The Impact of Copy Number Deletions on General Cognitive Ability and Ventricle Size in Patients with Schizophrenia and Healthy Control Subjects

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Background: General cognitive ability is usually lower in individuals with schizophrenia, partly due to genetic influences. However, the specific genetic features related to general cognitive ability are poorly understood. Individual variation in a specific type of mutation, uncommon genetic deletions, has recently been linked with both general cognitive ability and risk for schizophrenia.

Methods: We derived measures of the aggregate number of "uncommon" deletions (i.e., those occurring in 3% or less of our combined samples) and the total number of base pairs affected by these deletions in individuals with schizophrenia ($n = 79$) and healthy control subjects ($n = 110$) and related each measure to the first principal component of a large battery of cognitive tests, a common technique for characterizing general cognitive ability. These two measures of mutation load were also evaluated for relationships with total brain gray matter, white matter, and lateral ventricle volume.

Results: The groups did not differ on genetic variables. Multivariate general linear models revealed a group (control subjects vs. patients) × uncommon deletion number interaction, such that the latter variable was associated with lower general cognitive ability and larger ventricles in patients but not control subjects.

Conclusions: These data suggest that aggregate uncommon deletion burden moderates central features of the schizophrenia phenotype.

S chizophrenia and general cognitive ability are highly heritable, correlated human phenotypes whose genetic foundations remain poorly understood, despite decades of intensive study. Specific single nucleotide polymorphisms (SNPs) account for <1% of each type of phenotypic variation (1,2). The aggregate impact of many thousands of SNPs might be as high as 34% for schizophrenia (3) and 51% for general cognitive ability (4), although this remains controversial (5,6). Among the many ideas offered to account for the "missing heritability" (7), two seem especially promising. First, copy number variations (CNVs), a type of mutation, might represent another important source of genetic variation for schizophrenia and cognitive ability. Second, there is a growing realization that the effects of different genetic variations (SNPs or CNVs) might depend on "genetic background." The current study examined a specific type of CNV, "uncommon" deletions, for its possible impact on general cognitive ability in two groups presumed to differ in genetic background, healthy control subjects and individuals with schizophrenia.

Several lines of evidence suggest that CNVs can impact brain development. First, rare, large CNVs are more common among patients with diverse neurodevelopmental disorders, including schizophrenia (8), autism (9), epilepsy (10), and attention-deficit/hyperactivity disorder (11). The proportion of cases attributable to such anomalies remains uncertain. Second, CNVs capture much more nucleotide variation than single nucleotide substitutions (12) and have a higher de novo mutation rate (13,14). Their adverse impact is revealed by evidence of negative selection pressure (13). Third, CNVs might be related to general cognitive ability, a trait with a distinctive developmental trajectory that relies upon specific distributed cortical networks (15). Consistent with this proposition, in a study of individuals with attention-deficit/hyperactivity disorder, a subsample with intellectual disability had an especially high number of CNVs (11). Furthermore, we have also recently reported that uncommon deletions might be related to intelligence in individuals with alcohol abuse (16). In a relatively small sample of individuals originally recruited for a study designed to investigate genetic correlates of alcohol dependence, we found that the total length of uncommon deletions (i.e., those occurring in <5% of our sample) correlated with the full scale intelligence quotient from the Wechsler Abbreviated Scale of Intelligence at $r = -.30$ (16). The association of intelligence with uncommon deletions did not seem to be influenced by alcohol dependence per se, because covarying a common measure of dependence had no effect on results.

These results linking CNVs with intelligence are broadly consistent with a body of research that has used an indirect

marker of mutation load, fluctuating anatomic asymmetry, which refers to nondirectional asymmetries in physical body characteristics that are symmetric at the population level, such as the length of the two ears. The idea that fluctuating asymmetry partly, even if imperfectly, reflects mutation load is supported by studies linking increased fluctuating asymmetry with factors that increase the number of mutations, such as exposure to radiation (17) or heat (18), and factors that increase their functional impact (inbreeding depression [19]). A recent meta-analysis demonstrated small but reliable associations between aggregate measures of fluctuating asymmetry and intelligence (20).

The effects of mutations may depend upon "genetic background" has been highlighted in recent reports, focusing on both schizophrenia and intelligence. Mitchell (21) notes that it is extremely common for mutations to be modified by additional variants in the genetic background (see also Girirajan et al. [22]). Johnson (23) has argued, partly on the basis of the results of selective breeding studies, that the effects of specific genes depend upon other genetic variations such as mutations as well as variable environmental triggers.

