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Impaired Rich Club Connectivity in Unaffected Siblings of Schizophrenia Patients

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Schizophrenia has been conceptualized as a disorder of brain connectivity. Recent studies suggest that brain connectivity may be disproportionally impaired among the so-called rich club. This small core of densely interconnected hub regions has been hypothesized to form an important infrastructure for global brain communication and integration of information across different systems of the brain. Given the heritable nature of the illness, we hypothesized that connectivity disturbances, including abnormal rich club connectivity, may be related to familial vulnerability for schizophrenia. To test this hypothesis, both schizophrenia patients and unaffected siblings of patients were investigated. Rich club organization was examined in networks derived from diffusion-weighted imaging in 40 schizophrenia patients, 54 unaffected siblings of patients, and 51 healthy control subjects. Connectivity between rich club hubs was differentially reduced across groups ($P = .014$), such that it was highest in controls, intermediate in siblings (7.9% reduced relative to controls), and lowest in patients (19.6% reduced compared to controls). Furthermore, in patients, lower levels of rich club connectivity were found to be related to longer duration of illness and worse overall functioning. Together, these findings suggest that impaired rich club connectivity is related to familial, possibly reflecting genetic, vulnerability for schizophrenia. Our findings support a central role for abnormal rich club organization in the etiology of schizophrenia.

Key words: connectome/diffusion-weighted imaging/familial vulnerability/brain network

Introduction

Brain function depends on effective communication among regions linked within a complex system of white matter pathways, known as the human connectome.[1] Disruption or aberrant formation of this central system, be it through impaired connectivity or abnormal network architecture, has been proposed to lead to impaired processing and integration of information and, ultimately, brain dysfunction.[2,3] Schizophrenia, a severe psychiatric disorder characterized by hallucinations and delusions, decreased social interaction, and cognitive impairment, has long been hypothesized as a disorder of brain connectivity.[4–9] Indeed, imaging studies have provided extensive evidence for impaired white matter connectivity in schizophrenia.[10–13] In addition, graph analytical studies, examining the topology of the brain network at a systems level, have shown reduced efficiency and increased connection distance in schizophrenia, as well as a less central position of frontal and parietal hubs.[14–17]

In the context of a possible disruption of information integration in schizophrenia, the presence of so-called 'brain hubs' may be of particular interest because network hubs have been suggested to play a critical role in enabling efficient communication between distributed parts of a dynamic system.[18–22] Within the healthy brain, hub regions have been noted not only to be individually rich in connectivity but also to form a densely interconnected "core" or "rich club."[21,23,24] Due to its central position in the network, the rich club has been proposed to play an important role in enabling global neural signaling and interregional brain communication and integration.[20,25] In this context, impaired wiring of this central system may be hypothesized to have detrimental effects on global communication in the brain. Indeed, recent studies examining rich club topology in schizophrenia have demonstrated impaired rich club connectivity.[3,26] Because the importance of genetic influences in the etiology of schizophrenia has been firmly established[27,28] and structural brain abnormalities in schizophrenia[10,29,30] have been associated with genetic factors,[31–34] these disturbances in rich club connectivity may be related to familial risk for schizophrenia. Studying unaffected first-degree relatives

of schizophrenia patients, who share in the genetic risk for schizophrenia but lack the influence of psychosis or antipsychotic medication,[35,36] provides the opportunity to study such a hypothesis. In this study, rich club connectivity is examined in schizophrenia patients, unaffected siblings of patients, and healthy controls, to determine whether brain network disturbances are related to familial, possibly reflecting genetic, vulnerability for the disorder.

Methods

Participants

A total of 145 participants were included in this study, including 40 schizophrenia patients, 54 unaffected siblings of schizophrenia patients, and 51 healthy controls. The patient group was previously examined (as a replication data set), in a study on structural network connectivity in schizophrenia.[3] Extending this previous investigation, this study examines connectomic effects in unaffected siblings of patients. Participants were recruited at the University Medical Center Utrecht, during an ongoing longitudinal study in the Netherlands (Genetic Risk and Outcome of Psychosis). The affiliated medical ethics committee approved the study, and all subjects provided written informed consent prior to participation.

Demographics are described in table 1. For all participants, presence or absence of current and lifetime psychopathology was established using the Comprehensive Assessment of Symptoms and History interview.[37] Patients were eligible for the study if they met Diagnostic and Statistical Manual of Mental Disorders, fourth edition[38] criteria for schizophrenia or related spectrum disorders (see table 1 for details). Siblings were included if they had no diagnosis of a current or lifetime psychotic disorder, including bipolar disorder. Healthy controls were included if they had no current or lifetime psychotic disorder and no first- or second-degree family members with a lifetime psychotic disorder.

