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Genomic structural variation in psychiatric disorders

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Abstract

Copy number variants (CNVs) are submicroscopic deletions and duplications of genomic material that were previously thought to be rare phenomena. They have now been robustly associated with a variety of disorders such as autism, schizophrenia, and attention-deficit/hyperactivity disorder through an emerging research base in affective disorders. A complex picture is emerging of a polygenic, heterogeneous model of disease, with CNVs conferring broad susceptibility to a variety of neurodevelopmental disorders, rather than specific disorders per se. Although the insights gleaned thus far only represent a small piece of a much larger puzzle, progress has been rapid and new technologies promise even more insights into these hitherto opaque brain disorders. We will discuss CNVs, the current state of evidence for their role in the pathogenesis of classical psychiatric disorders, and the application of such knowledge in clinical settings.

Psychiatric disorders have been observed to run in families for hundreds of years. Taking unipolar depressive disorder as an example, first-degree relatives of probands with depressive disorder are 2.83 fold as likely to suffer from the disorder than controls (Sullivan, Neale, & Kendler, 2000). Likewise, the monozygotic co-twin of a case of schizophrenia has a risk of over 50% of also developing the disorder, with the risk increasing exponentially as a function of the degree of relatedness to the proband (Sullivan, Kendler, & Neale, 2003). Heritability estimates for psychiatric disorders are variable, but autism (Bailey et al., 1995), schizophrenia (Cardno & Gottesman, 2000), bipolar disorder (McGuffin et al., 2003), and recurrent and severe forms of unipolar depressive disorder (McGuffin, Katz, Watkins, & Rutherford, 1996) all have strikingly high heritability estimates. Clinical groups are usually defined according to the *International Classification of Diseases, 10th Edition* (World Health Organisation, 1992) and *Diagnostic and Statistical Manual of Mental Disorders, Fourth Edition, Text Revision* (American Psychiatric Association, 1994) clinical descriptions. Although these have reliability, they are limited in terms of validity. Within-group heterogeneity is a constant problem in genetic studies of psychiatric disorders, and many individuals can fit criteria for more than one clinical group. This suggests that the neurobiology underlying psychiatric disorders is complex and multifaceted.

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Human neurodevelopment and genetics are intimately related subjects. Although a small number of disorders (e.g., Huntington disease) have been conclusively linked to particular genetic abnormalities, the genetic etiology of the majority has remained obscure, probably because it is more complicated than a single-gene to single-phenotype relationship. At the turn of the millennium, the first draft of the human genome was published (Lander et al., 2001). At about the same time, microarray chips were developed that allowed thousands of genetic experiments to be performed at once on a single glass slide. These two advances allowed what previously had been a time-consuming and expensive process to be performed both rapidly and relatively cheaply in large numbers of samples (Schena, Shalon, Davis, & Brown, 1995). The Genome-Wide Association Study (GWAS) was born, and with it has come vast quantities of new genetic data allowing for association studies of more complex traits with large numbers of different genomic variants.

Deletions and duplications of genetic material occur within the human genome by various molecular mechanisms (Zhang, Gu, Hurles, & Lupski, 2009). Down syndrome represents the most obvious example, caused by a whole duplication of chromosome 21. However, until comparatively recently it was thought that such deletion and duplication events were rare phenomena, explaining only a handful of sporadic, infrequently occurring genetic diseases (Perry et al., 2008). The new wave of genetic data from GWAS showed that these events occurred in the normal population, and the extent of this was a surprise to many geneticists (Freeman et al., 2006; Iafrate et al., 2004; McCarroll et al., 2006; Sebat et al., 2004). Copy number variants (CNVs) may represent one of the largest and most dynamic forms of genetic variation and the formation of gametes with reciprocal deletions and duplications is likely to be an essential driver of species evolution (Locke et al., 2003; Lupski,

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2007). Within the field of psychopathology, this new realm of variation has rapidly become of considerable interest.

Definitions

A CNV is rather arbitrarily defined as a deletion or duplication of genomic material of more than 1,000 base pairs (Feuk, Carson, & Scherer, 2006; Iafrate et al., 2004; Sebat et al., 2004). Deletion CNVs remove one copy (a hemizygous deletion) or both copies (a homozygous deletion) of genetic material. A homozygous deletion is likely to be more deleterious than a hemizygous deletion, because both copies of a given genomic sequence, which may include genes, are deleted. Duplication CNVs can exist as three copies, four copies, or more than four copies.

CNVs occur with differing frequencies throughout populations, and they are divided for convenience into those defined as "common" (occurring in >1 in 100 individuals in a population) and "rare" (<1 in 100; Pinto et al., 2010). When studying associations with disease states, the rarest CNVs are logically of the most interest, because they are more likely to be evolutionarily under negative selection pressure (and therefore conferring disadvantage, possibly disease). De novo CNVs are defined as CNVs that are seen in children but not in their biological parents, and they are usually generated in the cells that give rise to gametes or, more rarely, in the immediate cell divisions after conception. CNVs may also be referred to as "singleton" CNVs, which refers to a CNV that is only seen once in any given data set. Assuming the data set is large enough, such CNVs are also rare in the general population.

Schizophrenia

Schizophrenia is a common (~0.8% lifetime incidence) psychiatric disorder with a substantial heritable component (estimated at around 80%), with first-degree relatives of sufferers having a 10-fold increased risk of also developing the disorder (for a review, see Gottesman, 1991). Although it can occur at any age, even in childhood, it is most commonly seen in early adulthood and is characterized by problems with perception, inferential thinking, goal-directed behavior, and emotional expression (Andreasen, 1995), and a critical lack of insight into the disorder. No one symptom or sign is common to all individuals diagnosed with the disorder. Many follow a chronic course of illness suffering substantial psychosocial deficits and increased morbidity and mortality for the rest of their lives, especially from suicide (Harris & Barraclough, 1998). Although past genetic studies involving linkage, single nucleotide polymorphisms (SNPs), and candidate genes have discovered some interesting regions in the genome, these have not been replicated often (for a review, see, e.g., Schwab & Wildenauer, 2009; and Sullivan, 2005). A substantial proportion of heritability remains to be explained.

Deserving special mention before a wider discussion of CNVs in schizophrenia is a deletion of a region of chromosome 22q11.2. This deletion syndrome, in addition to being associated with a variety of congenital and neurodevelopmental abnormalities that result in eponymous syndromes such as DiGeorge syndrome, velocardiofacial syndrome, and conotruncal anomaly face syndromes (Robin & Shprintzen, 2005), is particularly significant because it is associated with a notably high incidence of schizophrenia (Murphy, Jones, & Owen, 1999). Attention-deficit/hyperactivity disorder (ADHD), autism, and mood disorders have also been associated with the deletion (Karayiorgou, Simon, & Gogos, 2010). It will be mentioned frequently in this article.

The first CNV studies in schizophrenia were published in late 2007 and early 2008. The first report, which was by Kirov et al. (2007), used comparative genomic hybridization arrays to investigate 93 individuals with schizophrenia compared to 372 controls being investigated for disorders that the authors considered unrelated to schizophrenia. They filtered out all CNVs found in the control group or found in the Database of Genomic Variants (http://projects.tcag.ca/variation/), which left 13 CNVs for further analysis. They identified two CNVs that were very likely to be etiologically significant. The first was a 1.4 mb duplication in chromosome 15q13.1, covering the amyloid beta A4 precursor protein-binding (*APBA2*), necdin-like 2, and tight junction protein 1 genes, neither of which was found in either biological parent (a de novo event). The *APBA2* gene is particularly interesting because it is highly expressed in all areas of the brain, including the prefrontal cortex, cingulate cortex, and amygdala. The protein product of this gene is a neuronal adaptor protein that binds directly to neurexins as part of a multiprotein complex that may act as a facilitator of neurotransmitter release from synaptic vesicles (Biederer & Südhof, 2000; Dulubova et al., 2007). This CNV has also subsequently been associated with both autism (Babatz, Kumar, Sudi, Dobyns, & Christian, 2009; Guilmatre et al., 2009) and schizophrenia (Need et al., 2009). The second CNV was a 250-KB deletion of 2p16.3 covering the neurexin-1 gene (*NRXN1*). Although *NRXN1* is a putatively interesting gene, because it has a pivotal role in supporting synaptic function, the CNV was also found in the asymptomatic mother.

