

Sex differences in structural brain asymmetry of children with autism spectrum disorders

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Previous studies have confirmed the sex difference of gray matter asymmetry in typically developing controls and the abnormal gray matter asymmetry in autism spectrum disorders. However, whether and how sex differences of gray matter asymmetry exist in autism spectrum disorders remains studied. This paper analyzes the above issues and explores correlations between gray matter asymmetry and autistic symptoms. Data from 72 children (36 males and 36 females) with autism spectrum disorders and 72 typically developing-controls (36 males and 36 females) between 8 and 14 years were included and obtained from the autism brain imaging data exchange repository (autism brain imaging data exchange I and autism brain imaging data exchange II). The voxel-based morphometry approach was used to assess gray matter asymmetry in T1-weighted brain data, and gray matter asymmetry was quantified as asymmetry index. A 2×2 analysis of covariance was used to identify asymmetry index differences among the four groups. Pearson correlation analysis was performed for asymmetry index values extracted from the clusters with significant differences between the four groups and autistic symptoms (social impairments, communication difficulties, and restricted, repetitive behaviors) measured by the revised autism diagnostic interview scale. Results showed that specific brain regions showed significant main effects for diagnosis in which autism spectrum disorders patients had more leftward asymmetry than typically developing-controls for the parahippocampal gyrus and the post-central gyrus; specific brain regions showed significant main effects for sex in which females showed more rightward asymmetry for the middle temporal gyrus, inferior frontal gyrus, angular gyrus, and postcentral gyrus and minor rightward asymmetry for the superior frontal gyrus than males; significant diagnosis \times sex interaction effects were identified in the angular gyrus and middle occipital gyrus. Pearson correlation analysis showed that males with autism spectrum disorders had a positive association between the asymmetry index value in the middle occipital gyrus and more significant verbal impairment measured by the revised autism diagnostic interview ($r = 0.387$, $p = 0.026$). The asymmetry index value in the parahippocampal gyrus was positively associated with more severe social impairment in females with autism spectrum disorders ($r = 0.422$, $p = 0.020$). We identified that the sex difference of gray matter asymmetry in children with autism spectrum disorders is qualitative rather than quantitative, which is relatively novel. Our findings provide the theoretical basis for conducting separate studies and using sex-

specific diagnostic methods and treatments for males and females children with autism spectrum disorders.

Keywords

Autism spectrum disorders; Gray matter asymmetry; Asymmetry index; Sex factors; Brain imaging

1. Introduction

Autism spectrum disorders (ASD) are a set of pervasive neurodevelopmental conditions characterized by social impairments, communication difficulties, and restricted, repetitive behaviors [1]. It is one of the most common developmental disorders, affecting about 1 in 160 people worldwide [2]. Despite its prevalence, studies have not been able to determine its exact etiology [3]. The imbalanced male-to-female prevalence ratio is one of the most apparent characteristics of ASD [2], and the sex bias for males is even higher in neuroimaging studies [4, 5]. There are also symptom differences between males and females with ASD [6]; males have lower levels of social impairments and externalizing problems [7] but more significant restricted behaviors compared to females with ASD [8]. Thus, sex is increasingly being recognized as the source of neuropathophysiological heterogeneity in ASD.

One of the most striking features of the human brain is its structural or functional lateralization or asymmetry. In most typically developing-controls (TDs), the left hemisphere specializes in motor control and language, while the right hemisphere is responsible for visual-spatial attention [9]. And in TDs, studies have found that numerous brain regions showed asymmetry differences in structure (especially gray matter) or function between males and females [10, 11]. Alterations of lateralization or asymmetry have been found in various mental and neurocognitive disorders, including schizophrenia and dyslexia [12, 13]. And atypical brain asymmetry has long been hypothesized for ASD, a supposition that is supported by multiple neuroimaging studies [14–16]. Several structure-based studies reported hemispheric asymmetry abnormalities in ASD, but discoveries of potential structural alterations for ASD have been inconsistent. For example,

Groen *et al.* [17] reported increased hippocampal volume in ASD. On the other hand, Eilam-Stock *et al.* [18] found significantly decreased hippocampal volume in subjects with ASD compared with TDs. Therefore, further structure-based researches on the hemispheric brain asymmetry of ASD are necessary.

Due to the relatively high prevalence of ASD in males, almost all existing studies on the brain structure of ASD have focused on samples that are predominantly or exclusively male. However, there is growing evidence that males and females with ASD have different brain structural alteration patterns. For example, [19] reported that females might need higher detrimental loads, including brain structural alterations, before developing clinically relevant levels of autistic characteristics. In addition, ASD-associated neural anatomy in females may differ from that in males; Ecker *et al.* [20] detected that temporal lobe cortical thickness was decreased and increased in females and males with ASD, respectively. Unfortunately, potential sex differences in brain structure have often been ignored in ASD. This may be one of the main reasons for the inconsistent results of brain structure research in ASD. Thus, the inclusion of females with ASD is critical to help improve our understanding of brain structural alterations in ASD.

