

Rodent models of Genetic and Chromosomal Variations in Psychiatric disorders

Jun Nomura¹, Geetha Kannan², and Toru Takumi¹

¹ RIKEN Brain Science Institute, Wako, Saitama, 351-0198, Japan

² Department of Microbiology and Immunology, University of Michigan, Ann Arbor, MI, USA

Short Running Title: Rodent models in psychiatric disorders

Jun Nomura, PhD, jun.nomura@riken.jp

Geetha Kannan, PhD, gkannan@umich.edu

Toru Takumi, MD, PhD, toru.takumi@riken.jp

Corresponding Author: Toru Takumi, MD. PhD., 2-1 Hirosawa, Wako, Saitama 351-0198, Japan, Tel: +81-48-467-5906, Fax: +81-048-467-6079, E-mail: toru.takumi@riken.jp

This is the author manuscript accepted for publication and has undergone full peer review but has not been through the copyediting, typesetting, pagination and proofreading process, which may lead to differences between this version and the Version of Record. Please cite this article as doi: [10.1111/pcn.12524](https://doi.org/10.1111/pcn.12524)

Abstract

Elucidating the molecular basis of complex human psychiatric disorders is challenging due to the multitude of factors that underpin these disorders. Genetic and chromosomal changes are two factors that have been suggested to be involved in psychiatric disorders. Indeed, numerous risk loci have been identified in autism spectrum disorders (ASD), schizophrenia, and related psychiatric disorders. Here, we introduce genetic animal models that disturb excitatory-inhibitory (E/I) balance in the brain and animal models mirroring human chromosomal abnormalities, both of which may be implicated in ASD pathophysiology. In addition, we discuss recent unique translational research using rodent models, such as *Cntnap2* knockout (KO) mouse, *Mecp2* mutant mouse, *Pick1* KO mouse, and NVHL (Neonatal Ventral Hippocampal Lesion) rat. By using these models, several types of drugs are administered during the developmental period to see the effect on psychotic symptoms and neural activities in adults. The accumulating evidence from recent animal studies provides informative intervention strategy as a translational research.

Author Manuscript

Introduction

The pathophysiology of psychiatric disorders is complex. Factors that may contribute to the development of psychiatric disease include imbalance in excitatory and inhibitory (E/I) transmission and chromosomal changes. The E/I imbalance can be caused by modulation of receptors that in turn lead to abnormal activity of glutamatergic neurons and γ -aminobutyric acid (GABA)ergic interneurons. Indeed, genes involved in E/I transmission are reported to be dysregulated in postmortem brains of patients with schizophrenia and Rett Syndrome (RTT)¹². Chromosomal abnormalities are another factor associated with psychiatric disorders. Copy number variation (CNVs) are duplications or deletions of genomic loci, ranging from kilobase (Kb) to megabase (Mb), and can pertain to either a single gene or multiple genes³. Autism Spectrum Disorder (ASD), schizophrenia, and RTT are diseases attributed to CNVs.

Uncovering the mechanism whereby specific genetic abnormalities lead to psychiatric disease is difficult to do in clinical studies. However, rodent models can be a useful tool in understanding the potential role of E/I imbalance and chromosomal abnormalities on the development of psychiatric disorders. In addition, rodent models permit the systematic testing of therapeutics during different stages of disease onset to determine the efficacy of the timing of treatment interventions. This review will focus on rodent models of E/I imbalance and CNVs relevant to psychiatric disorders. Rodent models suggesting the usefulness of treatment early in disease progression are also discussed.

1. Rodent models of Excitatory and Inhibitory (E/I) imbalance

Patients with ASD and other psychiatric disorders are thought to have E/I imbalance in the brain⁴⁵. ASD patients frequently present with epileptic seizures, with a prevalence rate estimated to range between 8 and 25%⁶. Epileptic seizures may be due to excess excitatory neuronal inputs of the cortical circuit, suggesting E/I imbalance in the brains of ASD patients⁷. Indeed, a recent study showed that three-dimensional cultures (cortical organoids) derived from individuals with severe idiopathic ASD have normal levels of glutamatergic excitatory neurons and increased numbers of GABAergic inhibitory neurons⁸. Also, a functional genetic analysis on patients with schizophrenia has uncovered genes involved in glutamatergic and GABAergic neurotransmission, suggesting these patients also have E/I imbalance⁹. In addition, patients with the X-linked neurodevelopmental disorder, RTT, have been shown to have hypoexcitability in the forebrain, possibly indicating E/I imbalance³. Postmortem brain studies of RTT patients have shown a decrease in glutamatergic

±-amino-3-hydroxy-5-methylisoxazol-4-propionic acid receptor (AMPA) and *N*-methyl-*D*-aspartate receptor (NMDAR) densities in the putamen and a decrease in GABA receptors in the caudate¹⁰. These studies demonstrate the heterogeneity of E/I imbalance associated with mental illness. However, the mechanism of E/I imbalance induced psychiatric symptoms is difficult to tease this apart with human studies. Therefore, rodent models are useful tools in elucidating the behavioral, molecular, cellular, and electrophysiological outcomes of E/I imbalance associated with psychiatric diseases (Table 1).

Rodent models with E/I imbalance exhibit phenotypes reminiscent of patients with ASD and schizophrenia. For example, Yizhar *et al.* utilized optogenetics to elevate excitatory neurons in the medial prefrontal cortex (mPFC) of mice. These mice displayed social deficits and an increase in electroencephalogram frequency as compared to controls, reminiscent of deficits observed in patients with ASD and schizophrenia¹¹. Another mouse model is the *insulin receptor substrate protein 53 kDa (IRSp53)* knockout (KO) mouse. IRSp53 is an excitatory synaptic scaffolding molecule detected throughout the brain¹². *IRSp53* KO mice showed enhanced hippocampal NMDAR function and impaired social interaction, a behavior observed in patients with ASD as well as the optogenetics mouse model of elevated mPFC excitatory neurons. Interestingly, administration of the NMDAR antagonist memantine or the selective metabotropic glutamate receptor5 (mGluR5) antagonist 2-methyl-6-(phenylethynyl) pyridine (MPEP) was able to recover the social deficits observed in *IRSp53* KO mice¹³. Testing these drugs for the ablation of social deficits in other mouse models would be an interesting step towards efficacy in treating humans. While the optogenetics and *IRSp53* KO rodent models increase the E/I ratio in the brain by modifying excitatory neurons, other rodent models increase the E/I ratio by modulating inhibitory neurons. Brown *et al.* increased the E/I ratio in the neocortex and dentate gyrus of mice by knocking down *Gad1* in these brain regions. *Gad1* encodes the 67-kDa isoform of glutamic acid decarboxylase (Gad67) and is important for inhibitory neurons. *Gad1* knock-down mice exhibited a decrease in inhibitory parvalbumin (PV) -expressing interneurons and displayed sensorimotor gating deficits, increased novelty-seeking, and reduced fear extinction¹⁴. Another group ablated metabotropic glutamate receptor 5 (*mGluR5*) on PV-expressing interneurons, which resulted in reduced numbers of PV neurons, decreased inhibitory currents, and behavioral abnormalities in mice. These mice exhibited increased repetitive behavior, impaired recognition memory, and sensory motor gating abnormalities¹⁵. Treatment of conditional *mGluR5* KO (in forebrain glutamatergic neurons) mice with MPEP decreased mPFC firing of GABAergic neurons

