Received: 23 December 2016 | Revised: 2 June 2017 | Accepted: 10 August 2017
DOI: 10.1002/ajmg.a.38465

RESEARCH REVIEW

WILEY AMERICAN JOURNAL OF medical genetics PART A

Pharmacological interventions to improve cognition and adaptive functioning in Down syndrome: Strides to date

Sarah J. Hart[1] | Jeannie Visootsak[2] | Paul Tamburri[2] | Patrick Phuong[2] | Nicole Baumer[3,4,5] | Maria-Clemencia Hernandez[6] | Brian G. Skotko[5,7] | Cesar Ochoa-Lubinoff[8] | Xavier Liogier D'Ardhuy[6] | Priya S. Kishnani[1] | Gail A. Spiridigliozzi[9]

[1] Department of Pediatrics, Duke University Medical Center, Durham, North Carolina
[2] F. Hoffmann-La Roche, Roche Pharma Research and Early Development, Neuroscience, Roche Innovation Center New York, New York, New York
[3] Department of Neurology, Boston Children's Hospital, Boston, Massachusetts
[4] Down Syndrome Program, Developmental Medicine Center, Boston Children's Hospital, Boston, Massachusetts
[5] Harvard Medical School, Boston, Massachusetts
[6] F. Hoffmann-La Roche, Roche Pharma Research and Early Development, Neuroscience, Roche Innovation Center Basel, Basel, Switzerland
[7] Down Syndrome Program, Division of Medical Genetics, Department of Pediatrics, Massachusetts General Hospital, Boston, Massachusetts
[8] Section of Developmental-Behavioral Pediatrics, Department of Pediatrics, Rush University Medical Center, Chicago, Illinois
[9] Department of Psychiatry and Behavioral Sciences, Duke University Medical Center, Durham, North Carolina

Correspondence
Sarah J. Hart, Department of Pediatrics, Duke University Medical Center, 905 S. LaSalle St., Box 103857, Durham, NC 27710.
Email: sarah.hart@duke.edu

Although an increasing number of clinical trials have been developed for cognition in Down syndrome, there has been limited success to date in identifying effective interventions. This review describes the progression from pre-clinical studies with mouse models to human clinical trials research using pharmacological interventions to improve cognition and adaptive functioning in Down syndrome. We also provide considerations for investigators when conducting human clinical trials and describe strategies for the pharmaceutical industry to advance the field in drug discovery for Down syndrome. Future research focusing on earlier pharmaceutical interventions, development of appropriate outcome measures, and greater collaboration between industry, academia, advocacy, and regulatory groups will be important for addressing limitations from prior studies and developing potential effective interventions for cognition in Down syndrome.

KEYWORDS
clinical trials, cognition, Down syndrome, interventions

1 | INTRODUCTION

Down syndrome (DS, OMIM #190685) is the most common genetic cause of intellectual disability and results from extra genetic material from chromosome 21 (mostly, trisomy 21). The live birth prevalence is approximately 1 in 792 live births (de Graaf, Buckley, & Skotko, 2015), with population estimates indicating about 206,000 individuals with DS living in the United States as of 2010 (de Graaf, Buckley, & Skotko, 2017). Congenital and acquired medical complications are variable among individuals with DS, but many present at a higher frequency than that of the general population. Due to recent advances in medical treatment including surgical correction of congenital heart disease,

treatment of endocrine disease (e.g., hypothyroidism, diabetes) and hematologic malignancies, the population prevalence of DS has increased over time (de Graaf et al., 2017) with a significant increase in the life expectancy of people with DS (Bittles, Bower, Hussain, & Glasson, 2007). The increased risk of early development of the neuropathology of Alzheimer disease (AD) in individuals with DS is also well-documented with approximately half having AD-associated dementia by the age of 60 years (Head, Powell, Gold, & Schmitt, 2012).

Although studies of DS historically have described the condition as a homogenous group, there is significant inter-individual variability in the phenotype of DS at multiple levels, including genetics, cellular biology, cognition, and behavior (Karmiloff-Smith et al., 2016). The genetic mechanisms underlying the variability in DS are not well understood (Patterson, 2009). Chromosome 21 has 726 genes currently annotated by the National Center for Biotechnology Information (NCBI). The expression and dosage sensitivity of the genes on chromosome 21 varies, and some researchers have hypothesized that the most dosage-sensitive genes are most likely to contribute to the DS phenotype (Korenberg et al., 1990; Prandini et al., 2007). Alternatively or additionally, other researchers believe that the phenotype is due to extra genetic material that, as a whole, disrupts multiple developmental pathways (Shapiro, 1997). A study of monozygotic twins discordant for trisomy 21 revealed that differential gene expression between the twins was organized into domains along chromosome 21 that were either up- or down-regulated, suggesting that gene expression dysregulation domains may contribute to some phenotypes in DS (Letourneau et al., 2014). Given the complexity of the genetic mechanisms in DS and the roles of many genes on chromosome 21 in brain function and development, understanding the mechanisms underlying the variability in cognitive abilities and other features in DS remains a significant challenge.

Despite the long-standing and rich history in our understanding of the unique cognitive and adaptive profiles in individuals with DS, there is no FDA-approved pharmacological treatment to date to improve cognition or adaptive functioning. While an increasing number of clinical trials have focused on improving these areas, the scientific community has had limited success to date, possibly due to the challenges in capturing meaningful change with current neurocognitive measures and the scant investment in research funding for this population (Heller, Spiridigliozzi, Crissman, Sullivan-Saarela, Li, et al., 2006). In recent years, the academic community, advocacy organizations, and pharmaceutical companies have developed growing and collaborative interests in additional clinical trials for people with DS. For example, the Trisomy 21 Research Society, which was founded to promote translational research and interventions in DS (t21RS), held its first meeting in 2015, and the Keystone Symposia on Molecular and Cellular Biology held its first meeting focused on the biology of DS and promotion of translational research in 2016 (2016a). Premier national and international DS advocacy organizations (e.g., LuMind Foundation and Lejeune Foundation) have been instrumental in funding research to advance clinical trials. The National Institutes of Health (NIH) has also supported the development of clinical trials for DS through the Down Syndrome Research Plan (NICHD, 2014) and DS-Connect, a national registry to connect families with researchers conducting clinical trials and improve understanding of health in DS (DHHS, 2016).

The goal of this review is to describe the historical basis and current state of pharmacological interventions in DS, in addition to strategies for future research from the perspective of investigators and industry. We describe how pharmacologic treatment studies in DS have progressed from pre-clinical studies that led to targeted pharmacological clinical trials, which historically targeted drugs used to treat AD, but with a focus to improve cognition and adaptive functioning in DS. We outline strategies used in the DS and neurodevelopmental research community to conduct human clinical trials and describe current partnerships and collaborations between clinicians, researchers, advocacy groups, and the pharmaceutical industry that will be essential to advance the field in drug discovery for DS.

2 | NEURODEVELOPMENT IN DOWN SYNDROME

Although most individuals with DS have mild-moderate cognitive impairment (Nicham et al., 2003), certain cognitive domains such as language and memory appear to be affected disproportionately in comparison to other types of intellectual disability, resulting in a characteristic neurocognitive phenotype. People with DS have relative strengths in visuospatial processing and implicit long-term memory and more difficulty in working memory, episodic long-term memory, expressive language, and executive function (Liogier d'Ardhuy et al., 2015). Characteristic patterns of cognitive development may reflect unique structural and functional differences in brain development in DS (Fidler & Nadel, 2007; Nadel, 2003), with differences in neurodevelopment becoming more evident across the lifespan (Grieco, Pulsifer, Seligsohn, Skotko, & Schwartz, 2015). Although cognitive growth continues throughout childhood and into adolescence and young adulthood, learning and memory difficulty tends to have a greater impact with age (Nadel, 2003). The IQ of individuals with DS as measured on standardized tests of intelligence has also been shown to decline with age (Carr, 2005), reflecting slower rates of skill acquisition and a widening of the gap between chronological age and developmental age, rather than loss of skills or deterioration of cognitive abilities (Carr, 2005; Couzens, Cuskelly, & Haynes, 2011). However, there may be a difference in trajectories of verbal- compared to nonverbal-cognitive abilities, with plateau and decline in young adults predominantly evident in verbal scores (Carr, 2005). Across the age span, adaptive skills have been noted to increase with age until middle childhood, at which point a plateau in adaptive development is seen (Dykens, Hodapp, & Evans, 2006).

Although there are considerable individual differences, language has been considered among the most significantly affected domains of functioning in people with DS (Abbeduto, Warren, & Conners, 2007), with delays more evident in expressive than receptive language (Chapman, 2006; Grieco et al., 2015). Language impairments emerge early in life and are marked by delayed development of language

milestones, emergence of articulation difficulty, phonological and grammatical errors, decreased growth of vocabulary, and difficulty with syntax that becomes more prominent in adolescence and young adulthood (Abbeduto et al., 2007; Chapman & Hesketh, 2001; Fidler, Philofsky, & Hepburn, 2007; Martin, Klusek, Estigarribia, & Roberts, 2009). Low muscle tone, oral-motor skills, middle ear problems, and hearing impairment may contribute to some of the difficulty with language (Abbeduto et al., 2007; Martin et al., 2009). Language difficulties may also contribute to impairments in adaptive behavior, with caregivers reporting significant relative weaknesses in communication compared to daily living and socialization skills and significantly weaker expressive language than receptive language (Dykens et al., 2006).

