The Journal of Child Psychology and Psychiatry

Journal of Child Psychology and Psychiatry 62:9 (2021), pp 1140–1149 doi:10.1111/jcpp.13384

Characterizing neuroanatomic heterogeneity in people with and without ADHD based on subcortical brain volumes

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Background: Attention-deficit/hyperactivity disorder (ADHD) is a prevalent neurodevelopmental disorder. Neuroanatomic heterogeneity limits our understanding of ADHD's etiology. This study aimed to parse heterogeneity of ADHD and to determine whether patient subgroups could be discerned based on subcortical brain volumes. **Methods:** Using the large ENIGMA-ADHD Working Group dataset, four subsamples of 993 boys with and without ADHD and to subsamples of 653 adult men, 400 girls, and 447 women were included in analyses. We applied exploratory factor analysis (EFA) to seven subcortical volumes in order to constrain the complexity of the input variables and ensure more stable clustering results. Factor scores derived from the EFA were used to build networks. A community detection (CD) algorithm clustered participants into subgroups based on the networks. **Results:** Exploratory factor analysis revealed three factors (basal ganglia, limbic system, and thalamus) in boys and men with and without ADHD. Factor structures for girls and women differed from those in males. Given sample size considerations, we concentrated subsequent analyses on males. Male participants could be separated into four communities, of which one was absent in healthy men. Significant case–control differences of subcortical volumes were observed within communities in boys, often with stronger effect sizes compared to the entire sample. As in the entire sample, none were observed in men. Affected men in two of the communities presented comorbidities more frequently than those in other communities. There were no significant differences in ADHD symptom severity, IQ, and medication use between communities in either boys or men. **Conclusions:** Our results indicate that neuroanatomic heterogeneity in subcortical volumes exists, irrespective of ADHD diagnosis. Effect sizes of case–control differences appear more pronounced at least in some of the subgroups. **Keywords:** ADHD; subcortical volume; neuroanatomic heterogeneity; community detection; effect sizes.

Introduction

Attention-deficit/hyperactivity disorder (ADHD) is a prevalent behavioral disorder of neurodevelopmental origins, which is characterized by age-inappropriate inattention (IA) and/or hyperactivity and impulsivity (HI; Faraone et al., 2015). ADHD frequently persists from childhood into adulthood, with a prevalence of 3.4%–5.3% in childhood/adolescence and 2.5% in adulthood (Polanczyk, de Lima, Horta, Biederman, & Rohde, 2007; Salum et al., 2015; Simon, Czobor, Balint, Meszaros, & Bitter, 2009).

ADHD is a heterogeneous disorder on the clinical, behavioral, cognitive, genetic, and neuroanatomic level. Clinically and behaviorally, there is strong interindividual variation in psychiatric as well as somatic comorbidities across the life span (Franke et al., 2018). Most individuals with ADHD have deficits in one or more cognitive domains, but there is substantial overlap between ADHD and controls (Mostert et al., 2018; Mueller, Hong, Shepard, & Moore, 2017; Nigg, Willcutt, Doyle, & Sonuga-Barke, 2005). The estimated heritability of ADHD is 70%–80%; and common genetic variants with small effect size are the major contributors to genetic susceptibility to ADHD (Faraone & Larsson, 2019). Considerable heterogeneity is also present in structural and functional brain architecture. The most consistent findings were observed for structural brain alterations in subcortical regions (De La Fuente, Xia, Branch, & Li, 2013). To overcome the limitations of small sample size studies, the ENIGMA-ADHD Working Group conducted a large mega-analysis (1,713 cases and 1,529 controls) across the life span (Hoogman, Bralten, et al., 2017). This analysis confirmed earlier findings of smaller caudate nucleus, putamen, and total intracranial volumes in ADHD and identified smaller nucleus accumbens and amygdala volumes in individuals with ADHD compared with healthy controls. Volumetric case–control differences were most prominent in

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Conflict of interest statement: See Acknowledgments for full disclosures.

childhood. However, the effect sizes were small, possibly reflecting neurobiological heterogeneity of ADHD.

Classification methods have been used to investigate heterogeneity within groups (Reichardt & Bornholdt, 2007). In ADHD research, community detection (CD), a graph-theoretical measure, has been applied to identify clusters of children with different neuropsychological performance profiles across a battery of tasks (Fair, Bathula, Nikolas, & Nigg, 2012). A similar method was used to identify three subgroups of children with ADHD presenting with distinct profiles of emotional functioning associated with clinical outcome (Karalunas et al., 2014). Taximetric analysis was applied in a sample of adolescents with ADHD, resulting in three subgroups with different profiles of executive functioning and motor inhibition (Stevens, Pearlson, Calhoun, & Bessette, 2018). In combination with other studies on the heterogeneity of functional brain architecture in ADHD (Costa Dias et al., 2015; Gates, Molenaar, Iyer, Nigg, & Fair, 2014), the results of these investigations suggested that differences in clinical, behavioral, and neurobiological presentation and course of ADHD may be captured in distinct subpopulations. Moreover, while both cases and healthy controls were present in the same subgroups, affected individuals were more impaired when compared to controls within the same subgroup (Fair et al., 2012; Mostert et al., 2018; Stevens et al., 2018).

