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Experimental Model Systems for Rare and Common Variants

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Abstract

Resolving the target genes, pathways, cellular phenotypes, and circuit functions impacted by the hundreds of genomic loci significantly associated with psychiatric disorders is a major challenge. Applications of genomic engineering in human-induced pluripotent stem cells (hiPSCs) and mice are widely used to study the impact of psychiatric risk variants within defined cell types of the brain. As the scale and scope of functional genomic studies expands, so must our ability to resolve the complex interplay of the many risk variants linked to psychiatric disorders. Here we discuss the current state of the field, with particular emphasis on hiPSC and mouse models, which have facilitated efforts to understand the pathophysiology of psychiatric disorders and translate genetic findings into disease-relevant biology in the service of advancing diagnostics and therapeutic development.

Introduction

Mental illness is a major contributor to global disease burden (GBD 2016 Disease and Injury Incidence and Prevalence Collaborators 2017). Successful efforts toward improved diagnosis, prognosis, treatment, and prevention require integrated efforts in a multitude of disciplines ranging from governmental legislation and public health strategies to biomedical innovation and pathophysiological insights. Here we discuss experimental approaches widely utilized in uncovering the molecular mechanisms underlying neurodevelopment and pathophysiological dysfunction of the human brain, with an emphasis on leveraging human genetic variation to elucidate the biology of mental illness through experimental model systems. We consider two widely used model systems for investigating the biology of genetic risk factors for human psychiatric disorders: genetically modified mice and human-induced pluripotent stem cells (hiPSCs).

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Most pharmacotherapies currently used in psychiatry were introduced to the clinic in the second half of the twentieth century. In 1949, lithium was discovered as a psychotropic medication with mood stabilizing effects, and in the following decade, the major classes of other pharmacotherapies (antipsychotics, benzodiazepines, and antidepressants) were introduced (Hyman 2012). Like many medications used in medicine, these medications were not rationally designed based on a known mechanistic pathophysiology to specifically target disorder mechanisms for modifying causal biological processes. Rather, most therapeutic discovery in psychiatry has emerged from serendipitous observations. Many of the seminal mechanistic "insights" into psychiatric disorder pathophysiology derived from uncovering the molecular targets of these serendipitously discovered therapeutic compounds. For instance, the findings that monoamine oxidase inhibitors are efficacious antidepressants and that D2-receptor antagonists are effective antipsychotics offered entry points for studying the neurobiology of depression (Hirschfeld 2000) and schizophrenia (Kendler and Schaffner 2011), respectively. However, emerging psychiatric genetic findings have paved a novel road forward with regard to cell type-specific disease etiology (Skene et al. 2018).

Heritability of Psychiatric Disorders

A critically important insight into the etiology of psychiatric disorders became known a century ago when the tendency for mental illness to aggregate in families was observed. This observation was statistically confirmed in the second half of the twentieth century in a large number of family, twin, and adoption studies aimed at quantitatively distinguishing environmental from nonenvironmental (i.e., genetic) disorder risk (Polderman et al. 2015). These studies revealed a relatively high, but varying degree of heritability for the major psychiatric illnesses (Visscher et al. 2008). For example, major depressive disorder has an estimated twin heritability of 0.37, autism spectrum disorder (ASD) 0.75, bipolar disorder 0.72, and schizophrenia (SCZ) 0.81 (Sullivan and Geschwind 2019). Overall, psychiatric disorders reflect the additive impact of hundreds of risk variants, each of which confer only a tiny increase of risk and are common in the population at large, coupled with that of highly penetrant rare mutations that underlie a fraction of cases.

