Received: 22 September 2022 | Revised: 29 November 2022 | Accepted: 20 January 2023

DOI: 10.1111/gbb.12838

ORIGINAL ARTICLE

Genes, Brain and Behavior

Polygenic risk score for attention-deficit/hyperactivity disorder and brain functional networks segregation in a community-based sample

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Funding information
Conselho Nacional de Desenvolvimento Científico e Tecnológico, Grant/Award Number: 573974/2008-0; Fundação de Amparo à Pesquisa do Estado de São Paulo, Grant/Award Numbers: 2013/08531-5, 16/04983-7, 16/13737-0, 2018/04654-9, 2018/21934-5, 2021/05332-8

Abstract

Neuroimaging studies suggest that brain development mechanisms might explain at least some behavioural and cognitive attention-deficit/hyperactivity disorder (ADHD) symptoms. However, the putative mechanisms by which genetic susceptibility factors influence clinical features via alterations of brain development remain largely unknown. Here, we set out to integrate genomics and connectomics tools by investigating the associations between an ADHD polygenic risk score (ADHD-PRS) and functional segregation of large-scale brain networks. With this aim, ADHD symptoms score, genetic and rs-fMRI (resting-state functional magnetic resonance image) data obtained in a longitudinal community-based cohort of 227 children and adolescents were analysed. A follow-up was conducted approximately 3 years after the baseline, with rs-fMRI scanning and ADHD likelihood assessment in both stages. We

hypothesised a negative correlation between probable ADHD and the segregation of networks involved in executive functions, and a positive correlation with the default-mode network (DMN). Our findings suggest that ADHD-PRS is correlated with ADHD at baseline, but not at follow-up. Despite not surviving for multiple comparison correction, we found significant correlations between ADHD-PRS and segregation of cingulo-opercular networks and DMN at baseline. ADHD-PRS was negatively correlated with the segregation level of cingulo-opercular networks but positively correlated with the DMN segregation. These directions of associations corroborate the proposed counter-balanced role of attentional networks and DMN in attentional processes. However, the association between ADHD-PRS and brain networks functional segregation was not found at follow-up. Our results provide evidence for specific influences of genetic factors on development of attentional networks and DMN.

We found significant correlations between polygenic risk score for ADHD (ADHD-PRS) and segregation of cingulo-opercular networks and default-mode network (DMN) at baseline. ADHD-PRS was negatively correlated with the segregation level of cingulo-opercular networks but positively correlated with the DMN segregation.

KEYWORDS
ADHD, DMN, GWAS, network segregation, networks, polygenic risk score, rs-fMRI

1 | INTRODUCTION

Complex interplays between genetic and environmental factors concur to cause mental disorders in general, and attention-deficit/hyperactivity disorder (ADHD), in particular.[1,2] In fact, epigenetic mechanisms such as variation in DNA methylation,[3] have been implicated in the development of ADHD.[4–6] Specific structural and functional brain network alterations during development are thought to mediate the relationship between causal factors and the expression of ADHD symptoms (Faraone et al., 2015). Advances in human genomics and brain connectomics[7] have allowed crucial refinements of the ADHD neurobiological models.[8–11] Promising results have emerged from resting-state functional magnetic resonance imaging (rs-fMRI) studies focused on the organisation of large-scale brain functional networks during typical and atypical development.[12,13]

To date, several rs-fMRI studies have observed associations between altered functional connectivity within and between specific networks and ADHD. Uddin et al.[14] reported a decreased homogeneity of the default-mode network (DMN) connections, particularly within the precuneus in ADHD patients when compared with healthy controls. Similarly, Castellanos et al.[15] found diminished functional connectivity between the dorsal anterior cingulate and precuneus/posterior cingulate in paediatric ADHD patients. Sato et al.[16] replicated these findings in children and adults, further suggesting that a delayed DMN maturation may play a role in ADHD expression.[16,17] More recently, Qian et al.[18] demonstrated that altered organisation of functional networks was associated with symptomatic heterogeneity in a male paediatric ADHD sample. In that study, boys with the combined ADHD subtype but not those with the hyperactive/impulsive subtype presented altered connectivity within the DMN, but stronger inter-network connectivity between the salience and executive control networks when compared with typically developing children.