Based on the findings of the sex difference of gray matter (GM) asymmetry in TDs and the abnormality of GM asymmetry in ASD, quantitative or qualitative sex differences of the GM asymmetry alterations in ASD are probably to be expected [11, 21]. However, if the effects on GM asymmetry in males and females with ASD are present in only one sex or the opposite form, previous research on GM asymmetry of ASD that did not consider the sex factor may be unreliable, as the above effects may be offset in these researches. This significantly reduced the possibility of detecting actual effects. Therefore, a sex-specific analysis is urgently needed to determine whether and how the sex differences of brain structural asymmetry might be altered in ASD to better understand the neuroanatomy of the situation.

Recently, Kurth *et al.* [22] established a fully automated voxel-based morphometry (VBM) method to specifically analyze brain GM asymmetry. A key advantage over the standard VBM workflow is establishing the voxel-wise hemispheric correspondence, which is ensured by using Diffeomorphic Anatomical Registration Lie (DARTEL) algebra to convert spatial normalization into a symmetric space. Several studies have used this approach to investigate the effects of sex, meditation, alcohol dependence, and schizophrenia on GM asymmetry [22–24].

Here, we used the advanced VBM method [25] to conduct a sex-specific study of GM asymmetry in ASD. GM asymmetry was quantified as asymmetry index (AI). We also explored correlations between AI values extracted from the clusters with significant differences between the four groups and autistic symptoms (social impairments, communication difficulties, and restricted, repetitive behaviors) measured by

the revised autism diagnostic interview (ADI-R) scale.

2. Participants and methods

2.1 Participants

The participants included 72 children (36 males and 36 females) with ASD and 72 TDs (36 males and 36 females) between 8 and 14. We used Gpower (<https://www.psychologie.hhu.de/arbeitsgruppen/allgemeine-psychologie-und-arbeitspsychologie/gpower.html>) software to calculate the sample size. The power and the required effect size in this paper are 0.85 and 0.24, respectively. All participants were obtained from the autism brain imaging data exchange (ABIDE) repository (ABIDE I and ABIDE II). The dataset consists of 1060 individuals with ASD and 1166 TDs, aged 6–65, collected from 36 international sites. Data were collected from five sites (KKI, NYU, OHSU, YALE, GU) that included five or more females with ASD who met the criteria. All subjects with ASD had a clinician's Diagnostic and Statistical Manual of Mental Disorders (DSM)-IV-TR or DSM-V diagnosis. The majority of participants were assessed using Autism Diagnostic Observation Schedule (ADOS) modules 3 or 4, ADI-R, or both. The ABIDE database also provided information on medications and comorbidities. The female ASD group was formed first because most sites included a few females with ASD. To avoid biases due to the different sites, random subjects were selected to form age-, sex- and site-matched groups. All four groups were formed with the following criteria: (1) age range: 8–14 years, (2) full-scale intelligence quotient (FIQ) ≥ 70 , and (3) right-handedness. Those with poor image quality were excluded. The Wechsler Intelligence Scale measured the FIQ for Children (KKI, GU, OHSU), the Differential Ability Scales (YALE), or the four subtests of the Wechsler Abbreviated Scale of Intelligence (NYU). Handedness was measured by the 22-item Edinburgh Handedness Inventory (KKI, NYU, OHSU) or parent report (YALE, GU). All data were provided by the site and published on the ABIDE website.

2.2 MRI data acquisition

All included data were acquired from 3.0-Tesla scanners, but the scanner types and image acquisition parameters varied across sites. The scan parameters and acquisition protocols are provided at http://fcon_1000.projects.nitrc.org/indi/abide/.

2.3 Image analysis

2.3.1 MRI data processing

The high-resolution 3D T1-weighted images were preprocessed using the VBM8 toolbox (<http://dbm.neuro.uni-jena.de/vbm8>) package Statistical Parametric Mapping (SPM8, <http://www.fil.ion.ucl.ac.uk/spm>) software. Before preprocessing, visual checks were performed for all 3D T1-weighted images. Then, all preprocessing steps were performed following the protocol by Kurth *et al.* [22]: (1) The 3D T1-weighted images were segmented into GM, white

matter (WM), and cerebrospinal fluid; (2) the segmented images were flipped at the midline; (3) the original and flipped GM and WM versions of each participant were used to create a symmetric DARTEL anatomical template; (4) the original and flipped GM and WM tissue segments of all participants were warped into the created symmetric template to align the brain of each participant in a shared space; and (5) to limit further data analysis to the right hemisphere, we used the symmetric template space to create a right-hemispheric mask in MRICron (<http://people.cas.sc.edu/rorden/mricron/index.html>). First, in the sagittal window of MRICron, we selected one plane to the right in the midline line of GM. This plane was marked as a volume of interest using the drawing tool. Then, masking was repeated in the next plane until the right hemisphere was finished. Finally, the file was saved in NIfTI format.

2.3.2 Global GM asymmetry analysis

The AI value of GM volume was calculated with an automated procedure. This procedure used the ‘calculate’ script provided by [22]. First, type ‘calculate’ in MATLAB’s command window to run the script and select ‘Step 6’, then the script asks for the original warped GM images and the hemispheric mask generated in the above steps [22]. This step generates the masked AI images of each participant. The formula in this script is [22]:

$$AI = ((i1 - i2) / ((i1 + i2) \times 0.5)) \times i3$$

where $i1$ is the original warped image, $i2$ is the flipped warped image, and $i3$ is the right-hemispheric mask image.