In April 2008 Walsh et al. (2008) published the results of a study of 150 patients with schizophrenia and 268 ethnically matched controls, genotyped on comparative genomic hybridization arrays, and followed up with SNP microarrays, with a replication sample consisting of 92 patients with childhood-onset schizophrenia. They found a threefold enrichment of CNVs in gene-coding regions in the schizophrenia cohort and those with an onset of illness less than 18 years were more than 4 times as likely to carry a CNV as controls. Twenty-eight percent of the childhood-onset schizophrenia cohort (which is conceptualized to represent a particularly severe form of the disease) carried a CNV over a gene-coding region, again, significantly greater than the control sample. Of the CNVs that were detected, the authors found that they were significantly more likely than chance to cover genes known to be associated with central nervous system development and function. Neuregulin signaling, extracellular signal-regulated kinase/mitogen-activated protein kinase sig-

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naling, synaptic long-term potentiation, axonal guidance signaling, integrin signaling, and glutamate receptor signaling were particularly associated. In contrast, in the control group the CNVs observed covered genes involving any biological pathway. The authors looked at a number of gene-coding CNVs in the case cohort in more detail, for example, v-erb-a erythroblastic leukemia viral oncogene homolog 4, and showed that the resulting transcribed protein was aberrant and unlikely to be functional.

In July 2008 Xu et al. (2008) published an analysis of 152 individuals with schizophrenia and their biological parents compared against 159 controls drawn from the same population. The advantage of this approach is that, by looking at the biological parents of the cases, the group could determine whether the CNVs observed in the cases were de novo or inherited from the biological parents.

They found 15 cases with CNVs where the CNV was not seen in either parent (de novo) as opposed to 2 in controls, a significant enrichment of approximately eightfold. Two cases carried two de novo CNVs each. To assess whether or not de novo CNVs might be important in the etiology of schizophrenia, they went on to look for de novo CNVs in a further cohort of 48 cases of schizophrenia who had a first- or second-degree relative who also had the disorder. In contrast, they did not find de novo CNVs in this cohort. They also found that sporadic cases of schizophrenia were only 1.5 times as likely than unaffected controls to harbor an inherited rare CNV.

Taken together, this suggests that de novo CNVs are more likely to explain sporadic cases of schizophrenia than cases where there is a family history of the disease (a similar picture to autism). It also suggests that only a small proportion of rare inherited CNVs might be expected to contribute to the pathogenesis of sporadic cases. They did not find any association of rare CNV events with age of onset, presence or history of learning disability, or parental age at birth. Three cases of the 22q11.2 deletion were identified in this cohort, of which two had mild facial dysmorphism.

In September 2008 the International Schizophrenia Consortium (2008) reported results of their analysis in 3,391 patients with schizophrenia and 3,181 ethnically matched controls. They found that the overall burden of CNVs was increased 1.15 fold in their case cohort in comparison to the control cohort. They also found that large CNVs (>500 kb), CNVs that occurred rarely in the overall sample, and CNVs that covered genes, were particularly enriched in the cases of schizophrenia.

The general increase in burden of rare CNVs partly reflected the observation of large, rare deletion CNVs in a minority of cases but no controls. Although statistical tests of association of these large CNVs mostly failed to reach significance levels, probably because of a lack of statistical power, CNVs similar to those found in other disorders such as autism were observed, which, taken with other studies, is a significant finding.

The 22q11.2 deletion was found to be significantly associated, but the group also reported new associations with deletion CNVs at 15q13.3 and 1q21.1. The 15q13.3 deletion had also been associated with mental retardation and seizures in an article published earlier that year (Sharp et al., 2008), and the region at 1q21.1 had previously been associated with schizophrenia by linkage (Brzustowicz, Hodgkinson, Chow, Honer, & Bassett, 2000).

In a further large study, Stefansson et al. (2008) initially looked for de novo CNVs in an exploratory population sample, and then tested these 66 regions for association in a sample of 1,433 cases of schizophrenia against 33,250 controls. Those regions found to have a suggestive association were followed up in a replication cohort comprised 3,285 cases and 7,951 controls. CNV deletions at 1q21.1, 15q11.2, and 15q13.3 were identified as candidates for association in the first sample and all three of these deletions showed significant association in the follow-up sample.

The 22q11.2 deletion was present in 8 out of 3,838 cases, but was notably absent in all controls. This lends support to the idea that schizophrenia is polygenic, but that some genetic variants confer more risk than others. It is worth noting that, in this study, the authors also had access to clinical data, so they were able to study whether or not various clinical indicators were different in those individuals with CNVs. They found no differences in clinical response to treatment and there was no sex bias or tendency for those with a CNV to have a family history of schizophrenia. The authors noted that cases carrying the 1q21.1 deletion had learning disabilities, and of the controls that had the deletion, 2 had dyslexia.

A "multiple hit" hypothesis is hinted at here. No one individual variant is either necessary or sufficient to cause schizophrenia (although they may cause mild subclinical problems on their own), but together multiple variants may act to predispose an individual strongly toward developing the disorder.

McCarthy et al. (2009) investigated the role of recurrent CNVs at 16p11.2 in schizophrenia, working on the basis of past findings implicating this region in autism (Marshall et al., 2008; Weiss et al., 2008). This research group had also previously detected this variant in 2 cases of childhood-onset schizophrenia (Walsh et al., 2008). Two cohorts of schizophrenia cases were analyzed, matched against suitable control sets. In the first cohort, they found the 16p11.2 duplication CNV in 12 out of 1,906 cases (0.63%), but only 1 out of 3,971 controls (0.03%). In the second cohort, they found this duplication in 9 out of 2,645 (0.34%) cases, but only 1 out of 2,420 controls (0.04%). This represents a 14.5-fold increased risk of schizophrenia for those carrying the CNV.

It is interesting that the deletion CNV at 16p11.2 was not associated with schizophrenia. However, in a meta-analysis of previous reports the authors showed that the deletion was actually more associated with developmental disorders and autism rather than schizophrenia, whereas the duplication might be associated with autism, schizophrenia, and bipolar disorder.

At first glance the results of the analysis of this region of 16p seem confusing. Deletions and duplications at 16p11.2 appear to be associated with autism, whereas only the duplication is associated with schizophrenia and bipolar disorder. Some individuals with 16p11.2 CNVs are apparently normal.

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Deletion CNVs, because they reduce gene dosage by 50%, are more likely to affect overall function than duplication CNVs, which increase gene dosage by 33.3%. It is worth noting that duplication events appear to be critical in human evolution, where divergence of a duplicated gene from its original allows the formation of genes and proteins with different, but usually related, functions (Zhang, 2003). If a gene copy is deleted, this process cannot happen. This observation sheds an oblique light on why a more severe disorder such as autism is only associated with the deletion CNV. Schizophrenia and bipolar disorder, which have later onset and are usually treatable, can be considered less severe; thus, perhaps the association with the duplication is more likely. However, this does not explain why (a) normal controls are seen with the CNV and (b) why the duplication CNV is also seen in autism. For a complex disease such as autism, it is more likely from current evidence that multiple deleterious variants are required, each contributing a small, and probably clinically undetectable, liability. Thus, normal individuals may harbor the CNV, but because they do not possess other relevant variants, go clinically undetected. Those with an autism diagnosis probably manifest a range of unfavorable variants, perhaps with interacting effects with the 16p11.2 CNV. With this in mind, the original result is perhaps more explicable, and we simultaneously receive a glimpse of the genetic complexity of these disorders.