For patients, the age of onset of first psychosis was recorded and the total duration of illness calculated. The type and daily dose of antipsychotic therapy was also recorded, and the dose of antipsychotic medication was converted to a chlorpromazine equivalent using conversion rates.[39] Functioning was assessed using the Camberwell Assessment of Need.[40] Symptom severity was assessed with the Positive And Negative Syndrome Scale (PANSS).[41] In siblings and controls, the presence and severity of subclinical symptoms was assessed using the Community Assessment of Psychic Experiences (CAPE).[42] All subjects were interviewed with regard to lifetime cannabis abuse or dependency. In addition, urine samples were acquired from each participant at the time of scanning, and these were screened for the presence of cocaine, amphetamine, and/or cannabis. Group-differences in demographic and clinical characteristics were statistically tested using ANOVA for continuous and chi-squared tests for categorical variables (table 1).

Imaging and Preprocessing

Magnetic Resonance Imaging (MRI) was performed on a 1.5 Tesla clinical MRI scanner at the University Medical Center Utrecht, including an anatomical T1 scan for tissue classification and cortical parcellation and a diffusion-weighted scan (2×32 diffusion directions[17,43]) for the reconstruction of white matter pathways. Freesurfer software[44] was applied to the anatomical T1 scan, to parcellate the cortical surface into 68 distinct regions (ie, 34 cortical regions per hemisphere) for each individual subject. The diffusion-weighted images were realigned and corrected[45] and a tensor was fitted to the diffusion profile within each voxel.[46] Next, white matter pathways were reconstructed, based on the fiber assignment by continuous tracking algorithm.[47] The T1 scan was realigned with diffusion-weighted scan, for anatomical overlap between the cortical parcellation maps and the collection of reconstructed tractography streamlines. Acquisition and preprocessing details are described in the supplementary information.

Connectome Reconstruction

For each subject, a brain network was reconstructed from the total set of cortical grey matter regions and reconstructed white matter tracts. Each brain network was represented as a graph G = (V, E), consisting of a set of nodes V and a set of connections E between nodes. Cortical brain regions ($N = 68$) were represented as nodes (V). Connections E represented, from the total collection of white matter tracts, those streamlines interconnecting 2 nodes. To minimize possible false-positive fiber streamlines,[48] a connection e between two regions i and j was included if at least 5 streamlines were present in the total collection of streamlines that both touched cortical regions i and j.[49,50] For each subject, a matric representation of the brain network was populated with all connections E, weighted according to the number of reconstructed streamlines (NOS). In addition, taking into account the influence of region size on NOS, connections were weighted according to streamline density, by dividing the number of streamlines connecting two regions by the mean volume of the connected regions.

Overall Connectome Organization

Structural brain networks were examined in terms of a number of graph metrics, together providing information on the global and local organization of the networks.[9,51,52] Global metrics included (1) overall connectivity strength S, computed for each participant as the sum of all connections in the brain network; (2) global efficiency GE, computed as the average inverse shortest path length; and (3) overall

Table 1. Demographic and Clinical Characteristics

	Controls (N = 51)	Siblings (N = 54)	Patients (N = 40)	P
Age, mean (SD)	29.4 (8.6)	28.4 (6.8)	30.6 (6.1)	NS
Gender, M/F	22/29	19/35	36/4[a]	<.0001
Psychiatric diagnosis				
Schizophrenia, N (%)	—	—	31 (75.6)	—
Schizoaffective disorder, N (%)	—	—	6 (14.6)	—
Other[b], N (%)	—	—	3 (9.8)	—
PANSS symptoms				
Total, mean (SD) [range]	—	—	46.9 (12.4) [30–83]	—
Positive, mean (SD) [range]	—	—	10.6 (3.8) [7–24]	—
Negative, mean (SD) [range]	—	—	12.6 (3.9) [7–23]	—
General, mean (SD) [range]	—	—	23.9 (6.2) [16–42]	—
CAPE subclinical symptoms (frequency)				
Total, mean (SD) [range]	10.5 (7.6) [0–31]	11.0 (8.1) [0–38]	—	NS
Positive, mean (SD) [range]	1.8 (2.4) [0–9]	2.0 (2.1) [0–9]	—	NS
Negative, mean (SD) [range]	5.1 (3.8) [0–15]	5.3 (4.8) [0–25]	—	NS
Depressive, mean (SD) [range]	3.5 (2.6) [0–12]	3.6 (2.8) [0–13]	—	NS
Age of illness onset, mean (SD)	—	—	22.5 (4.9)	—
Duration of illness in years, mean (SD)	—	—	7.6 (4.0)	—
Global functioning[c], mean (SD)	—	—	5.8 (3.2)	—
Antipsychotic medication				
Typical/atypical/none/unknown, N[d]	—	—	3/30/2/5	—
Chlorpromazine equivalent dose, mean (SD) [range][e]	—	—	275.9 (161.0) [50–625]	—
Cannabis, lifetime abuse/dependency, N (%)	2 (3.9)	1 (1.9)	4 (10)	NS
Urine drug screening at time of scan				
Any, N (%)	3 (5.9)	3 (5.6)	3 (7.5)	NS
Cocaine, N (%)	0 (0)	1 (1.9)	2 (5.0)	NS
Amphetamine, N (%)	0 (0)	0 (0)	0 (0)	NS
Cannabis, N (%)	3 (5.9)	2 (3.7)	1 (2.5)	NS
Recent or lifetime substance use, N (%)	4 (7.8)	4 (7.4)	6 (15.0)	NS

