Atypical Functional Brain Connectivity during Rest in Autism Spectrum Disorders

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Objective: Connectivity atypicalities in autism spectrum disorders (ASD) have been extensively proposed. The default mode network (DMN) is critical in this study, given the insight it provides for long-distance connectivity, and the importance of regions in this network for introspection and social emotion processing, areas affected in ASD. However, study of this network has largely been limited to adults; research earlier in development is lacking. The objective of this study was to examine DMN connectivity in children/adolescents with ASD.
Methods: A total of 115 children/adolescents, aged 6 to 17 years (71 males with ASD and 44 group age-matched TD males) were included in these analyses. We examined group differences in (1) functional connectivity between the posterior cingulate cortex and regions across the brain, (2) connectivity within the DMN as a function of age and intelligence quotient (IQ), and (3) the association between DMN connectivity and empathic accuracy.
Results: Individuals with ASD, relative to controls, showed either stronger or weaker connectivity between the posterior cingulate cortex (PCC) and DMN regions, depending on the region, but also showed stronger connectivity with non-DMN regions. A significant group-by-age interaction was observed in functional connectivity between the PCC and medial prefrontal cortex; connectivity increased with age in controls, but decreased in individuals with ASD. No effects of IQ were found. There was a significant group difference in the relation between DMN connectivity and empathic accuracy.
Interpretation: Differences in functional connectivity may suggest the presence of neural atypicalities that impact the development of typical connectivity in ASD. In addition to affecting DMN dynamics, these atypicalities may also impact social–cognitive abilities.

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Converging lines of evidence indicate that autism spectrum disorders (ASD) may be a disorder of atypical brain connectivity.[1–8] In both task-specific[3,9–11] and idle neural networks,[12,13] individuals with ASD often show weaker long-range connections relative to typically developing individuals. Findings with respect to short-range connectivity are equivocal: reductions[3,9–11] and increases[13–17] have been reported. The default mode network (DMN) is a network of brain areas mostly along the midline of the brain, which is active during rest but decreases activity during externally directed, attention-demanding cognitive tasks.[18] Not only does it provide

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information on long-range connectivity in ASD, but it also shares many of the same regions implicated in social emotion processing and self-awareness that are impaired in ASD.[19] Findings about atypical connectivity within the DMN in ASD are largely limited to adult populations. Given the developmental nature of ASD, it is important to improve our knowledge about potential connectivity differences earlier in life. It is also important to examine how factors such as intelligence quotient (IQ) and severity of symptomatology, which vary across individuals with ASD, may be related to such differences.

From anterior to posterior, brain regions within the DMN include the medial prefrontal cortex (MPFC), the anterior cingulate cortex (ACC), the middle temporal gyrus (MTG), the precuneus, and the posterior cingulate cortex (PCC). Seminal papers suggest that the DMN is anchored in the PCC, and the PCC can be used successfully as a seed in both adults and children.[20–23] The PCC shows robust activation during rest, and in typical populations show high connectivity with other DMN regions.[24,25]

Behaviorally, the activity in the DMN is suggested to reflect self-reflective thought such as autobiographical memories of familiar faces and/or experiences,[18] inward thinking involving processes such as monitoring, evaluation, and integration,[12] mind wandering, or low-level monitoring of the external environment.[22,24,26–28] Additionally, it has been suggested that DMN activity may be related to homeostatic aspects of the central nervous system, such as maintaining the balance of excitatory and inhibitory inputs.[29,30] Although a consensus has not been reached on the precise role of the DMN, research has consistently shown in the typically developed population that DMN activity is suppressed during cognitive and attention-demanding tasks.[21,31,32] Its deactivation has been suggested to facilitate performance on these more demanding tasks.[33]

Functional magnetic resonance imaging (fMRI) investigations into the DMN in ASD show atypical activity and connectivity in adult samples. Studies have found that regions such as the MPFC, ACC, and PCC failed to reduce their activity during active tasks[18,34] in adults with ASD. Kennedy and Courchesne[35] found that relative to controls, adults with ASD showed lower levels of activity in the MPFC and ACC during rest when compared to the active task. Resting state fMRI studies have found weaker connectivity between anterior and posterior regions[12,28] or across the network[13] and stronger connectivity between short-range areas.[28]