All AI maps were spatially smoothed with an 8-mm Gaussian smoothing kernel. All results were limited in the right hemisphere, and a positive AI indicated rightward asymmetry (larger right-hemispheric GM volume in a given cluster). Conversely, a negative AI indicated leftward asymmetry (larger left-hemispheric GM volume in a given cluster) [22, 26].

2.4 Statistical analyses

A 2×2 analysis of covariance (ANCOVA) was used to assess the main effect of diagnosis, sex, and their interactions (Age, FIQ, and the site were covariates) for all AI maps SPM8. The Gaussian random field (GRF) theory correction was applied (voxel-level $p < 0.01$, cluster-level $p < 0.05$) to determine the significant regions [25].

The AI values were extracted using Data Preprocessing Assistant for rs-fMRI (DPARSF) software from the clusters with significant differences in the 2×2 ANCOVA. Then the simple-effect analysis was implemented with SPSS 20.0 software for extracted AI values of the interaction effect. Finally, Pearson correlation analysis was performed for all extracted AI values and ASD clinical symptoms (social impairments, communication difficulties, and restricted, repetitive behaviors) measured by ADI-R scale, $p < 0.05$ (uncorrected) was used to determine the statistically significant correlation.

3. Results

3.1 Demographic and clinical details

Descriptive statistics are presented in Table 1. The four groups did not significantly differ in age ($p > 0.05$), but the ASD had lower FIQ than TDs (diagnosis effect) ($F = 25.47$, $p < 0.001$). There were no significant differences between females and males with ASD concerning medications, comorbidities, and scores of ADI-R and ADOS modules 3 or 4 (all $p > 0.05$).

3.2 The result of 2×2 ANCOVA for AI

All brain regions in results were reported using the Automated Anatomical Labeling template.

3.2.1 Diagnosis effect

Specific brain regions showed significant main effects for diagnosis. The ASD patients had more leftward asymmetry than TDs for the parahippocampal gyrus and postcentral gyrus (GRF correction, voxel-level $p < 0.01$, cluster-level $p < 0.05$; Fig. 1).

3.2.2 Sex effect

Relative to males, females showed significantly more rightward asymmetry in the middle temporal gyrus, inferior frontal gyrus, angular gyrus, and postcentral gyrus and minor rightward asymmetry in the superior frontal gyrus (GRF correction, voxel-level $p < 0.01$, cluster-level $p < 0.05$; Fig. 2).

3.2.3 Diagnosis-by-sex interaction

Significant diagnosis \times sex interaction effects were identified in the angular gyrus and middle occipital gyrus (GRF correction, voxel-level $p < 0.01$, cluster-level $p < 0.05$; Fig. 3). Results of the simple-effect analysis were as follows: Females with ASD showed more, and males with ASD showed less rightward asymmetry relative to their respective TDs in the angular gyrus (all $p < 0.05$) and the middle occipital gyrus ($p < 0.05$ and $p > 0.05$ respectively). The detailed results of the simple-effect analysis are shown in Tables 2 and 3. All extracted AI values are listed in Table 4.

3.3 Brain-behavior relationships

We observed significant correlations between extracted AI values and clinical severity in the ASD group. Specifically, males with ASD showed a positive association between the AI value in the middle occipital gyrus and more significant verbal impairment measured by ADI-R ($r = 0.387$, $p = 0.026$; Fig. 4). In addition, the AI value in the parahippocampal gyrus was positively associated with more severe social impairment in females with ASD ($r = 0.422$, $p = 0.020$; Fig. 5 and Table 5).

4. Discussion

We mapped differences in GM asymmetry between ASD and TDs and between the male and female children with ASD. Previous studies on cortical asymmetry in ASD found variable differences or no differences [27]. Although Postema *et al.* [27] reported that total hemispheric results

Table 1. Group characteristics for the included male and female participants, separately for the ASD and TD groups.

	Males with ASD		Females with ASD		Males TDs		Females TDs		Diagnosis effect		Sex effect		Diagnosis × Sex	
	n	mean ± SD (rate)	n	mean ± SD (rate)	n	mean ± SD	n	mean ± SD	F-value	p-value	F(t)-value	p-value	F-value	p-value
Age (year)	36	10.88 ± 1.89	36	10.80 ± 2.03	36	10.82 ± 1.96	36	10.83 ± 1.96	0.002	0.965	0.008	0.930	0.021	0.885
FIQ	36	107.64 ± 17.35	36	102.61 ± 15.34	36	117.75 ± 10.65	36	116.64 ± 13.14	25.466**	0.000	1.647	0.201	0.670	0.414
ADI-R														
Social	33	19.52 ± 5.57	30	18.91 ± 6.16							0.251	0.677		
Verbal	33	15.36 ± 4.27	30	14.52 ± 4.83							−0.150	0.452		
RRB	33	5.68 ± 2.69	30	5.21 ± 2.52							−0.523	0.462		
ADOS														
Total	14	10.08 ± 4.27	18	12.28 ± 3.10							−1.665	0.107		
Social	14	3.46 ± 1.21	18	3.33 ± 0.77							0.282	0.782		
Communication	14	6.23 ± 3.06	18	8.11 ± 1.20							−2.073	0.067		
RRB	14	2.31 ± 1.60	18	2.39 ± 1.65							−0.137	0.892		
Comorbidity														
ADHD	26	10/26	26	9/26							0.083	0.773		
Medication	31	12/31	31	9/31							0.421	0.648		

Notes: ASD, autism spectrum disorders; TDs, typically developing controls; FIQ, full-scale intelligence quotient; SD, standard deviation; ADI-R, revised autism diagnostic interview scale; ADOS, autism diagnostic observation schedule; RRB, restricted repetitive behaviors; ADHD, attention deficit hyperactivity disorder, ** $p \leq 0.01$.