Common Variation in Psychiatric Disorders

Genome-wide association studies (GWASs) were a promising design to overcome some of the obstacles encountered in linkage and candidate gene designs. After their introduction, single nucleotide polymorphism (SNP) microarrays quickly became inexpensive, which enabled researchers to investigate relatively large cohorts of cases and controls. Furthermore, with SNP

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markers being distributed widely across the genome, loci could be investigated in an unbiased fashion. Despite these advantages, the first years of GWASs in psychiatry were characterized by failures to identify reproducible loci that reached the threshold for genome-wide statistical significance. For example, nine GWASs on major depressive disorder (published between 2010 and 2013 with sample sizes ranging between 1,000–9,500 cases) did not reveal any genome-wide significant loci (Flint and Kendler 2014).

Successes using the GWAS approach began to emerge as sample sizes increased (Hyde et al. 2016; Major Depressive Disorder Working Group of the Psychiatric GWAS Consortium 2013; Wray et al. 2018a), and phenotypes were more narrowly defined (Converge Consortium 2015). This development was facilitated by the formation of global research consortia, such as the Psychiatric Genomics Consortium, which allowed for the analysis of very large cohorts with consistent genotype quality control. To date, GWAS has yielded:

- 287 loci for SCZ (Pardinas et al. 2018; Trubetskoy et al. 2022).
- Over 100 for major depressive disorder (Howard et al. 2019; Levey et al. 2021; Wray et al. 2018b).
- 12 for ASD (Grove et al. 2019).

Biologically, risk variants linked to SCZ (Fromer et al. 2014; Marshall et al. 2017; Pardinas et al. 2018; Purcell et al. 2014), ASD (De Rubeis et al. 2014; Sanders et al. 2015; Satterstrom et al. 2020), and more broadly across the neuropsychiatric disorder spectrum (Cross-Disorder Group of the Psychiatric Genomics Consortium 2019; Lee et al. 2019d; Schork et al. 2019; Sey et al. 2020; Watanabe et al. 2019a) are enriched for genes involved in synaptic biology and transcriptional regulation (De Rubeis et al. 2014; Neale et al. 2012; O'Roak et al. 2012a, 2014; Sanders et al. 2012; Talkowski et al. 2012).

The GWAS design is suited to discover SNPs associated with diseases that are common in a population, where the minor allele frequency is >1%. Individually, each SNP typically confers only a small risk to disease status, with odds ratios ranging between 1.05–1.15, but in aggregate, common variants explain approximately 24% of the variance in SCZ liability (Trubetskoy et al. 2022). In recent years, an increasing number of publications have investigated multiple SNPs that are differentially present in cases versus controls, leading to the generation of polygenic risk scores for psychiatric disorders (International Schizophrenia Consortium et al. 2009) as predictors of diagnosis, clinical severity, and/or drug responsiveness (Hess et al. 2021; Ruderfer et al. 2016; Zhang et al. 2019). For SCZ, for example, when sorted on effect size (odds ratios), inheriting the top 10% versus the bottom 10% of SNP-risk variants increases one's chance for developing SCZ nearly fortyfold. Although informative with respect to the role of SNPs on the biology of SCZ, as well as the shared polygenic risk architecture across psychiatric disorders (Cross-Disorder Group of the Psychiatric Genomics Consortium 2019; Lee et al. 2019d; Schork et al. 2019), the sensitivity and specificity of these polygenic risk scores are

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currently too low to contribute meaningfully to individual-level psychiatric diagnoses (Zheutlin et al. 2019).

Rare Variation in Psychiatric Disorders

The discovery of the architecture of rare genetic variation in psychiatric disorders has progressed in parallel with the elucidation of common SNP associations. The first wave of findings implicating rare genetic variation in psychiatric disorder risk were large copy number variants (CNVs) that disrupted the function of one or multiple genes and were found in patients with ASD and SCZ (Iossifov et al. 2012; Karayiorgou et al. 1995; Marshall et al. 2017; Sebat et al. 2007). Following these early discoveries, whole-exome and whole-genome sequencing continued to reveal novel, rare, and highly penetrant variants implicated in psychiatric disorder risk. Rare variant discovery has been most successful for ASD, for which over 100 high-confidence genes have been described to date (Satterstrom et al. 2020); these are typically *de novo* protein-truncating variants (PTVs) that are individually rare, but jointly account for around 15% of individuals with ASD. For SCZ, ten genes have been identified thus far with the same degree of statistical confidence (Singh et al. 2016, 2022; Steinberg et al. 2017; Takata et al. 2014). Among rare variants, evidence of their association with SCZ risk has been strongest for PTVs. However, missense variants have also been implicated in ASD, although effect sizes are typically smaller than for PTVs (Ruzzo et al. 2019; Sanders et al. 2015). For major depressive disorder and bipolar disorder, very little is known about the contribution of rare variants to risk (Palmer et al. 2022).