The concept of segregation of brain functional networks provides a metric to investigate neurodevelopmental changes.[19,20] Functional segregation can be assessed by determining the community structure of a network and quantifying the relations within and between the identified specialised communities or modules. Most neuroimaging studies focus on the investigation of functional integration between neural modules. However, the specialisation within each module may also play a role in ADHD. Lin et al.[21] reported an increased global brain functional network segregation in ADHD. Ghaderi et al.[22] reported abnormal functional segregation in ADHD with combined subtype, but not in the inattentive one. Supporting several previous reports, Mills et al.[13] described reduced negative connectivity between task-positive and task-negative networks in children with ADHD. Interestingly, connections between the DMN, dorsal attention and cingulo-opercular networks explained most of the differences between ADHD and control groups and were mostly influenced by age.

At the same time, genomic architectures, genetic risk factors, and polygenic risk scores (PRS) associated with susceptibility to ADHD have recently been discovered.[8,23] Besides allowing the estimation of a high heritability (70%–80%) for ADHD, several twin studies have further suggested that a full-blown ADHD diagnosis represents the extreme of heritable quantitative or continuous traits.[23–25] Common variant risk of ADHD is indeed associated with continuous measures of ADHD symptoms.[26,27] The most recent published ADHD-GWAS evaluated 20,183 patients and 35,191 controls and found 12 independent genomic regions associated with ADHD, several of then near to genes

implicated in neurodevelopmental processes. PRS is an approach that uses the results from GWAS to evaluate the polygenic risk of each individual in an independent cohort. Hamshere et al.[28] have reported that patients with ADHD present higher polygenic risk score (ADHD-PRS) than healthy controls. Demontis et al.[8] estimated the liability-scale of SNP heritability to be around 0.21 and the current GWAS found that the maximum variance explained by the estimated PRS was 5.5%. Moreover, ADHD-PRS was shown to be correlated with ADHD symptoms in the general population.[29] Interestingly, ADHD-PRS is also associated not only with hyperactivity/impulsivity symptoms, but also with genetic vulnerability for a myriad of psychiatric symptoms and disorders in children and in the general population.[29] Moreover, great advances have been made in uncovering epigenetic mechanisms underlying ADHD development.[30,31] For instance, ADHD-PRS was also shown to be correlated with global and peripheral DNA methylation levels.[32–34]

Despite such remarkable advances in both genetics and neuroimaging investigations of ADHD, the putative mechanisms by which polygenic architecture influences clinical features via functional neurodevelopment remain largely speculative. Specifically, investigating associations between genetic susceptibility to ADHD and abnormalities on brain functional segregation may provide evidence for a plausible biological mechanism leading to ADHD. Thus, in this study, we investigated the correlation between the ADHD-PRS developed by Demontis et al.[8] and the functional segregation of brain networks in a community-based cohort of children longitudinally followed for 3 years. Based on the previous neuroimaging findings summarised above, we hypothesise that the ADHD-PRS would be associated with altered segregation metrics in the attentional networks (negative correlation) and DMN (positive correlation).

2 | MATERIALS AND METHODS

2.1 | Participants and assessments

All participants were enrolled as part of the Brazilian High-Risk Cohort Study for Mental Conditions (BHRCS). Children from the community were recruited in schools from two Brazilian cities (São Paulo and Porto Alegre). Magnetic resonance imaging (MRI) scans were acquired from a total of 309 children and adolescents at two time-points. The detailed demographic information is described in Table 1. Detailed information on cohort design, data collection procedures and quality controls implemented can be found in Salum et al.[35] and Pan et al.[36] The subjects are the same described in Salum et al.[35] with the same scanner and sequence parameters for both baseline and follow-up MRI acquisitions.

Although almost 700 children participants were scanned at baseline, only 309 subjects could be successfully scanned at follow-up (at the time this study was conducted). Due to the age range, many participants were not eligible due to teeth braces and some simply refused to participate or did not show-up. Sixty-six participants were excluded due to excessive head motion defined as mean frame-displacement >0.5 mm, and another 16 participants were excluded due to missing values on the genetic profile, resulting in a total of 227 subjects with imaging and genetic data included in the statistical analyses. Parents or legal guardians provided written consent for all children enrolled. In addition, all children provided verbal assent to participate in the study. All protocols and procedures were approved by the local ethics committees.