and increased firing of glutamatergic neurons¹⁶. These results elucidate a possible anti-psychotic action of MPEP and suggest that mGluR5 directly regulates E/I balance by dominantly regulating inhibitory neuronal input onto glutamatergic neurons. While memantine recovers deficits in a genetic KO, another NMDAR antagonist, ketamine, has also been assessed by Patrizi *et al.* They reported that chronic administration of ketamine from either postnatal day (P)15 or P30 to P55 increased neuronal activity, lifespan, respiratory function, and visual acuity of *Mecp2*-null (KO) mice. The authors confirmed that low-dose chronic ketamine administration (8 mg/kg) is rapidly detected in the brain without any adverse side effects. Intriguingly, the time points chosen for the start of ketamine administration correlate with when PV cortical circuits are altered (P15) and the onset of RTT phenotypes (P30). Since the spiking activities of cortical excitatory neurons are tightly regulated by PV-positive inhibitory neurons¹⁷, it is possible that ketamine preferentially modulates NMDAR on PV-positive cells, leading to disinhibition of pyramidal cells and thus renormalizing E/I balance in the cortex of the *Mecp2* KO mouse model of RTT. The pharmacological studies discussed in this section support previous findings that E/I imbalance in the mPFC affects sociability¹¹, and show that pharmacological intervention of receptors such as mGluR5 or NMDAR, may be effective against psycho-social deficits.

Table 1. Representative animal models of E/I imbalance

Animal model	E/I ratio	Phenotypes	Rescue	Reference
Optogenetic control	Increase in mPFC	Social deficits, high frequency electroencephalogram (30-80Hz)	Optogenetic	Yizar <i>et al.</i> (2011) ¹¹
<i>Gad1</i> knockdown	Increase	Sensorimotor gating deficits, increased novelty-seeking, and reduced fear extinction	-	Brown <i>et al.</i> (2015) ¹⁴
Postnatal ablation of <i>mGluR5</i> from PV-expressing interneuron	Increase	Impaired recognition memory, increased repetitive behaviors, and sensory motor gating abnormalities	-	Barnes <i>et al.</i> (2015) ¹⁵
<i>IRSp53</i> KO	Increase	Social deficits	Memantin	Chung <i>et al.</i>

			(NMDAR antagonist) MPEP (selective antagonist for the metabotropic glutamate receptor subtype mGluR5)	(2015) ¹³
<i>mGluR5</i> KO knockout in GABAergic neurons	Increase	Antidepressant-like behavior	-	Lee <i>et al.</i> (2015) ¹⁶
<i>mGluR5</i> KO in glutamatergic neurons	Decrease	Depression-like behavior	MPEP	Lee <i>et al.</i> (2015) ¹⁶
<i>Mecp2</i> KO	Decrease	Several neurological and psychological phenotypes ¹⁸	Ketamine (NMDAR antagonist)	Patrizi <i>et al.</i> (2016) ¹⁹

2. Rodent models of Copy Number Variations

CNV is defined as deletion or duplication of a genomic locus larger than 1 Kb in size. Some CNVs encompass dozens of genes that may include coding gene(s) or genes in regulatory regions²⁰ such as the promoter, enhancer, and silencer, while others pertain to only a single gene. Available mouse models of CNVs include 7q11.23 (deletion)²¹, 15q11-13 (duplication)²², 15q13.3 (deletion)^{23,24}, 16p11.2 (deletion and duplication)²⁵, 17p11.2 (deletion and duplication)²⁶, 22q11.2 (deletion)²⁷, and MECP2 (deletion and duplication). These mice are known to display behavioral and brain abnormalities paralleling what has been frequently reported in patients²⁸. In this review, we focus on CNV mouse models of 15q11-13, 22q11.2, and MECP2.

2.1. 15q11-13 duplication

A duplication in 15q11-13 is one of the most prevalent chromosomal abnormalities in

ASD. This region is syntenic to mouse chromosome 7qB5-qC. The 15q11-13 chromosomal duplication confers >85% risk of developing ASD, and is found in 1-3% of individuals with ASD²⁹. Mice with paternally inherited 6 Mb 15q11-13 duplication (6 Mb patDp/+) displayed social deficits, behavioral inflexibility, and abnormal ultrasonic vocalization (USV), which are typical autistic behavioral phenotypes in rodents. These behaviors were not present in mice with a maternally inherited duplication (matDp/+) ²². In addition to behavioral abnormalities, 6 Mb patDp/+ mice had decreased serotonin levels in the developing brain³⁰ and displayed late-onset obesity³¹, both of which resemble what is observed in patients with paternally inherited 15q11-13 duplication³². Recently, in order to identify a causal gene of obesity, Kishimoto *et al.* generated a new mouse model with a 3 Mb 15q11-13 paternal duplication (3 Mb patDp/+) ³¹. Transcriptome analysis on these mice revealed an up-regulation in the gene *secreted frizzled-related protein 5 (Sfrp5)*. Sfrp5 induces adipocyte differentiation and increases adipocytes during obesity. This gene may therefore be a possible target for combating or preventing obesity accompanied by autistic symptoms, but it may not be effective against cognitive dysfunction or social deficits.

Another hallmark of ASD and some behavioral changes such as deregulation of motor coordination and learning³³, is cerebellar dysfunction. 6 Mb patDp/+ mice exhibited impaired cerebellum-dependent motor learning and coordination, but normal cerebellar morphology²²³⁴³⁵. This is consistent with a magnetic resonance imaging (MRI) study in patDp/+ mice that revealed regional difference existing in the dentate gyrus, medial striatum, and dorsal raphe nucleus, but not in the cerebellar cortex³⁶³⁷³⁸. In addition, patDp/+ mice showed impaired long-term depression (LTD) at cerebellar parallel fiber-Purkinje cell synapses³⁹, which may reflect a cellular mechanism underlying motor learning impairment. Since the cerebellum is likely involved in motor and non-motor aspects of ASD phenotypes³³, cerebellar abnormalities may be involved in a portion of the ASD pathophysiology.

2.2. 22q11.2 deletion

The 22q11.2 deletion, a region syntenic to mouse chromosome 16qA3, is also known as DiGeorge syndrome and velo-cardio-facial syndrome. It is one of the most common chromosomal abnormalities, occurring in 1 out of every 4,000 live births⁴⁰. Most cases (90%) are of a 3 Mb microdeletion on the long arm of chromosome 22, but a 1.5 Mb microdeletion is also common and has been reported in 8% of cases⁴¹. The 22q11.2 microdeletion has been associated with congenital heart defects (CHD), palatal anomalies, hypoparathyroidism, hypocalcemia, facial dysmorphisms, and learning

disabilities. Additionally, this chromosomal deletion has been strongly associated with schizophrenia and other mental illnesses such as anxiety disorders, mood disorders, ASD, and attention deficit hyperactivity disorder (ADHD)⁴².