Rare variants are frequently pleiotropic and show robust overlap across psychiatric disorders (Rees et al. 2021), although different mutation types are implicated in some shared genes (Ben-Shalom et al. 2017). In addition, it is increasingly recognized that common and rare genetic risk factors converge at least partially on the same underlying pathogenic biological processes (Chang et al. 2018; Jia et al. 2018; Nehme et al. 2021; Singh et al. 2022).

Functional Genomic Studies of Psychiatric Disorder Pathophysiology

Understanding the heritability and genetic architecture of psychiatric disorders is only the first step in understanding the underlying pathophysiology and successfully translating this knowledge into clinically effective treatments. Although the contribution of common variation to the heritability of most psychiatric disorders appear collectively to exceed that of rare variation, mechanistic biological insights into rare variants has thus far been technically more feasible to investigate using gene targeting approaches in experimental model systems. However, increasingly sophisticated methods are yielding

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exciting new advances and insights into the biology of common variants and polygenic risk.

Mouse Models of Psychiatric Genetic Risk

Much has been learned from mouse models with modifications in genes associated with psychiatric disorders. For ASD, gene knockouts include the mouse homologues of *CHD8* (Katayama et al. 2016), *TSC1* (Goorden et al. 2007), *SCN2A* (Spratt et al. 2019), *SHANK3* (Peca et al. 2011), *NRXN1* (Etherton et al. 2009), *CNTNAP2* (Penagarikano et al. 2011) as well as *16p11.2* microdeletion (Brunner et al. 2015), all of which are causal to syndromic or non-syndromic ASD in humans (Satterstrom et al. 2020). These rodent models have partially overlapping behavioral phenotypes reminiscent of human ASD-related behavior, including impaired social interactions and vocal communication, repetitive grooming, and seizures. Commonalities in these models across the molecular, cellular, and circuit levels include neuronal morphology deficits as well as altered cellular signaling (PI3K, mTOR) and excitation/inhibition imbalance (de la Torre-Ubieta et al. 2016). Rodent models for ASD have been particularly useful because of their relatively high degree of genetic homology to humans, their practical advantages as experimental animals, the availability of phenotypic batteries for core behavioral phenotypes, and the possibility for experimental inquiry at multiple levels of organization, from molecule to circuit to behavior.

For SCZ, the number of valid and therefore useful genetic rodent models has been fewer than for ASD because of the time required to identify *high-confidence* risk genes or CNVs for SCZ. Moreover, given that historical candidate genes for SCZ have largely failed to validate in more recent genomic studies (Farrell et al. 2015; Sullivan 2013), many long-standing animal models of SCZ (notably *DISC1* KO) have questionable credibility today. The two genetic risk factors that confer the highest risk for SCZ—the *22q11.2* microdeletion (Sigurdsson et al. 2010) and *SETD1A* (Mukai et al. 2019; Nagahama et al. 2020)—have therefore yielded genetic rodent models with a higher *a priori* chance for robust face and construct validity. Moreover, a mouse model of the well-replicated *16p11.2* microduplication SCZ genetic risk variant has also yielded novel biological insights (Bristow et al. 2020; Horev et al. 2011). Overall, behavioral phenotypes for SCZ models include deficits in working memory and prepulse inhibition, hypothesized to be reminiscent of cognitive and negative SCZ symptoms.