Community detection methods have been widely applied to brain networks (Newman, 2006). Here, we applied this approach to parse heterogeneity in subcortical brain volume using data from the ENIGMA-ADHD Working Group (n = 2,493 in total). Our objectives were (a) to examine whether subgroups of participants could be defined based on subcortical volumes and whether this categorization was related to the clinical presentation of ADHD; and (b) to explore whether case–control effect sizes would be stronger within subgroups.

Methods

Participants and ADHD assessment

This study used available magnetic resonance imaging (MRI) data from the international ENIGMA-ADHD Working Group (http://enigma.ini.usc.edu/ongoing/enigma-adhd-working-group/). The group shares structural MRI scans from children and adults with ADHD, as well as phenotypic information, including comorbidities, IQ, age, and gender from over 35 cohorts across the world. With a rolling inclusion design, new cohorts can join the group at any time, but data freezes are set for each analysis. Each site verified the diagnosis of ADHD and assessment of comorbidities (Table S1). All participating sites had approval from local ethics committees.

To constrain heterogeneity in the ENIGMA-ADHD dataset, we stratified our sample by age and sex. Our subsamples comprised 993 boys (aged 4–14 years), 400 girls (aged 4–14 years), 653 adult men, and 447 adult women (aged > 22 years; Table 1). We first applied EFA and CD to the subsample of boys, which was the largest subsample within the dataset. The same method was subsequently applied to the other three subsamples to investigate whether similar subgroups exist in these subsamples.

Neuroimaging

Structural T1-weighted brain MRI data were collected at each site. All scans were subsequently analyzed using the standardized ENIGMA protocols based on FreeSurfer version 5.1 or 5.3. For each participant, we computed left and right volumes of the nucleus accumbens, putamen, pallidum, caudate nucleus, thalamus, amygdala, and hippocampus, as well as intracranial volume (ICV). For all analyses, we used the mean of the left and right subcortical volume. Outliers were identified as above or below three times the interquartile range, and participants with missing data were excluded from the analysis.

Factor analysis

Exploratory factor analysis (EFA) was applied to reduce the space of subcortical volume data by modeling latent factors, which in general requires 300 cases per analysis (Tabachnick & Fidell, 2006). In current study, we invited EFA to identify underlying brain organization based on subcortical brain volume. In considering nonlinear patterns of subcortical brain volumes across age, each subcortical volume was regressed individually with age, age^2, sex, ICV, and sampling site; this was done for children and adults separately. Residuals were used to construct covariance matrices. Squared multiple correlations were built as prior communality estimates. Maximum likelihood and oblique rotation were used to extract factors. The number of eigenvectors extracted was based on the scree plot. A variable was considered to load on one factor if the loading on the factor was 0.40 or more. Model fitness was evaluated based on Tucker–Lewis Index (TLI), Bayesian information criterion (BIC), and the root mean square error of approximation (RMSEA). Confirmation factor analysis (CFA) was used to test whether the factor structures generated in girls or women by EFA is superior than the factor structure observed in males, based on comparative fit index (CFI), TLI, BIC, Akaike information criterion (AIC), and RMSEA. The analyses were performed using the psych package in R programming 3.6.2.

Community detection

We applied CD to identify distinct communities of participants based on factor scores generated by the EFA of subcortical volumes. Applying a modularity algorithm, CD identifies clusters of individuals in a network by requiring strong correlation among them (Newman, 2006). CD was performed in three steps. First, $n \times n$ weighted, undirected networks were created by correlating participants with each other on their normalized factor scores to provide distance information between subject pairs. For this, a threshold of $r = .5$ was chosen, where reachability remained equal to 1 (using r values of 0.3 and 0.7 did not change results). Subsequently, a weight-conserving modularity algorithm was applied to identify distinct communities of participants in each network (Fair et al., 2012; Rubinov & Sporns, 2011). To obtain the most optimal partitioning of the network, this algorithm iteratively sorts nodes (participants in this study) into communities until the modularity (Q) reaches a maximum. Q is the number of edges (correlations between participants) falling within communities minus the expected number in a random network. Q ranges between −1 and 1, with positive values indicating that the strength of edges within communities is larger than expected at random. In the current study, all Q values ranged between 0.45 and 0.49 (Table 2), indicating that the strength of

Table 1 Characteristics of participants

	Boys		Girls		Men		Women	
Variables	Patients	Controls	Patients	Controls	Patients	Controls	Patients	Controls
N	563	430	135	265	412	241	223	224
Mean age (*SD*)	11.0 (1.9)	10.8 (1.9)	10.3 (1.9)	10.4 (1.8)	32.1 (8.9)	32.2 (8.7)	36.1 (10.1)	35.7 (11.0)
Mean IQ (*SD*)	103.2 (15.8)	110.4 (14.8)	103.1 (14.8)	112.5 (13.8)	108.4 (14.3)	113.3 (14.8)	108.5 (15.4)	110.5 (15.3)

IQ = Intelligence quotient; SD = Standard deviation. IQ was only available for subsample of patients and controls (see Table S1 for details).

correlations between participants within communities was very strong, underscoring the robustness of the network structure.