In recent years, through genetically modified mutant mice that carry multigene and single gene mutations, we have learned that a mutation in *Tbx1*, a member of T-box family transcription factor gene, is responsible for most of the congenital defects seen in patients and mouse models⁴³⁴⁴⁴⁵⁴⁶. Rodent models suggest that 22q11.2 deletion leads to behavioral changes and deficits in RNA processing. Mice carrying a hemizygous 1.3 Mb chromosomal deficiency (*Df(16)A+/-*)²² showed deficits in pre-pulse inhibition (PPI), a test of sensory information-processing of external stimuli. PPI deficits are frequently observed in several psychiatric and neurological diseases including schizophrenia, obsessive-compulsive disorder, ADHD, and Huntington's disease⁴⁷. In addition to PPI deficits, *Df(16)A+/-* mice showed dysregulation in miRNA biogenesis²⁷. In these mice, a subset of pri-miRNA was up-regulated and a smaller subset of the mature form of the miRNAs was down-regulated due to haploinsufficiency of the *DiGeorge syndrome critical region 8* gene (*Dgcr8*: also known as *Pasha*). Because *Dgcr8* is an essential component of pri-miRNA processing⁴⁸, haploinsufficiency of this gene may affect brain cognitive functions²⁷⁴⁹ due to dysregulation of critical downstream genes involved in neural development. For example, *insulin growth factor (IGF)* is one gene downstream of *Dgcr8*. Administration of IGF to *Dgcr8* KO mice rescued hippocampal functions⁵⁰. Recently, Mukai *et al.* reported that deletion of the 22q11 locus *zinc finger and Asp-His-His-Cys (DHHC) domain-containing protein 8 (Zdhhc8)* led to changes in axonal growth, dendrite arborization, synaptic connectivity, spatial working memory, and prefrontal-hippocampal synchrony⁵¹. Behavioral and neuronal abnormalities were consistent between *Df(16)A+/-* and *Zdhhc* KO mice. This interesting consequence gives rise to new insights into post-translational modification of proteins related to cognitive function and abnormal brain circuit formation.

2.3. *MECP2*

Mutations in the *MECP2* gene cause the neurodevelopmental disorder RTT. This syndrome is characterized by small head size (microcephaly), gait abnormalities, loss of language, breathing disturbances, and poor motor movements with repetitive stereotypies. *MECP2* is located on region Xq28 of the X chromosome where deletions, mutations, and duplications of this gene have been reported⁵². Classically, RTT has been recognized as a girl's disorder, with symptomatic women having either a *MECP2* point mutation or deletion. Women with a *MECP2* duplication remain healthy, likely due to

skewed X inactivation⁵³. In contrast, boys with a *MECP2* duplication present with clinical features of RTT. For instance, Meins *et al.* reported on a *MECP2* duplication of approximately 430 kb in a boy with hypotonia, mental retardation, lack of speech, and loss of motor movements⁵³. Other male patients with *MECP2* duplications have presented with infantile hypotonia, mental retardation, poor speech development, recurrent infections, epilepsy, progressive spasticity, developmental regression, and ASD⁵².

Mecp2 mutant mice are available to elucidate the role of *Mecp2* on neurological and psychological functions¹⁸. Sztainberg *et al.* generated a *Mecp2* duplication mouse model through conditional *Mecp2*-overexpression⁵⁴. These mice displayed hypoactivity, anxiety-like behavior, motor abnormalities, and social deficits, which are behaviors seen in patients with a *MECP2* duplication. Observed deficits were recovered by 1) deleting the conditional *Mecp2*-overexpression allele by using tamoxifen-inducible Cre recombination and 2) stereotactic injection of antisense oligonucleotides (ASOs) into the right ventricles of the brain, which then hybridized with target mRNA to silence *Mecp2*. These rescue experiments successfully ameliorated the abnormal cortical electroencephalography (EEG), molecular, and electrophysiological deficits displayed by *Mecp2* duplication mice. Since ASOs efficiently suppressed *Mecp2* gene expression in mouse brains as well as *MECP2* duplication patient-derived lymphoblastoids, it may be a suitable drug candidate for *MECP2* duplication patients. However, possible off-target effects still need to be determined.

3. Intervention during brain development may treat psychiatric symptoms

Abnormalities arising from a genetic mutation may effectively be recovered by intervention at a young age (Table 2). One study suggesting this was performed on contactin-associated protein-like 2 (*Cntnap2*) KO mice⁵⁵. *Cntnap2* is a member of the *NRXN* gene superfamily and encodes a neuronal transmembrane protein that acts as a cell adhesion molecule, implicating the protein in neural plasticity. *Cntnap2* KO mice showed abnormalities similar to patients with a *CNTNAP2* mutation⁵⁶, such as epileptic seizures and an abnormal EEG pattern⁵⁵. Indeed, *Cntnap2* is one of the best-replicated risk genes for ASD⁵⁷. It is therefore not surprising that *Cntnap2* KO mice display core features of ASD, including repetitive behaviors, behavioral inflexibility, and social deficits. Social deficits in neurodevelopmental disorders such as schizophrenia and ASD have been suggested to also be caused in part by an imbalance in the neurotransmitter oxytocin (OXT)⁵⁸⁵⁹⁶⁰. **OXT treatment of *Cntnap2* KO improved social deficits observed**

in the reciprocal social interaction and three-chamber social interaction tests. Interestingly, the recovery of behavioral deficits was dependent upon the age at which OXT was administered. Adult mice (6-8 weeks of age) acutely treated with OXT showed improvement in social interactions. Similarly, early postnatal mice (7-21 days of age) chronically treated with OXT showed improved social interaction up to one week after OXT cessation⁵⁶. It is therefore possible that OXT can be used to treat social deficits, particularly when administered early in life.