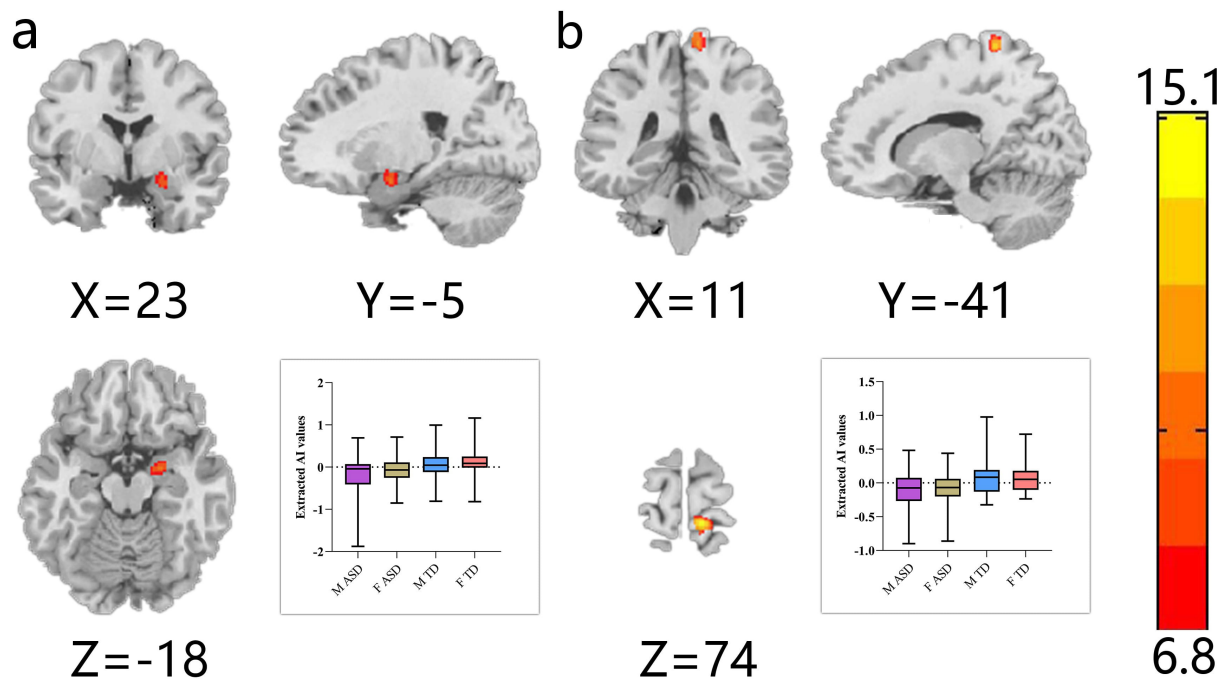


Fig. 1. The map for brain regions showing a statistically significant main effect of diagnosis in the 2 × 2 analysis of covariance for asymmetry index and the histograms for extracted AI values in those brain regions (GRF correction, voxel-level $p < 0.01$, cluster-level $p < 0.05$; (a) Parahippocampal gyrus; (b) Postcentral gyrus; The left side of the figure corresponds to the left cerebral hemisphere).

showed generalized less leftward asymmetry of cortical thickness in ASD, the direction of GM asymmetrical alterations in specific brain regions remains to be established. Using the advanced VBM approach, we found more leftward asymmetry of GM volumes in the parahippocampal gyrus and postcentral gyrus in ASD.

The parahippocampal gyrus is essential for ASD. Abnormalities in this brain region have been reported in previous

studies on ASD. Mei *et al.* [28] reported decreased density in the parahippocampal gyrus in ASD. Yang *et al.* [29] found that the GM volume in the parahippocampal gyrus in adults with ASD was significantly increased relative to TDs. In addition, the AI value in the parahippocampal gyrus was positively associated with more severe social impairment in females with ASD ($r = 0.422$, $p = 0.020$). This result is consistent with previous studies that linked structural alterations