Note: NS, not significant; PANSS, Positive and Negative Syndrome Scale; CAPE, Community assessment of psychic experiences; CAN, Camberwell Assessment of Need.
[a]Indicates the subject group that is statistically different from other subject groups.
[b]Other diagnoses include schizophreniform disorder and psychotic disorder not otherwise specified.
[c]Global functioning as measured by the total number of met and unmet needs on the CAN.
[d]"Typical" includes haloperidol, flupenthixol and perfenazine; "atypical" includes risperidone, olanzapine, quetiapine, clozapine, aripiprazole; "none" is no current antipsychotic treatment.
[e]Chlorpromazine equivalent doses were calculated using conversion rates (risperidone 66:1, olanzapine 20:1, quetiapine 1.3:1, clozapine 1:1, haloperidol 33:1, aripiprazole 13.3:1, flupenthixol 50:1, perfenazine 12.5:1).[39]

clustering C, computed as the average likelihood that the neighbors of a node are interconnected. In addition to global measures, node-specific graph metrics, such as node-specific levels of S_i, GE_i and C_i were computed. Network metrics were all based on NOS-weighted networks and computed using the MATLAB-based Brain Connectivity Toolbox (http://www.brain-connectivity-toolbox.net).[53]

Rich Club Organization

Rich club organization has been described in detail previously.[20,21,23,24,54] In short, rich club organization of a network is characterized by an above-average level of connectivity between the high-degree nodes of a network, over what can be expected by chance.[55] If a rich club is present, the high-degree ('rich') nodes of the network are tightly interconnected, forming a densely connected core or 'club' of nodes: the rich club.

Rich Club Definition. In this study, rich club definition was based on previous studies on rich club connectivity on both low- and high-resolution networks.[20,23,24] Using an a priori defined rich club selection ensured that the rich club was selected unbiased across the 3 subject groups. Based on the previous investigations, rich club regions included the superior frontal gyrus, precuneus, superior parietal gyrus, and insula, all bilateral (in total 11.8% of the network nodes). The participation of these brain regions in the rich club is based on degree (unweighted) and has been well validated by previous studies in human and nonhuman subjects.[3,20,21,23,24,56] Following this definition, the nodes of the network were classified into "rich club" nodes and "peripheral" (ie, nonrich club) nodes.

Classification of Connections: Rich Club, Feeder, and Local Connections. Classification of rich club nodes allowed for the categorization of the edges of the network into

"rich club connections," being those edges that link members of the rich club; "feeder connections," which are the edges that link rich club nodes to peripheral nodes; and "local connections," being those edges that interconnect peripheral nodes (figure 1). For each subject, "rich club," "feeder," and "local" connectivity were computed as the sum of connectivity strength of "rich club," "feeder," and "local" connections, respectively.

Verification of Rich Club Definition. In addition to the a priori rich club definition, the rich club was defined per subject, to allow for possible individual (and between-group) variation in rich club formation. Two separate approaches to define the rich club per individual subject were adopted: (1) by selecting the top 8 highest ranking nodes (ie, top 12% nodes with the highest strength) in the individual network and (2) by selecting the subset of nodes with a degree of 1.25 SD above the mean of the network, allowing for individual differences in rich club size.[3] Both approaches are discussed in more detail in the supplementary information.

Statistical Analysis

Statistical Assessment of Familial Effects. We hypothesized that network abnormalities that are related to familial factors would be most pronounced in patients and intermediate in siblings, compared with controls. Statistical testing involved Jonckheere-Terpstra permutation analysis (10 000 permutations), a nonparametric test for ordered differences in 3 or more study populations,[57] described in more detail in the supplementary information. In the assessment of rich club connectivity (ie, rich club, feeder, and local connectivity) and global network measures (ie, S, GE, and C), values of $P < .05$ were considered to indicate statistical significance. To correct for multiple testing in the analyses of the node-specific metrics S_i, GE_i, and C_i (68 tests were performed per metric), P-values were subjected to a false discovery rate threshold of $q = 0.05$.

A Network-Based Statistic (NBS)[58]-derived analysis was performed to explore components of connections showing ordered differences in connectivity strength such that controls > siblings > patients. Specifically, for each connection comprising the network, a Jonckheere-Terpstra test was performed and a test statistic signifying the extent of ordered differences in connectivity strength assigned. Next, in a binary matrix representation of the network, connections with a test-statistic Tjt < 2.575 were marked 1, and 0 otherwise. In the resulting difference matrix, the largest connected component was