Intelligence quotient (IQ) was estimated using the vocabulary and block design subtests of the Weschler Intelligence Scale for Children and the approach of Tellegen and Briggs.[37] Psychiatric symptoms were assessed using the parent-reported Development and Well-Being Assessment (DAWBA).[38] ADHD items of the DAWBA assessment were summed up to obtain a total score of ADHD symptoms. The ADHD-DAWBA module was used without the skip rules to allow a characterisation of the full 18 ADHD symptoms for the whole population. Response categories for each item were coded as 0 (None), 1 (A Little) and 2 (A Lot). Scores varied from 0 to 54, with higher scores representing higher probability of having ADHD. Thus, it is important to highlight that all analyses in this study were conducted from a dimensional framework (ADHD score) and not diagnostic (controls vs. patients).

2.2 | Polygenic score

For each participant, DNA was extracted from blood or saliva samples, and genotyping was conducted using the HumanOmniExpressV1

TABLE 1 Demographic information of participants.

	Site		
	Rio Grande do Sul	São Paulo	Total
N	120	107	237
Males	61	60	121
Females	59	47	106
Mean baseline age (SD)	131.34 (24.11)	127.21 (19.25)	130.00 (22.00)
Mean follow-up age (SD)	162.53 (23.96)	157.88 (18.86)	160.34 (21.78)
Mean IQ (SD)	103.27 (16.38)	105.03 (17.44)	104.10 (16.88)
Mean baseline DAWBA-ADHD	8.75 (8.42)	8.81 (8.98)	8.78 (8.67)
Mean follow-up DAWBA-ADHD	7.50 (7.06)	6.21 (7.16)	6.89 (7.12)

Note: *N* is the number of subjects at each subsample; the age is reported in months for both baseline and follow-up; DAWBA-ADHD is the ADHD scores from DAWBA questionnaire.

(Illumina) system. The ADHD polygenic risk scores (ADHD-PRS) were then calculated using the PRSice v2 package Choi and O'Reilly 2019[39] and considering the summary statistics of the genome-wide association study (GWAS) from the Psychiatric Genomics Consortium (PGC, see https://www.med.unc.edu/pgc/results-and-downloads)[8] based on 2189 DNA samples. The summary statistics of PGC and iPSYCH were calculated using the package described in Reference 8. In this sample, 321 were cases and 1868 were controls (no history of ADHD). Demontis et al.[8] considered a 0.8 imputation quality and 0.01 for minor allele frequency (MAF). Our sample has not been imputed and thus we used a MAF of 0.01. Before generating the scores, clumping was used to obtain single-nucleotide polymorphisms (SNPs) in linkage equilibrium, and those in linkage disequilibrium ($r^2 > 0.1$) within a 250 bp window were discarded. We included 35,445 SNPs, and the *p*-value threshold with the largest Nagelkerke's R2 (variance explained by the PRS) was considered the best-fit threshold (*p*-value threshold of 0.1412, explained variance of 1%). We considered the 10 principal components as potential covariates in further analysis. The following options were used in PRSice: PRSice_linux --all-score --bar-levels 0.001, 0.05, 0.1, 0.2, 0.3, 0.4, 0.5, 1 --base ADHD_GWAS.gz --binary-target T --clump-kb 250 kb --clump-p 1 --clump-r2 0.1 --cov BHRC_Probands_Final_pca10.eigenvec --print-snp; where ADHD_GWAS.gz. We used the option binary-target true as we have only cases and controls. The genetic variants, and respective effect sizes and weights in PRS are reported in the Supplementary Material.

2.3 | Image acquisition and preprocessing

Considering the challenges of acquiring quality rs-fMRI data in children and adolescents, all participants were included in recreational activities before scanning for desensitisation, decreasing head movement during scanning. The images were collected in two 1.5 T MRI scanners using the same parameters and protocols (GE, Signa HDX and HD at each site, i.e., geographical location of data acquisition). First, T1-weighted images were acquired for registration (repetition time [TR] = 10.916 ms, echo time [TE] = 4.2 ms, slice thickness = 1.2 mm, flip angle = 15°, matrix size = 256 × 192, field of view [FOV] = 245 mm, number of excitations [NEX] = 1, bandwidth = 122.109, number of slices up to 156 for whole-brain coverage). Then, rs-fMRI data were obtained in an eyes opened protocol (TR = 2000 ms, TE = 30 ms, slice thickness = 4 mm, gap = 0.5 mm, flip angle = 80°, matrix size = 80 × 80, FOV = 240 mm, reconstruction matrix = 128 × 128, NEX = 1, number of slices = 26, total of 180 volumes).