To assess robustness of the community structure, we examined variation of information (VOI). Briefly, a proportion of edges of a network was randomly perturbed (alpha). VOI was calculated as the variance between the original and perturbed networks over a range of alpha, which ranges between 0 and 1 (Karrer, Levina, & Newman, 2008).

All CD analyses were performed in Matlab (Mathworks) and the functions provided by Olaf Sporns, Mikail Rubinov, and collaborators (Rubinov & Sporns, 2011).

Statistical analyses

Age was compared between patients and controls using an independent samples *t*-test. Estimated IQ scores were compared between groups with analysis of variance (ANOVA) after regressing the effects of age, IQ assessment instrument, and sampling site. For each community, we compared subcortical factor scores between cases and controls using *t*-tests to link to the previous studies of the ENIGMA-ADHD Working Group (Hoogman, Bralten, et al., 2017). Starting with the subsample of boys, we also investigated whether ADHD symptom severity and IQ differed among the communities using ANOVAs; chi-square tests were used to compare medication use and the presence of comorbidities between communities. False discovery rate (FDR) was used to correct for multiple comparisons of case–control differences across age bins within the analyses of factor scores and the subcortical volumes, separately. All analyses were performed in IBM SPSS Statistics 25.

Results

Participant characteristics

Demographics of this sample are described in Table 1. Mean age did not differ between cases and controls for boys ($t = -1.7$, $p = 0.09$), girls ($t = 0.14$, $p = .89$), men ($t = 0.22$, $p = .83$), and women ($t = -0.37$, $p = .71$). Differences in IQ scores between cases and controls were significant in boys ($F = 44.1$, $df = 1$, $p = 5.6e\text{-}11$), girls ($F = 25.6$, $df = 1$, $p = 7.1e\text{-}7$), men ($F = 11.3$, $df = 1$, $p = .001$), but not in women ($F = 0.94$, $df = 1$, $p = .33$).

EFA on subcortical volumes

Aiming to limit heterogeneity and maximize power, we started with the largest subsample available, which was for boys, and performed EFA on residualized subcortical brain volumes. From the covariance matrix, we extracted three eigenvectors (Figure 1, Table 3). Volumes of caudate nucleus, globus pallidus, nucleus accumbens, and putamen loaded on the first factor. We interpreted this first factor as 'basal ganglia'. The second factor included hippocampus and amygdala and was interpreted as 'limbic system'. The third factor comprised only the thalamus. The three factors accounted for 25%, 16%, and 12% of the total shared variance, respectively (TLI = 0.92, BIC = −1.45, RMSEA = 0.07).

We next performed EFA in girls (Figure S1, Table 3). Volumes of caudate nucleus, nucleus accumbens, and putamen loaded on the first factor; the second factor only included the globus pallidus; the third factor comprised by the hippocampus, amygdala, and thalamus volume. The three factors accounted for 16%, 18%, and 20% of the total shared variance, respectively (TLI = 0.86, BIC = −3.43, RMSEA = 0.10). The comparison of model fitness indicated that this factor structure (CFI = 0.87, TLI = 0.77, BIC = 39117, AIC = 39053, RMSEA = 0.12) was superior to the one generated in boys (CFI = 0.82, TLI = 0.69, BIC = 39144, AIC = 39080, RMSEA = 0.14; chi-square difference = 26.4, $p = 2.2e\text{-}16$).

EFA was also run for adult men and women, separately. In men with and without ADHD, the same three eigenvectors as in boys were extracted, which accounted for 23%, 17%, and 17%, respectively, of the total shared variance (TLI = 0.98, BIC = −13.8, RMSEA = 0.04). In women, three eigenvectors were also found, but the factor structure differed from the others (Figure S2, Table 3). Volumes of nucleus accumbens and putamen loaded on the first factor. The second factor included caudate nucleus, globus pallidus, and thalamus. The third factor comprised hippocampus and amygdala volume. The three factors accounted for 17%, 18%, and 23% of the total shared variance, respectively (TLI = 0.98, BIC = −12.8, RMSEA = 0.01). Additional model comparison indicated that this factor structure was superior in the subsample of women (CFI = 0.51, TLI = 0.06, BIC = 44524, AIC = 44454, RMSEA = 0.29) to the factor structure we observed in subsamples of males (CFI = 0.43, TLI = −0.01, BIC = 44584, AIC = 44518, RMSEA = 0.30; chi-square difference = 66.6, $p = 3.4e\text{-}16$).