Long-term memory, involving the encoding, storage, and retrieval of information, is known to be preferentially affected in DS compared to individuals with other intellectual disabilities (Carlesimo, Marotta, & Vicari, 1997). Long-term memory can be divided into explicit or declarative memory (e.g., memory for facts, concepts, events, and experiences) and implicit or procedural memory (e.g., incidental learning and memory of skills and tasks). Explicit memory seems to be selectively impaired in individuals with DS (Carlesimo et al., 1997; Vicari, 2001) and likely contributes significantly to difficulties with academic learning. Working/short-term memory, which is important for holding, sorting, processing, and manipulating information, is also affected in DS, with greater difficulties in processing and remembering verbal/auditory information compared to visual–spatial information (Fidler & Nadel, 2007; Jarrold, Nadel, & Vicari, 2009). There is a significant interrelationship between verbal short-term memory and language development, with difficulties in these domains likely contributing to impairments in learning and overall functioning (Silverman, 2007).

Behavior and social-emotional functioning also impact learning in individuals with DS. Children with DS have relative strengths in social motivation and engagement, but they may struggle with social problem solving or decision making and higher order social cognition tasks (Fidler, 2006; Fidler & Nadel, 2007). Noncompliant behavior and difficulty with task persistence are also commonly seen in children with DS, with some researchers describing a personality-motivation style, characterized by strong-willed or stubborn temperament (Fidler, 2006; Fidler & Nadel, 2007; Kasari & Freeman, 2001). Some researchers have also suggested that children with DS may use a strategy of social distraction as a means of avoiding tasks (Kasari & Freeman, 2001). As difficulties in specific cognitive and behavioral domains in DS have a significant impact on overall adaptive function, development of targeted interventions is a critical goal for reducing barriers to further accomplishment for individuals with DS.

3 | PRE-CLINICAL WORK IN DOWN SYNDROME

The development of interventions that might improve cognition in people with DS has been predominantly based on pre-clinical work using mouse models. While multiple mouse models of DS have been developed, we focus our review on the Ts65Dn mouse model that has been used in the majority of pre-clinical studies. The Ts65Dn model has segmental trisomy of mouse chromosome 16, which shares a large homology with human chromosome 21 (Davisson, Schmidt, & Akeson, 1990). The Ts65Dn model has dosage imbalance for genes corresponding to human chromosome 21q21-22.3 (Reeves et al., 1995). While the Ts65Dn model does not have gene dosage imbalance for all genes on chromosome 21, Ts65Dn mice have been shown to express similar characteristics to humans with DS, including relative deficits in learning and memory and differences in neurodevelopment and neuronal morphology (Reeves et al., 1995). Over the past years, promising results in the Ts65Dn mouse model have revealed rescue of DS-associated associated learning and memory deficits with administration of drugs targeting various neurotransmitter systems or proteins linked to pathogenesis of neurodevelopment in DS. We review here several of the primary targets that have been investigated in intervention studies with mouse models of DS, including Aβ protein, serotonin, gamma-aminobutyric acid (GABA), dual-specificity tyrosine phosphorylation-regulated kinase 1a (DYRK1a) protein, and N-methyl-D-aspartate (NMDA) receptors. We refer the reader to several recent reviews on pharmacological interventions in mouse models of DS for further details on neurobiological mechanisms and evidence for using these targets to improve cognition (Bartesaghi et al., 2015; Costa & Scott-McKean, 2013; Gardiner, 2015).

Triplication of the *APP* gene on chromosome 21, which expresses β-amyloid precursor protein (APP), has been shown to be associated with degeneration of cholinergic neurons and with development of AD-like pathology in both the mouse model and in individuals with DS (Bartesaghi et al., 2015). The neuronal pathology in the Ts65Dn mouse includes presence of neurofibrillary tangles and accumulation of Aβ protein, which aggregates to form amyloid plaques. Mouse models have been used to investigate potential therapeutic avenues that target the cleavage and processing of the APP protein as part of AD-like pathology. As γ-secretase is the final step required for formation of Aβ protein and amyloid plaques, there has been a large effort to develop γ-secretase inhibitors as a potential therapeutic avenue for AD (Wolfe, 2008) and, more recently, as a potential intervention in DS. Studies using the Ts65Dn mouse model have demonstrated correction of learning deficits (Netzer et al., 2010) and restoration of neurogenesis defects (Giacomini et al., 2015) with γ-secretase inhibitors. While further research is needed to clarify the role of the triplication of APP in the pathogenesis for DS (Costa & Scott-McKean, 2013), pharmaceuticals targeting γ-secretase may hold promise for future clinical trials.

Fluoxetine has been investigated as a potential therapeutic target in DS primarily because it has been found be linked to neurogenesis in the hippocampus, a structure in the brain that is critical for learning and memory and that is characterized by continued neurogenesis in adulthood (Gardiner, 2015). Treatment with fluoxetine in adult mice has been demonstrated to rescue hippocampal neurogenesis (Clark, Schwalbe, Stasko, Yarowsky, & Costa, 2006), hippocampal expression of 5-hydroxytryptamine1A receptors and brain-derived neurotrophic

factor (Bianchi et al., 2010), learning deficits (Bianchi et al., 2010), and hippocampal dendritic pathology (Guidi et al., 2013). Prenatal administration of fluoxetine in the Ts65Dn mice has been associated with rescue of neurogenesis and neurodevelopment at postnatal day two, with effects still present at postnatal day 45 accompanied by rescue of behavioral performance (Guidi et al., 2014).

GABA is an inhibitory neurotransmitter that is significantly involved in cognition and memory and has been linked to deficits in DS. It is proposed that DS is characterized by an imbalance in the regulation of excitatory and inhibitory signaling, with excessive inhibitory GABA-mediated signaling linked to reduced long-term potentiation in the hippocampus in the Ts65Dn mice (Costa & Scott-McKean, 2013). Treatment with a $GABA_A$ antagonist in Ts65Dn mice has been found to reverse learning and memory deficits and normalize synaptic plasticity in the hippocampus (Fernandez et al., 2007). Due to the link between non-competitive $GABA_A$ antagonists and seizures (Wetmore & Garner, 2010), more specific GABAergic therapies have been developed targeting the $GABA_A$ α5 subunit. $GABA_A$ α-5 negative allosteric modulators (NAMs; also called inverse agonists) have been found to enhance learning and memory in control mice (Collinson et al., 2002) and are found to be highly expressed specifically in the hippocampus (Gardiner, 2015). Administration of $GABA_A$ α-5 NAMs have been associated with improvements in recognition memory, spatial learning and memory and rescue of hippocampal synaptic plasticity and adult neurogenesis in Ts65Dn mice (Braudeau et al., 2011; Martinez-Cue et al., 2013). $GABA_B$ receptor antagonists have also been investigated as potential therapeutic targets, with one study showing rescue in performance of object recognition, place recognition and contextual fear conditioning (Kleschevnikov et al., 2012).

Recently, the well-established concept of excessive GABAergic inhibition in DS has been challenged by a study suggesting that GABA might be excitatory rather than inhibitory in Ts65Dn mice (Deidda et al., 2015). The authors also reported that administration of the NKCC1 inhibitor bumetanide improved the performance of adult Ts65Dn mice in contextual fear-conditioning, object location and object recognition tasks. However, these data remain to be confirmed in additional preclinical studies.

A recent body of evidence also suggests a role of the chromosome 21 gene *DYRK1A* in developmental and cognitive deficits associated with DS. The gene encodes the protein DYRK1A, overexpression of which appears related to pathogenic mechanisms associated with intellectual deficits (Costa & Scott-McKean, 2013). The polyphenol epigallocatechin gallate (EGCG), found in high concentrations in green tea leaves, has been shown to be an inhibitor of DYRK1a (Bain, McLauchlan, Elliott, & Cohen, 2003). Pre-clinical studies of EGCG in Ts65Dn mice have shown potential rescue of learning and memory deficits (De la Torre et al., 2014) and normalization of long-term potentiation in the hippocampus (Xie, Ramakrishna, Wieraszko, & Hwang, 2008).

Therapies targeting the NMDA (glutamatergic) receptor system have also been investigated with Ts65Dn mouse model. Memantine is an NMDA antagonist that has previously been shown to rescue learning and memory deficits in mouse models of AD and stroke (Gardiner, 2015; Lipton, 2007). Costa, Scott-McKean, and Stasko (2008) showed that overexpression of genes on chromosome 21 was linked to excessive NMDA signaling in Ts65Dn mice, and found that administration of memantine was linked to normalization of NMDA receptor functioning and improved learning and memory performance.

Other pre-clinical studies in Ts65Dn mice have demonstrated improvements in learning and memory by targeting a variety of other neurotransmitter systems or proteins, including norepinephrine, estrogen, minocycline, lithium, melatonin, sonic hedgehog, antioxidants, and neuropeptides (LaFerla, Green, & Oddo, 2007). In general, administration of therapies prenatally or in early development has been associated with significant changes in rescue of neurogenesis and behavioral features, while therapies administered after the critical periods of neurogenesis and synaptogenesis have more limited effects (Stagni, Giacomini, Guidi, Ciani, & Bartesaghi, 2015). For example, treatment during embryogenesis with the precursor to acetylcholine leads to improvements in hippocampal neurogenesis and learning and memory in adult and aged trisomic mice (Ash et al., 2014; Moon et al., 2010; Velazquez et al., 2013). Other studies have also suggested similar significant effects on neurodevelopment using prenatal administration of compounds including fluoxetine, active peptide fragments of activity-dependent neuroprotective protein and activity-dependent neuroprotective factor, SGS-11 (an analog of piracetam), tocopherol, and EGCG (Guedj, Bianchi, & Delabar, 2014; Stagni et al., 2015). While the majority of pre-clinical studies have investigated pharmaceutical interventions in older mice, the fewer number of studies investigating prenatal therapies have demonstrated significant effects on neurogenesis, brain cellularity, connectivity, and behavior (Guedj et al., 2014; Stagni et al., 2015).