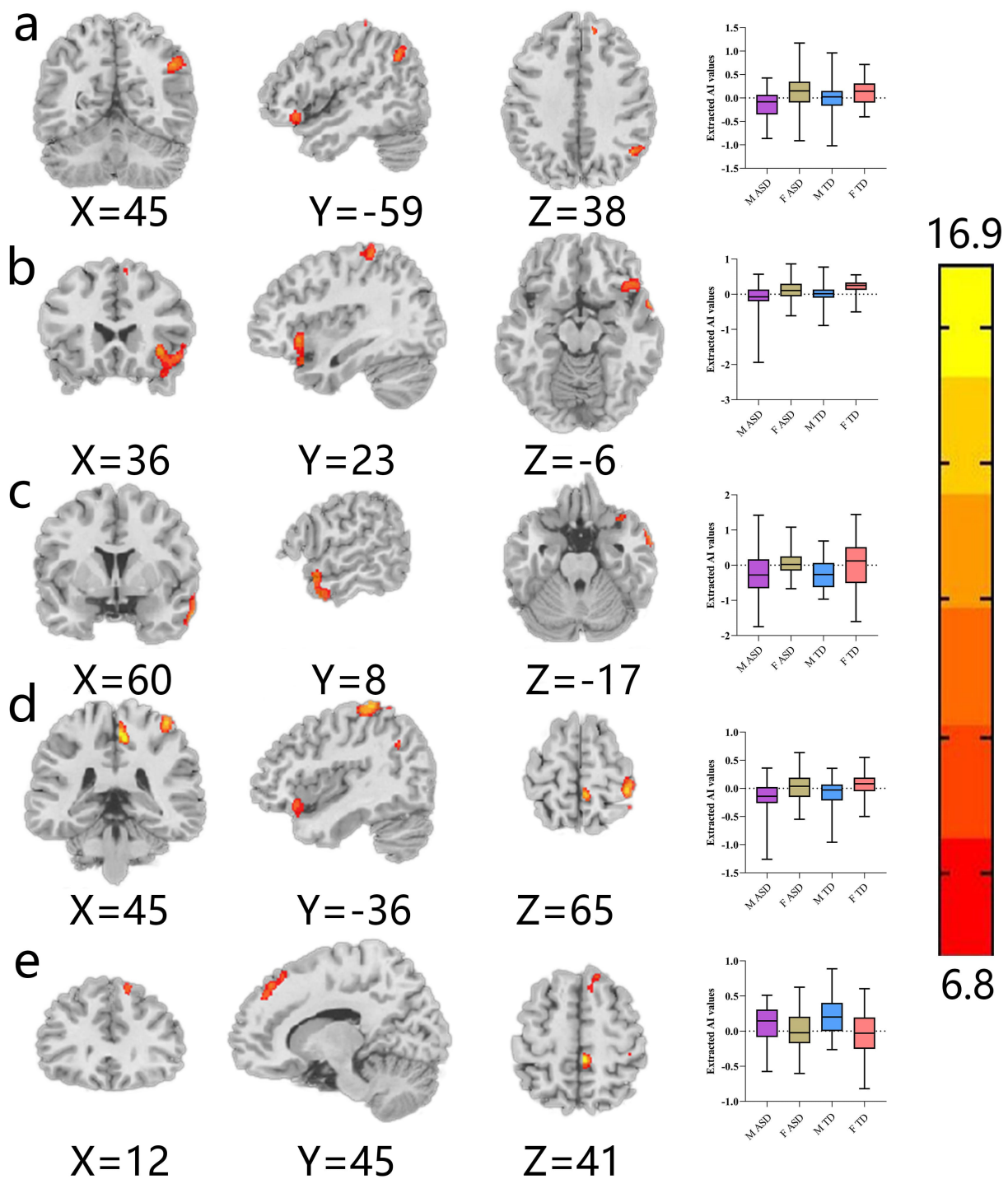


Fig. 2. The map for brain regions showing a statistically significant main effect of sex in the 2×2 analysis of covariance for asymmetry index and the histograms for extracted AI values in those brain regions (GRF correction, voxel-level $p < 0.01$, cluster-level $p < 0.05$; (a) Angular gyrus; (b) Inferior frontal gyrus; (c) Middle temporal gyrus; (d) Postcentral gyrus; (e) Superior frontal gyrus; The left side of the figure corresponds to the left cerebral hemisphere).

in the parahippocampal gyrus to social deficits in ASD [30], indicating that structural alterations of the parahippocampal gyrus indeed serve a junctional role in ASD etiology.

Concerning the postcentral gyrus, volume abnormalities reported in this brain region in ASD have been inconsistent

[31]. Mizuno *et al.* [31] reported decreased left postcentral gyrus volume in ASD, while Brieber *et al.* [32] described increased volume in this brain region in patients with ASD comparing with TDs. This may be due to the small sample sizes, the different proportions of females with ASD, and the