In the current study we evaluated the relationship between the overall burden of uncommon deletions and general cognitive ability in two samples that likely differ in underlying genetics, healthy control subjects, and individuals with schizophrenia. Our primary goals were to test the hypothesis that the number and length of rare deletions would correlate negatively with the first principal component (PC) emerging from a large battery of neuropsychological tests, a common way to operationalize general cognitive ability; and to determine whether the hypothesized negative relationship between uncommon deletions and general intellectual ability differed in individuals with schizophrenia and healthy control subjects. A secondary goal was to explore relationships between overall measures of cranial volumes—gray matter, white matter, and ventricle volumes—and measures of uncommon deletions in both groups.

Methods and Materials

Participants

The participants were recruited through the Mind Clinical Imaging Consortium. This consortium includes research teams at the Mind Research Network and University of New Mexico, Massachusetts General Hospital, the University of Minnesota, and the University of Iowa (see White et al. [24] for additional details). A total of 385 individuals were originally studied, although CNV, magnetic resonance imaging (MRI), and cognitive data were available only for 243 individuals. The current analysis is limited to the subset of these individuals who stated their racial background was "white." The CNVs—like single nucleotide polymorphisms—show population stratification, and use of a single reference genome might influence measurement of the total CNV burden. Hence, we could not accurately determine the "rareness" of CNVs in nonwhite groups using frequency data from a predominantly white sample, and our numbers of minority participants were not sufficient to perform within-group analyses of CNV frequencies. Analysis of PCs reflecting population stratification from SNP data revealed that four self-described "white" individuals did not cluster with the rest of the white sample, so these were eliminated from further analysis. The final sample included 79 individuals with schizophrenia (57 males, 22 females) and 110 control subjects (67 males, 43 females). The number of participants recruited from each site were: Albuquerque, New

Mexico: 19 patients/21 control subjects; Boston, Massachusetts: 13 patients/14 control subjects; Minneapolis, Minnesota: 18 patients/19 control subjects; and Iowa City, Iowa: 28 patients/57 control subjects.

A comprehensive clinical diagnostic assessment included either the Structured Clinical Interview for the DSM-IV (25) or the Comprehensive Assessment of Symptoms and History (26). The mean length of illness for patients with schizophrenia was 12.02 years (SD = 11.07, range 0–42). Most patients were currently being treated with antipsychotics (77 of 79; 96%), and very few were antipsychotic naïve (2 of 79; 2.5%). Symptoms of schizophrenia were evaluated with the Scale for the Assessment of Positive Symptoms (27) and the Scale for the Assessment of Negative Symptoms (28). Healthy control subjects were recruited from the general community through medical clinics and advertisements in local newspapers. Exclusionary criteria for the control group were presence of a physical or neurologic disorder affecting brain function, and lifetime history of any Axis I disorder, including substance abuse or dependence. Control subjects were not excluded if they had a first-degree relative with an Axis I psychiatric disorder. The institutional review board at each site approved this study.

Intellectual Assessment

All participants were administered a comprehensive battery of neuropsychological tests tapping these domains: reading ability, verbal fluency, working memory, verbal abstraction, nonverbal reasoning, attention, and visuomotor and executive skills. To facilitate accurate assessment of factor structure, the entire sample (n = 247) was used for a PC analysis, although subsequent analyses were limited to the white sample, as described in the preceding text (the first PC calculated for the subsample reported here with both MRI and genetic data correlated r = .96 with the first PC computed from the original, larger sample; all significant effects [see Results] were also significant with the first PC calculated on the smaller subset, but because the factor structure might be more accurately estimated in the larger, original sample, these results will be reported in the following text). A total of 25 test scores were entered for each participant for the PC analysis (see Table S1 in Supplement 1 for a list of specific test variables and factor loadings). The first component was used in all analyses as a measure of general cognitive ability. Across the total number of expected data points (247 participants with 25 variables), a total of 2.4% were missing values. To avoid discarding participants through list-wise deletion, missing values of specific tests were replaced by the individual group mean. Because the total proportion of total individual test scores imputed in this way was very small, this procedure likely had negligible effects on the results.

Genetic Analyses

The DNA extracted from blood samples was genotyped with Illumina HumanOmni1-quad chip (Illumina, San Diego, California), including 1,140,419 markers. The intensity values (log R ratio [LRR]) and β allele frequency for the markers in autosomes were used for CNV detection. The details of CNV detection were described in Carter et al. (18) and Chen et al. (29,30). Briefly, outlier correction was applied to replace the isolated large LRR values with median ± 2 SD, and PC-based correction was performed to eliminate variation induced by experimental or GC (guanine-cytosine) content factors. Two PCs were corrected here. Data from all participants passed the quality control on LRR (LRR d < .28) (31). The data were then segmented with a circular