The majority of studies have used a relatively small, adult male sample when examining DMN connectivity. Recently, some have begun to look at the DMN in individuals with ASD younger than 21 years; however, these papers are still few and the sample sizes have been small.[13,15,36,37] These studies report both hypoconnectivity[13,15,36,37] and hyperconnectivity[37] within the DMN. It may be possible that DMN properties change across developmental stages.[37] More studies are needed to delineate the effect of age and other factors on the DMN in children and adolescents with ASD. As part of a large, multisite project, we collected data from 115 children and adolescents between the ages of 6 and 17 years; 71 children with ASD and 44 age-matched typically developing controls. In this sample, we examined (1) functional connectivity between the posterior cingulate cortex and regions across the brain, (2) connectivity within the DMN as a function of age and IQ, and (3) the association between DMN connectivity and empathic accuracy. We hypothesized that children and adolescents would show atypical connectivity relative to age-matched controls across the brain and within DMN. We expected atypical relations between connectivity and age, IQ, and empathic accuracy.

Subjects and Methods

Participants

A total of 115 participants between the ages of 6 and 17 years were imaged as part of the NeuroDevNet Autism Demonstration Project, a multisite initiative to study brain structural and behavioral development in ASD children[38] (ASD: $n = 71$, age $= 12.3 \pm 3.1$ years, IQ $= 97.8 \pm 19.7$; typically developing: $n = 44$, age $= 12.2 \pm 3.8$ years, IQ $= 117.2 \pm 9.7$). ASD participants had a clinical diagnosis according to the Diagnostic and Statistical Manual of Mental Disorders, 4th Edition[39] and were not on psychotropic medications at the time of the scan. Diagnoses were confirmed using the Autism Diagnostic Observational Schedule–Generic[40] and the Autism Diagnostic Interview–Revised.[41] Any participants who had a primary psychiatric condition (other than ASD in the ASD group), a history of head injury, epilepsy, neuromotor impairment, or a genetic disorder associated with ASD were excluded. Additionally, controls were excluded if they had a first-degree relative with ASD. Intelligence was assessed in all participants by either the Wechsler Abbreviated Scale of Intelligence[42] or the Wechsler Abbreviated Scale of Intelligence, 2nd Edition.[43]

fMRI Acquisition

Magnetic resonance (MR) data were acquired at the Hospital for Sick Children and Montreal Neurological Institute, using the same 3T MR device (Tim Trio, VB17; Siemens, Erlangen, Germany) and identical imaging sequences. Anatomical images were acquired using sagittal 3-dimensional (3D) magnetization-prepared rapid gradient echo (field of view [FOV] $= 192 \times 240 \times 256$mm, 1mm isovoxels, repetition time [TR]/echo time [TE]/inversion time $= 2{,}300/2.96/900$ milliseconds, flip angle [FA] $= 9°$). Resting state fMRI was acquired using an Axial 2D

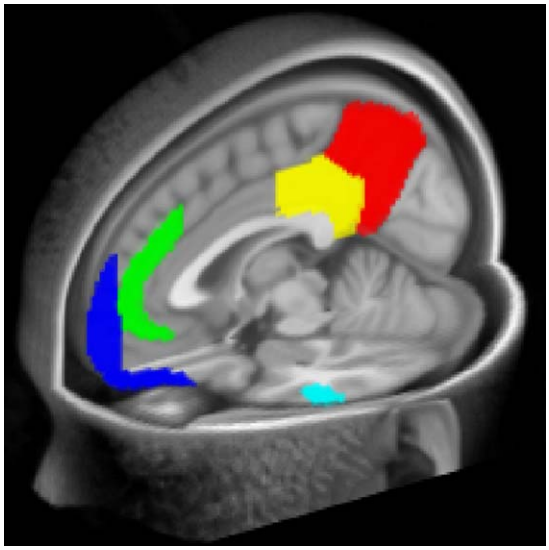

FIGURE 1: Seed regions based on the Harvard–Oxford cortical atlas.