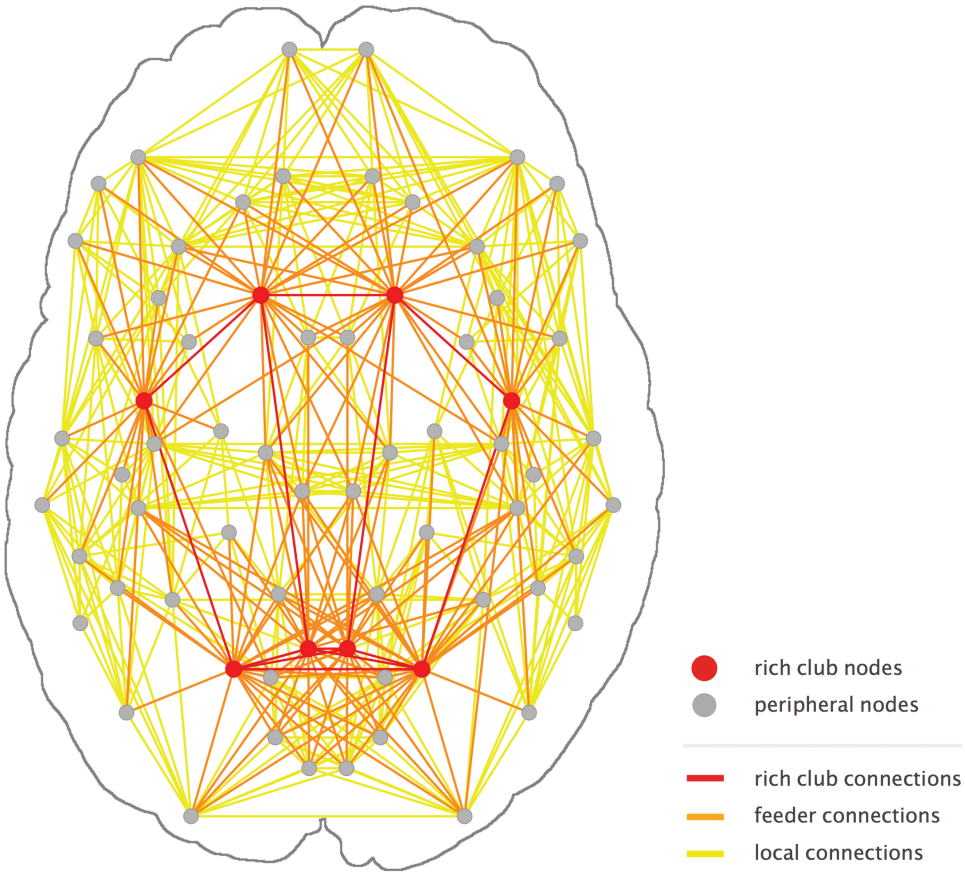

Fig. 1. Rich club. Schematic representation of a group-averaged reconstructed structural brain network. Nodes are categorized into rich club and nonrich club peripheral nodes and connections are color coded to indicate rich club, feeder, or local connections.

identified and its size stored. Permutation testing (10 000 permutations, randomizing group assignment) was used to assign a *P*-value to the observed effect, as the proportion of random permutations resulting in a component of equal or greater size than the component in the original computation.

Statistical Assessment of Clinical Correlates. The relationship between rich club connectivity and clinical measures was examined by linear regression analyses, with the level of connectivity within the rich club (ie, NOS count between rich club members) as the dependent variable and clinical measures as the independent variable. To examine rich club connectivity corrected for individual variation in global brain network strength S and age, these values were included as covariates in the regression analysis. Rich club connectivity was examined for a possible link with duration of illness, global functioning, PANSS symptoms in patients, CAPE subclinical symptoms in siblings, and the daily dose of antipsychotic medication in a chlorpromazine equivalent.

Results

Rich Club Organization

Jonckheere-Terpstra permutation analysis revealed a significant ordered difference in rich club connectivity strength across subject groups (P = .014, figure 2), such that rich club connectivity (ie, NOS) was highest in controls, intermediate in siblings (7.9% reduced relative to controls), and lowest in patients (19.6% reduced compared with controls). Weighting of connections according to streamline density (NOS divided by the mean volume of the connected regions), confirmed this finding (P = .039), suggesting that the observed effect is independent of possible group-differences in region size. Definition of the rich club on the individual level confirmed these findings (supplementary information). The effects were stable in bootstrap validation and across different threshold parameters (supplementary information and figure 2). Notably, high consistency was found in the regions participating in rich club formation between a priori and individual rich club selection, and across subject groups in individual rich club selection (supplementary information).

In addition, because overall connectivity strength S can have a strong influence on other network measures,[17] an additional analysis was performed in which the proportion of overall connectivity strength within the rich club (ie, NOS count between rich club regions divided by S) was assessed, to correct for a possible influence of individual differences in S. Again, this analysis revealed reductions in rich club connectivity across groups (P = .049).

Connectivity values of feeder connections did not show differential reductions (P = .187). This suggests that the observed reduction in rich club connectivity does not result from a general reduction in connectivity of high-degree regions, but rather that connections spanning those regions appear particularly vulnerable. Indeed, connectivity of rich club edges relative to the total strength of hub regions (ie, NOS count of rich club edges divided by the total strength—ie, rich club + feeder edges—of hub regions) was significantly reduced across subject groups (P = .016). Similarly, local connections did show a differential reduction in connectivity strength (P = .047), but with a less substantial effect (2.5% reduced in siblings | 12.8% reduced in patients) compared with rich club connections. These findings were supported by analyses using individual rich club definition, in which no significant differential reductions in feeder or local connectivity were observed (supplementary information).