All rs-fMRI data preprocessing was carried out using the CONN toolbox v.18a (https://www.nitrc.org/projects/conn/),[40] SPM12 (http://www.fil.ion.ucl.ac.uk/spm/software/spm12/), and standard MNI-space pipeline based on the following steps: EPI images unwarping and head motion correction, T1 segmentation and spatial normalisation to MNI ICBM152 template, functional normalisation, denoising,[41] motion censoring (displacement >0.5 mm and global signal $z > 3$), and spatial smoothing (functional width half maximum [FWHM] = 8 mm), linear detrending and signal bandpass filtering (0.008–0.09 Hz), and nuisance regression (white matter and cerebrospinal fluid signals, six head motion parameters and their respective derivatives, and motion censoring) based on the simult approach.[42]

Regions-of-interest (ROI) were defined using Gordon cortical parcellation.[43] This atlas is comprised of 333 ROIs subdivided into 12 subnetworks: auditory, cingulo-opercular, cingulo-parietal, DMN, dorsal and ventral attention, frontoparietal, retrosplenial-temporal, salience, somatomotor of hand, somatomotor of mouth, visual networks, and 47 unlabeled ROIs. For each ROI, we extracted the average (across voxels) preprocessed blood oxygenation level dependent (BOLD) signal as the ROI representative. Finally, based on these signals, the functional connectivity matrices among all ROIs of each subject were calculated using bivariate Pearson correlations and transformed to *z*-scores. All further analyses were carried out in R Platform for Statistical Computing (www.r-project.org) version 3.5.0.

2.4 | Segregation metric

For each subject, the segregation measure of each subnetwork was calculated by the ratio between the mean functional connectivity within the subnetwork and the mean connectivity between its ROIs and all other remaining ROIs. We chose this metric since it is dependent on both intra-network (numerator) and inter-network (denominator) strengths. Such a metric is specific to intra-network segregation since a normalisation on the whole brain connectivity is implemented.

2.5 | Statistical analyses

A demographic model was built using the General Linear Model (GLM) and considering the ADHD-PRS as the dependent variable. The demographic model's independent variables included the site of acquisition, age at baseline, gender, and estimated IQ. Additionally, a clinical model was built with ADHD-PRS as the dependent variable and ADHD-DAWBA scores, site of acquisition, age, and gender as independent variables. Separate models were computed for the two timepoints independently. For consistency evaluation, correlation analyses between ADHD-DAWBA scores at baseline and follow-up were conducted using the Pearson correlation coefficient. Analogous analyses were conducted regarding the segregation measures.

For the baseline rs-fMRI data, segregation analyses were conducted using the GLM considering the ADHD-PRS as the dependent variable and the segregation measure of each subnetwork as the independent variables. In order to consider possible confounding effects of ancestrality on ADHD-PRS, we added the first 10 principal components of the genotyping data as covariates in all models involving the ADHD-PRS. The same procedure was carried out independently on the follow-up data. Finally, association analyses between ADHD-DAWBA and brain networks segregation were performed using the same previously described GLM model, but

replacing the ADHD-PRS as the dependent variable by the ADHD-DAWBA total score. Type I Error was set at 5% (FDR correction for 13 multiple comparisons).

3 | RESULTS

3.1 | Demographic and clinical models

Demographic information is presented in Table 1. The distributions of the demographic (age at each data collection and inters-scan intervals; IQ) and clinical variables (ADHD-DAWBA scores) of interest in both time points of data collection are represented in histograms (Figure 1). As expected, the frequency of children with higher scores in the ADHD-DAWBA items was strictly smaller in the follow-up when compared with the baseline, with rather fewer subjects with scores above 20. Despite this difference in the frequency of children with higher scores, ADHD-DAWBA scores at baseline and follow-up were moderately consistent ($r = 0.51$; $p < 0.001$).