Another study suggesting intervention prior to adulthood may better recover deficits was performed on *protein interacting with C kinase-1 (Pick1)* KO mice. Pick1 is a postsynaptic protein involved in synaptic plasticity and affects AMPAR trafficking. Indeed, hippocampal and cerebellar LTD are significantly impaired in *Pick1* KO mice, likely due to impaired internalization, recycling, or intracellular retention of AMPAR⁶¹. During development, neonatal *Pick1* KO mice have decreased levels of the NMDAR co-agonist D-serine in the prefrontal cortex and hippocampus⁶². As D-serine levels recover later in life, one may be able to use *Pick1* KO mice as a conditional D-serine knockdown model. Interestingly, adult *Pick1* KO mice, who have levels of D-serine comparable to wild-type mice, exhibited impaired synaptic plasticity and behavioral abnormalities including learning and memory, PPI, and cognitive deficits^{63,64}. This suggests that decreased levels of D-serine during development adversely affect the brain during adulthood. To test whether rescuing D-serine levels during development would prevent behavioral deficits during adulthood, *Pick1* KO mice were treated with D-serine during the neonatal development period (P3-P17). In the treated mice, abnormal NMDA-elicited firing in prefrontal pyramidal neurons and PPI deficits were rescued in adulthood⁶⁴. Interestingly, these electrophysiological and behavioral deficits were rescued only with D-serine treatment of neonates (P3-P17), and not with D-serine treatment of adults (P56-P70)⁶⁴. These results indicate that neonatal NMDAR activation seems indispensable for adult psychopathology.

Behavioral abnormalities of *Mecp2* KO mice can also be recovered by intervention during brain development with the chronic NMDAR antagonist ketamine¹⁹. In this study, the authors treated mice at two different time windows in order to identify the critical time period to recover physiological abnormalities. Ketamine was administered from either 1) before the onset of RTT symptoms (P15) to P55 (40 days total administration) or 2) at the onset of RTT symptoms (P30) until P55 (25 days total administration). Interestingly, both ketamine treatment starting before the onset of RTT symptoms (P15) and at the onset of RTT symptoms (P30) rescued respiratory function, visual activity, neuronal evoked and spontaneous activities, and PV

immunofluorescence intensity in the visual cortex of *Mecp2* KO mice. Pharmacokinetic analysis revealed the low dose of ketamine (8 mg/kg) administered was promptly absorbed in the brain without any detrimental side effects. Therefore, ketamine administration might be a suitable pharmacotherapy for RTT patients. However, whether ketamine can recover existing symptoms of RTT adult patients remains to be elucidated.

Intervention prior to adulthood has also been shown to recover deficits induced by oxidative stress. Oxidative stress affects neuronal synapses, myelin, and interneuron integrity. It is one of the known causal factors of neurodegenerative disorders⁶⁵, and may also be involved in the development of psychiatric disorders⁶⁶. Accumulating clinical and epidemiological studies suggest an imbalance in genes involved in oxidative and redox signaling in patients with psychiatric disorders. Specifically, patients show changes in *glutathione (GSH)*, *microsomal glutathione S-transferase 1 (MGST1)*, *superoxide dismutase (SOD)*, and *catalase (CAT)*. One rodent model of oxidative stress is the neonatal ventral hippocampal lesion (NVHL) rat model. This rat model develops behavioral, neurochemical and electrophysiological deficits relevant to schizophrenia during adolescence^{67,68}. Using NVHL rats, Cabungcal *et al.* assessed whether pre-symptomatic oxidative stress can lead to schizophrenic-like symptoms in adults. Indeed, they identified increased oxidative stress in the prefrontal cortex (PFC) of both P21 (juvenile) and P61 (adult) NVHL rats. In addition, there was decreased PV immunopositive neuron in the PFC of P61 rats, which may reflect cortical disinhibition that is frequently seen in patients with schizophrenia. Treatment of NVHL rats from P5 to P50 with the antioxidant *N*-acetyl-cysteine (NAC), a glutathione precursor, prevented the reduction of cortical PV interneurons, electrophysiological changes, and behavioral deficits. These findings suggest that the redox pathway may be a potential drug target for patients with schizophrenia, particularly when manipulated during brain development.

Table 2. Phenotypic rescue by early intervention

Animal model	Drug/main effect	Time period	Rescued behavioral phenotypes	Reference
<i>Cntnap2</i> KO mouse	Oxytocin (Hormone: modulate social behavior)	P7-P21	Social behaviors (three-chamber test, Reciprocal social interaction test)	Penagarikano <i>et al.</i> (2015) ⁵⁶
<i>Pick1</i> KO	D-serine	P3-P14	Sensory motor	Nomura <i>et</i>

mouse	(Endogenous coagonist for NMDAR)		gating (PPI)	<i>al.</i> (2016) ⁶⁴
<i>Mecp2</i> mouse	KO Ketamine (Non-competitive NMDAR antagonist)	P15-P55 P30-P55	RTT-like phenotypes (hindlimb-clasping, apnea episode, visual acuity)	Patrizi <i>et al.</i> (2016) ¹⁹
NVHL rat	NAC (<i>N</i> -acetyl cysteine: Antioxidant)	P5-P50	Sensory motor gating (PPI)	Cabungcal <i>et al.</i> (2014) ⁶⁸

Discussion

The best way to ultimately elucidate psychopathological conditions in the brain is to use samples from human patients. Yet, acquiring brain tissues from living patients is impossible, postmortem brain samples are limited, and non-invasive methods to analyze brain structure and activities, such as functional magnetic resonance imaging (fMRI) and MRI, have limited outcomes. To overcome these hurdles, some scientists have turned to induced pluripotent stem cells (iPSCs) and induced neurons (iNs), which are derived from patient samples and therefore helpful in understanding the cellular dysfunction of patients. Several types of iPSCs have been generated from patients with various diseases to be used in disease modeling and drug screening of therapeutics⁶⁹. For example, iPSCs from three independent patients with *MECP2* duplication have recently been established⁷⁰. Expression analysis revealed changes in markers for neural progenitors, migration, and development of different brain regions. **The cellular morphology of neurons derived from iPSCs with *MECP2* duplication showed that synaptogenesis and dendritic complexity were increased compared with controls, suggesting hyper-connected neural networks, which are observed in children with ASD^{71,72}.** Furthermore, patient-derived neurons displayed higher numbers of action potentials than controls, which may underlie abnormal glutamatergic neurotransmission. Deficits observed in *MECP2* iPSCs were ameliorated by the administration of the histone deacetylase (HDAC) inhibitor, NCH-51. This study reveals the usefulness of iPSCs as a disease model, as well as a tool in drug screening at the cellular level. In addition to iPSCs, fibroblasts can now be directly converted into either serotonergic, glutamatergic, GABAergic, or dopaminergic neurons⁷³, aiding in unraveling how specific cellular systems are affected in certain diseases. The recent success of culturing

organoids will accelerate human disease modeling. While iPSCs, iNs, and organoids are useful tools because they are derived from patients, they are unable to provide a complete picture of deficits throughout the brain.