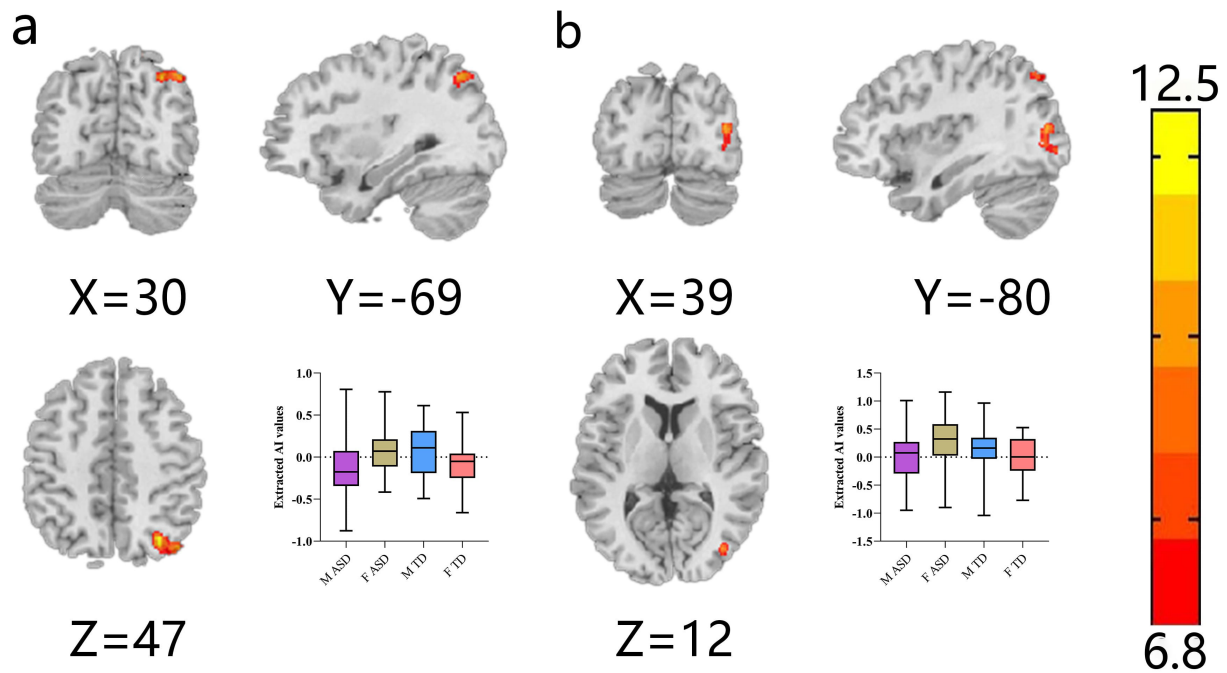


Fig. 3. The map for brain regions showing a statistically significant main effect of diagnosis \times sex in the 2×2 analysis of covariance for asymmetry index and the histograms for extracted AI values in those brain regions (GRF correction, voxel-level $p < 0.01$, cluster-level $p < 0.05$; (a) Angular gyrus; (b) Middle occipital gyrus; The left side of the figure corresponds to the left cerebral hemisphere).

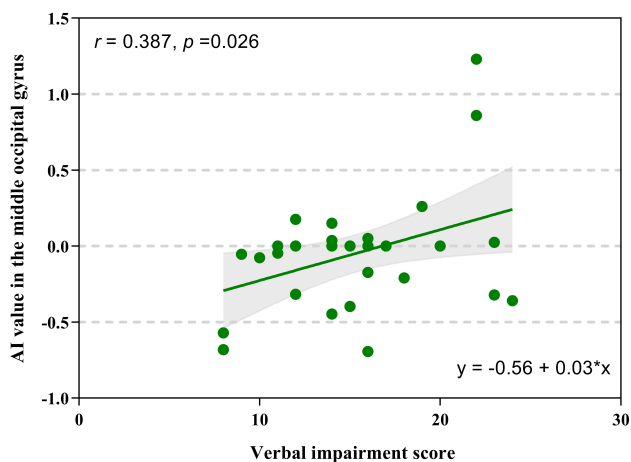


Fig. 4. The correlation between the asymmetry index value in the middle occipital gyrus and the verbal impairment score measured by the revised autism diagnostic interview scale in males with autism spectrum disorders ($r = 0.387$, $p = 0.026$).

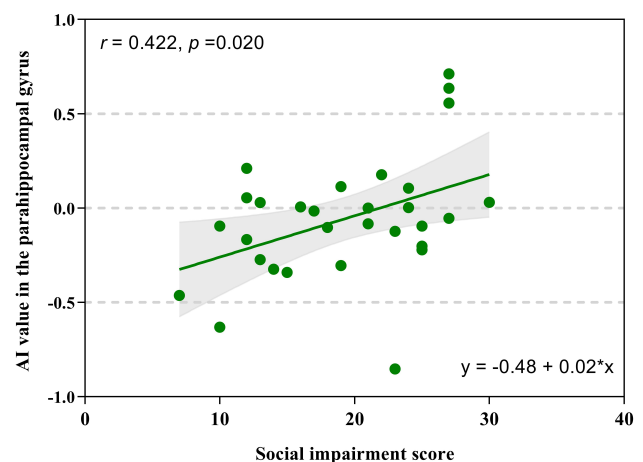


Fig. 5. The revised autism diagnostic interview scale measured the correlation between the asymmetry index value in the parahippocampal gyrus and social impairment score in females with autism spectrum disorders ($r = 0.422$, $p = 0.020$).

different image processing techniques among different studies. We used an advanced VBM method specifically designed to assess brain structural asymmetry, had a relatively large sample size, and considered the sex factor. Therefore, our findings are more reliable and can provide more substantial evidence to clarify the inconsistent findings reported earlier. The postcentral gyrus participates in somatosensory functions. Abnormal sensory processing can lead to social impairments, communication difficulties, and restricted, repetitive behaviors, which are core characteristics of ASD [33]. There-

fore, the abnormality of the postcentral gyrus may cause the core symptoms of ASD.