For the demographic model, which considered the ADHD-PRS as the dependent variable and site of acquisition, age at baseline, gender, and estimated IQ as independent variables, we found a significant negative correlation only with estimated IQ ($b = -1.03 \times 10^{-06}$; $p = 0.001$). The coefficients for all other variables were not statistically significant ($p > 0.05$). The results remain unchanged when the 10 first principal components of GWAS are included as covariates. For clinical analyses, post hoc significant positive correlation between ADHD-PRS and ADHD-DAWBA ($b = 1.35 \times 10^{-06}$; $p = 0.030$) at baseline were observed. This finding is not statistically significant ($p = 0.25$) when considering a model in which the 10 first principal components of GWAS are included as

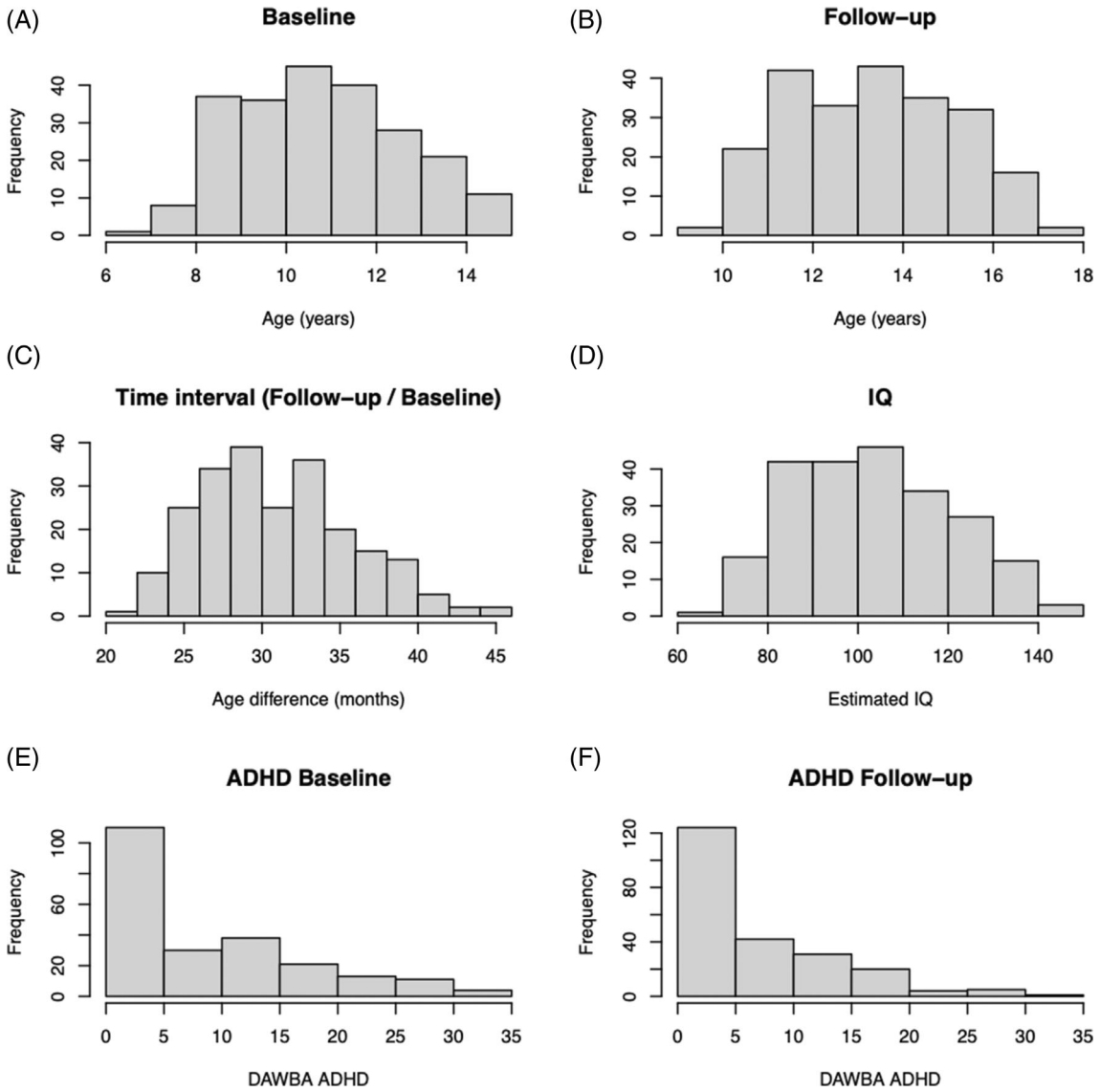

FIGURE 1 Panels A and B: Histogram for participant age (in years) at baseline and follow-up assessment. Panel C: Histogram for the time interval (in months) between the MRI scanning. Panel D: Histogram for estimated IQ (standard scale) at baseline. Panels E and F: Histogram for the ADHD scores (DAWBA) at baseline and follow-up assessments.

covariates. At follow up, this same correlation was significantly marginal at follow-up ($b = 1.55 \times 10^{-06}$; $p = 0.049$) and not statistically significant if the 10 principal components are included ($p = 0.23$).

3.2 | Segregation analyses

The correlation analyses of the segregation measures of each network between the baseline and follow-up is shown in Table 2. Note that the correlation values were modest (but mostly statistically significant), with higher values for the somatomotor of the hand and cingulo-parietal networks.

As our main result regarding the associations between ADHD-PRS and brain networks segregation measures of DMN and attentional networks, the GLM analyses reported statistically significant findings for baseline but not at follow-up. The GLM results are presented in Table 3. Despite not surviving for multiple comparison adjustment (FDR), of importance to our main hypothesis, the segregation metrics at baseline of cingulo-opercular network was negatively correlated (uncorrected-$p = 0.027$, FDR-$p = 0.351$) with ADHD-PRS, while the segregation of the DMN was marginally positively correlated (uncorrected-$p = 0.046$, FDR-$p = 0.552$) with ADHD-PRS.