4 | CLINICAL TRIALS IN DOWN SYNDROME

The development of clinical trials for people with DS has progressed from originally being primarily focused on AD to targeting pediatric populations and investigating early pharmacological interventions. Pharmacological studies in DS began in the 1960s and continued throughout the 1980s with trials investigating vitamins and supplements (Table 1). These early studies were often based on anecdotal case reports with no clear mechanistic rationale and were typically small, single-center trials, sometimes open-label, making it difficult to draw valid conclusions regarding efficacy. Beginning in the 1990s and into the 2000s, pre-clinical research using the Ts65Dn mouse model and other translational research made it possible to target molecular mechanisms in the brain to address the cognitive and functional deficits associated with DS (Table 1).

The earliest clinical trials of pharmaceutical interventions in DS were focused on the cholinergic system (Kishnani et al., 1999). DS has been associated with abnormalities in peripheral and central cholinergic function (Beccaria et al., 1998; Florez, del Arco, Gonzalez, Pascual, & Pazos, 1990; Sacks & Smith, 1989) and with reductions in cholinergic neurons (Casanova, Walker, Whitehouse, & Price, 1985), which may affect cortical neuronal connectivity and maturation during early

TABLE 1 History of clinical trials in Down syndrome

Before 1989	1990s	2000s	2010s	Ongoing
Vitamins and supplements	**Cognition**			
Pituitary extract (Berg, Kerman, Stern, & Mittwoch, 1961)	Donepezil (Kishnani et al., 1999)	Piracetam (Lobaugh et al., 2001)	Folinic acid (Blehaut et al., 2010)	Folinic acid and thyroid hormone (NCT01576705, Jerome Lejeune Inst.)
Niacin (Heaton-Ward, 1962)		Donepezil (Heller et al., 2004)	Donepezil (Kishnani et al., 2010)	PTZ (COMPOSE study, Balance therapeutics)
U series vitamin (Bumbalo, 1964)		Thyroxine (van Trotsenburg et al., 2005)	Rivastigmine (Heller et al., 2010)	Memantine (NCT02304302, University Hospitals Cleveland Medical Center)
Vitamin B6, 5-hydroxytryptophan (Pueschel, Reed, Cronk, & Goldstein, 1980)		Rivastigmine (Heller, Spiridigliozzi, Crissman, Sullivan, Eells, et al., 2006)	Donepezil (Kondoh et al., 2011)	Intranasal glulisine (NCT02432716, HealthPartners Institute)
Megavitamins and thyroid hormone (Harrell et al.,1981)		Vitamins, minerals and folinic acid (Ellis et al., 2008)	Memantine (Boada et al., 2012)	ACI-24 (NCT02738450, AC Immune)
Vitamins and minerals (Bennett, McClelland, Kriegsmann, Andrus, & Sells, 1983)		Donepezil (Kishnani et al., 2009)	EGCG (De la Torre et al., 2014)	
Vitamins and minerals (Weathers, 1983)			Rivastigmine (Spiridigliozzi et al., 2016)	
Megavitamins (Smith, Spiker, Peterson, Cicchetti, & Justine, 1984)			EGCG and cognitive training (de la Torre et al., 2016)	
Vasopressin (Eisenberg, Hamburger-Bar, & Belmaker, 1984)			ELND005 (Rafii et al., 2017)	
Vitamin B6 (Coleman et al., 1985)			Basmisanil (NCT01436955, Hoffmann-La Roche)	
Thiamine (Lonsdale & Kissling, 1986)			Basmisanil (NCT02024789, Hoffmann-La Roche)	
Vitamins and minerals (Bidder, Gray, Newcombe, Evans, & Hughes, 1989)			Basmisanil NCT02484703, Hoffmann-La Roche)	
			Donepezil (NCT02094053, Eisai Inc.)	
	Dementia in DS			
			Antioxidants (Lott et al., 2011)	Nicotine (NCT01778946, Vanderbilt)
			Memantine (Hanney et al., 2012)	
			Vitamin E (NCT01594346/NCT00056329, New York State Institute for Basic Research)	

EGCG, epigallocatechin gallate; PTZ, pentylenetetrazole.

development (Becker, Mito, Takashima, & Onodera, 1991; Berger-Sweeney, 2003). Cholinesterase inhibitors have been used to investigate potential effects of enhancing cholinergic function on cognition. Donepezil, a reversible inhibitor of acetylcholinesterase, is approved for use in people with AD in the general population. Several recently completed randomized double-blind, placebo-controlled trials revealed donepezil to be generally safe and well tolerated but provided no significant benefit as a cognitive enhancer in children or adults with DS (Kishnani et al., 2009, 2010). A recent Cochrane Collaboration review concluded that in adults with DS, there was no difference in cognitive functioning or behavior between individuals with DS treated with donepezil and placebo, although the likelihood of experiencing an adverse event was significantly higher for subjects with DS on donepezil (Livingstone, Hanratty, McShane, & Macdonald, 2015).

Rivastigmine is approved for the treatment of mild to moderate AD and dementia due to Parkinson's disease. Rivastigmine has been shown to have benefit on the cognitive, functional and behavioral problems commonly associated with AD (Corey-Bloom, Anand, & Veach, 1998; Finkel, 2004; Rosler et al., 1999; Rosler, Retz, Retz-Junginger, & Dennler, 1998) and Parkinson's disease dementias (Emre et al., 2004). However, no clinical trials to date have demonstrated significant benefits in these domains in individuals with DS. A recent randomized double-blind, placebo-controlled trial of rivastigmine in children and adolescents with DS suggested potential improvement in a subset of participants for expressive language, but overall was not associated with significant effects on adaptive function, executive function, language or memory measures (Keeling et al., in press; Spiridigliozzi et al., 2016). A major challenge noted in these studies has been the choice of neurocognitive measures that are sensitive to change in cognition and overall function, as many of the measures used have been standardized for a neurotypically developing population or may be associated with ceiling or floor effects in the study population with DS (Heller, Spiridigliozzi, Crissman, Sullivan-Saarela, Li, et al., 2006).

Piracetam is a member of the class of drugs known as nootropics, which are generally thought to enhance cognitive function by influencing vascular and neuronal functions in instances of brain dysfunction (Winblad, 2005). It has been found to be a positive allosteric modulator of the α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid (AMPA) receptor (Ahmed & Oswald, 2010). A Phase 2 placebo-controlled, 2-period crossover study by Lobaugh et al. (2001) of children with DS (ages 6–13) was conducted to evaluate the effect of piracetam on a range of cognitive functioning (including attention, memory, and learning). The study concluded that piracetam therapy did not significantly improve cognitive performance over placebo, but was associated with adverse events of the central nervous system in 7 of the 18 children who completed the study.

As multiple genes involved in folate metabolism are located on chromosome 21 and folate deficiency has been linked to intellectual disability, folinic acid has been investigated as a potential intervention for cognition in DS. A randomized controlled trial of antioxidants and folinic acid did not find significant effects on development or long-term communication abilities in infants with DS (Ellis et al., 2008). However, a double-blind, placebo-controlled, single-center study in a pediatric DS population (3–30 months old) found significant improvement in global developmental age for those taking 1 ± 0.3 mg/kg folinic acid daily when compared to placebo (Blehaut et al., 2010). This effect was larger in a sub-analysis of subjects taking concomitant thyroid hormone. It was also noted that the dosage of folinic acid in the earlier study (Ellis et al., 2008) (0.1 mg daily) may have been too low to significantly impact cognition (Blehaut et al., 2010). Folinic acid and thyroid hormone are currently being investigated in combination in a 4-arm, placebo-controlled trial for improvement of psychomotor development in young children with DS, ages 6–18 months (ClinicalTrials.gov identifier NCT01576705).

Memantine is a low-affinity uncompetitive antagonist for glutamatergic NMDA receptors (Chen et al., 1992; Chen & Lipton, 1997). It is approved by the U.S. FDA and the European Medicines Agency for treatment of moderate-to-severe AD. A study reported by Boada et al. (2012) in individuals with DS ages 18–32 showed that while there were no significant differences in the two primary measures, a significant improvement was seen in a secondary measure test score (California Verbal Learning Test-II- supraspan word learning) related to hippocampus-dependent function. However, a study in adults with DS over age 40 showed that a 1-year treatment with memantine (at a lower dose of 10 mg/d) was well tolerated, but no significant improvement was seen in primary or secondary measures of cognition or adaptive function (Hanney et al., 2012). Costa and Scott-McKean recently initiated a Phase 2 memantine trial in young adults with DS age 15–32 years to evaluate if a 16-week treatment will have an effect on learning and memory (clinicaltrials.gov NCT02304302).