The ability to spatiotemporally manipulate genetically defined cell types and neuronal circuits *in vivo* has been a critical advance for understanding the complexity of network connectivity, perception, and behavior. Genetically engineered mouse lines and viral vectors utilizing common strategies for cell type-specific expression of transgenes has enabled a diverse set of tools to monitor, label, and manipulate genetically defined brain cell types. The

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Cre/loxP system, in which Cre recombinase efficiently catalyzes recombination at genomically integrated *loxP* sites, is the most widely employed strategy for cell type-specific transgene expression and endogenous genome manipulation, enabling cell type-, temporal-, and region-specific control through cell type-specific gene promoters combined with an ever-expanding suite of fluorescent proteins, optogenetic tools, and genetically encoded calcium indicators. Increasing numbers of well-characterized Cre driver lines with CNS expression have been generated through large-scale initiatives such as GENSAT (Gong et al. 2007), NIH Neuroscience Blueprint Cre Driver Network (Taniguchi et al. 2011), and the Allen Institute (Madisen et al. 2012), as well as by independent investigators. Limited effort has been given, however, to implement next-generation genome-wide screens for novel cell type-specific enhancers (Shima et al. 2016). Thus, the full potential and utility of the various driver lines and enhancer elements currently available for providing genetic access to defined cell populations across the entire brain have not yet been fully realized. Continued efforts to discover cell type-specific gene promoter driver lines will provide increasing precision to the classification of cell types, dissection of neural circuit function, and pathophysiological understanding of psychiatric disorder risk variants.

Modeling psychiatric disorders in rodents also comes with caveats and limitations, the foremost being the complex phenotype of mental disorders. Despite high hopes and expectations, other than rare pathogenic mutations for SCZ and ASD (which only account for ~2% and ~10% of cases, respectively, based on current best estimates), psychiatry still lacks objectively measurable biomarkers with sufficiently high positive and negative predictive values to contribute meaningfully to clinical diagnosis for the majority of common psychiatric disorders. Therefore, we remain dependent on cognitive and behavioral phenotypes as outcome measures. Psychiatric disorders typically affect higher-order brain processes, including cognition, emotion, and perception for which the current diagnostic criteria are heavily reliant on self-report of subjective experiences. Accordingly, a substantial proportion of mouse models of genetic risk for neurodevelopmental and psychiatric disorders generated to date have struggled to yield robust behavioral phenotypes that have been reliably replicated across multiple independent laboratories. Although many aspects of brain development appear to be conserved across mammalian species (Defelipe 2011), mice and humans are separated by an estimated 85 million years of evolution. Moreover, species-specific differences in neurobiology appear to be especially apparent in the cerebral cortex. For example, the human neocortex has approximately a thousandfold increase in both neuronal numbers and surface area compared to mice (Herculano-Houzel et al. 2006). At the tissue-organizational level, superficial layer neurons appear to be expanded in numbers in humans (Hill and Walsh 2005), as is the case for outer radial glial cells that are scarcely present in mice (Hansen et al. 2010; Shitamukai et al. 2011; Wang et al. 2011). Single nucleus RNA sequencing has recently

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corroborated some of these histological observations and has revealed other notable transcriptomic differences across putatively homologous cell types (Hodge et al. 2019). Furthermore, although coding sequence and linear gene sequence are reasonably well conserved between mice and humans, noncoding sequence is highly divergent, which has proven highly problematic for mouse modeling of common variant polygenic risk. Moreover, the recent emergence of molecular genetic therapeutics, such as antisense oligonucleotides, might also be difficult to model in mice for modalities that have very stringent genomic target sequence requirements (e.g., Angelman syndrome mouse models; Milazzo et al. 2021; see also Bearden et al., this volume).