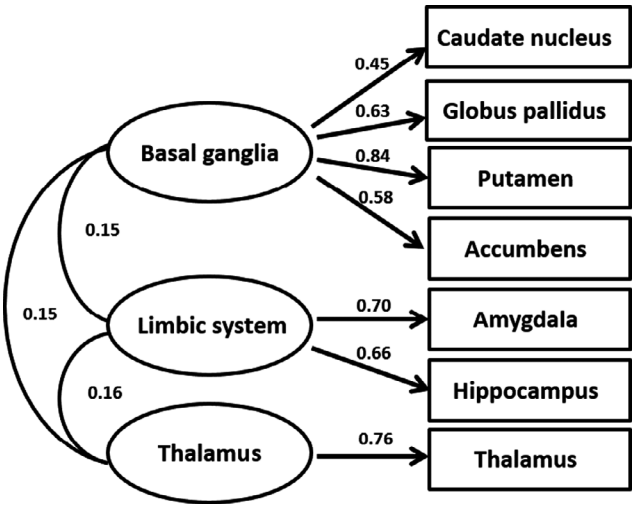

Figure 1 The three-factor model that was generated by EFA in the boys with estimated factor loadings of the latent factors. Note: Similar factor models were generated in boys with and without ADHD separately

CD on factor scores of subcortical volumes in boys and men

Given that factor structures differed between males and females, subsequent CD results would have been incomparable between them. For the subsequent CD analyses, we therefore focused exclusively on boys and adult men, where sample sizes were most appropriate for CD-type analyses.

In all boys (with and without ADHD), we observed four distinct communities, each comprising 20%–30% of the sample (Figure 2; Table 2). Community 1 was characterized by larger volume in basal ganglia, normal volume in limbic system, and smaller volume of thalamus compared to the average volume of the whole sample. Community 2 showed opposite characteristics for basal ganglia and thalamus to Community 1. Community 3 had smaller basal ganglia and thalamus and larger volume in the limbic system, whereas Community 4 showed the reverse pattern compared to Community 3. Repeating the analysis in boys with and without ADHD separately resulted in largely similar findings (Figure 2).

Quality control measures, that is, the quality index (Q value) and VOI (Table 2, Figure S3), showed that these communities were significantly different from subgroups generated from random networks, and the networks were robust against chance variation. Furthermore, although the distribution of cases and controls across communities differed among cohorts (Table S2), leave-one-out analyses of the five largest cohorts showed no evidence for specific cohorts driving the community structure, and the same four communities were found in each analysis.

Community detection in adult men (with and without ADHD) resulted in four communities similar to those observed in boys, each accounting for 15%–31% of the sample (Figure 2; Table 2; Figure S4). Cases were distributed across all four communities, but the controls were only present in three communities, with no healthy men in Community 3. The distribution of cases and controls over communities is shown in Table S3 for each cohort.

Comparison of subcortical factor scores between patients and controls in each community

Within each of the four unique communities observed in boys and men, we investigated whether subjects with and without ADHD showed group differences in volumes for each of the subcortical factor structures (Table 4, Figure 2). Boys with ADHD in Community 1 and Community 3 had smaller subcortical volumes in basal ganglia compared to controls; boys with ADHD in Community 1 also had larger volumes in the limbic system than controls. Those with ADHD in Community 2 had smaller volumes in this system than controls. Boys with ADHD in Community 2 and Community 3 also showed larger volumes for thalamus, and those in Community 4 had smaller thalamus volume. Effect sizes for boys ranged from $d = -0.90$ (95%CIs [−1.17, −0.62]) to $d = 0.65$ (95%CIs [0.39, 0.90]; Table 4). In men, no case–control differences at the factor score level survived FDR correction (Table 4). As a

Table 2 The distribution of participants in subsamples in communities

Subsamples	Total	Patients	Controls
Boys (N)	**992**	**563**	**430**
Community 1	220 (22.2%)	119 (21.1%)	130 (30.3%)
Community 2	270 (27.2%)	167 (29.7%)	95 (22.1%)
Community 3	234 (23.6%)	122 (21.8%)	103 (24.0%)
Community 4	268 (27.0%)	154 (27.4%)	101(23.5%)
Q values	0.45	0.45	0.46
Men (N)	**653**	**412**	**241**
Community 1	201 (30.8%)	127 (30.8%)	102 (42.3%)
Community 2	166 (25.4%)	90 (21.8%)	70 (29.0%)
Community 3	97 (14.9%)	79 (19.2%)	0
Community 4	189 (28.9%)	116 (28.2%)	69 (28.6%)
Q values	0.47	0.46	0.49

