children. Testing these mechanisms in both sexes and across multiple models during the neonatal period, together with earlier identification of at-risk infants, would allow the two literatures to be connected more directly.

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