Need et al. (2009) studied CNVs in 1,013 cases of schizophrenia compared to 1,084 controls. They initially looked at CNVs larger than 2 mb and found eight deletions in the cases and nine duplications in 6 cases and 3 in controls. Large deletions were statistically more likely to occur in cases than controls, but not duplications. They then went on to look for deletion events larger than 2 mb in a further cohort of 1,547 controls screened with an extensive battery of cognitive tests. They did not find any deletions larger than 2 mb in this group, further underlining the association with the schizophrenia cohort. They observed four of the large deletions in the 22q11.2 region, three in the 1q21.1 region, and two new CNVs, one in 16p13.11 (and within which 2 further cases had a smaller CNV), and one on 8p22. They also saw duplications larger than 2 mb in 15q11.2-13.3, extending across the Prader–Willi/Angelman syndrome critical region and the gene *APBA2*, which had previously been reported in another case of schizophrenia by Kirov et al. (2007). They commented that although duplications were also seen in controls, very large duplications (>3 mb) were not, and were segregated entirely with cases, suggesting that although duplications may not be as deleterious as deletions, very large duplication events may be significant in schizophrenia. They also investigated common CNVs, which were arbitrarily defined, using an approach capitalizing on the fact that some SNPs tag CNVs (i.e., a particular SNP allele that segregates with the CNV). They did not find any significant associations in their data.

Levinson et al. (2011) studied 3,945 subjects with schizophrenia or schizoaffective disorder compared to 3,611 screened controls, and compared the rates of rare CNVs between groups. The use of screened controls in this study adds power to the groups' ability to detect an association, because it may be reasonable to expect that controls screened for an absence of medical problems harbor fewer CNVs than cases. The group confirmed previous associations found for deletions at 15q13.3, 1q21.1, and 22q11.2, as well as duplications at 16p11.2. They identified a new association region at 3q29. Seven cases had a deletion in this region, but no controls. This CNV had previously been found in 14 individuals in a sample of 14,698 individuals with mild to moderate mental retardation, and a variety of other phenotypes, including autism (Ballif et al., 2008). Overall, this article provides strong independent support for the role of certain rare CNVs in schizophrenia.

Buizer-Voscamp et al. (2011) investigated CNVs over gene coding regions in 834 Dutch schizophrenia patients and 672 Dutch control subjects screened for an absence of psychiatric symptoms and history. They found that their cases had a similar burden of CNVs (2.1 per sample) to controls. When they stratified their CNVs by type (deletion or duplication) and size (50–500 kb, 500 kb to 1 mb, and ≥1 mb), however, they found significantly more deletion CNVs in cases than controls across size ranges. They also found CNVs overlapping with those previously found in other studies (1q42 and 22q11.2) and a new locus at 5q35.1.

The CNVs discussed so far are large, usually spanning hundreds of thousands or millions of base pairs. The advantage of studying such CNVs is that they are easy to detect with current array technology, and stand out as candidates for disease association. The disadvantage of studying such large variants is that they usually encompass large numbers of genes. One or more than one gene may be relevant to a particular disorder, but without more focused study it is difficult to tell which gene may be playing an etiological role. In this case, smaller CNVs that cover or interrupt individual genes are more informative, although they can be harder to detect.

NRXN1 encodes the presynaptic neuronal cell surface protein that mediates neurotransmission by interacting with neuroligins. Disruptions of this gene impair normal synaptic function (Sudhof, 2008). CNVs in this gene were shown by Kirov et al. (2007) in two siblings with schizophrenia, and an analysis of this gene within a subcohort of schizophrenia patients in 2008 found significant associations between CNVs affecting exons of this gene and schizophrenia (Rujescu et al., 2009). Walsh et al. (2008) also found a deletion of *NRXN1* affecting identical twins with early onset schizophrenia. Marshall et al. (2008) and Weiss et al. (2008), among others, have also linked variants in *NRXN1* to autism (see, e.g., Feng et al., 2006; Kim et al., 2008; Morrow et al., 2008; Szatmari et al., 2007), and a further study has linked this gene to mental retardation (Friedman et al., 2006). *NRXN1* is large, and the CNVs demonstrated in the gene (usually deletions) variously affect different regions. CNVs that disrupt exons (i.e., the protein-coding parts of genes) appear to be most associated with disease, as might be expected, but other CNVs may also be relevant.

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A complicated picture for even a single gene emerges here. *NRXN1* encodes alpha and beta subunit proteins formed from transcription via different promoters, and multiple isoforms of each alpha and beta variant are produced by splicing the messenger RNA. A disrupting CNV within *NRXN1* is likely to disrupt the balance of splice variants produced at different sites in the CNS, and probably subtly affects synaptic neurotransmission, with the extent of disruption being dependent on the precise nature of the CNV involved. In a review of this gene and meta-analysis of association studies Kirov, Rujescu, Ingason, Collier, O'Donovan, and Owen (2009) showed the CNV to be present in 17 out of 8,798 cases (0.19%) but only 17 out of 42,054 controls (0.04%; $p = 1.3 \times 10^{-5}$) with deletions of more than 100 KB even more strongly associated with disease ($p = 3.7 \times 10^{-6}$). Thus, although there is compelling evidence of association, *NRXN1* CNVs only account for a tiny proportion of overall liability to the disorder, and most cases will not exhibit a CNV in this gene. Nonetheless, here is an important small piece of a much larger puzzle.

ADHD

ADHD is a common childhood psychiatric disorder that in a substantial proportion of cases continues into adulthood (Biederman, 1998). It is more often seen in boys and comprises extreme degrees of impulsivity, inattentiveness, and hyperactivity. Although it remains a somewhat controversial diagnosis in some circles, a recent meta-analysis estimated the worldwide pooled prevalence at 5.29% (Polanczyk, de Lima, Horta, Biederman, & Rohde, 2007). The term *hyperkinetic disorder* is sometimes used outside of North America, but the diagnostic criteria are similar (World Health Organisation, 1992). ADHD has substantial heritability estimates from twin and adoption studies of around 76% (Franke, Neale, & Faraone, 2009). However, the genetic etiology remains obscure.

Elia et al. (2010) genotyped 335 cases of ADHD and their parents and 2,026 healthy controls to study CNVs. They detected 222 inherited CNVs in cases that were not present in any of the controls. Twenty-eight covered 22 genes previously implicated in other disorders such as autism and schizophrenia. Eleven CNVs had also previously been associated with autism and schizophrenia. Although in a pathway analysis the authors found an overrepresentation of genes involved in learning, central nervous system development, hindbrain development, and cell adhesion, they found no difference in mean CNV count and CNV size between cases and controls. This suggests that cases of ADHD, although not having a higher CNV burden in general, do have a focused set of CNVs occurring over genes occurring in particular biological pathways.

In a further large study of large, rare CNVs in 410 children with ADHD compared to 1,156 controls, Williams et al. (2010) detected a highly significant difference of 57 CNVs in their case set, equivalent to 15.6% of cases, and 78 in the control set, equivalent to 7.5%. Of particular note in this study was that some children had intellectual disability (IQ $<$ 70). Even when cases with intellectual disability were excluded, however, 12.5% of the cases still harbored a CNV, maintaining statistical significance. The average IQ of children in the case group was 89, perhaps suggesting that more CNVs than this might actually be attributable to general intellectual ability. However, the overlap between different neurocognitive problems (e.g., ADHD) and general intellectual ability (measured by IQ) is particularly hard to measure and may in any event not be a meaningful distinction. This reinforces the notion that CNVs can confer a broadly increased risk of neuropsychiatric illness (that may be partially mediated by general intellectual ability) rather than contributing to any particular disorder on its own.

Lesch et al. (2011), in a smaller and more focused study, considered 99 children and adolescents with severe ADHD. Two-thirds of these children came from families with at least 2 members affected with ADHD, 8 patients were from extended families with high numbers of sufferers of ADHD, and the remainder (24 cases) were sporadic. In contrast to Williams et al. (2010), they excluded any case with an IQ of less than 80 in this study and any case with a comorbid diagnosis of autism or schizophrenia. They found 17 CNVs in their cohort: 4 deletions and 13 duplications. Two CNVs were de novo. Many of the CNVs contained genes that may be associated with ADHD etiology, and the study provides a good basis for more focused research on the neurobiological changes that these CNVs may elicit.