EPI (FOV = 224 × 224mm, matrix = 64 × 64, 40 × 3.5mm interleaved slices, TR/TE = 2,340/30 milliseconds, FA = 70°, 120 acquisitions). Total scan time was 4 minutes and 43 seconds. Participants were told to look at a cross in the center of the screen for the duration of the scan. The resting state sequence was 1 sequence acquired as part of a larger multimodal imaging protocol.

fMRI Preprocessing

fMRI data were processed using a combination of Analysis of Functional NeuroImages (AFNI), FMRIB Software Library, and locally developed software tools. Processing steps included motion correction and slice timing correction, followed by linear registration to the Montreal Neurological Institute 152 template. Data were smoothed using a 2D 7mm full width at half maximum Gaussian kernel, and bandpass filtered with lower and upper cutoff frequencies of 0.01 and 0.2Hz, respectively. Motion signals generated from maximum displacement motion estimates as well as whole brain, cerebrospinal fluid, and white matter signals were removed. Volumes with a maximum displacement exceeding 1.75mm were omitted from analyses. Subjects with >33% of their volumes omitted were excluded from the study.

fMRI Connectivity Analyses: DMN and Control Network

Inter-region connectivity was calculated as the mean correlation of all voxels within a brain region, relative to the mean signal within the eroded source region—the left or right PCC. An individual's PCC was defined using the Harvard–Oxford cortical atlas. The erosion process followed the method developed by Golestani and Goodyear.[44] Each participant's PCC was iteratively eroded until a mean intervoxel cross-correlation of 0.7 was obtained. For each participant, the mean signal within the left and right PCC was calculated after erosion, and used to measure connectivity between the PCC and a target region. Prior to all statistical analyses, correlation coefficients were converted to a normal distribution using Fisher z transformation. The same approach was used to define the occipital pole in each individual, for the purpose of carrying out control analyses in the visual network.

Primary Analysis: Whole Brain Connectivity with the PCC

The mean signal with the left and right PCC were correlated on a voxel-by-voxel basis for the whole brain. As a control, we carried out the same analysis with the left and right occipital pole as seed points. Regions of significant correlation were identified using cluster-based thresholding ($p < 0.01$, $\alpha < 0.01$).

Secondary Analysis: DMN Connectivity as a Function of Age and IQ

We investigated DMN inter-regional connectivity, using the eroded PCC as the source region, and the left and right MPFC, ACC, MTG, and precuneus as target regions. These regions were selected based on their consistent reporting as regions within the DMN across several studies,[12,13,18,28,34–37] as defined using the Harvard–Oxford cortical atlas (Fig 1). A linear model was used to estimate the interactions between diagnosis, IQ, and age.

As a control, we examined connectivity with respect to age within the visual system. The left and right occipital poles were used as seed points. Regions of interest included the lingual gyrus, cuneal, calcarine, and occipital pole. We additionally included mean maximum displacement as a covariate in the mixed effects analyses.

Results were not subject to a test for multiple corrections

Exploratory Analysis: Correlations between DMN Connectivity and Reading the Mind and the Eyes Test Scores

As an exploratory analysis, we examined the relation between DMN connectivity and empathic accuracy as measured by the Reading the Mind in the Eyes Test (RMET)[45] in a subset of the population where RMET scores were measured (Table 1). The linear model used for this analysis included diagnosis, IQ, age, and RMET as factors of interest. Results were not subject to a test for multiple corrections.

All brain images were generated using AFNI and are displayed with full intensity range, unless otherwise noted using color bars. GNU Image Manipulation Program was used to crop and combine images, ensuring correct size and resolution. No adjustments were made to image contrast or brightness. Correlation plots were generated directly from R, with proper sizing and resolution.