hiPSC-Based Modeling of Psychiatric Disorder Risk

Human embryonic stem cell lines, first grown in 1998, were immediately recognized as a nearly limitless source of material for studies of human disease (Thomson et al. 1998). Studies to uncover the molecular networks regulating pluripotency (i.e., their capability to differentiate into every cell type in the human body) found four genes that, when overexpressed, are sufficient to reprogram adult somatic cells into hiPSCs (Takahashi et al. 2007), making it possible to generate donor-specific cells. This opened novel avenues of human disease modeling, in particular for neuropsychiatric disorders, that otherwise have limited opportunity for physiological studies with patient-derived CNS cell types and tissue. Early studies of psychiatric disorders largely focused on idiopathic cohorts of ASD (Mariani et al. 2015) and SCZ (Brennand et al. 2011). To reduce heterogeneity between cases, hiPSC-based studies frequently contrast hiPSC-derived neurons generated from cases that share a highly penetrant rare mutation with, for example, *MECP2* (Marchetto et al. 2010), *FMR1* (Zhang et al. 2018), *CACNA1C* (Paşca et al. 2011), *SHANK3* (Shcheglovitov et al. 2013), *NRXN1* (Flaherty et al. 2019), *NLGN4* (Marro et al. 2019), and *22q11.2* microdeletion (Khan et al. 2020). These studies report cellular phenotypes such as altered neuronal development and/or synaptic function. Likewise, engineered deletions of risk genes—such as *FMR1* (Zhang et al. 2018), *SHANK3* (Yi et al. 2016), and *NRXN1* (Pak et al. 2015)—yield similar cellular phenotypes compared to those derived from patients carrying equivalent mutations. Despite the relatively subtle effects predicted for SCZ-associated GWAS SNPs, several have been validated using case-control cohort designs; for example, *C4* (Sellgren et al. 2019) and *CACNA1C* (Yoshimizu et al. 2015).

Neural differentiation from hiPSCs can yield all major cell types of the brain:

- glutamatergic neurons (Yu et al. 2014),
- GABAergic neurons (Shao et al. 2019),
- dopaminergic neurons (Hook et al. 2014),
- neural progenitor cells (Brennand et al. 2015),

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- astrocytes (Windrem et al. 2017),
- oligodendrocytes (McPhie et al. 2018), and
- microglia (Sellgren et al. 2019).

These hiPSC-derived brain cells recapitulate many aspects of gene expression (Brennand et al. 2015; Hoffman et al. 2017; Mariani et al. 2012; Nicholas et al. 2013; Paşca et al. 2015; Qian et al. 2016), cellular diversity (Velasco et al. 2019), and micro-architectural features of the developing human brain (Kadoshima et al. 2013; Paşca 2019), but have thus far shown resistance to *in vitro* differentiation into later neurodevelopmental stages. Notably, the rare and common risk genes associated with psychiatric disorders are enriched for those expressed during fetal cortical development (Gulsuner et al. 2013; Loohuis et al. 2015; Schork et al. 2019; Talkowski et al. 2012). Both ASD and SCZ involve pathophysiological mechanisms with antecedents substantially earlier than the onset of clinical symptoms. In conclusion, hiPSC-derived models are particularly well-positioned to investigate the neurodevelopmental impact of psychiatric risk variants, particularly those predicted to influence fetal cortical development.

CRISPR Methods for hiPSC-Based Psychiatric Risk Modeling

CRISPR-associated proteins (Cas) use short, easily synthesized, clustered regularly interspaced short palindromic repeats (CRISPR) sequences to guide RNAs to recognize specific complementary strands of DNA. Together, they form the basis of a technology known as CRISPR/Cas genome engineering with which it is possible to edit DNA sequences efficiently (Anzalone et al. 2019; Hsu et al. 2014), activate or repress endogenous gene expression (Ho et al. 2017), cleave RNA (Konermann et al. 2018), methylate DNA (Liu et al. 2018b), modify histones (Hilton et al. 2015; Thakore et al. 2015), or alter chromatin interactions (Liu et al. 2018a) on a near genome-wide scale (Sanson et al. 2018). Altogether, this makes it possible to comprehensively introduce or correct the risk architecture associated with psychiatric disorders, alone or in combination, across the major cell types of the brain.