Rodent models of genetic mutations found in humans with mental illness have been useful in understanding complex brain abnormalities. However, it is obvious that rodents are vastly different from humans, and therefore rodent disease models may not always reflect human pathophysiological conditions. A good example is the mouse model of RTT. Although some exceptions have been reported in clinical studies, most MECP2 mutations are lethal in males. However, *Mecp2* null mutation in male mice is not lethal although they die around 10 weeks of age. Similarly, female *Mecp2* heterozygous mice appear normal at a young age although RTT is characterized as early neurological regression⁷⁴. In a mouse model of Fragile X Syndrome (FXS), it was found that a mGluR5 antagonist potently ameliorates phenotypic abnormalities^{75,76}. When the pharmaceutical companies Roche and Novartis went into Phase II trials of the mGluR5 antagonist, they found no beneficial effect. This is a common theme for many drugs that show promise in rodent models, but not in humans. In addition, as psychiatric disorders are multifactorial disorders and may be due to changes in many systems throughout the body (e.g. brain vs microbiome and immune system), analysis of the whole body seems to be essential to understand pathophysiology of psychiatric disorders. Despite differences between rodents and humans, animal models are still valuable tools to study human diseases because we can study rodents at the system level, within a living organism, and assess drugs in different ways in real time, which are all difficult and more costly to do in human subjects.

As we have discussed, human, rodent, and cell culture studies, all have their strengths and weaknesses in understanding the mechanism and treatment of genetic disorders as it pertains to the brain. Studies combining all tools may therefore result in the most promising avenues to follow through to clinical drug trials. One step in this direction is exemplified by a study reported by Hao *et al.* This group applied deep brain stimulation (DBS), which is commonly used in the clinical treatment of humans with movement disorders such as Parkinson's and dystonia⁷⁷, to work on a RTT mouse model. Using DBS, the authors successfully rescued neurological and cognitive deficits in the RTT mice⁷⁸. As suggested by this work, combining human studies (postmortem samples, iPSCs or iNs, brain imaging) with animal models of human disease may be necessary to truly understand multifactorial psychiatric disorders such as ASD and schizophrenia.

Acknowledgments

The authors appreciate all members of our laboratory for useful discussions and comments. This work was supported by Japan Society for the Promotion of Science (JSPS) KAKENHI Grant Numbers 26870397 (J.N.), 16H06316, 16K13110 (T.T.), The Ministry of Education, Culture, Sports, Science and Technology (MEXT) Grant Number 16H06463 (T.T.), Intramural Research Grant for Neurological and Psychiatric Disorders of NCNP (T.T.), Takeda Science Foundation (T.T.) and Takeda Pharmaceutical Company Limited (T.T).

Disclosure Statement

All authors declare that they have no conflicts of interest.

Author's Contribution

JN drafted the manuscript. All authors edited it.

References

1. Akbarian S, Sucher NJ, Bradley D, Tafazzoli A, Trinh D, Hetrick WP, et al. Selective alterations in gene expression for NMDA receptor subunits in prefrontal cortex of schizophrenics. *J. Neurosci.* 1996; 16: 19–30. Available from <http://www.ncbi.nlm.nih.gov/pubmed/8613785>
2. Colantuoni C, Jeon OH, Hyder K, Chenchik A, Khimani AH, Narayanan V, et al. Gene expression profiling in postmortem Rett Syndrome brain: differential gene expression and patient classification. *Neurobiol. Dis.* 2001; 8: 847–65. Available from <http://www.sciencedirect.com/science/article/pii/S0969996101904280>
3. Sebat J, Lakshmi B, Troge J, Alexander J, Young J. Large-scale copy number polymorphism in the human genome. *Science (80-.).* 2004; 525: 525–8. Available from <http://www.sciencemag.org/cgi/content/abstract/sci;305/5683/525>
4. Eichler SA, Meier JC. E-I balance and human diseases - from molecules to networking. *Front. Mol. Neurosci.* 2008; 1: 2. Available from <http://www.pubmedcentral.nih.gov/articlerender.fcgi?artid=2526001&tool=pmcentrez&rendertype=abstract>
5. Nelson SB, Valakh V. Review Excitatory / Inhibitory Balance and Circuit Homeostasis in Autism Spectrum Disorders. *Neuron.* 2015; 87: 684–98. Available from <http://dx.doi.org/10.1016/j.neuron.2015.07.033>
6. Argyropoulos A, Gilby KL, Hill-Yardin EL. Studying Autism in Rodent Models:

- Reconciling Endophenotypes with Comorbidities. *Front. Hum. Neurosci.* 2013; 7: 1–10. Available from <http://journal.frontiersin.org/article/10.3389/fnhum.2013.00417/abstract>
7. Stafstrom CE. Epilepsy: a review of selected clinical syndromes and advances in basic science. *J. Cereb. Blood Flow Metab.* 2006; 26: 983–1004. Available from <http://jcb.sagepub.com/content/26/8/983.full>
 8. Mariani J, Coppola G, Zhang P, Abyzov A, Provini L, Tomasini L, et al. FOXP1-Dependent Dysregulation of GABA/Glutamate Neuron Differentiation in Autism Spectrum Disorders. *Cell.* 2015; 162: 375–90. Available from <http://linkinghub.elsevier.com/retrieve/pii/S009286741500759X>
 9. Pocklington AJ, Rees E, Walters JTR, Han J, Kavanagh DH, Chambert KD, et al. Novel Findings from CNVs Implicate Inhibitory and Excitatory Signaling Complexes in Schizophrenia. *Neuron.* 2015; 86: 1203–14. Available from <http://linkinghub.elsevier.com/retrieve/pii/S0896627315003724>
 10. Blue ME, Naidu S, Johnston M V. Altered development of glutamate and GABA receptors in the basal ganglia of girls with Rett syndrome. *Exp. Neurol.* 1999; 156: 345–52.
 11. Yizhar O, Fenno LE, Prigge M, Schneider F, Davidson TJ, O’Shea DJ, et al. Neocortical excitation/inhibition balance in information processing and social dysfunction. *Nature.* 2011; 477: 171–8. Available from <http://dx.doi.org/10.1038/nature10360>
 12. Burette AC, Park H, Weinberg RJ. Postsynaptic distribution of IRSp53 in spiny excitatory and inhibitory neurons. *J. Comp. Neurol.* 2014; 522: 2164–78.
 13. Chung W, Choi SY, Lee E, Park H, Kang J, Park H, et al. Social deficits in IRSp53 mutant mice improved by NMDAR and mGluR5 suppression. *Nat. Neurosci.* 2015; 18: 435–43. Available from <http://www.ncbi.nlm.nih.gov/pubmed/25622145>
 14. Brown JA, Ramikie TS, Schmidt MJ, Báldi R, Garbett K, Everheart MG, et al. Inhibition of parvalbumin-expressing interneurons results in complex behavioral changes. *Mol. Psychiatry.* 2015; 20: 1499–507. Available from <http://www.nature.com/doi/10.1038/mp.2014.192>
 15. Barnes S, Pinto-Duarte A, Kappe A, Zembrzycki A, Metzler A, Mukamel E, et al. Disruption of mGluR5 in parvalbumin-positive interneurons induces core features of neurodevelopmental disorders. *Mol. Psychiatry.* 2015; 20: 1161–72.