Several recent clinical trials in DS have focused on targeting the GABA system. Following pre-clinical studies showing improvements in learning and memory with a $GABA_A$ antagonist (Fernandez et al., 2007) and selective $GABA_A$ α5 NAM (Martinez-Cue et al., 2013), pentylenetetrazole (PTZ) and Basmisanil (developed by Hoffmann-La Roche) have been investigated in clinical trials for possible pro-cognitive effects associated with their antagonism/inverse agonism of the $GABA_A$ receptor. PTZ is a $GABA_A$ antagonist that was previously approved by the FDA for the treatment of various cognitive impairments. PTZ has also been linked to seizures in animal models at higher doses, leading to safety concerns in human clinical trials (Gardiner, 2015). Though the FDA has since revoked PTZ approval due to lack of evidence for clinical efficacy, PTZ is currently under investigation for cognitive enhancement in individuals with DS. A placebo-controlled study of adolescents and young adults (ages 13–35) with DS investigated the pro-cognitive effects of PTZ up to 12 weeks (COMPOSE study—Australian New Zealand Clinical Trials Registry ID ACTRN12612000652875). Cognitive function was assessed in the domains of language, executive function, and adaptive behavior. Currently, the study has completed enrollment and follow-up assessments, however, study results have not yet been published. Basmisanil (Hoffmann-La Roche Pharmaceuticals) is a selective $GABA_A$ α5 negative allosteric modulator (NAM) that has been investigated in two multi-center, Phase 2, randomized, double-blind, placebo-controlled studies to improve cognition in DS in a 26-week treatment study of adolescents and adults ages 12–30 years

(CLEMATIS study, ClinicalTrials.gov identifier NCT02024789) and a pediatric population between 6 and 11 years of age (ClinicalTrials.gov identifier NCT02484703). Unpublished results from the CLEMATIS study showed that Basmisanil was not associated with significant impacts on cognition or adaptive behavior in young adults and adolescents with DS, leading to early discontinuation of the study in the pediatric population (age 6–11 years) (Statement on CLEMATIS trial 2016c).

ELND005 (scyllo-Inositol) is an amyloid anti-aggregation agent purported to have two potential benefits for people with DS: (1) prevent the accumulation of plaques that might contribute to AD and (2) improve working memory and cognitive functioning by regulating myo-inositol levels in the brain. A clinical trial of ELND005 in neurotypically developed adults with AD did not demonstrate significant effects on cognition or adaptive function (Salloway et al., 2011). A recent phase II study in young adults with DS and without dementia showed that ELND005 was determined to have an acceptable safety and tolerability profile, and there were no serious adverse events reported in the study (Rafii et al., 2017). Of the 15 subjects who had neuropsychiatric symptoms at baseline, improvements (decreased Cumming's NPI-total score) were observed at 4 weeks as follows: in 1 of 3 placebo subjects, in 0 of 4 subjects receiving 250 mg daily of ELND005, and in 7 of 8 subjects receiving 250 mg twice daily of ELND005. There were no significant overall treatment group-related trends on cognitive or behavioral measures.

EGCG, a compound found in green tea leaves, has also been investigated in human clinical trials for DS. A recent randomized, placebo-controlled pilot study by De la Torre et al. (2014) tested the effects of EGCG from green tea extract on cognition in young adults with DS. The authors concluded that treatment with EGCG for 3 months reversed cognitive deficits in memory recognition, working memory and quality of life. A second, Phase 2 study by the same group has been completed and revealed that a combination of EGCG and cognitive training for 12 months was more effective than placebo and cognitive training at improving visual recognition memory, inhibitory control, and adaptive behavior (de la Torre et al., 2016). Phase 3 trials with a larger population of individuals with DS will be needed to assess and confirm the long-term efficacy of EGCG and cognitive training.

Several additional interventions targeting Alzheimer pathogenesis are currently being explored in clinical trials for DS. Recently, a vaccine targeting Aβ protein has been developed (ACI-24) that is designed to stimulate the immune system to prevent accumulation of amyloid plaques and enhance clearance (2015). This vaccine is currently being investigated in people with DS in a Phase 1 study (ClinialTrials.gov identifier NCT02738450). Intranasal glulisine, a rapid-acting insulin, has been investigated in AD due to the role of insulin signaling with Alzheimer pathogenesis (Rosenbloom et al., 2014). Intranasal glulisine is currently being investigated in adults with DS to determine safety, feasibility, and cognitive effect on memory measures (ClinicalTrials.gov identifier NCT02432716). Transdermal nicotine is also being investigated as a treatment for cognitive decline in adults with DS to establish safety, tolerability and efficacy for cognitive performance (ClinicalTrials.gov identifier NCT01778946).

Finally, a pilot study has begun at the University of Texas Southwestern Medical Center investigating effects of prenatal fluoxetine treatment in pregnant mothers with a fetal diagnosis of DS or positive screen for DS with non-invasive prenatal testing (2016b). Participants' children with DS will then receive postnatal treatment with fluoxetine until 2 years of age. Primary outcomes of this trial will be feasibility and safety, with efficacy measured using a broad neurodevelopmental scale at 6 months, 1 year, and 2 years of age.

The history of clinical trials in DS indicates that the majority of intervention studies conducted so far have focused on adolescent or adult populations. While the initial focus on this age range was required to establish safety of pharmacological interventions, these studies have had limited success in demonstrating efficacy. This highlights the need for future studies to investigate potential therapeutic effects of earlier interventions on cognitive function. As pharmacological interventions are likely to have the greatest impact during the critical times of brain development, directing future studies toward younger populations may hold greater promise for influencing cognition in people with DS (Stagni et al., 2015). Additionally, future studies may be able to use targeted approaches to identify and analyze potential sub-groups of responders to interventions, as individual differences may contribute significant variability in cognitive measures and potential responses to medication. Finally, defining appropriate measures for children of various age ranges will be important for future studies to address, as lack of generally accepted endpoints to assess efficacy in individuals with DS and other intellectual disabilities has been a major challenge in previous clinical trials.

4.1 | Considerations for clinical trials

Conducting clinical trials research in DS can be associated with inherent challenges in recruitment, retention, consenting, logistics of conducting study procedures, and assessments of safety and efficacy. We describe here some of the current challenges and potential strategies to address them from the perspective of investigators experienced with research in this population (see Table 2).

Despite the rich history in clinical trials research in DS, the limitations in the prior studies highlight a unique opportunity for industry to make a significant contribution to future investigations of interventions for cognition in DS. While Ts65Dn pre-clinical findings in mouse models of DS have been primarily focused on hippocampal-specific interventions and measures, translation of these findings to clinical trials may be limited by the more complex cognitive and behavioral phenotype seen in humans. The choice of efficacy measures also presents a significant challenge, as no single measure that could be used to evaluate treatment efficacy currently exists that meets all criteria for ideal clinical and regulatory endpoints. When selecting outcome measures, important considerations are test–retest reliability, suitability of measures for specific age ranges, and measures that do not exhibit large practice, ceiling, or floor effects (Liogier d'Ardhuy et al., 2015). Liogier d'Ardhuy et al. (2015) from Hoffmann-La Roche Pharmaceuticals assessed reliability and

TABLE 2 Considerations for clinical trials in Down syndrome

Recruitment and communication with potential participants:	**Promoting participant comfort:**
○ Establish realistic expectations through telephone screening and informing parents of the inclusion/exclusion criteria and tasks.	○ Prepare a written/visual schedule and review the schedule after each task.
○ In situations where families have participated in prior trials of interventions that have been found to lack efficacy, communication and sensitivity to concerns from families is important to facilitate their understanding of the research process and prevent discouragement from enrolling in future studies.	○ Consider creating Social Stories (visual storybooks breaking down each procedure into incremental steps).
Informed consent:	○ Potential anxiety during safety assessments like electrocardiogram (EKG) or electroencephalogram (EEG) may be managed by allowing participants to review a visual depiction of the procedures and to touch and familiarize themselves with equipment.
○ Allow ample time and use simple, clear and age-appropriate language in materials.	○ Give ample time to acclimate to new settings and avoid rushing participants, particularly during the initial visits.
○ Have caregivers provide co-consent even when an adult participant with DS is his/her own legal guardian.	○ Provide consistent environments and try to have the same raters and study coordinators at each visit.
○ Investigators should assess for dissent from the participant with DS and discontinue their participation if the dissent is deemed significant.	○ Take short breaks with rewards or prizes.
	○ Encourage parents to take an active role in preparing and soothing their children. However, allowing participants to have some degree of autonomy (i.e., time away from their parents) may also give them greater confidence to tolerate testing like EKGs and blood draws.
Study design:	**Data analysis:**
○ Consider the order of procedure administration, such as effects of participating in neuropsychological testing directly after anxiety-producing procedures (e.g., having blood drawn). Some outcome measures could be affected by requirements for breaks, such as memory measures.	○ Researchers should consider the significant heterogeneity and complexity in the phenotype among individuals with neurodevelopmental disorders. Future studies may require analysis of sub-groups of participants that may show greater response to intervention. Larger study samples may be needed in future studies to help identify and assess these potential sub-groups.
○ Minimize potential practice effects on memory measurements, as repeating measurements may lead to false improvements in memory scores. Additionally, if different memory measures are administered sequentially, recall may be confounded by intrusion errors from another subtest. Administration of memory measures less frequently or spacing testing temporally may help address these issues.	○ Investigation of outliers in the data or careful stratification of sub-groups may provide important insights on individual factors that may contribute to the variability in response to intervention.
○ Consider modifying materials to make them more applicable to the study population. Some frequently used study measures in clinical trials of adolescents or adults are standardized for typically developing young children and include wording targeted to young children (e.g., "preschool version"). Parents could potentially perceive an assumption by investigators that their child should be functioning at a level lower than their chronological age.	
○ Consider using tools or technologies participants are familiar with, to avoid potential underestimation of cognitive ability.	

suitability of several measures of IQ, memory, executive function, and language in adolescents and adults DS (12–30 years) and provide recommendations on use of these measures in clinical trials for each age range. While this work represents an important initial advance, additional studies are needed to assess appropriate outcome measures for future clinical trials.