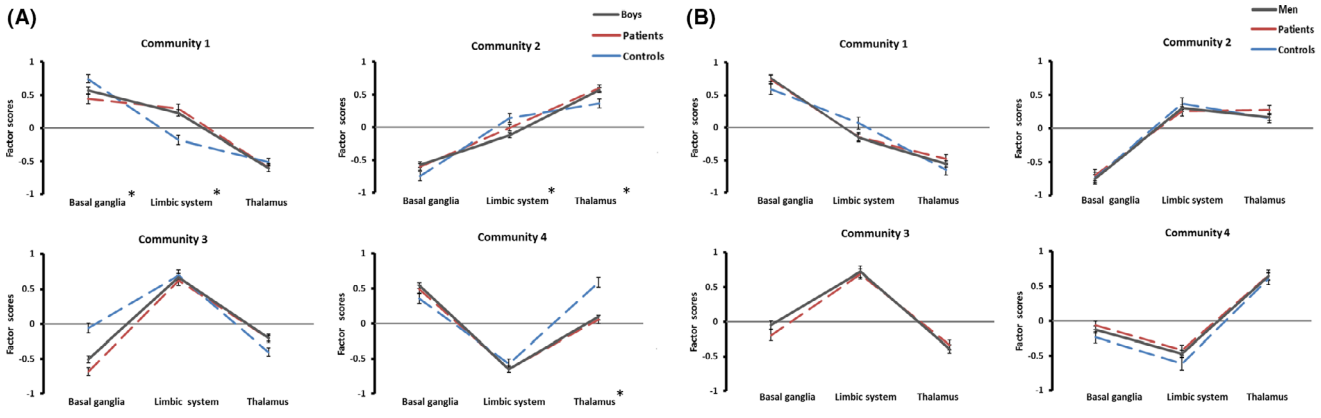

Figure 2 Communities generated by CD. A: Communities in boys; B: Communities in men. Note: Lines represent participants in each community from CD. Y-axis indicates the mean factor scores for each factor. Error bars: standard error of the mean. * indicates the difference of factor scores between patients and controls are significant

Table 3 The model fitness of EFA in each subsample

	Boys	Men	Girls	Women
TLI	0.92	0.98	0.86	0.98
BIC	−1.45	−13.8	−3.43	−12.8
RMSEA	0.07	0.04	0.10	0.04

supplementary analysis step, we also performed case–control comparisons for each individual subcortical volume in each community and in the entire sample, for both boys (see Table S4) and men (see Table S5). We observed several significant case–control differences in boys, and the effect size ranged from $d = -0.69$ (95%CIs [−0.95 to −0.62]) to $d = 0.57$ (95%CIs [0.31, 0.82]). In men, pallidum and amygdala volume for Community 1 survived FDR correction (pallidum: $d = 0.43$ (95%CIs [0.16 to 0.69]); amygdala: $d = -0.34$ (95%CIs [−0.60 to −0.07])). Importantly, the effect sizes of case–control differences within communities were larger than those of the whole subsample (Table 4, Tables S4 and S5).

ADHD clinical profiles and comorbidities in communities

Among boys with ADHD, information on the severity of IA and HI symptoms was available for $n = 355$ (63.0%) and $n = 358$ (63.5%), respectively (Table S6). This information was also available for 135 men with ADHD (32.8%; Table S7). Neither total ADHD symptoms nor IA/HI symptom levels differed between communities in either boys or men (not shown). For 491 (87.2%) boys with ADHD and 270 (65.5%) men with ADHD, we had information on IQ available (Tables S6 and S7); no association with the communities was observed for IQ (boys with ADHD: $F = 0.63$, $df = 3$, $p = .60$; men with ADHD: $F = 0.80$, $df = 3$, $p = .49$). For 517 (91.8%) boys with ADHD and 365 (88.6%) men with ADHD, information was also available on mediation use (Tables S6 and S7). There was not significant association between medication use and the communities (boys with ADHD: $\chi^2 = 0.85$, $p = .84$; men with ADHD: $\chi^2 = 4.08$, $p = .25$). For the analysis of comorbidities, we concentrated only on the presence or absence of common psychiatric comorbidities in ADHD, since the assessment of psychiatric comorbidities had been done using varied instruments across cohorts. Information was available for 311 (55.2%) boys with ADHD. Among them, 120 (38.6%) reported comorbid psychiatric disorders (Table S6). Anxiety and oppositional defiant disorder (ODD) were most frequently reported, occurring in 9.6% and 16.4%, respectively. There was neither a difference in the presence of (any) comorbidity between communities ($\chi^2 = 0.98$, $p = .81$), nor were anxiety or ODD more frequently reported in one community compared to any other (anxiety: $\chi^2 = 4.95$, $p = .18$; ODD: $\chi^2 = 5.09$, $p = .17$). In men with ADHD, 205 (49.8%) had available information; among them, 113 (55.1%) reported comorbid psychiatric disorders (Table S7). Mood disorder and substance use disorder (SUD) were most frequently reported, occurring in 32.4% and 22.9% of men with ADHD, respectively. The presence of (any) comorbidity was more frequent in Community 1 and Community 4 than in the other two communities ($\chi^2 = 15.63$, $p = .001$). Mood disorder and SUD were most frequent in Community 1 and Community 4 (mood disorder: $\chi^2 = 9.35$, $p = .02$; SUD: $\chi2 = 23.08$; $p = 2.0e\text{-}05$).