Finally, GLM analyses between ADHD-DAWBA and brain networks segregation metrics did not show any statistically significant association, neither at baseline (F-test $p = 0.979$) nor at follow-up (F-test $p = 0.068$).

As a complementary result, by using the previous GLM with IQ as dependent variable, we found that the segregation of the ventral attention network was statistically significant as independent variable at baseline (beta = −10.8581; SD = 3.5941; uncorrected-$p = 0.003$; fdr-$p = 0.03$) but not at follow-up.

TABLE 2 Pearson correlation analyses of the segregation measures between baseline and follow-up.

Network	Correlation	p-value
Auditory	0.265	<0.001
Cingulo-opercular	0.277	<0.001
Cingulo-parietal	0.339	<0.001
Default mode	0.284	<0.001
Dorsal attention	0.264	<0.001
Frontoparietal	0.306	<0.001
Other	0.123	0.06
Retrosplenial-temporal	0.171	0.009
Salience	0.15	0.022
Somatomotor hand	0.356	<0.001
Somatomotor mouth	0.253	<0.001
Ventral attention	0.259	<0.001
Visual	0.197	0.003

4 | DISCUSSION

Rs-fMRI findings suggest that diversions on the development of specific functional networks segregation constitute a possible mechanism for at least some ADHD symptoms.[13,44,45] Among those specific networks, the DMN and cingulo-opercular network are usually implicated. Further, multiple common genetic variants contributing to the risk of developing ADHD have been discovered, and a PRS has been used to aggregate its effects. Here, we set out to integrate these lines of investigation by testing the hypothesis that ADHD-PRS is associated with segregation of specific brain functional networks in a community-based cohort of children and adolescents. Moreover, we investigated whether such a hypothetical association would be stable during the developmental process. As hypothesised, although not surviving to multiple comparisons, the segregation of cingulo-opercular network and DMN was associated with the ADHD polygenic score at

TABLE 3 GLM coefficients of segregation analysis at baseline considering ADHD-PRS as the dependent variable (including the first 10 principal components of the genotyping data as covariates).