Although use of a randomized placebo-controlled design is critical to address potential intervention effects, available funding is often limited to support these study designs in developmental disabilities (Heller, Spiridigliozzi, Crissman, Sullivan-Saarela, Li, et al., 2006). Collaboration of investigators with industry is needed to support larger, multi-site randomized placebo-controlled designs to address

these limitations. Additionally, it is critical for industry to partner with clinicians who have expertise in cognition and DS in order to choose measures that are likely to be appropriate for the study population. Finally, as research avenues for different developmental disabilities (such as, Fragile X syndrome) often proceed in parallel, greater collaboration between investigators studying different conditions in both academia and industry is needed for the field to benefit from lessons learned within each group.

Industry-led studies provide an opportunity to address the key challenges inherent in clinical trials for DS in multiple ways. First, industry-led longitudinal non-interventional studies similar to the study by Liogier d'Ardhuy et al. (2015) provide an opportunity to establish the suitability of measures and characterizing variability, learning and practice effects across different stages of neuro-development. Industry-led patient-centered outcomes research through early engagement with families can also help to establish criteria for what constitutes a treatment benefit in individuals with DS. Additionally, collaboration of industry with the U.S. FDA, European Medicines Agency, key opinion leaders and advocacy groups will be important to obtain feedback and agreement on the clinical endpoints.

To facilitate research efforts, it will also be important for industry to collaborate with the Down Syndrome Medical Interest Group, which was created to help ensure state-of-the-art medical care for individuals with DS in the United States (DSMIG-DSMIG-USA). If a potential new pharmacological treatment is developed for DS, it will be essential for pharmaceutical companies to work with the DSMIG to collaborate with DS clinics across the country. Collaboration of industry with the NIH Down Syndrome Working Group will be important for identifying additional outcome measures for clinical trials. This working group created a research plan highlighting major goals for future research in DS, including a focus on creating a common set of measures for use across studies, age groups, and developmental domains (Eunice Kennedy Shriver National Institutes of Child Health and Development and NIH Down Syndrome Working Group 2014). Additional long-term objectives in this area are to develop better measures targeting specific brain regions and circuits to enhance cognitive batteries at specific developmental stages. As individuals with DS have shown differences in neural connectivity related to cognitive function (Anderson et al., 2013; Pujol et al., 2015), the NIH Research Plan on Down Syndrome has also emphasized future use of technologies like functional MRI, EEG, and PET scanning for detecting brain functional changes associated with potential interventions. Use of measures like the PROMIS program (Patient-Reported Outcomes Measurement Information System), a self-reported health assessment system created by the NIH (Gershon, Rothrock, Hanrahan, Bass, & Cella, 2010), could also potentially be adapted for individuals with disabilities and explored for use in future studies of interventions. Finally, continued work on validating the NIH Toolbox Cognitive Battery for intellectual disabilities to provide common clinical endpoints for cognition in individuals with DS and other conditions (Hessl et al., 2016) will be critical for future studies in both academia and industry.

5 | CONCLUSIONS

Clinical trials in DS have shifted from historically focusing on interventions that were targeted for AD and improving learning/memory toward the current focus on potential interventions to improve multiple facets of cognition and adaptive behavior in children and adults with DS. Despite the increased knowledge about cognition in DS, the body of research, to date, has significant limitations, including a focus on older study participants, limited information about reliability or suitability of study measures, and heterogeneity among individuals in study populations. Future research focusing on earlier interventions, development of appropriate outcome measures, identification of potential sub-groups of responders to interventions, and collaboration between industry, academia, advocacy, and regulatory groups will be important for addressing limitations and moving toward development of potential effective interventions for cognition in DS.

ACKNOWLEDGMENTS

The authors would like to thank Casey Evans, Margaret Pulsifer, and Alexandra Silva for conducting part of this research review, writing portions of this manuscript, and reviewing draft versions.

CONFLICTS OF INTEREST

Nicole Baumer has a sister with Down syndrome. Dr. Baumer also serves in a non-paid capacity on the Medical and Scientific Advisory Board for the Massachusetts Down Syndrome Congress and has received research support for conducting clinical trials from Hoffmann-La Roche. Maria-Clemencia Hernandez, Xavier Liogier d'Ardhuy, Paul Tamburri, and Patrick Phuong are employees of F. Hoffmann-La Roche Ltd. Priya S. Kishnani and Gail A. Spiridigliozzi have received research support for conducting clinical trials from Hoffmann-La Roche. Brian G. Skotko occasionally consults on the topic of Down syndrome through Gerson Lehrman Group. He receives remuneration from Down syndrome non-profit organizations for speaking engagements and associated travel expenses. Dr. Skotko receives annual royalties from Woodbine House, Inc., for the publication of his book, *Fasten Your Seatbelt: A Crash Course on Down Syndrome for Brothers and Sisters*. Within the past 2 years, he has received research funding from F. Hoffmann-La Roche, Inc. and Transition Therapeutics to conduct clinical trials on study drugs for people with Down syndrome. Dr. Skotko is occasionally asked to serve as an expert witness for legal cases where Down syndrome is discussed. He serves in a non-paid capacity on the Honorary Board of Directors for the Massachusetts Down Syndrome Congress, the Board of Directors for the Band of Angels Foundation, and the Professional Advisory Committee for the National Center for Prenatal and Postnatal Down Syndrome Resources. Dr. Skotko has a sister with Down syndrome. Jeannie Visootsak is currently a full-time employee of Ovid Therapeutics. Dr Visootsak's work on this

manuscript began when she was a full-time employee of Hoffmann La-Roche and prior to her employment at Ovid Therapeutics. Sarah J. Hart and Cesar Ochoa-Lubinoff have no conflicts of interest to declare.

ORCID

Sarah J. Hart http://orcid.org/0000-0003-0974-3209

REFERENCES

(2015). AC Immune: AD treatment and prevention (11/1/2016). Available online at: http://www.acimmune.com/en/ad-treatment-and-prevention/

(2016a). Biology of Down syndrome: Impacts across the biomedical spectrum (10/25/2016). Available online at: https://www.keystonesymposia.org/16A4

(2016b). A Pilot Feasibility Trial of Prenatal and Early Postnatal Fluoxetine Treatment for Intellectual Impairments of Down Syndrome (12/22/2016). Available online at: http://www.utsouthwestern.edu/research/fact/detail.html?studyid=STU%20032014-006

(2016c). Statement on CLEMATIS Trial. June 28, 2016. (10/15/2016). Available online at: http://www.roche.com/media/store/statements.htm

Abbeduto, L., Warren, S. F., & Conners, F. A. (2007). Language development in Down syndrome: From the prelinguistic period to the acquisition of literacy. *Mental Retardation and Developmental Disabilities Research Review*, *13*, 247–261.

Ahmed, A. H., & Oswald, R. E. (2010). Piracetam defines a new binding site for allosteric modulators of alpha-amino-3-hydroxy-5-methyl-4-isoxazole-propionic acid (AMPA) receptors. *Journal of Medicinal Chemistry*, *53*, 2197–2203.

Anderson, J. S., Nielsen, J. A., Ferguson, M. A., Burback, M. C., Cox, E. T., Dai, L., … Korenberg, J. R. (2013). Abnormal brain synchrony in Down syndrome. *Neuroimage Clinical*, *2*, 703–715.

Ash, J. A., Velazquez, R., Kelley, C. M., Powers, B. E., Ginsberg, S. D., Mufson, E. J., & Strupp, B. J. (2014). Maternal choline supplementation improves spatial mapping and increases basal forebrain cholinergic neuron number and size in aged Ts65Dn mice. *Neurobiology of Disease*, *70*, 32–42.

Bain, J., McLauchlan, H., Elliott, M., & Cohen, P. (2003). The specificities of protein kinase inhibitors: An update. *Biochemical Journal*, *371*, 199–204.

Bartesaghi, R., Haydar, T. F., Delabar, J. M., Dierssen, M., Martinez-Cue, C., & Bianchi, D. W. (2015). New perspectives for the rescue of cognitive disability in Down syndrome. *Journal of Neuroscience*, *35*, 13843–13852.

Beccaria, L., Marziani, E., Manzoni, P., Arvat, E., Valetto, M. R., Gianotti, L., … Chiumello, G. (1998). Further evidence of cholinergic impairment of the neuroendocrine control of the GH secretion in Down's syndrome. *Dementia and Geriatric Cognitive Disorders*, *9*, 78–81.

Becker, L., Mito, T., Takashima, S., & Onodera, K. (1991). Growth and development of the brain in Down syndrome. *Progress in Clinical and Biological Research*, *373*, 133–152.

Bennett, F. C., McClelland, S., Kriegsmann, E. A., Andrus, L. B., & Sells, C. J. (1983). Vitamin and mineral supplementation in Down's syndrome. *Pediatrics*, *72*, 707–713.