Discussion

In this study, we set out to investigate whether previously reported small effect sizes of case–control brain volume differences in ADHD might be explained by (structured) heterogeneity and whether parsing heterogeneity could identify behaviorally and/or clinically meaningful subgroups. Factor analysis of volumetric covariance indicated that the latent structure of subcortical volumes consists of basal ganglia, limbic system, and thalamus in male participants. Different latent factors seemed to underlie subcortical organization in females. Given

Table 4 Comparison of the mean of three-factor scores between ADHD patients and controls in each community

	Basal ganglia					Limbic system					Thalamus				
	Mean factor scores		*p* value	Adjusted *p* value	Cohen's *d* (95% CIs)	Mean factor scores		*p* value	Adjusted *p* value	Cohen's *d* (95% CIs)	Mean factor scores		*p* value	Adjusted *p* value	Cohen's *d* (95% CIs)
Community	Patients	Controls				Patients	Controls				Patients	Controls			
Boys	−0.10 (0.90)	0.13 (0.89)	**6.1e-05**	**4.1e-04**	−0.26 (−0.38 to −0.13)	−0.01 (0.83)	0.01 (0.84)	.74	.74	−0.02 (−0.15 to 0.10)	0.02 (0.81)	−0.03 (0.82)	.29	.43	0.07 (−0.06 to 0.19)
1	0.44 (0.74)	0.75 (0.71)	**1.8e-03**	**5.0e-03**	−0.42 (−0.67 to −0.17)	0.30 (0.72)	−0.18 (0.75)	**2.8e-06**	**6.3e-06**	0.65 (0.39 to 0.90)	−0.60 (0.67)	−0.51 (0.68)	.40	.44	−0.13 (−0.37 to 0.13)
2	−0.61 (0.64)	−0.75 (0.73)	.11	.23	0.21 (−0.04 to 0.46)	−0.10 (0.64)	0.14 (0.66)	**4.0e-03**	**.02**	−0.37 (−0.63 to −0.12)	0.60 (0.65)	0.37 (0.65)	**.01**	**.04**	0.37 (0.11 to 0.62)
3	−0.68 (0.72)	−0.06 (0.66)	**1.8e-10**	**4.8e-09**	−0.90 (−1.17 to −0.62)	0.62 (0.77)	0.70 (0.72)	.43	.50	−0.11 (−0.37 to 0.16)	−0.21 (0.63)	−0.40 (0.65)	**.02**	.08	0.30 (0.04 to 0.57)
4	0.49 (0.73)	0.36 (0.70)	.15	.26	0.18 (−0.07 to 0.43)	−0.65 (0.67)	−0.58 (0.70)	.41	.50	−0.10 (−0.35 to 0.15)	0.06 (0.77)	0.59 (0.71)	**6.6e-08**	**8.9e-07**	−0.71 (−0.97 to −0.45)
Cohen's *d* effect sizes*															
Men [a]	0.02 (0.88)	−0.03 (0.93)	.47	.53	0.06 (−0.10 to 0.22)	0.02 (0.85)	−0.04 (0.86)	.34	.44	0.08 (−0.08 to 0.24)	0.03 (0.85)	−0.06 (0.88)	.20	.34	0.10 (−0.06 to 0.26)
1	0.74 (0.72)	0.59 (0.77)	.13	.26	0.20 (−0.06 to 0.46)	−0.15 (0.80)	0.07 (0.83)	.05	.14	−0.27 (−0.53 to 0.00)	−0.48 (0.76)	−0.65 (0.77)	.09	.21	0.22 (−0.04 to 0.49)
2	−0.70 (0.71)	−0.74 (0.77)	.72	.74	0.06 (−0.26 to 0.37)	0.26 (0.63)	0.37 (0.74)	.30	.43	−0.16 (−0.48 to 0.15)	0.27 (0.68)	0.15 (0.59)	.24	.38	0.19 (−0.13 to 0.51)
3	−0.20 (0.71)	NA	NA	NA	NA	0.69 (0.83)	NA	NA	NA	NA	−0.34 (0.63)	NA	NA	NA	NA
4	−0.07 (0.65)	−0.24 (0.66)	.09	0.21	0.26 (−0.04 to 0.56)	−0.42 (0.72)	−0.62 (0.71)	.07	.19	0.27 (−0.02 to 0.58)	0.66 (0.73)	0.61 (0.70)	.65	.70	0.07 (−0.23 to 0.37)
Cohen's *d* effect sizes*															

Adjusted *p* value: FDR correction across age bins. Significant difference in bold. 95% CIs: 95% Confidence intervals. NA = not available.

[a]Community 3 is absent in men, because no healthy controls were presented.

*Cohen's s effect sizes come from *t*-test that compared mean factor scores between ADHD patients and healthy controls within each community.

sample sizes considerations, we concentrated all subsequent analyses on males. Among them, we discerned four distinct communities, one of which did not comprise any healthy adult males. In the subsample of boys, effect sizes of several case–control differences were larger within specific communities than in the total sample. The substructure of the brain volumes did not seem to have a behavioral correlate at the level of ADHD symptom severity, but men with ADHD in two communities more frequently reported the presence of comorbidities than those within the other two communities.

Similar factor structures of subcortical brain volumes existed in boys and men, regardless of ADHD status. The observed three-factor structure—basal ganglia, limbic system, and thalamus—is consistent with functional neuroanatomy and neurodevelopmental connections (Hibar et al., 2015). Interestingly, factor structures differed between male and female participants and also among females across the life span. Sex differences in subcortical brain volumes have consistently been reported in previous studies. Some studies reported larger volumes of amygdala, pallidum, and putamen in males (Cheng et al., 2009; Rijpkema et al., 2012). Other studies observed larger hippocampus, caudate nucleus, and thalamus in females (Kiraly et al., 2016; Luders, Gaser, Narr, & Toga, 2009; Takahashi, Ishii, Kakigi, & Yokoyama, 2011). However, this is the first paper to report on different correlations between subcortical structures in the two sexes. It is interesting to speculate whether such differences in subcortical brain volume organization may be related to differences in ADHD presentation and comorbidity profiles between sexes.