Bipolar Affective Disorder

Bipolar affective disorder occurs in about 1% of the general population, affecting males and females equally, and is characterized by episodic periods of mania and depression (or, less commonly, mania alone) usually accompanied by disturbances in behavior and thinking (World Health Organisation, 1992). In contrast to schizophrenia, where residual deficits are common, recovery is usually complete between episodes (Judd & Schettler, 2010). Most twin and adoption studies indicate a heritability of about 80% and the lifetime risk for the disorder in monozygotic co-twins of a sufferer of bipolar disorder is about 40%–70%, and first-degree relatives about 5%–10% (Craddock & Jones, 1999). SNP GWAS of bipolar disorder have met with some success, with the Psychiatric GWAS Consortium Bipolar Disorder Working Group, in a meta-analysis 11,974 cases of bipolar disorder compared to 51,792 controls, confirming an association with the alpha subunit of the L-type voltage gated calcium channel and a new SNP in an intron of a human homolog of the Drosophila pair-rule gene 10-m (Sklar et al., 2011). Previous studies had also identified SNPs in the neurocan gene (Cichon et al., 2011) and the ankyrin 3 gene (Schulze et al., 2009).

Zhang, Cheng, et al. (2009) compared the frequency of rare CNVs detected in 1,001 bipolar affective disorder cases against 1,034 controls. Looking specifically at singleton dele-

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tions (deletion CNVs occurring only once in the entire sample) they found that 16.2% of cases contained a singleton deletion CNV compared to 12.3% in controls. Some singleton CNVs covered genes already implicated in schizophrenia, specifically the metabotropic glutamate receptor 7 gene and *LARGE*. Bipolar disorder and schizophrenia are considered to be distinct yet overlapping disorders, and so this is an interesting and plausible finding. The metabotropic glutamate receptor 7 gene encodes a metabotropic glutamate receptor expressed in the brain, and *LARGE* encodes a gene that has previously been associated with mental retardation (congenital muscular dystrophy type 1D gene; Longman, 2003).

Singleton CNVs are an interesting group of variants, and a logical group to focus on. They are defined as occurring only once in a data set, and they thus are more likely to represent variants that occur rarely in a population. Rarely occurring variants are more likely to be functional (i.e., they have some sort of effect, usually negative, on the organism) simply because their rarity suggests that evolution is placing them under negative selection pressure.

Grozeva et al. (2010) have also studied CNVs in a sample of 1,697 bipolar cases compared with 2,806 unscreened controls. In contrast to Zhang, Cheng, et al. (2009), they found significantly *fewer* deletion CNVs in the cases of bipolar disorder than controls. However, it is notable that the control group in this study is unscreened whereas Zhang, Cheng, et al. screened their control group for an absence of neuropsychiatric disorder. The two studies are difficult to compare in this context.

McQuillin et al. (2011) focused on 546 cases of bipolar disorder compared to 517 controls screened for lifetime absence of psychiatric disorder. Similar to Grozeva et al. (2010), but in contrast to Zhang, Cheng, et al. (2009) they found no evidence to suggest an increased burden of rare CNVs, including singleton CNVs. They also found that, overall, bipolar cases tended to have a lower burden of CNVs compared to, in this case, screened controls. Nevertheless, they found instances of CNVs previously seen in other CNV association studies. Five cases had a CNV on chromosome 19 covering a variety of zinc finger domain containing proteins that had previously been identified by the International Schizophrenia Consortium (2008). Three cases had a CNV on chromosome 1 identified by Zhang, Cheng, et al. (2009); 4 cases had CNVs on chromosome 10 that had also been seen by Grozeva et al. (2010).

Priebe et al. (2011) studied 882 cases of bipolar disorder compared to 872 population controls. They found that the frequency of samples with rare duplications and the average total length of singleton deletions was significantly increased in cases with an age of onset of disorder of $\leq$21 years when compared to controls. They also found evidence for a significant overrepresentation in cases of two common CNV regions in 10q11 and 6q27. Comparing the frequency of CNVs in regions previously implicated in psychiatric disorders, they did not find a significant increase when cases were compared to controls in any region. In contrast, like many studies, the rarity of the CNVs themselves suggests a lack of power to detect an association.

Malhotra et al. (2011) recently published a study looking at a cohort of 788 trios, including 185 with bipolar diagnoses and 177 with schizophrenia diagnoses. They found that the frequency of de novo CNVs was significantly higher in the bipolar diagnosis cohort, with an odds ratio of 4.8, and a particular enrichment in those with a younger age of onset ($<$18). Within the schizophrenia cases, a similar picture for de novo CNVs was also observed.

Unipolar Depressive Disorder

Unipolar depressive disorder is a variously estimated to have a prevalence of between 4.4% (Weissman et al., 1988) and 17.1% (Kessler et al., 1994), with estimates depending on the severity cutoff used to define the disorder. Whatever criteria are used, unipolar depression has one of the highest morbidity burdens of any physical or mental disorder (World Health Organisation, n.d.). An episode is usually defined by a pervasive lowering of mood, reduction of energy, and decrease in activity with a reduced capacity for enjoyment, interest, and concentration lasting for at least 2 weeks (World Health Organisation, 1992). Biological symptoms including sleep and appetite disturbance are commonly seen. The above symptoms are almost always associated with a lowering of self-esteem and self-confidence in conjunction with ideas of guilt and unworthiness. Although a meta-analysis of depressive disorder estimated the heritability to be 37% (Sullivan et al., 2000), this figure increases to approximately 70% when samples with recurrent and severe forms of the disease are analyzed (Kendler, Neale, Kessler, Heath, & Eaves, 1992; McGuffin et al., 1996). Like schizophrenia, a gradation of risk is seen as genetic distance from the proband increases, with monozygotic co-twins of a sufferer having a risk of about 50% of also suffering from the disorder (Craddock & Forty, 2006). Although three linkage studies of depressive disorder all implicated a region of chromosome 15 (Camp et al., 2005; Holmans et al., 2004; McGuffin et al., 2005), no genome-wide association studies of the disorder have yet generated a replicated locus that survived correction for multiple testing (Lewis et al., 2010; Muglia et al., 2010; Rietschel et al., 2010; Shi et al., 2011; Sullivan et al., 2000; Terracciano et al., 2010; Wray et al., 2012).

Within the sphere of CNVs, three studies have been reported. Glessner et al. (2010) analyzed 1,693 cases of depression and 4,506 controls genotyped on the Perlegen 600k platform. They found significant association between cases and a duplication at the SLIT1 gene on chromosome 5. This study was, unfortunately, hampered by the microarray platform that, because the polymerase chain reaction in the assay is run to saturation, tends to have a high signal-to-noise ratios that make calling CNVs difficult.

Rucker et al. analyzed large, rare CNVs in 3,106 cases of recurrent depression, 459 controls screened for lifetime-absence of psychiatric disorder, and 5,619 unscreened popula-

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tion controls from Phase 2 of the Wellcome Trust Case Control Consortium (Rucker et al., 2011). Within these groups, they found an ordered increase in the frequency of samples with deletion CNVs, but not duplication CNVs, with the screened control sample showing the lowest frequency of deletion CNVs, cases of depression the highest frequency, and unscreened controls falling in the middle. Further analysis showed that CNVs deleting protein coding regions of the genome were largely responsible for the association, and, within an analysis of regions previously implicated in schizophrenia, there was an overall enrichment of CNVs in cases when compared to the screened controls, although no individual area was significant. One individual was found to harbor the 22q11.2 deletion and five individuals had CNVs at 16p11.2. Although this study highlights that part of the etiology of recurrent depression is likely to involve rare CNVs, as it does in autism and schizophrenia, it also suggested that screened control groups may have a particularly low frequency of deletion CNVs. That a relatively uniform frequency of duplication CNVs was observed throughout the cohorts within this study may suggest that duplication CNVs are less likely to be pathogenic.