Berg, J. M., Kerman, B. H., Stern, J., & Mittwoch, U. (1961). Treatment of mongolism with pituitary extract. *Journal of Mental Science*, *107*, 475–480.

Berger-Sweeney, J. (2003). The cholinergic basal forebrain system during development and its influence on cognitive processes: Important questions and potential answers. *Neuroscience and Biobehavioral Reviews*, *27*, 401–411.

Bianchi, P., Ciani, E., Guidi, S., Trazzi, S., Felice, D., Grossi, G., … Bartesaghi, R. (2010). Early pharmacotherapy restores neurogenesis and cognitive performance in the Ts65Dn mouse model for Down syndrome. *Journal of Neuroscience*, *30*, 8769–8779.

Bidder, R. T., Gray, P., Newcombe, R. G., Evans, B. K., & Hughes, M. (1989). The effects of multivitamins and minerals on children with Down syndrome. *Developmental Medicine and Child Neurology*, *31*, 532–537.

Bittles, A. H., Bower, C., Hussain, R., & Glasson, E. J. (2007). The four ages of Down syndrome. *European Journal of Public Health*, *17*, 221–225.

Blehaut, H., Mircher, C., Ravel, A., Conte, M., de Portzamparc, V., Poret, G., … Sturtz, F. G. (2010). Effect of leucovorin (folinic acid) on the developmental quotient of children with Down's syndrome (trisomy 21) and influence of thyroid status. *PLoS ONE*, *5*, e8394.

Boada, R., Hutaff-Lee, C., Schrader, A., Weitzenkamp, D., Benke, T. A., Goldson, E. J., & Costa, A. C. (2012). Antagonism of NMDA receptors as a potential treatment for Down syndrome: A pilot randomized controlled trial. *Translational Psychiatry*, *2*, e141.

Braudeau, J., Delatour, B., Duchon, A., Pereira, P. L., Dauphinot, L., de Chaumont, F., … Potier, M. C. (2011). Specific targeting of the GABA-A receptor alpha5 subtype by a selective inverse agonist restores cognitive deficits in Down syndrome mice. *Journal of Psychopharmacology*, *25*, 1030–1042.

Bumbalo, T. S. (1964). Negative results: Treatment of down syndrome with the "U" series. *Journal of the American Medical Association*, *361*.

Carlesimo, G. A., Marotta, L., & Vicari, S. (1997). Long-term memory in mental retardation: Evidence for a specific impairment in subjects with Down's syndrome. *Neuropsychologia*, *35*, 71–79.

Carr, J. (2005). Stability and change in cognitive ability over the life span: A comparison of populations with and without Down's syndrome. *Journal of Intellectual Disability Research*, *49*, 915–928.

Casanova, M. F., Walker, L. C., Whitehouse, P. J., & Price, D. L. (1985). Abnormalities of the nucleus basalis in Down's syndrome. *Annals of Neurology*, *18*, 310–313.

Chapman, R. S. (2006). Language learning in Down syndrome: The speech and language profile compared to adolescents with cognitive impairment of unknown origin. *Down's Syndrome, Research and Practice: The journal of the Sarah Duffen Centre*, *10*, 61–66.

Chapman, R. S., & Hesketh, L. J. (2001). Language, cognition, and short-term memory in individuals with Down syndrome. *Down's Syndrome, Research and Practice: The journal of the Sarah Duffen Centre*, *7*, 1–7.

Chen, H. S., & Lipton, S. A. (1997). Mechanism of memantine block of NMDA-activated channels in rat retinal ganglion cells: Uncompetitive antagonism. *Journal of Physiology*, *499*(Pt 1), 27–46.

Chen, H. S., Pellegrini, J. W., Aggarwal, S. K., Lei, S. Z., Warach, S., Jensen, F. E., & Lipton, S. A. (1992). Open-channel block of N-methyl-D-aspartate (NMDA) responses by memantine: Therapeutic advantage `against NMDA receptor-mediated neurotoxicity. *Journal of Neuroscience*, *12*, 4427–4436.

Clark, S., Schwalbe, J., Stasko, M. R., Yarowsky, P. J., & Costa, A. C. (2006). Fluoxetine rescues deficient neurogenesis in hippocampus of the Ts65Dn mouse model for Down syndrome. *Experimental Neurology*, *200*, 256–261.

Coleman, M., Sobel, S., Bhagavan, H. N., Coursin, D., Marquardt, A., Guay, M., & Hunt, C. (1985). A double blind study of vitamin B6 in Down's syndrome infants. Part 1–Clinical and biochemical results. *Journal of Mental Deficiency Research*, *29*(Pt 3), 233–240.

Collinson, N., Kuenzi, F. M., Jarolimek, W., Maubach, K. A., Cothliff, R., Sur, C., … Rosahl, T. W. (2002). Enhanced learning and memory and altered GABAergic synaptic transmission in mice lacking the alpha 5 subunit of the GABAA receptor. *Journal of Neuroscience*, *22*, 5572–5580.

Corey-Bloom, J., Anand, R., & Veach, J. (1998). A randomized trial evaluating the efficacy and safety of ENA 713 (rivastigmine tartrate), a new acetylcholinesterase inhibitor, in patients with mild to moderately severe Alzheimer's disease. *International Journal of Geriatric Psychopharmacology*, *1*, 55–65.

Costa, A. C., & Scott-McKean, J. J. (2013). Prospects for improving brain function in individuals with Down syndrome. *CNS Drugs*, *27*, 679–702.

Costa, A. C., Scott-McKean, J. J., & Stasko, M. R. (2008). Acute injections of the NMDA receptor antagonist memantine rescue performance deficits of the Ts65Dn mouse model of Down syndrome on a fear conditioning test. *Neuropsychopharmacology*, *33*, 1624–1632.

Couzens, D., Cuskelly, M., & Haynes, M. (2011). Cognitive development and Down syndrome: Age-related change on the Stanford-Binet test (fourth edition). *American Journal on Intellectual and Developmental Disabilities*, *116*, 181–204.

Davisson, M. T., Schmidt, C., & Akeson, E. C. (1990). Segmental trisomy of murine chromosome 16: A new model system for studying Down syndrome. *Progress in Clinical and Biological Research*, *360*, 263–280.

de Graaf, G., Buckley, F., & Skotko, B. G. (2015). Estimates of the live births, natural losses, and elective terminations with Down syndrome in the United States. *American Journal of Medical Genetics Part A*, *167A*, 756–767.

de Graaf, G., Buckley, F., & Skotko, B. G. (2017). Estimation of the number of people with Down syndrome in the United States. *Genetics in Medicine*, *19*, 439–447.

de la Torre, R., de Sola, S., Hernandez, G., Farre, M., Pujol, J., Rodriguez, J., … Group, T. S. (2016). Safety and efficacy of cognitive training plus epigallocatechin-3-gallate in young adults with Down's syndrome (TESDAD): A double-blind, randomised, placebo-controlled, phase 2 trial. *Lancet Neurology*, *15*, 801–810.

De la Torre, R., De Sola, S., Pons, M., Duchon, A., de Lagran, M. M., Farre, M., … Dierssen, M. (2014). Epigallocatechin-3-gallate, a DYRK1A inhibitor, rescues cognitive deficits in Down syndrome mouse models and in humans. *Molecular Nutrition & Food Research*, *58*, 278–288.

Deidda, G., Parrini, M., Naskar, S., Bozarth, I. F., Contestabile, A., & Cancedda, L. (2015). Reversing excitatory GABAAR signaling restores synaptic plasticity and memory in a mouse model of Down syndrome. *Nature Medicine*, *21*, 318–326.

DHHS. (2016). National Institutes of Health: Turning discovery into health (10/25/2016).

DSMIG-USA. Down Syndrome Medical Interest Group—USA. (2016). Available online at: http://www.dsmig-usa.wildapricot.org/

Dykens, E. M., Hodapp, R. M., & Evans, D. W. (2006). Profiles and development of adaptive behavior in children with Down syndrome. *Down's Syndrome, Research and Practice: The journal of the Sarah Duffen Centre*, *9*, 45–50.

Eisenberg, J., Hamburger-Bar, R., & Belmaker, R. H. (1984). The effect of vasopressin treatment on learning in Down's syndrome. *Journal of Neural Transmission*, *60*, 143–147.

Ellis, J. M., Tan, H. K., Gilbert, R. E., Muller, D. P., Henley, W., Moy, R., … Logan, S. (2008). Supplementation with antioxidants and folinic acid for children with Down's syndrome: Randomised controlled trial. *BMJ*, *336*, 594–597.

Emre, M., Aarsland, D., Albanese, A., Byrne, E. J., Deuschl, G., De Deyn, P. P., … Lane, R. (2004). Rivastigmine for dementia associated with Parkinson's disease. *New England Journal of Medicine*, *351*, 2509–2518.

Fernandez, F., Morishita, W., Zuniga, E., Nguyen, J., Blank, M., Malenka, R. C., & Garner, C. C. (2007). Pharmacotherapy for cognitive impairment in a mouse model of Down syndrome. *Nature Neuroscience*, *10*, 411–413.

Fidler, D. J. (2006). The emergence of a syndrome-specific personality profile in young children with Down syndrome. *Down's Syndrome, Research and Practice: The journal of the Sarah Duffen Centre*, *10*, 53–60.