Both boys and men could be separated into communities based on subcortical volume modularity. The community structure observed was similar in cases and controls, as has been observed also in cognitive investigations of ADHD (Fair et al., 2012; Mostert et al., 2018), providing further evidence that heterogeneity among individuals with ADHD is 'nested' in normal variation (Fair et al., 2012). In the present study, four communities were observed in boys with and without ADHD, and in men with ADHD, while in healthy men, only three communities were present. It seemed like community structure in healthy men simplified from four to three communities, while patients retained a four-community distribution. Using the same methodology in the ENIGMA-ASD cohorts, we found a similar community structure in individuals with ASD and healthy controls; the number of communities went from four in boys to three in male adolescents and men (Li et al., 2020). The retention of the four-community structure in ADHD may thus be consistent with findings of delayed maturation in ADHD (Hoogman, Bralten, et al., 2017; Hoogman et al., 2019; Shaw et al., 2018), but more research in longitudinal samples is clearly needed.

Effect sizes for case–control differences reported for subcortical volumes have always been small. The largest study of subcortical brain volumes in ADHD, performed by the ENIGMA-ADHD Working Group, reported effect sizes ranging from $d = -0.19$ to -0.10 across the life span, with largest effects in children (Hoogman, Bralten, et al., 2017). Case–control differences within each community showed that (a) not every community had significant differences for a specific volume, and (b) among those communities showing significant differences at the factor level, effect sizes ranged from $d = -0.90$ (95%CIs [−1.17, −0.62]) to 0.65 (95%CIs [0.39, 0.90]), which were considerably larger than the largest effect size observed in the full cohort, which was −0.26 (95% CIs [−0.38, −0.13]; Table 2). Similar trends were also present for individual subcortical brain volumes (Tables S4 and S5). The current results highlight the neuroanatomical heterogeneity in the population and suggest that brain-based ADHD subtypes may exist.

As in the ENIGMA-ADHD and previous meta-analyses, case–control differences in the basal ganglia factor all pointed to smaller volumes in ADHD patients (Hoogman, Bralten, et al., 2017). More differentiated results were observed for the limbic system and thalamus. For the limbic system (and its components, amygdala, and hippocampus), larger volumes were seen in boys with ADHD in Community 1, whereas the cases in Community 2 had smaller volumes. For the thalamus, we observed larger volumes in individuals with ADHD in Community 2 and Community 3, whereas those with ADHD in Community 4 had smaller volumes than healthy controls. Such findings indicate that the direction of ADHD effects on subcortical volumes may differ between communities. The effects may (partially) be canceled out if analyzing the whole sample containing such subgroups. Looking at the inconsistent findings reported in literatures, different studies may have thus included different proportions of specific ADHD communities. Our finding may therefore reconcile inconsistencies in the direction of ADHD effects reported on subcortical volumes in previous studies. Case–control differences were not significant in adult males. This result corroborates the earlier findings that developmental brain-structural differences observed with MRI in ADHD may normalize in adulthood (Hoogman, Bralten, et al., 2017; Hoogman et al., 2019; Shaw et al., 2018). To analyze the significance of the brain structure-based communities for clinical presentation of ADHD, we explored potential differences between ADHD patients in the different communities. The communities did not appear to be associated with the severity of ADHD symptoms, IQ, and medication use. This might have been a result of our limited sample size for these analyses, but our earlier study did not reveal significant association between subcortical volumes and ADHD symptoms

score using meta-analysis (Hoogman, Bralten, et al., 2017). Similarly, our earlier study also found IQ did not account for case–control difference in subcortical volumes and no effect of medication use on subcortical volumes in patients (Hoogman, Buitelaar, et al., 2017). We did find some indication of clinical relevance of the communities when analyzing the presence of comorbidity: adult males with ADHD in Community 1 and Community 4 more frequently reported comorbidities than those in the other two communities, in particular mood disorder and SUD. Community 1 and Community 4 were characterized by relatively larger basal ganglia across the entire sample, which may be consistent with a previous study reporting increased basal ganglia volume in long-term substance abusers (Moreno-Alcázar et al., 2018). The lack of significant associations with symptom severity and the limited findings for comorbidities may be due to insufficient power of the analyses in individual communities. Replication in independent samples with larger sample sizes is needed.