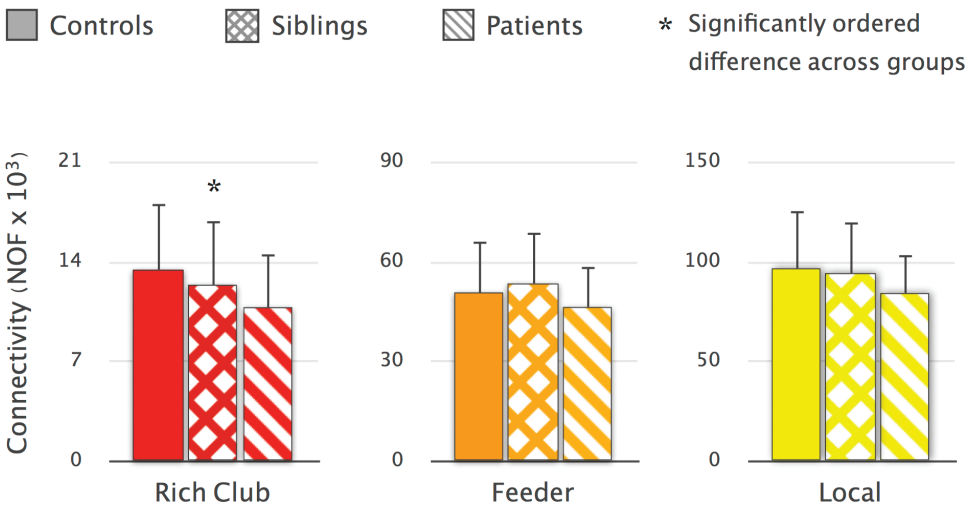

Fig. 2. Rich club, feeder, and local connectivity. Bar graphs indicate connectivity strength (ie, sum of reconstructed fibers), for rich club, feeder, and local connections. A significant ordered difference, such that controls > siblings > patients, was found for rich club connectivity (P = .014).

Global Network Metrics

Consistent with previous reports on aberrant global network organization in schizophrenia patients,[14,16,17] bivariate comparison showed that *S*, *GE*, and *C* were significantly reduced in patients relative to controls. Extending these findings, Jonckheere-Terpstra testing revealed a differential reduction of *C*, such that controls > siblings > patients ($P = .009$). No ordered differences were found for *S* or *GE*, suggesting intact overall levels of strength and global network efficiency in unaffected siblings (supplementary table 1).

It has been noted that the presence of a rich club may underlie organizational attributes, including high clustering, in many real-world networks.[59] To explore the relationship between impaired rich club connectivity and the reductions in *C*, this effect was reexamined after correcting *C* for rich club connectivity using linear regression. In this analysis, differential reductions in *C* were attenuated ($P = .079$). Moreover, bivariate comparison indicated that the reduction in clustering in schizophrenia patients, compared with controls, was also weakened ($P = .072$). These findings suggest that the observed reductions in *C* may, in part, be due to impaired rich club connectivity.

Node-Specific Network Metrics

Analysis of local network topology revealed FDR-significant differential reductions in node-specific metrics across subject groups, such that controls > siblings > patients (all $q < 0.05$, figure 3): S_i was reduced in patients and intermediate in siblings for the bilateral superior frontal gyrus and left inferior temporal gyrus; E_i was differentially reduced for the bilateral rostral anterior cingulate, left superior frontal, and right precentral gyri; C_i of bilateral superior frontal and rostral anterior cingulate gyrus, left medial orbitofrontal, right precentral gyrus and right insula was reduced in patients and intermediate in siblings, compared with controls.

Subnetwork of Differentially Reduced Connectivity

NBS-derived analysis identified a subnetwork of connections showing ordered differences in connectivity