Moreover, because CRISPR engineering yields comparisons across a shared genetic ("isogenic") background, the mechanism of action of SCZ SNPs can be resolved in a cell type-specific and even context-dependent manner, through enhancer-promoter looping (*CACNA1C*; Roussos et al. 2014), 3D-genome folding (*PCDH*; Rajarajan et al. 2018), miRNA abundance (*miR-137*; Forrest et al. 2017), and mRNA levels (*FURIN*; Schrode et al. 2019).

High-throughput methods facilitate the comprehensive survey of the loci associated with psychiatric disease (Townsley et al. 2020). Massively parallel reporter assays evaluate the regulatory activity of the thousands of loci associated with psychiatric disorder risk (Mulvey and Dougherty 2021; Myint et al. 2020; Uebbing et al. 2021) and/or human cortical development (Geller

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et al. 2019; Inoue et al. 2019), albeit not at endogenous loci. For examining regulatory elements within their original genomic context, CRISPR expression quantitative trait loci (eQTL) mapping can identify SNPs that differentially regulate proximal (Gasperini et al. 2019) and distal (Fulco et al. 2019) target gene expression. Likewise, population-scale village-in-a-dish experiments—whereby single-cell RNA sequencing is applied to pooled populations of hiPSC-derived neurons or glia from dozens of genotyped donors (Cuomo et al. 2020; Jerber et al. 2021; Mitchell et al. 2020; Neavin et al. 2021a)—are well-powered to uncover transcriptomic and/or phenotypic impacts of common and rare variants.

Traditional functional genomic approaches assess one variant or gene at a time in a hypothesis-driven manner. Large-scale screens systematically manipulate many variants or genes in an arrayed format, characterizing those that result in specific changes, but requiring substantial investments of time and resources. In contrast, pooled designs can identify variants or genes that impact phenotypes such as proliferation, survival, or fluorescence; this allows for isolation of "hits" from the cellular population using fluorescent-activated cell sorting. Moreover, coupling CRISPR-based pooled perturbations to single-cell RNA sequencing (Dixit et al. 2016; Mimitou et al. 2019) expands the questions that can be queried. Altogether, this makes possible the comprehensive interrogation of candidate disease genes (Cederquist et al. 2020) or gene lists (Liu et al. 2018c; Lu et al. 2019; Tian et al. 2019).

Fundamental Questions Lacking Answers

A major challenge in the field is connecting disease-associated risk genes to their respective pathways and functions (Sullivan and Geschwind 2019). Risk variants are predicted to converge at the pathway level (Ballouz and Gillis 2017). Might this represent a novel point of therapeutic intervention? Perturbation of a dozen SCZ risk genes in human neurons revealed convergent changes in gene expression among a subset of genes and subnetworks involved in synaptic function (Townsley et al. 2022). Evaluation of an overlapping set of ASD genes *in vivo* (35 genes) in fetal mouse brains (Jin et al. 2020) and *Xenopus tropicalis* (10 genes) (Willsey et al. 2021), and *in vitro* in human neural progenitor cells (27 genes) (Cederquist et al. 2020) and human brain organoids (3 genes) (Paulsen et al. 2022) revealed convergent impacts on gene expression (Jin et al. 2020; Paulsen et al. 2022), WNT signaling (Cederquist et al. 2020), and neurogenesis (Cederquist et al. 2020; Jin et al. 2020; Paulsen et al. 2022; Willsey et al. 2021). Taken together, multifaceted evidence suggests that asynchronous development of inhibitory GABAergic neurons and deep-layer glutamatergic projection neurons contribute meaningfully to the etiology of severe psychiatric illness through distinct molecular pathways (Paulsen et al. 2022).