Available from <http://www.nature.com/doi/10.1038/mp.2015.113>

16. Lee K-W, Westin L, Kim J, Chang JC, Oh Y-S, Amreen B, et al. Alteration by p11 of mGluR5 localization regulates depression-like behaviors. *Mol. Psychiatry*. 2015; 20: 1546–56.
17. Thankachan S, Mckenna JT, McNally JM, Yang C, Choi JH, Chen L, et al. Correction for Kim et al., Cortically projecting basal forebrain parvalbumin neurons regulate cortical gamma band oscillations. *Proc. Natl. Acad. Sci.* 2015; 112: 3535–40. Available from <http://www.pnas.org/lookup/doi/10.1073/pnas.1507465112>
18. Lyst MJ, Bird A. Rett syndrome: a complex disorder with simple roots. *Nat. Rev. Genet.* 2015; 16: 261–75. Available from <http://www.nature.com/doi/10.1038/nrg3897>
19. Patrizi A, Picard N, Simon AJ, Gunner G, Centofante E, Andrews NA, et al. Chronic Administration of the N-Methyl-D-Aspartate Receptor Antagonist Ketamine Improves Rett Syndrome Phenotype. *Biol. Psychiatry*. 2016; 79: 755–64. Available from <http://linkinghub.elsevier.com/retrieve/pii/S0006322315006836>
20. Malhotra D, Sebat J. CNVs: Harbingers of a Rare Variant Revolution in Psychiatric Genetics. *Cell*. 2012; 148: 1223–41. Available from <http://linkinghub.elsevier.com/retrieve/pii/S0092867412002772>
21. Li HH, Roy M, Kuscuoglu U, Spencer CM, Halm B, Harrison KC, et al. Induced chromosome deletions cause hypersociability and other features of Williams-Beuren syndrome in mice. *EMBO Mol. Med.* 2009; 1: 50–65. Available from <http://www.pubmedcentral.nih.gov/articlerender.fcgi?artid=3378107&tool=pmcentrez&rendertype=abstract>
22. Nakatani J, Tamada K, Hatanaka F, Ise S, Ohta H, Inoue K, et al. Abnormal Behavior in a Chromosome- Engineered Mouse Model for Human 15q11-13 Duplication Seen in Autism. *Cell*. 2009; 137: 1235–46. Available from <http://linkinghub.elsevier.com/retrieve/pii/S0092867409004498>
23. Fejgin K, Nielsen J, Birkenow MR, Bastlund JF, Nielsen V, Lauridsen JB, et al. A Mouse Model that Recapitulates Cardinal Features of the 15q13.3 Microdeletion Syndrome Including Schizophrenia- and Epilepsy-Related Alterations. *Biol. Psychiatry*. 2014; 76: 128–37. Available from

- <http://linkinghub.elsevier.com/retrieve/pii/S0006322313007671>
24. Kogan JH, Gross AK, Featherstone RE, Shin R, Chen Q, Heusner CL, et al. Mouse Model of Chromosome 15q13.3 Microdeletion Syndrome Demonstrates Features Related to Autism Spectrum Disorder. *J. Neurosci.* 2015; 35: 16282–94. Available from <http://www.jneurosci.org/cgi/doi/10.1523/JNEUROSCI.3967-14.2015>
 25. Horev G, Ellegood J, Lerch JP, Son YE, Muthuswamy L, Vogel H, et al. Dosage-dependent phenotypes in models of 16p11.2 lesions found in autism. *Proc. Natl. Acad. Sci.* 2011; 108: 17076–81. Available from <http://www.pnas.org/cgi/content/abstract/108/41/17076>
 26. Walz K, Caratini-Rivera S, Bi W, Fonseca P, Mansouri DL, Lynch J, et al. Modeling del(17)(p11.2p11.2) and dup(17)(p11.2p11.2) contiguous gene syndromes by chromosome engineering in mice: phenotypic consequences of gene dosage imbalance. *Mol. Cell. Biol.* 2003; 23: 3646–55.
 27. Stark KL, Xu B, Bagchi A, Lai W-S, Liu H, Hsu R, et al. Altered brain microRNA biogenesis contributes to phenotypic deficits in a 22q11-deletion mouse model. *Nat. Genet.* 2008; 40: 751–60.
 28. Nomura J, Takumi T. Animal models of psychiatric disorders that reflect human copy number variation. *Neural Plast.* 2012; 2012: 589524. Available from <http://www.pubmedcentral.nih.gov/articlerender.fcgi?artid=3414062&tool=pmcentrez&rendertype=abstract>
 29. Cook EH, Scherer SW. Copy-number variations associated with neuropsychiatric conditions. *Nature.* 2008; 455: 919–23.
 30. Tamada K, Tomonaga S, Hatanaka F, Nakai N, Takao K, Miyakawa T, et al. Decreased exploratory activity in a mouse model of 15q duplication syndrome; implications for disturbance of serotonin signaling. *PLoS One.* 2010; 5: e15126. Available from <http://journals.plos.org/plosone/article?id=10.1371/journal.pone.0015126>
 31. Kishimoto R, Tamada K, Liu X, Okubo H, Ise S, Ohta H, et al. Model mice for 15q11–13 duplication syndrome exhibit late-onset obesity and altered lipid metabolism. *Hum. Mol. Genet.* 2015; 24: 4559–72. Available from <http://www.hmg.oxfordjournals.org/lookup/doi/10.1093/hmg/ddv187>
 32. Marini C, Ceconi A, Contini E, Pantaleo M, Metitieri T, Guarducci S, et al. Clinical and genetic study of a family with a paternally inherited 15q11-q13

- duplication. *Am. J. Med. Genet. Part A*. 2013; 161: 1459–64. Available from <http://doi.wiley.com/10.1002/ajmg.a.35907>
33. Wang SSH, Kloth AD, Badura A. The Cerebellum, Sensitive Periods, and Autism. *Neuron*. 2014; 83: 518–32. Available from <http://dx.doi.org/10.1016/j.neuron.2014.07.016>
34. Piochon C, Kloth AD, Grasselli G, Titley HK, Nakayama H, Hashimoto K, et al. Cerebellar plasticity and motor learning deficits in a copy-number variation mouse model of autism. *Nat. Commun*. 2014; 5: 5586. Available from <http://www.nature.com/doi/10.1038/ncomms6586>
35. Kloth AD, Badura A, Li A, Cherskov A, Connolly SG, Giovannucci A, et al. Cerebellar associative sensory learning defects in five mouse autism models. *Elife*. 2015; 4: 1–26. Available from <http://elifesciences.org/lookup/doi/10.7554/eLife.06085>
36. Ellegood J, Anagnostou E, Babineau B a, Crawley JN, Lin L, Genestine M, et al. Clustering autism: using neuroanatomical differences in 26 mouse models to gain insight into the heterogeneity. *Mol. Psychiatry*. 2015; 20: 118–25. Available from <http://www.nature.com/doi/10.1038/mp.2014.98>
37. Ellegood J, Anagnostou E, Babineau BA, Crawley JN, Lin L, Genestine M, et al. 3D visualization of the regional differences. *Mol. Psychiatry*. 2015; 20: 1–1. Available from <http://www.nature.com/doi/10.1038/mp.2014.168>
38. Ellegood J, Nakai N, Nakatani J, Henkelman M, Takumi T, Lerch J. Neuroanatomical phenotypes are consistent with autism-like behavioral phenotypes in the 15q11-13 duplication mouse model. *Autism Res*. 2015; 8: 545–55.
39. Ito M, Yamaguchi K, Nagao S, Yamazaki T. *Long-term depression as a model of cerebellar plasticity*. 1st ed. Elsevier B.V.; 2014. Available from <http://www.ncbi.nlm.nih.gov/pubmed/24916287>
40. Burnside RD. 22q11.21 Deletion Syndromes: A Review of Proximal, Central, and Distal Deletions and Their Associated Features. *Cytogenet. Genome Res*. 2015; 146: 89–99.
41. Michaelovsky E, Frisch A, Carmel M, Patya M, Zarchi O, Green T, et al. Genotype-phenotype correlation in 22q11.2 deletion syndrome. *BMC Med. Genet*. 2012; 13: 1–11. Available from BMC Medical Genetics