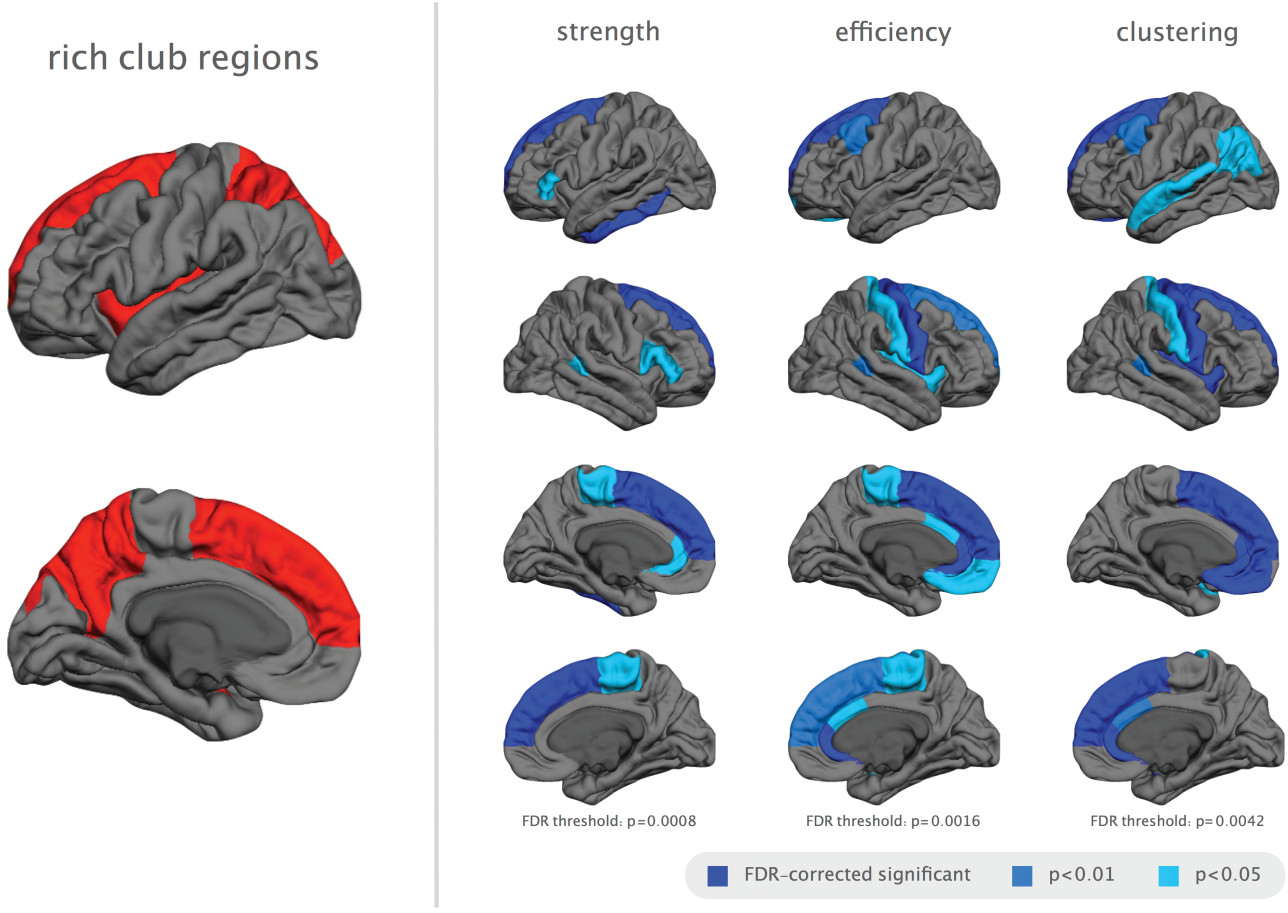

Fig. 3. Node-specific abnormalities. Cortical regions for which differential reductions (ie, controls > siblings > patients) in S_i, E_i, and C_i were found. Regions are color-coded according to *P*-value, with dark blue regions surviving FDR-correction, marking the bilateral superior frontal and rostral anterior cingulate gyri, left medial orbitofrontal and inferior temporal gyri, and right precentral and insular gyri, (all $q < .05$).

strength such that controls > siblings > patients (P = .007; 10 000 permutations). The component comprised 19 differentially reduced connections between 17 brain regions, including 5 rich club regions (figure 4).

Abnormalities in Rich Club Regions

While rich club nodes account for ~12% of network nodes, a large proportion of differential reductions in node-specific metrics were found among the rich club. A post-hoc permutation analysis testing the null-hypothesis that all nodes are at equal risk to be affected indicated that the disproportionate number of differential reductions in nodal measures for rich club nodes was statistically significant for nodal strength (67% of findings, P = .033) and clustering (43%, P = .029), but not efficiency (25%, P = .399). Similarly, the probability that at least 5 out of the 17 regions in the subnetwork identified in the NBS are rich club regions is unlikely to occur under the null-hypothesis (P = .021).