Fidler, D. J., & Nadel, L. (2007). Education and children with Down syndrome: Neuroscience, development, and intervention. *Mental Retardation and Devevelopmental Disabilities Research Review*, *13*, 262–271.

Fidler, D. J., Philofsky, A., & Hepburn, S. L. (2007). Language phenotypes and intervention planning: Bridging research and practice. *Mental Retardation and Devevelopmental Disabilities Research Review*, *13*, 47–57.

Finkel, S. I. (2004). Effects of rivastigmine on behavioral and psychological symptoms of dementia in Alzheimer's disease. *Clinical Therapeutics*, *26*, 980–990.

Florez, J., del Arco, C., Gonzalez, A., Pascual, J., & Pazos, A. (1990). Autoradiographic studies of neurotransmitter receptors in the brain of newborn infants with Down syndrome. *American Journal of Medical Genetics Supplement*, *7*, 301–305.

Gardiner, K. J. (2015). Pharmacological approaches to improving cognitive function in Down syndrome: Current status and considerations. *Drug Design, Development and Therapy*, *9*, 103–125.

Gershon, R. C., Rothrock, N., Hanrahan, R., Bass, M., & Cella, D. (2010). The use of PROMIS and assessment center to deliver patient-reported outcome measures in clinical research. *Journal of Applied Measurement*, *11*, 304–314.

Giacomini, A., Stagni, F., Trazzi, S., Guidi, S., Emili, M., Brigham, E., … Bartesaghi, R. (2015). Inhibition of APP gamma-secretase restores Sonic Hedgehog signaling and neurogenesis in the Ts65Dn mouse model of Down syndrome. *Neurobiology of Disease*, *82*, 385–396.

Grieco, J., Pulsifer, M., Seligsohn, K., Skotko, B., & Schwartz, A. (2015). Down syndrome: Cognitive and behavioral functioning across the lifespan. *American Journal of Medical Genetics Part C: Seminars in Medical Genetics*, *169*, 135–149.

Guedj, F., Bianchi, D. W., & Delabar, J. M. (2014). Prenatal treatment of Down syndrome: A reality? *Current Opinion in Obstetrics and Gynecology*, *26*, 92–103.

Guidi, S., Stagni, F., Bianchi, P., Ciani, E., Giacomini, A., De Franceschi, M., … Bartesaghi, R. (2014). Prenatal pharmacotherapy rescues brain development in a Down's syndrome mouse model. *Brain*, *137*, 380–401.

Guidi, S., Stagni, F., Bianchi, P., Ciani, E., Ragazzi, E., Trazzi, S., … Bartesaghi, R. (2013). Early pharmacotherapy with fluoxetine rescues dendritic pathology in the Ts65Dn mouse model of down syndrome. *Brain Pathology*, *23*, 129–143.

Hanney, M., Prasher, V., Williams, N., Jones, E. L., Aarsland, D., Corbett, A., … Researchers, M. T. (2012). Memantine for dementia in adults older than 40 years with Down's syndrome (MEADOWS): A randomised, double-blind, placebo-controlled trial. *Lancet*, *379*, 528–536.

Harrell, R. F., Capp, R. H., Davis, D. R., Peerless, J., & Ravitz, L. R. (1981). Can nutritional supplements help mentally retarded children? An exploratory study. *Proceedings of the National Academy of Sciences of the United States of America*, *78*, 574–578.

Head, E., Powell, D., Gold, B. T., & Schmitt, F. A. (2012). Alzheimer's disease in down syndrome. *European Journal of Neurodegenerative Disease*, *1*, 353–364.

Heaton-Ward, W. A. (1962). Inference and suggestion in a clinical trial (Niamid in mongolism). *The British Journal of Psychiatry*, *108*, 865–870.

Heller, J. H., Spiridigliozzi, G. A., Crissman, B. G., McKillop, J. A., Yamamoto, H., & Kishnani, P. S. (2010). Safety and efficacy of rivastigmine in adolescents with Down syndrome: Long-term follow-up. *Journal of Child and Adolescent Psychopharmacology*, *20*, 517–520.

Heller, J. H., Spiridigliozzi, G. A., Crissman, B. G., Sullivan-Saarela, J. A., Li, J. S., & Kishnani, P. S. (2006). Clinical trials in children with Down syndrome: Issues from a cognitive research perspective. *American Journal of Medical Genetics Part C: Seminars in Medical Genetics*, *142C*, 187–195.

Heller, J. H., Spiridigliozzi, G. A., Crissman, B. G., Sullivan, J. A., Eells, R. L., Li, J. S., … Kishnani, P. S. (2006). Safety and efficacy of rivastigmine in adolescents with Down syndrome: A preliminary 20-week, open-label study. *Journal of Child and Adolescent Psychopharmacology*, *16*, 755–765.

Heller, J. H., Spiridigliozzi, G. A., Doraiswamy, P. M., Sullivan, J. A., Crissman, B. G., & Kishnani, P. S. (2004). Donepezil effects on language in children with Down syndrome: Results of the first 22-week pilot clinical trial. *American Journal of Medical Genetics Part A*, *130A*, 325–326.

Hessl, D., Sansone, S. M., Berry-Kravis, E., Riley, K., Widaman, K. F., Abbeduto, L., … Gershon, R. C. (2016). The NIH Toolbox Cognitive Battery for intellectual disabilities: Three preliminary studies and future directions. *Journal of Neurodevelopmental Disorders*, *8*, 35.

Jarrold, C., Nadel, L., & Vicari, S. (2009). Memory and neuropsychology in Down syndrome. *Down Syndrome Research and Practice*, *12*, 68–73.

Karmiloff-Smith, A., Al-Janabi, T., D'Souza, H., Groet, J., Massand, E., Mok, K., … Strydom, A. (2016). The importance of understanding individual differences in Down syndrome. *F1000Research*, *5*, 389.

Kasari, C., & Freeman, S. F. (2001). Task-related social behavior in children with Down syndrome. *American Journal of Mental Retardation*, *106*, 253–264.

Keeling, L. A., Spiridigliozzi, G. A., Hart, S. J., Baker, J. A., Jones, H. N., & Kishnani, P. S. (in press). Challenges in measuring the effects of pharmacological interventions on cognitive and adaptive functioning in individuals with Down syndrome: A systematic review. *American Journal of Medical Genetics Part A*.

Kishnani, P. S., Heller, J. H., Spiridigliozzi, G. A., Lott, I., Escobar, L., Richardson, S., … McRae, T. (2010). Donepezil for treatment of cognitive dysfunction in children with Down syndrome aged 10–17. *American Journal of Medical Genetics Part A*, *152A*, 3028–3035.

Kishnani, P. S., Sommer, B. R., Handen, B. L., Seltzer, B., Capone, G. T., Spiridigliozzi, G. A., … McRae, T. (2009). The efficacy, safety, and tolerability of donepezil for the treatment of young adults with Down syndrome. *American Journal of Medical Genetics Part A*, *149A*, 1641–1654.

Kishnani, P. S., Sullivan, J. A., Walter, B. K., Spiridigliozzi, G. A., Doraiswamy, P. M., & Krishnan, K. R. (1999). Cholinergic therapy for Down's syndrome. *Lancet*, *353*, 1064–1065.

Kleschevnikov, A. M., Belichenko, P. V., Faizi, M., Jacobs, L. F., Htun, K., Shamloo, M., & Mobley, W. C. (2012). Deficits in cognition and synaptic plasticity in a mouse model of Down syndrome ameliorated by GABAB receptor antagonists. *Journal of Neuroscience*, *32*, 9217–9227.

Kondoh, T., Kanno, A., Itoh, H., Nakashima, M., Honda, R., Kojima, M., … Moriuchi, H. (2011). Donepezil significantly improves abilities in daily lives of female Down syndrome patients with severe cognitive impairment: A 24-week randomized, double-blind, placebo-controlled trial. *International Journal of Psychiatry in Medicine*, *41*, 71–89.

Korenberg, J. R., Kawashima, H., Pulst, S. M., Ikeuchi, T., Ogasawara, N., Yamamoto, K., … Epstein, C. J. (1990). Molecular definition of a region of chromosome 21 that causes features of the Down syndrome phenotype. *American Journal of Human Genetics*, *47*, 236–246.

LaFerla, F. M., Green, K. N., & Oddo, S. (2007). Intracellular amyloid-beta in Alzheimer's disease. *Nature Reviews: Neuroscience*, *8*, 499–509.

Letourneau, A., Santoni, F. A., Bonilla, X., Sailani, M. R., Gonzalez, D., Kind, J., … Antonarakis, S. E. (2014). Domains of genome-wide gene expression dysregulation in Down's syndrome. *Nature*, *508*, 345–350.

Liogier d'Ardhuy, X., Edgin, J. O., Bouis, C., de Sola, S., Goeldner, C., Kishnani, P., … Khwaja, O. (2015). Assessment of cognitive scales to examine memory, executive function and language in individuals with down syndrome: Implications of a 6-month observational study. *Frontiers in Behavioral Neuroscience*, *9*, 300.

Lipton, S. A. (2007). Pathologically-activated therapeutics for neuro-protection: Mechanism of NMDA receptor block by memantine and S-nitrosylation. *Current Drug Targets*, *8*, 621–632.

Livingstone, N., Hanratty, J., McShane, R., & Macdonald, G. (2015). Pharmacological interventions for cognitive decline in people with Down syndrome. *The Cochrane Database of Systematic Reviews*, *10*, CD011546.