Degenhardt et al. (2012) genotyped 604 cases of major depressive disorder and 1,643 controls on Illumina SNP arrays. They found evidence that CNVs in four regions of the genome, 7p21.3, 15q26.3, 16p11.2, and 18p11.32, were associated with their depressed cases. Reciprocal CNVs at 16p11.2 were already discussed in this article, which are associated with many different psychiatric disorders, and were observed in the CNV study of depressive disorder by Rucker et al. (2011). The statistically significant association of this particular CNV with depressive disorder is novel, however.

Common CNVs

Common CNVs, which are usually called copy number polymorphisms (CNPs), are defined variably by different groups, but usually as occurring in more than 1%, 5%, or 10% of the population (Conrad et al., 2010; Craddock et al., 2010; Grozeva et al., 2010; McQuillin et al., 2011; Rucker et al., 2011). Depending on the definition used, they account for a varying but significant proportion of variation within the genome, but have not been as intensively studied as rare and de novo CNVs, probably because they are intuitively less attractive candidates for explaining disease susceptibility. Some groups have argued that CNPs may in fact account for most of the susceptibility to complex diseases (Risch & Merikangas, 1996). Others propose that both CNPs of small effect and rarer CNVs of larger effect play interacting roles (Uher, 2009).

Previous work in genome-wide association studies using SNPs observed that most CNPs are "tagged" by SNPs (Craddock et al., 2010). In other words, the presence of a CNP can be inferred if a particular allele at a particular SNP is seen. Using this observation, Conrad et al. (2010) were able to analyze the origins and impact of CNPs and show that, at least with current methods for analysis and detection, that they are unlikely to contribute to a substantial proportion of heritability in neuropsychiatric disorders. This is not a surprising result, because no particular form of genetic variation has yet been shown to substantially contribute to complex disease susceptibility. However, it does place CNPs in context and inform us that, as with CNVs, SNPs, and other forms of genetic variation, our expectations should be modest.

Conclusion

Multiple lines of evidence exist that CNVs, especially those that are rare and de novo, contribute to susceptibility for complex psychiatric diseases such as schizophrenia and ADHD, and possibly, to a lesser extent, bipolar disorder and unipolar depressive disorder. Most data sets in existence have been collected based on the aggregation of groups of individuals who share similar diagnoses or characteristics. In this context, the etiological association of CNV with disease state is natural, but it runs the risk of oversimplifying a complex picture. Nature is unlikely to respect our own phenotypic delineations, and this appears to be the case at this level of genetic variation. It may be more reasonable to conceive from the evidence so far that rare and de novo CNVs are associated with a general liability to psychiatric and other outcomes rather than specific diseases. Large, rare deletion CNVs may be associated with more severe disorders than duplications, but even here the spectrum of phenotypic variation is surprisingly wide, suggesting an interacting concert of genetic variants in addition to those observed.

De novo events may partly explain some cases of sporadic disease occurring in families with no prior history, although transmissible events may also contribute to disease running through families. The observation of similar variants in the normal parents of affected probands reinforces the concept that CNVs have mostly low penetrance values for individual disorders, and lends evidence to a "multiple-hit" hypothesis of etiology.

Even with these new findings, in a field with a modest number of replicated genetic associations, the proportion of heritability accounted for by CNVs is likely to be low. Genetic association studies are currently underpowered to detect interaction effects or the effects of variants with a small effect size, although large consortiums of collaborating research centers may help with this problem (Cichon et al., 2009). Untangling the interacting effects of different genetic variants is likely to be particularly tricky, but could pave the way for a new etiological model for susceptibility to psychiatric disorders. This is much cause for celebration in a field with, hitherto, a lack of replicated evidence to explain the large heritability estimates from twin and adoption studies. We must add, however, a cautionary note. The human brain is the least amenable organ to study, and research in the genetics of mental disorders is currently far removed from clinical reality. Psychiatric diagnoses in particular are notable for their lack of validating objective biomarkers and, more so in the past,

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have been notorious for their lack of reliability. What is abundantly clear, in a field where many research findings of the past have not stood the test of time, is that the gap between a genetic variant and observed behavior is vast and we should be cautious about inferring causation from association.

Are we yet at a stage where knowledge about CNVs may be safely translated into the clinic? Although CNVs are clearly associated with human disease states, they also represent normal genetic variability within the population (the mechanisms of CNV formation are most probably an essential driver of normal species evolution). Separating benign from pathogenic CNVs is difficult, especially when diagnostic boundaries are blurred and clinical measurements of neurocognitive function are difficult to make. Nonetheless, from the evidence presented thus far, some CNVs are clearly associated with disorders such as schizophrenia. If a deleterious variant is detected, then there are implications for genetic counseling and family planning, because such variants may or may not be passed on to offspring, mediated largely by chance. Thus, although the detection of a variant may not directly benefit the patient, it may benefit future generations.

Could it be possible to predict development of disease by screening for CNVs? The answer to this is almost certainly "no." Frequently observed in the studies contributing to this article are apparently normal individuals harboring supposedly pathogenic CNVs. Even in individuals with the 22q11.2 deletion, currently associated with the highest risk of development of psychiatric disorders of any CNV, only a minority will go on to develop schizophrenia. Vassos et al. (2010) analyzed the penetrance of variants associated with schizophrenia occurring in the genomic regions 15q13.3, 1q21.1, 15q11.2, 17p12, 2p16.3, 16p13.1, and 16p11.2. Estimates for these variants, themselves already having been identified as significantly associated with schizophrenia in various different studies (Ingason et al., 2011; International Schizophrenia Consortium, 2008; Kirov et al., 2007; Kirov, Grozeva, et al., 2009; McCarthy et al., 2009; Rujescu et al., 2009; Stefansson et al., 2008; Vrijenhoek et al., 2008), ranged between 2% and 9%. In other words, 91%–98% of individuals with one of these variants will *not* go on to develop schizophrenia. Although they may go on to develop another disorder (the penetrance level for "any disorder" is probably higher than for individual disorders alone) the levels of penetrance seen are likely to be much below levels seen in Mendelian disorders (Vassos et al., 2010).

Taken together this suggests that screening for CNVs is unlikely to be informative. For those who are free of disease and do not harbor a CNV, there will be no reassurance that they will not go on to develop disease. Likewise for those who are free of disease but found to harbor a CNV, there is no certainty that they would go on to develop it; indeed, the likelihood is that they would not. The limitations of screening are starkly seen with these examples. Even if strong interactional effects are found between individual CNVs or between CNVs and other forms of genetic variation, clinical tests for diagnosis and prediction demand high sensitivities and specificities, which are unlikely to be achieved in the near future.

What of treatment? Although no study has demonstrated that those harboring CNVs show any differences in terms of treatment outcome, relatively few studies have been designed to search for such effects. The study of CNVs and the proteins produced by the genes that they involve may lead to new, targeted therapies in the future. This picture has already become a clinical reality in cancer therapeutics for some individuals. In psychiatry, however, many barriers exist for a similar aspiration to be realized.

CNVs represent the first replicated evidence in the search for the genetic etiology of psychiatric disorders. Disorders with high heritability scores appear to be particularly associated with CNVs, as would be expected. More research is required to clarify the contribution of CNVs to different disorders. Even so, from a broad perspective, it is unlikely that CNVs will account for a large amount of unexplained heritability, which will probably be filled by other genetic variants, interaction effects, epigenetic effects, and as yet unknown factors. Large, rare CNVs represent the tip of the etiological iceberg. Taken together, the evidence strongly argues for a polygenic, complex model of disease of the type Gottesman and Shields (1967, based on the work of Falconer (1965), argued for over 40 years ago. The effect of CNVs with smaller effect sizes, and particularly the interaction effects between different CNVs, have yet to be defined and may require novel research and analysis strategies.