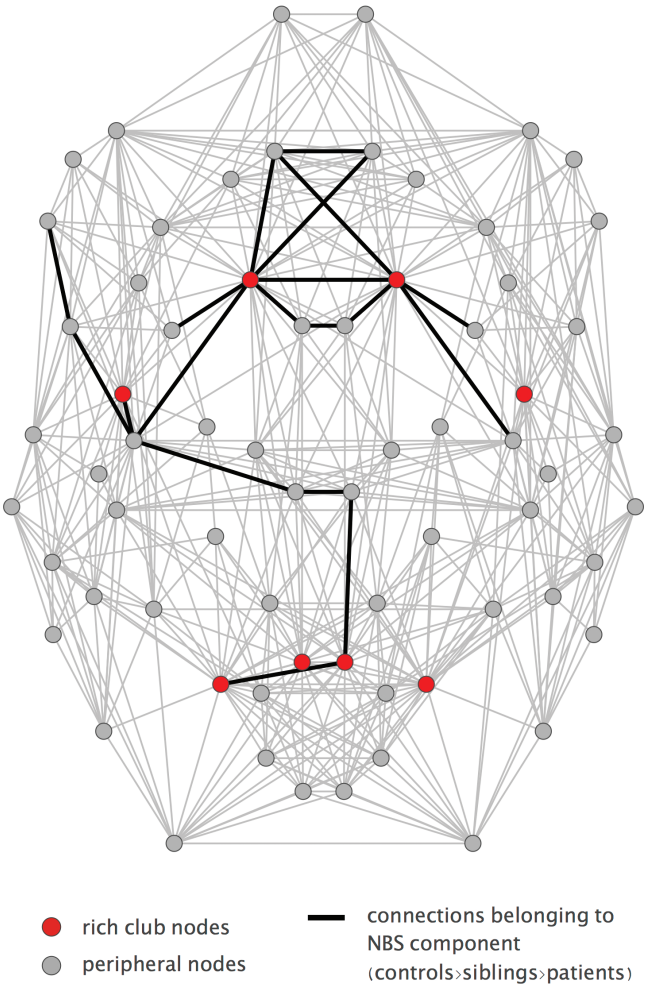

Fig. 4. Component of differentially affected connections. Schematic representation of a group-averaged brain network, showing a component of connections with differentially reduced connectivity strength, ie, controls > siblings > patients (P = .007). Cortical regions are represented as nodes, with node coloring indicating rich club or peripheral nodes; bold black lines indicating the connections belonging to the subnetwork. Importantly, no individual connection can be declared significant alone, only the subnetwork as a whole.[57]

Clinical Correlates

Disease Outcome. In patients, a negative linear relationship between rich club connectivity and duration of illness (β = −0.27, P = .033), as well as global functioning (β = −0.21, P = .036), was observed (supplementary figures 1A and B, respectively).

Symptoms. No significant correlation between rich club connectivity and clinical symptoms as measured by the PANSS was found in patients, nor with positive and negative subscale scores. Similarly, in siblings, no significant association with total subclinical symptoms as measured by the CAPE was observed, nor with positive, negative, and depressive subscales (all P > .05).

Medication. No significant relationship between the chlorpromazine equivalent dose of antipsychotic medication and rich club connectivity was observed (β = .09, P = .492).

Substance Abuse. No significant differences in rich club connectivity were found between subjects with current or lifetime substance use, compared with those without (all P > .05). Nevertheless, to exclude a possible confounding effect of substance use on rich club connectivity, rich club connectivity was reexamined after excluding subjects with current or lifetime substance use, confirming our main findings (P = .020).

Discussion

The findings of this study suggest that abnormal brain network organization, in particular impaired rich club connectivity, is related to familial predisposition for schizophrenia. Previous structural network studies in schizophrenia have shown an altered overall network organization[9] and decreased connectivity density of the brain's central rich club.[3] Extending previous findings, this study now reveals intermediate levels of rich club connectivity in unaffected siblings of schizophrenia patients, suggesting that impaired rich club connectivity in patients is likely to have a familiar, possibly genetic, component. In addition to reductions in rich club connectivity, our study shows differentially reduced levels of global clustering in schizophrenia patients and unaffected siblings. In patients, impaired rich club connectivity was associated with illness effects, in that lower rich club connectivity was associated with longer duration

of illness and worse clinical functioning. In all, our findings suggest that rich club dysconnectivity may be a core aspect of schizophrenia, both prior to and after the onset of illness.

Region-specific connectivity strength S_i, clustering C_i, and efficiency E_i of particularly frontal cortical regions were also found to show differential reductions across subject groups, such that values were highest in controls, intermediate in siblings, and lowest in patients. While rich club members account for only ~12% of the nodes in the network, abnormalities in S_i and C_i appear to be overrepresented among members of the rich club. Similarly, the NBS-derived component of differentially reduced connections (ie, most pronounced reductions in patients, intermediate values in siblings) included an above chance proportion of the rich club. These findings may be indicative of high-degree regions being generally more susceptible (both biologically and statistically) to disease-related changes, or of rich club connectivity being particularly vulnerable for familial effects in schizophrenia.

It has been shown that various measures of (brain) network organization are interrelated.[15,60] The presence of a rich club has, eg, been proposed to underlie important organizational attributes, including high clustering, in many real-world networks.[59] Putatively, the observed reductions in global clustering in schizophrenia patients,[61] which this study suggests to be related to familial predisposition for schizophrenia, may relate to aberrant rich club organization. Indeed, the attenuated reductions in C after correction for rich club connectivity seem to suggest that the observed differences in global clustering may, in part, be due to impaired rich club connectivity. Future studies examining the relationship between various brain network measures are of particular interest.