The strengths of the current study include the use of the large sample size of the ENIGMA-ADHD dataset to explore neuroanatomic subgroups, which provides us with the opportunity to better understand the small effect sizes of case–control differences in ADHD. The ENIGMA-ADHD dataset converged brain imaging data from over 35 cohorts, which were analyzed using standardized protocols to harmonize segmentation and quality control processes. The large sample size and harmonized procedures may blend random fluctuations in each cohort. A potential limitation is the arbitrariness of using the modularity algorithm; the application of different classification methodologies could result in different communities. However, in this study, we applied a widely used technique and obtained a consistent approximation across two subsamples (boys and men). Our group has applied the same modularity algorithm for neuropsychological performance, which included six input variables (Mostert et al., 2018). From that study, we learned that CD may lead to instable results when there are too many degrees of freedom. In the current study, we thus explored the latent structure in subcortical brain volumes by applying EFA, which enabling us to constrain the complexity of input variables before running CD. EFA reduced the brain structure dimensions from 7 to 3, which was necessary to allow stabilization of the CD and allowed us to identify similar communities in each subgroup. EFA also provided interesting information resulted in the basic organization of the subcortical brain volumes and served as a check of underlying correlations of subcortical structures correlations between groups. A second limitation was the heterogeneity of the ENIGMA-ADHD dataset, where several different diagnostic instruments had been used, and the fact that sample sizes were dramatically lower when we examined associations within single communities. Third, we only focused on subcortical brain volumes in this study, as these have been most consistently associated with ADHD. However, differences between ADHD patients and controls are also observed in cortical measures, especially in surface area (Hoogman et al., 2019). Therefore, the clinical relevance of communities might be increased when cortical features are taken into account. Lastly, because factor structures differed between males and females, we only applied CD analyses in the male samples which provided more power than the female samples due to their larger sample sizes. The neuroanatomic profiles of subcortical brain volumes in females and the heterogeneity among genders require further study.

To conclude, using subcortical MRI data from the ENIGMA-ADHD Working Group, we were able to stratify our sample into neuroanatomically more homogeneous subgroups with preliminary links to the clinical presentation of ADHD. Our study may provide groundwork for future studies that parse neuroanatomical heterogeneity to increase our understanding of ADHD biology and pathology.

Supporting information

Additional supporting information may be found online in the Supporting Information section at the end of the article:

Table S1. Additional information of methods in each participating site.
Table S2. The distribution of boys (patients/controls) in each community.
Table S3. The distribution of adult men (patients/controls) in each community.
Table S4. Comparison of subcortical brain volumes of each community in subsample of boys.
Table S5. Comparison of subcortical brain volumes of each community in subsample of adult men.
Table S6. ADHD symptoms, IQ, comorbidities, and medication information in each cohort in boys with ADHD.
Table S7. ADHD symptoms, IQ, comorbidities and mediation information in each cohort in adult men with ADHD.
Figure S1. The three-factor model that was generated by EFA in girls with estimated factor loadings of the latent factors.
Figure S2. The three-factor model that was generated by EFA in adult women with estimated factor loadings of the latent factors.
Figure S3. Variation of Information in each subsample.
Figure S4. Scree plot of EFA in each subsample.

Acknowledgements

T.L. is supported by China Scholarship Council (CSC) under the Grant CSC n° 201507720006. ENIGMA received funding from the National Institutes of Health

(NIH) Consortium grant U54 EB020403, supported by a cross-NIH alliance that funds Big Data to Knowledge Centers of Excellence (BD2K). Support was also received from the European Community's Horizon 2020 Programme (H2020/2014–2020) under grant agreements no. 667302 (CoCA) and no. 728018 (Eat2beNICE). M.H. and B.F. were supported by personal grants from the Netherlands Organization for Scientific Research (NWO) Innovation Program (Veni grant 91619115 to MH; Vici grant 016-130-669 to BF). Lastly, the authors also gratefully acknowledge support from the European College for Neuropsychopharmacology (ECNP) for the ECNP Network ADHD across the Lifespan.

J.K.B. has been in the past 3 years a consultant to / member of advisory board of / and/or speaker for Shire, Roche, Medice, and Servier. He is not an employee of any of these companies, and not a stock shareholder of any of these companies. He has no other financial or material support, including expert testimony, patents, royalties. In the past year, S.V.F., a contributing member of the ENIGMA-ADHD Working Group, received income, potential income, travel expenses continuing education support and/or research support from Tris, Otsuka, Arbor, Ironshore, Shire, Akili Interactive Labs, Enzymotec, Sunovion, Supernus, and Genomind. With his institution, he has US patent US20130217707 A1 for the use of sodium-hydrogen exchange inhibitors in the treatment of ADHD. He also receives royalties from books published by Guilford Press: Straight Talk about Your Child's Mental Health, Oxford University Press: Schizophrenia: The Facts, and Elsevier: ADHD: Non-Pharmacologic Interventions. He is Program Director of www.adhdinadults.com. B.F. has received educational speaking fees from Medice. The remaining authors have declared that they have no competing or potential conflicts of interest.

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Key points

- Neuroanatomic heterogeneity limits our understanding of the etiology of attention-deficit/hyperactivity disorder (ADHD). The objective is to parse neuroanatomic heterogeneity of ADHD and determine whether subgroups could be discerned in patients based on subcortical volumes.
- The study indicate that neuroanatomic heterogeneity in subcortical volumes exists, with ADHD patients and controls showing similar patterns. Effect sizes of case–control differences appear more pronounced in some of the four observed subgroups.

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Appendix 1

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Accepted for publication: 9 December 2020