Network	Coefficient	Std. dev.	t-statistics	Uncorr. p	fdr-p
Auditory	4.03×10^{-06}	1.24×10^{-05}	0.326	0.745	1
Cingulo-opercular	-2.84×10^{-05}	1.28×10^{-05}	−2.224	**0.027**	**0.351**
Cingulo-parietal	-4.41×10^{-06}	3.77×10^{-06}	−1.170	0.243	1
Default mode	2.10×10^{-05}	1.05×10^{-05}	2.007	**0.046**	**0.552**
Dorsal attention	-4.79×10^{-06}	1.26×10^{-05}	−0.381	0.703	1
Frontoparietal	9.78×10^{-06}	1.21×10^{-05}	0.806	0.421	1
Other	-3.12×10^{-05}	3.21×10^{-05}	−0.971	0.333	1
Retrosplenial-temporal	4.23×10^{-06}	6.55×10^{-06}	0.646	0.519	1
Salience	-8.73×10^{-06}	6.24×10^{-06}	−1.398	0.164	1
Somatomotor hand	-1.42×10^{-05}	1.02×10^{-05}	−1.397	0.164	1
Somatomotor mouth	8.92×10^{-06}	6.02×10^{-06}	1.482	0.140	1
Ventral attention	-1.27×10^{-05}	1.51×10^{-05}	−0.837	0.403	1
Visual	2.21×10^{-06}	9.36×10^{-06}	0.236	0.813	1

Note: Statistically significant coefficients (uncorrected Type I Error set at 5%) are highlighted in bold.

baseline. However, these associations were not observed at a 3-year follow-up evaluation.

First, it is important to highlight that the *p*-values in Table 3 are uncorrected and, if so, the DMN and cingulo-opercular networks findings are not statistically significant. However, since this was a hypothesis driven analysis, focused on DMN and attentional networks given the physiopathology of ADHD, we believe the results raise relevant discussions and insights to the field. To the best of our knowledge, this is the first study to investigate associations between ADHD-PRS and brain modularity.

Corroborating current knowledge on the nature and meaning of interactions between large-scale functional networks, DMN segregation was shown to be positively correlated with ADHD-PRS, while segregation measures of cingulo-opercular networks were negatively correlated. It is well established that spontaneous BOLD activity of DMN nodes is anti-correlated with spontaneous activity in cingulo-opercular networks.[14] Moreover, task-related activation of cingulo-opercular network nodes leads to the deactivation of the DMN. A proposed functional implication of these counterbalanced activities of DMN and attentional networks includes the instantiation of task-specific psychological processes. Cingulo-opercular networks are indeed thought to instantiate different aspects of attentional processes,[43,46] with the cingulo-opercular network supporting tonic alertness and task-set maintenance.[47,48]

However, the lack of evidence for an association between ADHD-PRS and functional networks segregation in the same sample after 3 years might indicate a developmental stage-dependent influence of ADHD genetic susceptibility factors on brain functional organisation. In a comprehensive review on the development of functional connectivity and networks, Grayson and Fair[49] found compelling evidence for an evolving community structure of functional networks, with a trend to strengthen connections (i.e., diminishing segregation). Anderson et al.[50] reported that strengthening between networks connectivity from adolescence to early adulthood is associated with diminishing segregation of the DMN and cingulo-opercular network. The negative correlation between the DMN and attentional network was also shown to become more negative with age. A lag on the development of functional networks, particularly the cingulo-opercular network, has indeed been proposed as a mechanism for attentional problems.[17] A specific lag on the lessening segregation of attentional networks and DMN was indeed observed in patients with ADHD when compared with typically developing controls. Additionally, as expected, we observed an age-dependent decline of ADHD symptoms in our sample, which may partially contribute to a lack of association between ADHD-PRS and functional segregation at follow-up.[51]

An apparently puzzling finding was that, although we have found a correlation between ADHD-PRS and brain networks segregation, and between the former and an increased risk for ADHD, we have found no significant dependence between brain networks segregation and ADHD-DAWBA score. This result disagrees with our initial hypothesis, and we speculate two possible conjectures. The first is that functional segregation may not be the cause or the mediator of the symptoms and, both may be consequences of another process driven by the genetic profile. The second explanation is that the signal-to-noise ratio of the data is insufficient to yield significant statistical findings. It is well-known that the noise level is associated with low field scanners (1.5 T), short scanning time (only 5 min), head motion and systemic artefacts, and measurement error in both rs-fMRI and clinical assessment.