References

American Psychiatric Association. (1994). *Diagnostic and statistical manual of mental disorders* (4th ed., text revision). Arlington, VA: Author.

Andreasen, N. (1995). Symptoms, signs, and diagnosis of schizophrenia. *Lancet*, *346*(8973), 477–481.

Babatz, T. D., Kumar, R. A., Sudi, J., Dobyns, W. B., & Christian, S. L. (2009). Copy number and sequence variants implicate *APBA2* as an autism candidate gene. *Autism Research*, *2*, 359–364.

Bailey, A., Le Couteur, A., Gottesman, I., Bolton, P., Simonoff, E., Yuzda, E., et al. (1995). Autism as a strongly genetic disorder: evidence from a British twin study. *Psychological Medicine*, *25*, 63–77.

Ballif, B. C., Theisen, A., Coppinger, J., Gowans, G. C., Hersh, J. H., Madan-Khetarpal, S., et al. (2008). Expanding the clinical phenotype of the 3q29 microdeletion syndrome and characterization of the reciprocal microduplication. *Molecular Cytogenetics*, *1*, 8.

Biederer, T., & Südhof, T. C. (2000). Mints as adaptors. Direct binding to neurexins and recruitment of munc18. *Journal of Biological Chemistry*, *275*, 39803–39806.

Biederman, J. (1998). Attention-deficit/hyperactivity disorder: A life-span perspective. *Journal of Clinical Psychiatry*, *59*(S7), 4–16.

Brzustowicz, L. M., Hodgkinson, K. A., Chow, E. W., Honer, W. G., & Bassett, A. S. (2000). Location of a major susceptibility locus for familial schizophrenia on chromosome 1q21-q22. *Science*, *288*(5466), 678–682.

Buizer-Voskamp, J. E., Muntjewerff, J.-W., Genetic Risk and Outcome in Psychosis (GROUP) Consortium Members Strengman, E., Sabatti, C., Stefansson, H., et al. (2011). Genome-wide analysis shows increased frequency of copy number variation deletions in Dutch schizophrenia patients. *Biological Psychiatry*, *70*, 655–662.

https://doi.org/10.1017/S0954579412000740 Published online by Cambridge University Press

Camp, N., Lowry, M., Richards, R., Plenk, A., Carter, C., Hensel, C., et al. (2005). Genome-wide linkage analyses of extended Utah pedigrees identifies loci that influence recurrent, early-onset major depression and anxiety disorders. *American Journal of Medical Genetics: Part B Neuropsychiatric Genetics*, *135B*, 85–93.

Cardno, A. G., & Gottesman, I. I. (2000). Twin studies of schizophrenia: From bow-and-arrow concordances to Star Wars Mx and functional genomics. *American Journal of Medical Genetics*, *97*, 12–17.

Cichon, S., Craddock, N., Daly, M., Faraone, S., Gejman, P., Kelsoe, J., et al. (2009). Genomewide association studies: history, rationale, and prospects for psychiatric disorders. *American Journal of Psychiatry*, *166*, 540–556.

Cichon, S., Mühleisen, T. W., Degenhardt, F. A., Mattheisen, M., Miró, X., Strohmaier, J., et al. (2011). Genome-wide association study identifies genetic variation in neurocan as a susceptibility factor for bipolar disorder. *American Journal of Medical Genetics*, *88*, 372–381.

Conrad, D. F., Pinto, D., Redon, R., Feuk, L., Gokcumen, O., Zhang, Y., et al. (2010). Origins and functional impact of copy number variation in the human genome. *Nature*, *464*(7289), 704–712.

Craddock, N., & Forty, L. (2006). Genetics of affective (mood) disorders. *European Journal of Human Genetics*, *14*, 660–668.

Craddock, N., & Jones, I. (1999). Genetics of bipolar disorder. *Journal of Medical Genetics*, *36*, 585–594.

Craddock, N., Hurles, M. E., Cardin, N., Pearson, R. D., Plagnol, V., Robson, S., et al. (2010). Genome-wide association study of CNVs in 16,000 cases of eight common diseases and 3,000 shared controls. *Nature*, *464*(7289), 713–720.

Degenhardt, F., Priebe, L., Herms, S., Mattheisen, M., Mühleisen, T. W., Meier, S., et al. (2012). Association between copy number variants in 16p11.2 and major depressive disorder in a German case-control sample. *American Journal of Medical Genetics: Part B Neuropsychiatric Genetics*, *159B*, 263–273.

Dulubova, I., Khvotchev, M., Liu, S., Huryeva, I., Südhof, T. C., & Rizo, J. (2007). Munc18-1 binds directly to the neuronal SNARE complex. *Proceedings of the National Academy of Sciences*, *104*, 2697–2702.

Elia, J., Gai, X., Xie, H. M., Perin, J. C., Geiger, E., Glessner, J. T., et al. (2010). Rare structural variants found in attention-deficit hyperactivity disorder are preferentially associated with neurodevelopmental genes. *Molecular Psychiatry*, *15*, 637–646.

Falconer, D. (1965). The inheritance of liability to certain diseases, estimated from the incidence among relatives. *Annals of Human Genetics*, *29* 51–76.

Feng, J., Schroer, R., Yan, J., Song, W., Yang, C., Bockholt, A., et al. (2006). High frequency of neurexin 1β signal peptide structural variants in patients with autism. *Neuroscience Letters*, *409*, 10–13.

Feuk, L., Carson, A., & Scherer, S. (2006). Structural variation in the human genome. *Nature Reviews Genetics*, *7*, 85–97.

Franke, B., Neale, B. M., & Faraone, S. V. (2009). Genome-wide association studies in ADHD. *Human Genetics*, *126*, 13–50.

Freeman, J. L., Perry, G. H., Feuk, L., Redon, R., McCarroll, S. A., Altshuler, D. M., et al. (2006). Copy number variation: new insights in genome diversity. *Genome Research*, *16*, 949–961.

Friedman, J. M., Baross, A., Delaney, A. D., Ally, A., Arbour, L., Asano, J., et al. (2006). Oligonucleotide microarray analysis of genomic imbalance in children with mental retardation. *American Journal of Medical Genetics*, *79*, 500–513.

Glessner, J., Wang, K., Sleiman, P., Zhang, H., Kim, C., Flory, J., et al. (2010). Duplication of the SLIT3 locus on 5q35.1 predisposes to major depressive disorder. *PLoS One*, *5*, e15463.

Gottesman, I. I. (1991). *Schizophrenia genesis: The origins of madness*. New York: W. H. Freeman.

Gottesman, I., & Shields, J. (1967). A polygenic theory of schizophrenia. *Proceedings of the National Academy of Sciences*, *58*, 199–205.

Grozeva, D., Kirov, G., Ivanov, D., Jones, I., Jones, L., Green, E., et al. (2010). Rare copy number variants: A point of rarity in genetic risk for bipolar disorder and schizophrenia. *Archives of General Psychiatry*, *67*, 318–327.

Guilmatre, A., Dubourg, C., Mosca, A. L., Legallic, S., Goldenberg, A., Drouin-Garraud, V., et al. (2009). Recurrent rearrangements in synaptic and neurodevelopmental genes and shared biologic pathways in schizophrenia, autism, and mental retardation. *Archives of General Psychiatry*, *66*, 947–956.

Harris, E., & Barraclough, B. (1998). Excess mortality of mental disorder. *British Journal of Psychiatry*, *173* 11–53.

Holmans, P., Zubenko, G., Crowe, R., Depaulo, J. J., Scheftner, W., Weissman, M., et al. (2004). Genomewide significant linkage to recurrent, early-onset major depressive disorder on chromosome 15q. *American Journal of Medical Genetics*, *74*, 1154–1167.

Iafrate, A., Feuk, L., Rivera, M., Listewnik, M., Donahoe, P., Qi, Y., et al. (2004). Detection of large-scale variation in the human genome. *Nature Genetics*, *36*, 949–951.