42. Schneider M, Debbané M, Bassett AS, Chow EWC, Fung WLA, van den Bree MBM, et al. Psychiatric Disorders From Childhood to Adulthood in 22q11.2 Deletion Syndrome: Results From the International Consortium on Brain and Behavior in 22q11.2 Deletion Syndrome. *Am. J. Psychiatry*. 2014; 171: 627–39. Available from <http://www.ncbi.nlm.nih.gov/pubmed/24577245>
43. Jerome L, Papaioannou V. DiGeorge syndrome phenotype in mice mutant for the T-box gene, *Tbx1*. *Nat. Genet*. 2001; 27: 286–91.
44. Paylor R, Glaser B, Mupod A, Ataliotis P, Spencer C, Sobotka A, et al. *Tbx1* haploinsufficiency is linked to behavioral disorders in mice and humans: Implications for 22q11 deletion syndrome. *Proc. Natl. Acad. Sci*. 2006; 103: 7729–34.
45. Hiramoto T, Kang G, Suzuki G, Satoh Y, Kucherlapati R, Watanabe Y, et al. *Tbx1*: identification of a 22q11.2 gene as a risk factor for autism spectrum disorder in a mouse model. *Hum. Mol. Genet*. 2011; 20: 4775–85.
46. Takahashi T, Okabe S, Broin P, Nishi A, Ye K, Beckert M, et al. Structure and function of neonatal social communication in a genetic mouse model of autism. *Mol. Psychiatry*. 2016; 21: 1208–14.
47. Javitt DC, Freedman R. Sensory processing dysfunction in the personal experience and neuronal machinery of schizophrenia. *Am. J. Psychiatry*. 2015; 172: 17–31.
48. Burger K, Gullerova M. Swiss army knives: non-canonical functions of nuclear Drosha and Dicer. *Nat. Rev. Mol. Cell Biol*. 2015; 16: 417–30. Available from <http://www.nature.com/doi/10.1038/nrm3994>
49. Xu B, Hsu PK, Stark KL, Karayiorgou M, Gogos JA. Derepression of a neuronal inhibitor due to miRNA dysregulation in a schizophrenia-related microdeletion. *Cell*. 2013; 152: 262–75. Available from <http://dx.doi.org/10.1016/j.cell.2012.11.052>
50. Ouchi Y, Banno Y, Shimizu Y, Ando S, Hasegawa H, Adachi K, et al. Reduced adult hippocampal neurogenesis and working memory deficits in the *Dgcr8*-deficient mouse model of 22q11.2 deletion-associated schizophrenia can be rescued by IGF2. *J. Neurosci*. 2013; 33: 9408–19. Available from <http://www.ncbi.nlm.nih.gov/pubmed/23719809>
51. Mukai J, Tamura M, Fénelon K, Rosen AM, Spellman TJ, Kang R, et al. Molecular Substrates of Altered Axonal Growth and Brain Connectivity in a

- Mouse Model of Schizophrenia. *Neuron*. 2015; 86: 680–95. Available from <http://www.ncbi.nlm.nih.gov/pubmed/25913858>
52. Ramocki MB, Tavyev YJ, Peters SU. The *MECP2* duplication syndrome. *Am. J. Med. Genet. Part A*. 2010; 152A: 1079–88. Available from <http://doi.wiley.com/10.1002/ajmg.a.33184>
53. Meins M, Lehmann J, Gerresheim F, Herchenbach J, Hagedorn M, Hameister K, et al. Submicroscopic duplication in Xq28 causes increased expression of the *MECP2* gene in a boy with severe mental retardation and features of Rett syndrome. *J. Med. Genet.* 2005; 42: e12.
54. Sztainberg Y, Chen H, Swann JW, Hao S, Tang B, Wu Z, et al. Reversal of phenotypes in *MECP2* duplication mice using genetic rescue or antisense oligos. *Nature*. 2015; 528: 123–6.
55. Peñagarikano O, Abrahams BS, Herman EI, Winden KD, Gdalyahu A, Dong H, et al. Absence of *CNTNAP2* Leads to Epilepsy, Neuronal Migration Abnormalities, and Core Autism-Related Deficits. *Cell*. 2011; 147: 235–46. Available from <http://linkinghub.elsevier.com/retrieve/pii/S0092867411010105>
56. Peñagarikano O, Lázaro MT, Lu X-H, Gordon A, Dong H, Lam H a., et al. Exogenous and evoked oxytocin restores social behavior in the *Cntnap2* mouse model of autism. *Sci. Transl. Med.* 2015; 7: 271ra8-271ra8. Available from <http://stm.sciencemag.org/content/7/271/271ra8>
57. Poot M. Connecting the *CNTNAP2* Networks with Neurodevelopmental Disorders. *Mol. Syndromol.* 2015; 6: 7–22. Available from <http://www.karger.com/?doi=10.1159/000371594>
58. Yamasue H. Promising evidence and remaining issues regarding the clinical application of oxytocin in autism spectrum disorders. *Psychiatry Clin. Neurosci.* 2016; 70: 89–99. Available from <http://doi.wiley.com/10.1111/pcn.12364>
59. Feifel D, Shilling PD, MacDonald K. A Review of Oxytocin's Effects on the Positive, Negative, and Cognitive Domains of Schizophrenia. *Biol. Psychiatry*. 2016; 79: 222–33. Available from <http://linkinghub.elsevier.com/retrieve/pii/S0006322315006502>
60. Sandi C, Haller J. Stress and the social brain: behavioural effects and neurobiological mechanisms. *Nat. Rev. Neurosci.* 2015; 16: 290–304. Available from <http://www.nature.com/doifinder/10.1038/nrn3918>

