Commentary

Biological Psychiatry

Linking Neurogenetics and Functional Connectivity in Autism

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The large neurobiological heterogeneity of autism spectrum disorder (ASD) and its genetic diversity have challenged our ability to identify how genetic risk translates into changes of brain structure and function and, finally, into behavioral symptoms. Various lines of evidence, stemming from genetics (1,2), neuroimaging [reviewed in (3)], and behavioral phenotyping (4), have highlighted the subtle and multifactorial mechanisms underlying the disease, which have resulted in limited success developing novel therapeutics. In the current issue of *Biological Psychiatry*, Rasero *et al.* (5) attempt to characterize neural heterogeneity in ASD in a data-driven way by clustering large-scale resting-state functional brain connectivity profiles, searching for subtypes and links to phenotypic variation. Following previous lines of work linking neuroimaging findings to transcriptomic patterns (6), the study used high-resolution transcriptomic data from the Allen Human Brain Atlas (7) to identify genes enriched in brain regions with altered functional connectivity characteristic for each subtype.

Motivated by previous studies on changes of resting-state connectivity in ASD, the authors examine large-scale brain connectivity patterns common within groups of individuals to search for subtypes in ASD. The study applies consensus clustering strategies to multivariate connectivity patterns of brain regions for associating connectivity-based ASD subtypes with their neurogenetic profiles. Here, the authors hypothesize that different biological features characterize the underlying neurodevelopmental and maturation brain connectivity profiles of potential unknown subtypes. Starting from an initial set of 2156 participants from the Autism Brain Imaging Data Exchange repository, rigorous data harmonization obtained 657 ASD and 884 control datasets.

After neuroimaging preprocessing and data harmonization, 82 regions (68 cortical, 34 in each hemisphere, and 14 subcortical) were identified in each brain from the Desikan-Killiany atlas and segmented using FreeSurfer. Then, to cluster the functional connectivity matrices, the authors leveraged a computational approach they previously developed to detect communities in complex networks. The method is summarized as follows: 1) for each node, define a distance matrix for the set of subjects by comparing the connectivity pattern of that node in all pairs of subjects; 2) cluster the distance matrix for each node; 3) build the consensus network from the corresponding partitions; and finally 4) extract groups of subjects by finding the communities of the consensus network thus obtained. This approach is different from previous implementations of consensus clustering, in that the consensus strategy combines the information arising from the connectivity patterns of each node. The consensus clustering was then applied to brain connectivity matrices. Because connectivity matrices may contain nuisance variables and effects of no interest (e.g., age), prior to subtyping, the authors regressed out age, sex, and motion from each connectivity entry of the participants with ASD.

By clustering the functional connectivity matrices using the above graph network approach, 2 major stable connectivity-based subtypes were found: the first, subtype 1, exhibited global hypoconnectivity (less average connectivity than typically developing control participants), and subtype 2 exhibited global hyperconnectivity. The 2 subtypes showed distinct regional aberrations of connectivity from typically developing subjects. Subtype 1 displayed altered connectivity patterns compared with control individuals in the superior temporal gyrus, posterior cingulate gyrus, and insula, regions typically involved in the default mode and salience networks. In contrast, subtype 2 had higher differences in the thalamus, putamen, and precentral gyrus. Curiously, with respect to cognitive and behavioral performance, the 2 subtypes were highly similar to each other, because among 10 different scores compared, only 2 of them gave uncorrected statistical differences (Autism Diagnostic Observation Schedule total $p = .03$, Social Responsiveness Scale total $p = .05$), which became nonsignificant after correcting for multiple comparisons (Autism Diagnostic Observation Schedule total false discovery rate–corrected $p = .27$, Social Responsiveness Scale total false discovery rate–corrected $p = .27$). Furthermore, no significant structural differences between subtypes 1 and 2 were found in region volume or thickness.

ASD is a polygenic, highly heterogeneous condition, with multitudes of common and rare variants contributing to the genetic architecture (8). Here, the authors leveraged the Allen Human Brain Atlas (7) to extract the transcriptomic profiles of each subtype. They used spatial autoregressive models to identify genes with enriched expressions in implicated brain regions. This strategy is known to reduce the correlation bias produced by the similar transcriptomic expression in proximal brain regions. Rasero *et al.* (5) then computed the association between functional connectivity alterations represented by pseudo-R^2 maps. Genes with expression patterns that were associated with the 2 biotypes were analyzed for gene enrichment in distinct pathways or biological processes. Surprisingly, genes associated with the first subtype did not show any enrichment, although several known ASD risk genes were on the list.

The second subtype, however, was associated with genes mapping to glutamatergic neurotransmission and synapse organization, indicating an excitation/inhibition imbalance, a

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leading well-known primary mechanism in the pathophysiology of ASD. These results suggest a link between excitation/inhibition imbalance and functional connectivity alterations, but only in one ASD subtype, that is characterized by global brain hyperconnectivity and major alterations in the somatomotor and default mode networks. Previous studies have suggested that an excitation/inhibition imbalance during development may be an essential mechanism, yet specific factors driving this imbalance are not well understood. Therapeutic interventions aiming to restore the excitation/inhibition balance in ASD have been proposed repeatedly, but the current study shows that only a subgroup of patients might benefit from this strategy—and that this patient group could be identified via neuroimaging.

The Rasero *et al.* (5) study is an important contribution to a newly emerging field aiming to bridge the so-far existent gap between systems neurosciences and molecular psychiatry, which can be followed up in several directions. First, while this work breaks new ground in the identification of ASD biotypes, it would be intriguing to follow up associations between connectivity patterns and clinical variables in even larger cohorts. These approaches might allow even more intricate analyses and could provide novel insight into how symptomatology or other factors, such as psychotropic medications, impact neural biotypes. Larger cohorts should also provide an opportunity to follow up on age-dependent effects on biotypes, as brain growth trajectories have been robustly described (9,10). Finally, the authors used the Allen Human Brain Atlas as a key tool to link neuroimaging findings to transcriptomic profiles. While this approach has proven to be a major translational tool, it remains opaque to gene expression changes in the brains of subjects who are on the autism spectrum. Correlating neuroimaging parameters with actual transcriptomic profiles in ASD will provide an important new angle once gene expression atlases for ASD individuals become available. While these open questions remain, Rasero *et al.* open an important new avenue into identifying ASD biotypes and might even provide a basis for a more personalized therapeutic approach for this group of individuals.

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Article Information

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