Ingason, A., Rujescu, D., Cichon, S., Sigurdsson, E., Sigmundsson, T., Pietilainen, O. P. H., et al. (2011). Copy number variations of chromosome 16p13.1 region associated with schizophrenia. *Molecular Psychiatry*, *16*, 17–25.

International Schizophrenia Consortium. (2008). Rare chromosomal deletions and duplications increase risk of schizophrenia. *Nature*, *455*(7210), 237–241.

Judd, L. L., & Schettler, P. J. (2010). The long term course and clinical management of bipolar I and bipolar II disorders. In L. N. Yatham & M. Maj (Eds.), *Bipolar disorder: Clinical and neurobiological foundations*. Hoboken, NJ: Wiley.

Karayiorgou, M., Simon, T. J., & Gogos, J. A. (2010). 22q11.2 microdeletions: Linking DNA structural variation to brain dysfunction and schizophrenia. *Nature Reviews Neuroscience*, *11*, 402–416.

Kendler, K. S., Neale, M. C., Kessler, R. C., Heath, A. C., & Eaves, L. J. (1992). A population-based twin study of major depression in women: The impact of varying definitions of illness. *Archives of General Psychiatry*, *49*, 257–266.

Kessler, R. C., McGonagle, K. A., Zhao, S., Nelson, C. B., Hughes, M., Eshleman, S., et al. (1994). Lifetime and 12-month prevalence of *DSM-III-R* psychiatric disorders in the United States: Results from the National Comorbidity Survey. *Archives of General Psychiatry*, *51*, 8.

Kim, H.-G., Kishikawa, S., Higgins, A. W., Seong, I.-S., Donovan, D. J., Shen, Y., et al. (2008). Disruption of neurexin 1 associated with autism spectrum disorder. *American Journal of Medical Genetics*, *82*, 199–207.

Kirov, G., Grozeva, D., Norton, N., Ivanov, D., Mantripragada, K., Holmans, P., et al. (2009). Support for the involvement of large copy number variants in the pathogenesis of schizophrenia. *Human Molecular Genetics*, *18*, 1497–1503.

Kirov, G., Gumus, D., Chen, W., Norton, N., Georgieva, L., Sari, M., et al. (2007). Comparative genome hybridization suggests a role for *NRXN1* and *APBA2* in schizophrenia. *Human Molecular Genetics*, *17*, 458–465.

Kirov, G., Rujescu, D., Ingason, A., Collier, D. A., O'Donovan, M. C., & Owen, M. J. (2009). Neurexin 1 (*NRXN1*) deletions in schizophrenia. *Schizophrenia Bulletin*, *35*, 851–854.

Lander, E., Linton, L., Birren, B., Nusbaum, C., Zody, M., Baldwin, J., et al. (2001). Initial sequencing and analysis of the human genome. *Nature*, *409*(6822), 860–921.

Lesch, K.-P., Selch, S., Renner, T. J., Jacob, C., Nguyen, T. T., Hahn, T., et al. (2011). Genome-wide copy number variation analysis in attention-deficit/hyperactivity disorder: Association with neuropeptide Y gene dosage in an extended pedigree. *Molecular Psychiatry*, *16*, 491–503.

Levinson, D. F., Duan, J., Oh, S., Wang, K., Sanders, A. R., Shi, J., et al. (2011). Copy number variants in schizophrenia: Confirmation of five previous findings and new evidence for 3q29 microdeletions and VIPR2 duplications. *American Journal of Psychiatry*, *168*, 302–316.

Lewis, C., Ng, M., Butler, A., Cohen-Woods, S., Uher, R., Pirlo, K., et al. (2010). Genome-wide association study of major recurrent depression in the U.K. population. *American Journal of Psychiatry*, *167*, 949–957.

Locke, D., Segraves, R., Carbone, L., Archidiacono, N., Albertson, D., Pinkel, D., et al. (2003). Large-scale variation among human and great ape genomes determined by array comparative genomic hybridization. *Genome Research*, *13*, 347–357.

Longman, C. (2003). Mutations in the human *LARGE* gene cause *MDC1D*, a novel form of congenital muscular dystrophy with severe mental retardation and abnormal glycosylation of alpha-dystroglycan. *Human Molecular Genetics*, *12*, 2853–2861.

Lupski, J. (2007). An evolution revolution provides further revelation. *Bioessays*, *29*, 1182–1184.

Malhotra, D., McCarthy, S., Michaelson, J. J., Vacic, V., Burdick, K. E., Yoon, S., et al. (2011). High frequencies of de novo CNVs in bipolar disorder and schizophrenia. *Neuron*, *72*, 951–963.

Marshall, C., Noor, A., Vincent, J., Lionel, A., Feuk, L., Skaug, J., et al. (2008). Structural variation of chromosomes in autism spectrum disorder. *American Journal of Medical Genetics*, *82*, 477–488.

McCarroll, S. A., Hadnott, T. N., Perry, G. H., Sabeti, P. C., Zody, M. C., Barrett, J. C., et al. (2006). Common deletion polymorphisms in the human genome. *Nature Genetics*, *38*, 86–92.

https://doi.org/10.1017/S0954579412000740 Published online by Cambridge University Press

McCarthy, S. E., Makarov, V., Kirov, G., Addington, A. M., McClellan, J., Yoon, S., et al. (2009). Microduplications of 16p11.2 are associated with schizophrenia. *Nature Genetics*, *41*, 1223–1227.

McGuffin, P., Katz, R., Watkins, S., & Rutherford, J. (1996). A hospital-based twin register of the heritability of DSM-IV unipolar depression. *Archives of General Psychiatry*, *53*, 129–136.

McGuffin, P., Knight, J., Breen, G., Brewster, S., Boyd, P. R., Craddock, N., et al. (2005). Whole genome linkage scan of recurrent depressive disorder from the depression network study. *Human Molecular Genetics*, *14*, 3337–3345.

McGuffin, P., Rijsdijk, F., Andrew, M., Sham, P., Katz, R., & Cardno, A. (2003). The heritability of bipolar affective disorder and the genetic relationship to unipolar depression. *Archives of General Psychiatry*, *60*, 497–502.

McQuillin, A., Bass, N., Anjorin, A., Lawrence, J., Kandaswamy, R., Lydall, G., et al. (2011). Analysis of genetic deletions and duplications in the University College London bipolar disorder case control sample. *European Journal of Human Genetics*, *19*, 588–592.

Morrow, E. M., Yoo, S.-Y., Flavell, S. W., Kim, T.-K., Lin, Y., Hill, R. S., et al. (2008). Identifying autism loci and genes by tracing recent shared ancestry. *Science*, *321*(5886), 218–223.

Muglia, P., Tozzi, F., Galwey, N. W., Francks, C., Upmanyu, R., Kong, X. Q., et al. (2010). Genome-wide association study of recurrent major depressive disorder in two European case-control cohorts. *Molecular Psychiatry*, *15*, 589–601.

Murphy, K., Jones, L., & Owen, M. (1999). High rates of schizophrenia in adults with velo-cardio-facial syndrome. *Archives of General Psychiatry*, *56*, 940–945.

Need, A., Ge, D., Weale, M., Maia, J., Feng, S., Heinzen, E., et al. (2009). A genome-wide investigation of SNPs and CNVs in schizophrenia. *PLoS Genetics*, *5*, e1000373.

Perry, G. H., Ben-Dor, A., Tsalenko, A., Sampas, N., Rodriguez-Revenga, L., Tran, C. W., et al. (2008). The fine-scale and complex architecture of human copy-number variation. *American Journal of Medical Genetics*, *82*, 685–695.

Pinto, D., Pagnamenta, A. T., Klei, L., Anney, R., Merico, D., Regan, R., et al. (2010). Functional impact of global rare copy number variation in autism spectrum disorders. *Nature*, *466*(7304), 368–372.