This study was conducted in a previously collected dataset with a parent-reported assessment of psychopathology. Hence clinical relevance and children's perception of the symptoms were not assessed. However, a complementary analysis on IQ, which is a more objective measure on the child, was associated with the segregation of the ventral attention network. Interestingly, IQ was found to be negatively associated with PRS. Moreover, head movement artefacts are an inherent issue when rs-fMRI is used to describe developmental changes since the amount of movement varies with age and psychopathology. Though careful measures were taken both during data acquisition, preprocessing and analysis to minimise motion-related artefacts, changes of measurement errors of segregation with age[49] could have partially influenced the results, and replications of these findings in other samples are desirable. Further replications in larger samples would also be of importance to confirm whether the association of ADHD-PRS and functional segregation is generalizable, and to better characterise differences in the relations between ADHD susceptibility and brain function according to developmental stage. Unfortunately, it is not possible to disentangle the impact of these various factors in the current data and these will remain as possible conjectures. Furthermore, Brazilian population is very admixed and genomic imputation in admixed cohort adds more errors than usual. On the other hand, the implications of not imputing are not very large since one of the key PRS steps is to select only one variant of the linkage disequilibrium block, thus, it would not substantially increase the number of SNPs in the PRS calculation. Finally, 66 subjects were excluded in this study due to excessive head motion. This is a limitation, since these subjects are more prone to present ADHD symptoms.

In sum, we found associations between ADHD-PRS and segregation of DMN and cingulo-opercular network in children and adolescents in the community. These associations were not observed in the same sample after approximately 3 years, suggesting that the relations between genetic susceptibility and functional networks may differ during the developmental process. Finally, opposite directions of correlations between ADHD-PRS and DMN versus cingulo-opercular networks agree with the currently proposed functioning of these networks and with plausible mechanisms for attentional problems.

ACKNOWLEDGMENTS

The opinions, hypotheses, conclusions, and recommendations of this study are those of the authors and do not necessarily represent the opinions of the funding agencies. The authors are grateful to FAPESP (grants 2013/08531-5; 16/04983-7; 16/13737-0; 2018/04654-9; 2018/21934-5; 2021/05332-8) and the National Institute of Developmental Psychiatry for Children and Adolescents, a science and

technology institute funded by CNPq (573974/2008-0). Claudinei Eduardo Biazoli has received funding from the EU Horizon 2020 research and innovation program under the Marie-Skłodowska-Curie grant agreement (896021). João Ricardo Sato receives research support from UNIEMP-Brazil.

CONFLICT OF INTEREST STATEMENT

Pedro Mario Pan received payment or honaria for lectures and presentations in educational eventos for Sandoz, Daiichi Sankyo, Eurofama, Abbot, Libbs, Instituto Israelita de Pesquisa e Ensino Albert Einstein, Instituto de D'Or de Pesquisa e Ensino. Luis Augusto Rohde has received grant or research support from, served as a consultant to, and served on the speakers' bureau of Abbott, Aché, Bial, Medice, Novartis/Sandoz, Pfizer/Upjohn, and Shire/Takeda in the last 3 years. The ADHD and Juvenile Bipolar Disorder Outpatient Programs chaired by Dr Rohde have received unrestricted educational and research support from the following pharmaceutical companies in the last 3 years: Novartis/Sandoz and Shire/Takeda. Dr Rohde has received authorship royalties from Oxford Press and ArtMed. Dr. Rodrigo Affonseca Bressan has been on the speakers' bureau/advisory board of AstraZeneca, Bristol, Janssen, and Lundbeck. Dr. Bressan has also received research grants from Janssen, Eli-Lilly, Lundbeck, Novartis, Roche, FAPESP, CNPq, CAPES, Fundação E.J. Safra, and Fundação ABAHDS. He is also a shareholder in Biomolecular Technology Ltd. Dr. Edson Amaro Jr. has received research grants from FAPESP, CNPq, CAPES, Fundação E.J. Safra, and Fundação ABAHDS. All other authors reported no biomedical financial interests or potential conflicts of interest.

DATA AVAILABILITY STATEMENT

Data available on request due to privacy/ethical restrictions.

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SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

How to cite this article: Sato JR, Biazoli CE, Bueno APA, et al. Polygenic risk score for attention-deficit/hyperactivity disorder and brain functional networks segregation in a community-based sample. *Genes, Brain and Behavior*. 2023; 22(2):e12838. doi:10.1111/gbb.12838