The sex difference of GM asymmetry was found in the angular gyrus in ASD. Males and females with ASD showed more minor and more rightward asymmetry in the angular gyrus, respectively, compared with their sex-matched TDs. A similar reduction in the angular gyrus was previously detected in [34] with the mixed-sex ASD sample (mainly males with ASD). Thus, less rightward asymmetry in the angular

Table 2. The result of the simple-effect analysis for the interaction effect in the angular gyrus.

Diagnosis	(I)sex	(J)sex	Mean Difference (I-J)	Std. Error	Sig ^b
ASD	Male	Female	-0.239*	0.068	0.001
	Female	Male	0.239*	0.068	0.001
TD	Male	Female	0.162*	0.068	0.019
	Female	Male	-0.162*	0.068	0.019
Sex	(I)diagnosis	(J) diagnosis	Mean Difference (I-J)	Std. Error	Sig ^b
Male	ASD	TD	-0.245*	0.068	0.000
	TD	ASD	0.245*	0.068	0.000
Female	ASD	TD	0.155*	0.068	0.024
	TD	ASD	-0.155*	0.068	0.024

Notes: Based on estimated marginal means. * The mean difference is significant at the 0.05 level. ^b Adjustment for multiple comparisons: Sidak. ASD, autism spectrum disorder; TDs, typically developing controls.

Table 3. The result of the simple-effect analysis for the interaction effect in the middle occipital gyrus.

Diagnosis	(I)sex	(J)sex	Mean Difference (I-J)	Std. Error	Sig ^b
ASD	Male	Female	-0.316*	0.092	0.001
	Female	Male	0.316*	0.092	0.001
TD	Male	Female	0.168	0.092	0.070
	Female	Male	-0.168	0.092	0.070
Sex	(I)diagnosis	(J) diagnosis	Mean Difference (I-J)	Std. Error	Sig ^b
Male	ASD	TD	-0.179	0.092	0.054
	TD	ASD	0.179	0.092	0.054
Female	ASD	TD	0.305*	0.092	0.001
	TD	ASD	-0.305*	0.092	0.001

Notes: Based on estimated marginal means. * The mean difference is significant at the 0.05 level. ^b Adjustment for multiple comparisons: Sidak. ASD, autism spectrum disorders; TDs, typically developing controls.

gyrus seems to be unique to males with ASD. Interestingly, this brain area is involved in the theory of mind, which is essential to help the human brain reason about others and effectively communicate and navigate in the social world [35]. There are sex differences for empathy in ASD [36], and the theory of mind is necessary for empathy. Therefore, sex differences of GM asymmetry in the angular gyrus may be why males are less empathetic than females with ASD.

The sex difference of GM asymmetry was also observed in the middle occipital gyrus in subjects with ASD. Like the angular gyrus, females with ASD showed more, and males with ASD showed less rightward asymmetry relative to their respective TDs in the middle occipital gyrus. This is the first work to find that males and females with ASD exhibited GM asymmetry alterations in the opposite direction relative to their sex-matched TDs. Moreover, males and females with ASD showed different correlativity between ASD symptom severity and AI value in several brain regions, including the middle occipital gyrus. This again suggests that the relationship between ASD and GM asymmetry partly varies by sex. The middle occipital gyrus mainly involves constructing visual and motorial perception, and aberrant perceptual processing might be associated with social dysfunction in ASD [37]. Considering the novelty of these results, they should be interpreted carefully, and they need to be replicated in future

studies.

The results do not support the “Female Protective Effect” theory, which states that females need higher detrimental loads, including brain structural alterations, before developing clinically relevant levels of autistic characteristics [19]. Instead, we detected opposite sex-associated effects in ASD for both two brain regions. Similarly, [38] found that neither behavioral nor neural data supported the idea that females with ASD had more severe abnormalities. Thus, our research provides evidence that sex differences of GM asymmetry in ASD may be qualitative rather than quantitative. Based on these findings, we suggest that researchers should not conduct cross-sex neuroimaging studies for ASD because they may raise the risk of offsetting present results in only one sex or opposite directions in males and females.

There are limitations in the current research. First, it was based on multi-site datasets, which may introduce additional sources of variance associated with participant characteristics or scan parameters. Secondly, our sample size was still insufficient; further studies with larger sample sizes are needed to confirm sex-specific brain structure alterations in subjects with ASD. Finally, participants’ clinical information was also limited, with ADOS and ADI-R scores available for only participants.

Table 4. Coordinates and AI values of clusters with significant differences between the four groups.