The examination of unaffected siblings of schizophrenia patients provides the opportunity to investigate whether brain abnormalities that are observed in overt illness exist in individuals at increased genetic risk for schizophrenia, in the absence of the effects of psychosis and antipsychotic medication.[35,36] The currently observed abnormalities in rich club organization in these individuals seem to suggest that rich club dysconnectivity includes a neurodevelopmental vulnerability for the illness, which may be mediated by genetic factors. If lower levels of rich club connectivity indeed reflect vulnerability for disease, how should the differential reduction in rich club connectivity—with the strongest reductions in patients and intermediate levels in siblings—be interpreted? One possible explanation may be that the observed reductions in rich club connectivity reflect a gene dosage effect, with a higher number, or expression, of schizophrenia risk alleles in patients than siblings, resulting in lower levels of rich club connectivity. In addition, the currently observed association between rich club dysconnectivity and duration of illness and clinical functioning (supplementary figure 1) may signify that the lower levels in patients might include an added effect of illness on a preexisting vulnerability. Given the cross-sectional nature of our study and small effect size of the clinical correlates, the relationship between rich club connectivity and illness dynamics should currently be interpreted as preliminary. Future longitudinal studies examining brain connectivity, and rich club changes in particular, in patients and/or individuals at high risk for psychosis are of particular interest in this regard.

A putative genetic influence on aberrant brain network architecture in schizophrenia is supported by twin- and family studies showing genetic control of structural and functional brain network organization.[62–64] Furthermore, recent studies suggest reduced functional connectivity,[65] decreased global efficiency, longer connection distance, and a lower number of hub regions[66] in offspring of schizophrenia patients. Moreover, in healthy individuals, the presence of schizophrenia risk gene DISC1 has been associated with lower levels of global network efficiency.[67] These studies and our current findings converge on the notion that connectome abnormalities in schizophrenia reflect, at least in part, an inherited susceptibility to the disease.[9]

Some issues should be considered when interpreting the findings of this study. First, the majority of patients received antipsychotic treatment, which may potentially influence structural brain connectivity. Altered white matter connectivity has, however, been also shown in medication-naive patients,[43] and this study shows deficits of rich club connectivity in never-medicated siblings of patients. Furthermore, across patients, no relationship between the dose of antipsychotic medication and rich club connectivity was found. Together, these findings suggest that our current results are unlikely to be due to antipsychotic medication alone. Second, there was a preponderance of men in the patient group. Post-hoc bivariate comparison of rich club connectivity between male and female healthy subjects indicated no significant difference ($P = .15$, 10 000 permutations), suggesting no gender difference in rich club formation in health. Furthermore, analyzing rich club connectivity in male subjects only ($N = 22$ controls; $N = 19$ siblings; $N = 36$ patients) did not change the nature of our results (3.9% reduction in siblings | 12.3% reduction in patients, both compared with controls, $P = .088$), suggesting that our findings were not an effect of gender. Third, no relationship was observed between dysconnectivity and symptom severity, which may relate to the variability of symptoms over time. Fourth, data on possible nonpsychotic comorbid disorders in the patient sample was not recorded. However, comorbid substance abuse was examined as a potential confounder and did not explain our findings.

In addition to these considerations, we note that our results are limited by the inherent properties of the acquired data. Diffusion imaging relies on water diffusion

as an indirect probe of axon geometry and, as such, suffers from a number of practical limitations (for review see Jbabdi and Johansen-Berg),[68] but it is the only currently available tool to study brain anatomical brain connectivity in humans in vivo. In this study, connectome reconstruction was performed on 1.5T, 32 diffusion direction, data, based on single-tensor reconstruction of the diffusion data and deterministic fiber tractography. High-field imaging, acquisition of more diffusion directions, and application of advanced white matter pathway reconstruction algorithms (eg, multi tensor and/or probabilistic fiber tracking approaches) may result in better detection of crossing, diverging, or converging fibers.[69–71] In addition, connectivity strength was measured using the number of reconstructed streamlines between brain regions. Several factors influence streamline count, including white matter volume, local white matter microstructure, and fiber length. Specifically, streamline count may artificially increase with length because more streamlines are started with increasing track length. At the same time, however, longer streamlines are more difficult to complete, resulting in a potential underestimation of fiber count. While there may be differences in the length of white matter connections between patients and controls, as a result of brain volume differences, studies do not suggest reduced total brain volume in relatives of patients,[72] and this study shows no differences in the overall number of reconstructed streamlines between siblings and controls. Moreover, significant reductions in rich club connectivity were again found across the three subject groups when the ratio of rich club connectivity (ie, rich club connectivity divided by *S*) was examined. In all, it is unlikely that our current findings result from simple differences in fiber length and/or count. In addition, rich club dysconnectivity as found in this study is consistent with reports based on high-field diffusion data,[23,24] and rich club organization has also been demonstrated for the macaque[21] and cat[53,55] connectome based on anatomical tract tracing, underscoring the biological validity of our current results.

In conclusion, our findings suggest that connectome abnormalities, including impaired rich club connectivity, are related to a familial, possibly reflecting genetic, predisposition for schizophrenia. Our findings emphasize a central role for abnormal rich club organization in the etiology of schizophrenia.

Supplementary Material

Supplementary material is available at http://schizophreniabulletin.oxfordjournals.org.

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