Lobaugh, N. J., Karaskov, V., Rombough, V., Rovet, J., Bryson, S., Greenbaum, R., … Koren, G. (2001). Piracetam therapy does not enhance cognitive functioning in children with down syndrome. *Archives of Pediatrics and Adolescent Medicine*, *155*, 442–448.

Lonsdale, D., & Kissling, C. D. (1986). Clinical trials with thiamine tetrahydrofurfuryl disulfide (TTFD) in Down's syndrome. *Journal of Orthomolecular Medicine*, *1*, 169–175.

Lott, I. T., Doran, E., Nguyen, V. Q., Tournay, A., Head, E., & Gillen, D. L. (2011). Down syndrome and dementia: A randomized, controlled trial of antioxidant supplementation. *American Journal of Medical Genetics Part A*, *155A*, 1939–1948.

Martin, G. E., Klusek, J., Estigarribia, B., & Roberts, J. E. (2009). Language characteristics of individuals with down syndrome. *Topics in Language Disorders*, *29*, 112–132.

Martinez-Cue, C., Martinez, P., Rueda, N., Vidal, R., Garcia, S., Vidal, V., … Hernandez, M. C. (2013). Reducing GABAA alpha5 receptor-mediated inhibition rescues functional and neuromorphological deficits in a mouse model of down syndrome. *Journal of Neuroscience*, *33*, 3953–3966.

Moon, J., Chen, M., Gandhy, S. U., Strawderman, M., Levitsky, D. A., Maclean, K. N., & Strupp, B. J. (2010). Perinatal choline supplementation improves cognitive functioning and emotion regulation in the Ts65Dn mouse model of Down syndrome. *Behavioral Neuroscience*, *124*, 346–361.

Nadel, L. (2003). Down's syndrome: A genetic disorder in biobehavioral perspective. *Genes, Brain, and Behavior*, *2*, 156–166.

NCBI. (2016). NCBI Map Viewer (11/17/2016). Available online at: https://www.ncbi.nlm.nih.gov/mapview/maps.cgi?taxid=9606&chr=21

Netzer, W. J., Powell, C., Nong, Y., Blundell, J., Wong, L., Duff, K., … Greengard, P. (2010). Lowering beta-amyloid levels rescues learning and memory in a Down syndrome mouse model. *PLoS ONE*, *5*, e10943.

Nicham, R., Weitzdorfer, R., Hauser, E., Freidl, M., Schubert, M., Wurst, E., … Seidl, R. (2003). Spectrum of cognitive, behavioural and emotional problems in children and young adults with Down syndrome. *Journal of Neural Transmission Supplementum*, *67*, 173–191.

NICHD. (2014). Down syndrome directions: The National Institutes of Health research plan on Down syndrome (10/25/2016). Available online at: https://www.nichd.nih.gov/publications/pubs/Documents/DSResearchPlan_2014.pdf

Patterson, D. (2009). Molecular genetic analysis of Down syndrome. *Human Genetics*, *126*, 195–214.

Prandini, P., Deutsch, S., Lyle, R., Gagnebin, M., Delucinge Vivier, C., Delorenzi, M., … Antonarakis, S. E. (2007). Natural gene-expression variation in Down syndrome modulates the outcome of gene-dosage imbalance. *American Journal of Human Genetics*, *81*, 252–263.

Pueschel, S. M., Reed, R. B., Cronk, C. E., & Goldstein, B. I. (1980). 5-hydroxytryptophan and pyridoxine. Their effects in young children with Down's syndrome. *American Journal of Diseases of Children*, *134*, 838–844.

Pujol, J., del Hoyo, L., Blanco-Hinojo, L., de Sola, S., Macia, D., Martinez-Vilavella, G., … de la Torre, R. (2015). Anomalous brain functional connectivity contributing to poor adaptive behavior in Down syndrome. *Cortex*, *64*, 148–156.

Rafii, M. S., Skotko, B. G., McDonough, M. E., Pulsifer, M., Evans, C., Doran, E., … Group, E. D. S. (2017). A randomized, double-blind, placebo-controlled, phase II study of oral ELND005 (scyllo-inositol) in young adults with Down syndrome without dementia. *Journal of Alzheimer's Disease*, *58*, 401–411.

Reeves, R. H., Irving, N. G., Moran, T. H., Wohn, A., Kitt, C., Sisodia, S. S., … Davisson, M. T. (1995). A mouse model for Down syndrome exhibits learning and behaviour deficits. *Nature Genetics*, *11*, 177–184.

Rosenbloom, M. H., Barclay, T. R., Pyle, M., Owens, B. L., Cagan, A. B., Anderson, C. P., … Hanson, L. R. (2014). A single-dose pilot trial of intranasal rapid-acting insulin in apolipoprotein E4 carriers with mild-moderate Alzheimer's disease. *CNS Drugs*, *28*, 1185–1189.

Rosler, M., Anand, R., Cicin-Sain, A., Gauthier, S., Agid, Y., Dal-Bianco, P., … Gharabawi, M. (1999). Efficacy and safety of rivastigmine in patients with Alzheimer's disease: International randomised controlled trial. *BMJ*, *318*, 633–638.

Rosler, M., Retz, W., Retz-Junginger, P., & Dennler, H. J. (1998). Effects of two-year treatment with the cholinesterase inhibitor rivastigmine on behavioural symptoms in Alzheimer's disease. *Behavioural Neurology*, *11*, 211–216.

Sacks, B., & Smith, S. (1989). People with Down's syndrome can be distinguished on the basis of cholinergic dysfunction. *Journal of Neurology, Neurosurgery and Psychiatry*, *52*, 1294–1295.

Salloway, S., Sperling, R., Keren, R., Porsteinsson, A. P., van Dyck, C. H., Tariot, P. N., . . . Investigators, E. A. (2011). A phase 2 randomized trial of ELND005, scyllo-inositol, in mild to moderate Alzheimer disease. *Neurology*, *77*, 1253–1262.

Shapiro, B. L. (1997). Whither Down syndrome critical regions? *Human Genetics*, *99*, 421–423.

Silverman, W. (2007). Down syndrome: Cognitive phenotype. *Mental Retardation and Devevelopmental Disabilities Research Review*, *13*, 228–236.

Smith, G. F., Spiker, D., Peterson, C. P., Cicchetti, D., & Justine, P. (1984). Use of megadoses of vitamins with minerals in Down syndrome. *Journal of Pediatrics*, *105*, 228–234.

Spiridigliozzi, G. A., Hart, S. J., Heller, J. H., Schneider, H. E., Baker, J. A., Weadon, C., . . . Kishnani, P. S. (2016). Safety and efficacy of rivastigmine in children with Down syndrome: A double blind placebo controlled trial. *American Journal of Medical Genetics Part A*, *170*, 1545–1555.

Stagni, F., Giacomini, A., Guidi, S., Ciani, E., & Bartesaghi, R. (2015). Timing of therapies for Down syndrome: the sooner, the better. Frontiers in Behavioral Neuroscience, 9, 265. t21RS. T21RS International Conference 2015: Changing paradigms in Down syndrome (10/25/2016). Available online at: http://www.t21rs.org/news-meetings/t21rs-international-conference-2015-paris

van Trotsenburg, A. S., Vulsma, T., van Rozenburg-Marres, S. L., van Baar, A. L., Ridder, J. C., Heymans, H. S., . . . deVijlder, J. J. (2005). The effect of thyroxine treatment started in the neonatal period on development and growth of two-year-old Down syndrome children: A randomized clinical trial. *The Journal of Clinical Endocrinology and Metabolism*, *90*, 3304–3311.

Velazquez, R., Ash, J. A., Powers, B. E., Kelley, C. M., Strawderman, M., Luscher, Z. I., . . . Strupp, B. J. (2013). Maternal choline supplementation improves spatial learning and adult hippocampal neurogenesis in the Ts65Dn mouse model of Down syndrome. *Neurobiology of Disease*, *58*, 92–101.

Vicari, S. (2001). Implicit versus explicit memory function in children with Down and Williams syndrome. *Down's Syndrome, Research and Practice: The journal of the Sarah Duffen Centre*, *7*, 35–40.

Weathers, C. (1983). Effects of nutritional supplementation on IQ and certain other variables associated with Down syndrome. *American Journal of Mental Deficiency*, *88*, 214–217.

Wetmore, D. Z., & Garner, C. C. (2010). Emerging pharmacotherapies for neurodevelopmental disorders. *Journal of Developmental and Behavioral Pediatrics*, *31*, 564–581.

Winblad, B. (2005). Piracetam: A review of pharmacological properties and clinical uses. *CNS Drug Reviews*, *11*, 169–182.

Wolfe, M. S. (2008). Inhibition and modulation of gamma-secretase for Alzheimer's disease. *Neurotherapeutics*, *5*, 391–398.

Xie, W., Ramakrishna, N., Wieraszko, A., & Hwang, Y. W. (2008). Promotion of neuronal plasticity by (−)-epigallocatechin-3-gallate. *Neurochemical Research*, *33*, 776–783.

How to cite this article: Hart SJ, Visootsak J, Tamburri P, et al. Pharmacological interventions to improve cognition and adaptive functioning in Down syndrome: Strides to date. *Am J Med Genet Part A*. 2017;1–13. https://doi.org/10.1002/ajmg.a.38465