Polanczyk, G., de Lima, M. S., Horta, B. L., Biederman, J., & Rohde, L. A. (2007). The worldwide prevalence of ADHD: A systematic review and metaregression analysis. *American Journal of Psychiatry*, *164*, 942–948.

Priebe, L., Degenhardt, F. A., Herms, S., Haenisch, B., Mattheisen, M., Nieratschker, V., et al. (2011). Genome-wide survey implicates the influence of copy number variants (CNVs) in the development of early-onset bipolar disorder. *Molecular Psychiatry*, 1–12.

Rietschel, M., Mattheisen, M., Frank, J., Treutlein, J., Degenhardt, F., Breuer, R., et al. (2010). Genome-wide association-, replication-, and neuroimaging study implicates HOMER1 in the etiology of major depression. *Biological Psychiatry*, *68*, 578–585.

Risch, N., & Merikangas, K. (1996). The future of genetic studies of complex human diseases. *Science*, *273*(5281), 1516–1517.

Robin, N., & Shprintzen, R. (2005). Defining the clinical spectrum of deletion 22q11.2. *Journal of Pediatrics*, *147*, 90–96.

Rucker, J. J. H., Breen, G., Pinto, D., Pedroso, I., Lewis, C. M., Cohen-Woods, S., et al. (2011). Genome-wide association analysis of copy number variation in recurrent depressive disorder. *Molecular Psychiatry.* Advance online publication. doi:10.1038/mp.2011.144

Rujescu, D., Ingason, A., Cichon, S., Pietiläinen, O. P. H., Barnes, M. R., Toulopoulou, T., et al. (2009). Disruption of the neurexin 1 gene is associated with schizophrenia. *Human Molecular Genetics*, *18*, 988–996.

Schena, M., Shalon, D., Davis, R., & Brown, P. (1995). Quantitative monitoring of gene expression patterns with a complementary DNA microarray. *Science*, *270*(5235), 467–470.

Schulze, T. G., Detera-Wadleigh, S. D., Akula, N., Gupta, A., Kassem, L., Steele, J., et al. (2009). Two variants in Ankyrin 3 (*ANK3*) are independent genetic risk factors for bipolar disorder. *Molecular Psychiatry*, *14*, 487–491.

Schwab, S. G., & Wildenauer, D. B. (2009). Update on key previously proposed candidate genes for schizophrenia. *Current Opinion in Psychiatry*, *22*, 147–153.

Sebat, J., Lakshmi, B., Troge, J., Alexander, J., Young, J., Lundin, P., et al. (2004). Large-scale copy number polymorphism in the human genome. *Science*, *305*(5683), 525–528.

Sharp, A., Mefford, H., Li, K., Baker, C., Skinner, C., Stevenson, R., et al. (2008). A recurrent 15q13.3 microdeletion syndrome associated with mental retardation and seizures. *Nature Genetics*, *40*, 322–328.

Shi, J., Potash, J. B., Knowles, J. A., Weissman, M. M., Coryell, W., Scheftner, W. A., et al. (2011). Genome-wide association study of recurrent early-onset major depressive disorder. *Molecular Psychiatry*, *16*, 193–201.

Sklar, P., Ripke, S., Scott, L. J., Andreassen, O. A., Cichon, S., Craddock, N., et al. (2011). Large-scale genome-wide association analysis of bipolar disorder identifies a new susceptibility locus near *ODZ4*. *Nature Genetics*, *43*, 977–983.

Stefansson, H., Rujescu, D., Cichon, S., Pietilainen, O., Ingason, A., Steinberg, S., et al. (2008). Large recurrent microdeletions associated with schizophrenia. *Nature*, *455*(7210), 232–236.

Sudhof, T. (2008). Neuroligins and neurexins link synaptic function to cognitive disease. *Nature*, *455*(7215), 903–911.

Sullivan, P. F. (2005). The genetics of schizophrenia. *PLoS Medicine*, *2*, e212.

Sullivan, P. F., Neale, M. C., & Kendler, K. S. (2000). Genetic epidemiology of major depression: review and meta-analysis. *American Journal of Psychiatry*, *157*, 1552–1562.

Sullivan, P., Kendler, K., & Neale, M. (2003). Schizophrenia as a complex trait: Evidence from a meta-analysis of twin studies. *Archives of General Psychiatry*, *60*, 1187–1192.

Szatmari, P., Paterson, A., Zwaigenbaum, L., Roberts, W., Brian, J., Liu, X., et al. (2007). Mapping autism risk loci using genetic linkage and chromosomal rearrangements. *Nature Genetics*, *39*, 319–328.

Terracciano, A., Sanna, S., Uda, M., Deiana, B., Usala, G., Busonero, F., et al. (2010). Genome-wide association scan for five major dimensions of personality. *Molecular Psychiatry*, *15*, 647–656.

Uher, R. (2009). The role of genetic variation in the causation of mental illness: An evolution-informed framework. *Molecular Psychiatry*, *14* 1072–1082.

Vassos, E., Collier, D. A., Holden, S., Patch, C., Rujescu, D., St Clair, D., et al. (2010). Penetrance for copy number variants associated with schizophrenia. *Human Molecular Genetics*, *19*, 3477–3481.

Vrijenhoek, T., Buizervoskamp, J., Vanderstelt, I., Strengman, E., Sabatti, C., Geurstvankessel, A., et al. (2008). Recurrent CNVs disrupt three candidate genes in schizophrenia patients. *American Journal of Medical Genetics*, *83*, 504–510.

Walsh, T., McClellan, J., McCarthy, S., Addington, A., Pierce, S., Cooper, G., et al. (2008). Rare structural variants disrupt multiple genes in neurodevelopmental pathways in schizophrenia. *Science*, *320*(5875), 539–543.

Weiss, L. A., Shen, Y., Korn, J. M., Arking, D. E., Miller, D. T., Fossdal, R., et al. (2008). Association between microdeletion and microduplication at 16p11.2 and autism. *New England Journal of Medicine*, *358*, 667–675.

Weissman, M. M., Leaf, P. J., Tischler, G. L., Blazer, D. G., Karno, M., Bruce, M. L., et al. (1988). Affective disorders in five United States communities. *Psychological Medicine*, *18*, 141–153.

Williams, N. M., Zaharieva, I., Martin, A., Langley, K., Mantripragada, K., Fossdal, R., et al. (2010). Rare chromosomal deletions and duplications in attention-deficit hyperactivity disorder: A genome-wide analysis. *Lancet*, *376*(9750), 1401–1408.

World Health Organisation. (1992). *The ICD-10 for Mental and Behavioural Disorders: Clinical descriptions and diagnostic guidelines*. Geneva: Author.

World Health Organisation. (n.d.). *Depression*. Retrieved November 8, 2011, from http://www.who.int/mental_health/management/depression/definition/en/

Wray, N. R., Pergadia, M. L., Blackwood, D. H. R., Penninx, B. W. J. H., Gordon, S. D., Nyholt, D. R., et al. (2012). Genome-wide association study of major depressive disorder: New results, meta-analysis, and lessons learned. *Molecular Psychiatry*, *17*, 36–48.

Xu, B., Roos, J. L., Levy, S., van Rensburg, E. J., Gogos, J. A., & Karayiorgou, M. (2008). Strong association of de novo copy number mutations with sporadic schizophrenia. *Nature Genetics*, *40*, 880–885.

Zhang, D., Cheng, L., Qian, Y., Alliey-Rodriguez, N., Kelsoe, J., Greenwood, T., et al. (2009). Singleton deletions throughout the genome increase risk of bipolar disorder. *Molecular Psychiatry*, *14*, 376–380.

Zhang, F., Gu, W., Hurles, M., & Lupski, J. (2009). Copy number variation in human health, disease, and evolution. *Annual Reviews in Genomics and Human Genetics*, *10* 451–481.

Zhang, J. (2003). Evolution by gene duplication: An update. *Trends in Ecology and Evolution*, *18*, 292–298.

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