61. Volk L, Chiu S-L, Sharma K, Huganir RL. Glutamate Synapses in Human Cognitive Disorders. *Annu. Rev. Neurosci.* 2015; 38: 127–49. Available from <http://www.annualreviews.org/doi/abs/10.1146/annurev-neuro-071714-033821%5Cnhttp://www.ncbi.nlm.nih.gov/pubmed/25897873>
62. Hikida T, Mustafa AK, Maeda K, Fujii K, Barrow RK, Saleh M, et al. Modulation of d-Serine Levels in Brains of Mice Lacking PICK1. *Biol. Psychiatry.* 2008; 63: 997–1000.
63. Volk L, Kim C-H, Takamiya K, Yu Y, Huganir RL. Developmental regulation of protein interacting with C kinase 1 (PICK1) function in hippocampal synaptic plasticity and learning. *Proc. Natl. Acad. Sci. U. S. A.* 2010; 107: 21784–9.
64. Nomura J, Jaaro-Peled H, Lewis E, Nuñez-Abades P, Huppe-Gourgues F, Cash-Padgett T, et al. Role for neonatal D-serine signaling: prevention of physiological and behavioral deficits in adult. *Mol. Psychiatry.* 2016; 21: 386–93.
65. Nakamura T, Lipton SA. Protein S-Nitrosylation as a Therapeutic Target for Neurodegenerative Diseases. *Trends Pharmacol. Sci.* 2015; 37: 73–84. Available from <http://linkinghub.elsevier.com/retrieve/pii/S0165614715002114>
66. Landek-Salgado MA, Faust TE, Sawa A. Molecular substrates of schizophrenia: homeostatic signaling to connectivity. *Mol. Psychiatry.* 2016; 21: 10–28. Available from <http://dx.doi.org/10.1038/mp.2015.141>
67. O’Donnell P. Cortical disinhibition in the neonatal ventral hippocampal lesion model of schizophrenia: new vistas on possible therapeutic approaches. *Pharmacol. Ther.* 2012; 133: 19–25. Available from <http://www.sciencedirect.com/science/article/pii/S016372581100163X>
68. Cabungcal JH, Counotte DS, Lewis EM, Tejeda HA, Piantadosi P, Pollock C, et al. Juvenile Antioxidant Treatment Prevents Adult Deficits in a Developmental Model of Schizophrenia. *Neuron.* 2014; 83: 1073–84. Available from <http://dx.doi.org/10.1016/j.neuron.2014.07.028>
69. Velasco I, Salazar P, Giorgetti A, Ramos-Mejía V, Castaño J, Romero-Moya D, et al. Concise Review: Generation of Neurons From Somatic Cells of Healthy Individuals and Neurological Patients Through Induced Pluripotency or Direct Conversion. *Stem Cells.* 2014; 32: 2811–7. Available from <http://www.pubmedcentral.nih.gov/articlerender.fcgi?artid=4282532&tool=pmcentrez&rendertype=abstract>
70. Nageshappa S, Carromeu C, Trujillo CA, Mesci P, Pasciuto E, Vanderhaeghen P.

- Altered neuronal network and rescue in a human MECP2 duplication model. *Mol. Psychiatry*. 2016; 21: 178–88. Available from <http://dx.doi.org/10.1038/mp.2015.128>
71. Supekar K, Uddin LQ, Khouzam A, Phillips J, Gaillard WD, Kenworthy LE, et al. Brain Hyperconnectivity in Children with Autism and its Links to Social Deficits. *Cell Rep*. 2013; 5: 738–47. Available from <http://dx.doi.org/10.1016/j.celrep.2013.10.001>
72. Di Martino A, Yan C-G, Li Q, Denio E, Castellanos FX, Alaerts K, et al. The autism brain imaging data exchange: towards a large-scale evaluation of the intrinsic brain architecture in autism. *Mol. Psychiatry*. 2014; 19: 659–67. Available from <http://www.ncbi.nlm.nih.gov/pubmed/23774715>
73. Vadodaria KC, Mertens J, Paquola A, Bardy C, Li X, Jappelli R, et al. Generation of functional human serotonergic neurons from fibroblasts. *Mol. Psychiatry*. 2016; 21: 49–61. Available from <http://www.nature.com/doi/10.1038/mp.2015.161>
74. Smeets EEJ, Pelc K, Dan B. Rett Syndrome. *Mol. Syndromol*. 2011; 2: 113–27.
75. Mullard A. Fragile X disappointments upset autism ambitions. *Nat. Rev. Drug Discov*. 2015; 14: 151–3. Available from <http://www.nature.com/doi/10.1038/nrd4555>
76. Mullard A. Fragile X drug development flounders. *Nat. Rev. Drug Discov*. 2016; 15: 77–77. Available from <http://www.nature.com/doi/10.1038/nrd.2016.18>
77. Ferenczi E, Deisseroth K. Illuminating next-generation brain therapies. *Nat. Neurosci*. 2016; Available from <http://www.nature.com/doi/10.1038/nn.4232>
78. Hao S, Tang B, Wu Z, Ure K, Sun Y, Tao H, et al. Forniceal deep brain stimulation rescues hippocampal memory in Rett syndrome mice. *Nature*. 2015; 526: 430–4. Available from <http://www.nature.com/doi/10.1038/nature15694> <http://www.ncbi.nlm.nih.gov/pubmed/26469053>

Please wait...

If this message is not eventually replaced by the proper contents of the document, your PDF viewer may not be able to display this type of document.

You can upgrade to the latest version of Adobe Reader for Windows®, Mac, or Linux® by visiting http://www.adobe.com/go/reader_download.

For more assistance with Adobe Reader visit <http://www.adobe.com/go/acrreader>.

Windows is either a registered trademark or a trademark of Microsoft Corporation in the United States and/or other countries. Mac is a trademark of Apple Inc., registered in the United States and other countries. Linux is the registered trademark of Linus Torvalds in the U.S. and other countries.

Author Manuscript

Please wait...

If this message is not eventually replaced by the proper contents of the document, your PDF viewer may not be able to display this type of document.

You can upgrade to the latest version of Adobe Reader for Windows®, Mac, or Linux® by visiting http://www.adobe.com/go/reader_download.

For more assistance with Adobe Reader visit <http://www.adobe.com/go/acrreader>.

Windows is either a registered trademark or a trademark of Microsoft Corporation in the United States and/or other countries. Mac is a trademark of Apple Inc., registered in the United States and other countries. Linux is the registered trademark of Linus Torvalds in the U.S. and other countries.

Author Manuscript

Please wait...

If this message is not eventually replaced by the proper contents of the document, your PDF viewer may not be able to display this type of document.

You can upgrade to the latest version of Adobe Reader for Windows®, Mac, or Linux® by visiting http://www.adobe.com/go/reader_download.

For more assistance with Adobe Reader visit <http://www.adobe.com/go/acrreader>.

Windows is either a registered trademark or a trademark of Microsoft Corporation in the United States and/or other countries. Mac is a trademark of Apple Inc., registered in the United States and other countries. Linux is the registered trademark of Linus Torvalds in the U.S. and other countries.

Author Manuscript