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Because psychiatric disorders arise through iterative pathological changes in circuit function (Südhof 2017a), an important challenge is to assemble more physiologically relevant *in vitro* models. How do risk variants impact cellular function within circuits? To answer this, we must incorporate critical aspects of neuronal circuitry (Birey et al. 2017; Xiang et al. 2019), glial support (Abud et al. 2017; Dezonne et al. 2017; Marton et al. 2019), vasculature (Cakir et al. 2019; Mansour et al. 2018), and blood–brain barrier functions (Vatine et al. 2019) but retain the capacity to resolve cell type-specific effects. For example, mutations in *CACNA1C* associated with Timothy syndrome cause deficits in calcium signaling in both glutamatergic (Birey et al. 2017; Paşca et al. 2011) and GABAergic (Birey et al. 2017) neurons, which specifically lead to perturbations of interneuron migration into cortical assembloids (Birey et al. 2022).

Risk variants linked to psychiatric disorders show cumulative effects (Tansey et al. 2016; Weiner et al. 2017). Will understanding how risk variants combine to yield a greater impact in aggregate improve genetic diagnosis and/or indicate new drug targets? Despite evidence to the contrary at the population level (Visscher et al. 2008; Wray et al. 2018b), within individual patients, genetic risk factors may combine in patterns dependent on whether their target genes are co-expressed in the same cell types or act within the same biological functions. For example, when SCZ risk genes are manipulated together in neurons, an unexpected combinatorial effect occurs that cannot be predicted from single gene perturbations alone, and this effect is concentrated within synaptic function and psychiatric disorder risk genes (Schrode et al. 2019). Likewise, interactions of multiple target genes at a single SCZ GWAS risk locus result in synergistic contributions to molecular and synaptic phenotypes observed in neurons (Zhang et al. 2021).

Disease-associated variants may regulate their target genes in context-dependent manners. How do risk variants interact with the environment across the diverse cell types that comprise the human brain? Gene–environment interactions in GWAS may be especially critical in studies of those psychiatric disorders that require specific exposures (e.g., substance use disorders) or stressors (e.g., posttraumatic stress disorder). For example, glucocorticoid signaling is highly associated with trauma response (Daskalakis et al. 2014), for which *in vitro* studies reveal that brain organoids exposed to excessive glucocorticoid show impaired neuronal maturation (Cruceanu et al. 2021), and hiPSC neurons derived from cases with posttraumatic stress disorder can be distinguished from controls by glucocorticoid hypersensitivity (Breen et al. 2021).

Finally, the extent to which clinical drug responsiveness is heritable and/or stable throughout the lifetime requires further investigation, but promising examples, such as lithium-responsive bipolar disorder (Hou et al. 2016), have been identified (Stern et al. 2018). Is clinical treatment response predictable? In hippocampal neurons derived from lithium-responsive (but not nonresponsive) patients with bipolar disorder, hyperexcitability is ameliorated following lithium treatment (Mertens et al. 2015). A similar phenotypic analysis was

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able to predict with 92% accuracy whether hippocampal granule cell neurons were derived from a patient with or without clinical responsiveness to lithium. An improved drug-screening strategy would better recapitulate disease pathophysiology and integrate advances in psychiatric genetics. Moving toward this, proof-of-concept application of transcriptomic drug screening, using hiPSC-based models, have demonstrated that drug-induced differences in SCZ patient-derived neural progenitor cells could reverse postmortem SCZ transcriptional signatures and were enriched for genes related to SCZ biology (Readhead et al. 2018). Altogether, these studies reveal major advantages of incorporating cell type- and patient-specific platforms in drug discovery.

Summary

Each patient represents a unique aggregation of risk factors with distinct expression patterns, biological convergence, and cumulative effects. A functional genomics approach that integrates stem cell and animal models with genome engineering might resolve some of the major questions regarding the impact of patient-specific variants across cell types, genetic backgrounds, and environmental conditions. Striving to translate risk "variants to genes," "genes to pathways," and "pathways to circuits" has the potential to reveal unexpected causal relationships between risk factors within and between the cell types of the brain. These insights could identify therapeutics tailored to an individual's specific risk profile and fuel the development of novel, personalized approaches to mental health care.

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