Brodmann area	MNI coordinates of peak			Cluster size (voxels)	F-value	ASD		TDs	
	x	y	z			Male	Female	Male	Female
The main effect of diagnosis									
Parahippocampal gyrus	23	−5	−18	173	10.17	−0.15 ± 0.48	−0.06 ± 0.33	0.08 ± 0.36	0.12 ± 0.36
Postcentral gyrus	11	−41	74	244	15.11	−0.11 ± 0.30	−0.10 ± 0.27	0.09 ± 0.27	0.07 ± 0.22
The main effect of sex									
Middle temporal gyrus	60	8	−17	214	13.18	−0.21 ± 0.62	0.09 ± 0.40	−0.24 ± 0.43	0.07 ± 0.69
Inferior frontal gyrus	36	23	−6	550	13.47	−0.07 ± 0.41	0.11 ± 0.27	−0.01 ± 0.28	0.21 ± 0.20
Angular gyrus	45	−59	38	224	12.59	−0.13 ± 0.29	0.16 ± 0.42	−0.03 ± 0.38	0.12 ± 0.28
Superior frontal gyruss	12	45	41	229	11.05	0.11 ± 0.26	−0.01 ± 0.27	0.21 ± 0.28	−0.04 ± 0.32
Postcentral gyrus	45	−36	65	287	14.90	−0.17 ± 0.30	0.04 ± 0.25	−0.06 ± 0.22	0.07 ± 0.23
Interaction effect									
Angular gyrus	30	−69	47	311	12.53	−0.17 ± 0.32	0.07 ± 0.27	0.08 ± 0.30	−0.08 ± 0.26
Middle occipital gyrus	39	−80	12	395	10.64	−0.01 ± 0.43	0.30 ± 0.43	0.16 ± 0.36	−0.00 ± 0.33

Notes: ASD, autism spectrum disorders; TDs, typically developing controls.

Table 5. Pearson correlation analysis for AI values extracted from the clusters with significant differences between the four groups and autistic symptoms measured by the ADI-R scale.

ADI-R	Middle Temporal Gyrus		Inferior Frontal Gyrus		Angular gyrus ^b		Superior Frontal Gyrus		Postcentral gyrus ^b		Parahippocampal gyrus		Postcentral gyrus ^a		Angular gyrus ^c		Middle occipital gyrus	
	r-value	p-value	r-value	p-value	r-value	p-value	r-value	p-value	r-value	p-value	r-value	p-value	r-value	p-value	r-value	p-value	r-value	p-value
Male																		
Social	0.277	0.118	-0.106	0.556	-0.123	0.497	0.033	0.853	0.264	0.137	0.117	0.516	0.054	0.767	-0.230	0.198	0.219	0.220
Verbal	0.233	0.192	-0.270	0.128	-0.040	0.825	-0.030	0.869	0.145	0.421	0.032	0.681	0.006	0.975	-0.249	0.163	0.387*	0.026
RRB	-0.047	0.797	-0.122	0.499	0.035	0.846	-0.048	0.972	0.299	0.091	-0.147	0.413	-0.029	0.874	-0.182	0.311	0.295	0.095
Female																		
Social	-0.293	0.116	-0.022	0.907	-0.223	0.237	0.337	0.069	-0.330	0.075	0.422*	0.020	0.130	0.494	-0.217	0.248	-0.100	0.600
Verbal	-0.125	0.511	0.161	0.394	-0.097	0.610	0.101	0.595	-0.260	0.160	0.168	0.376	0.217	0.250	-0.018	0.925	-0.085	0.650
RRB	0.112	0.555	-0.085	0.655	0.257	0.171	0.271	0.148	-0.018	0.924	0.045	0.815	0.202	0.283	0.120	0.528	0.193	0.308

Notes: AI, asymmetry index; ADI-R, revised autism diagnostic interview scale; RRB, restricted repetitive behaviors; * $p < 0.05$ (uncorrected); ^a the main effect of diagnosis; ^b the main effect of sex; ^c the main effect of interaction.

Using an advanced VBM approach, we identified diagnosis \times sex effects of GM asymmetry in ASD. These effect patterns were qualitative rather than quantitative by showing that males and females with ASD exhibited GM asymmetry alterations in the opposite direction, which is relatively novel. Our findings highlight the necessity of separate studies for males and females with ASD and the potentials for sex-specific diagnostic methods and treatments.

Abbreviations

GM, gray matter; TDs, typically developing controls; ASD, autism spectrum disorders; ABIDE, autism brain imaging data exchange; AI, asymmetry index; ADI-R, revised autism diagnostic interview; VBM, voxel-based morphometry; DARTEL, Diffeomorphic Anatomical Registration; DSM, Diagnostic and Statistical Manual of Mental Disorders; ADOS, Autism Diagnostic Observation Schedule; FIQ, full-scale intelligence quotient; SPM, Statistical Parametric Mapping; WM, white matter; ANCOVA, analysis of covariance; GRF, Gaussian random field; DPARSF, Data Preprocessing Assistant for rs-fMRI; ADHD, attention deficit hyperactivity disorder; GU, Georgetown University; KKI, Kennedy Krieger Institute; NYU, New York University; OHSU, Oregon Health and Science University; YALE, Yale Child Study Center.

Author contributions

CCL, MMN, PYF and HBX conceived and designed the experiments; CCL and MMN analyzed the data; CCL and HBX wrote the paper.

Ethics approval and consent to participate

The written informed consent was obtained from the site where they conducted the tests. The research was conducted with the Ethics Committee of the University of Zhongnan Hospital of Wuhan University (Ethical approval number: 2019072).

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Conflict of interest

The authors declare no conflict of interest.

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