Results

Participants

For our primary and secondary analyses, individuals with ASD and typically developing controls did not differ on

TABLE 1. Demographics

Characteristic	ASD, n = 58, mean ± SD, range	TD, n = 37, mean ± SD, range	Probability
Age distribution			
Age 7–18 years	12.5 ± 2.7, 6–17	12.7 ± 3.2, 7–17	$t(93) = 0.38$, $p = 0.71$
FSIQ	98.7 ± 19.2, 53–147	116.9 ± 8.8, 96–139	$t(93) = 5.40$, $p < 0.01$
ADI-R			
Social	18.7 ± 5.8		
Communication	15.5 ± 4.1		
Stereotyped behaviors	6.7 ± 1.6		
ADOS			
Social	7.3 ± 2.4		
Communication	3.0 ± 1.6		
Repetitive behaviors	2.5 ± 1.7		
RMET subgroup			
No.	58	16	
Age 7–18 years	12.5 ± 2.7, 6–17	12.9 ± 2.2, 7–17	$t(72) = 0.50$, $p = 0.62$
FSIQ	98.7 ± 19.2, 53–147	115.9 ± 7.5, 96–139	$t(72) = 3.50$, $p < 0.01$
RMET score	16.6 ± 4.5, 6–25	20.0 ± 2.9, 14–25	$t(72) = 2.86$, $p < 0.01$

ADI-R = Autism Diagnostic Interview–Revised; ADOS = Autism Diagnostic Observational Schedule; ASD = autism spectrum disorders; FSIQ = full scale intelligence quotient; RMET = Reading the Mind in the Eyes Test; SD = standard deviation; TD = typically developing.

age ($p = 0.71$); however, groups were significantly different on full scale IQ ($p < 0.01$). The subgroup included in our exploratory analysis did not differ on age ($p = 0.71$), but did on IQ ($p < 0.01$) and empathic accuracy ($p < 0.01$; see Table 1 for more details).

TABLE 2. Size of Eroded Seed Region

Region	ASD, mean, SD	TD, mean, SD	*p*
PCC L	173.67, 101.42	191.17, 131.31	0.50
PCC R	208.69, 97.82	208.67, 95.63	1.0
OP L	243.19, 130.56	230.44, 158.98	0.69
OP R	179.85, 102.51	170.78, 93.64	0.67

Size represents the number of 3.5 × 3.5 × 3.5mm voxels. ASD = autism spectrum disorders; L = left; OP = occipital pole; PCC = posterior cingulate cortex; R = right; SD = standard deviation.

Seed Region Erosion

There was also no significant difference between groups in the size of the seed region of interest (ROI; Table 2).

Motion

Fifteen (of 71) participants with ASD and 5 (of 44) typically developing participants were excluded for excessive motion. Mean maximum displacement (± standard deviation) was 0.37 ± 0.29mm for participants with ASD and 0.27 ± 0.29mm for typically developing controls. Although the average number of volumes omitted was higher in ASD (6.8 ± 10.3) than in typically developing controls (3.0 ± 8.3), this difference was not significant ($p = 0.06$). Irrespective of diagnosis, connectivity was not found to be significantly correlated with participant motion.

Primary Analysis: Voxelwise Analysis—Group Differences

Typically developing participants had higher connectivity compared to individuals with ASD between the left PCC and the left medial frontal gyrus (Brodmann area [BA] 11), the left and right angular gyri (BA 39), and the

TABLE 3. Primary Analysis: Between-Group Results

Comparison	No. of Voxels	Mean *z* (difference)	X, Y, Z	Side	BA	Region
PCC-L						
TD > ASD	1,981	−0.21	−4, −48, 18	L	30	Posterior cingulate
	1,790	−0.12	−2, 54, −14	L	11	Medial frontal gyrus
	1,028	−0.14	48, −70, 40	R	39	Angular gyrus
	529	−0.08	66, −14, −16	R	21	Inferior temporal gyrus
	471	−0.12	−54, −64, 36	L	39	Angular gyrus
ASD > TD	828	0.13	−62, −34, 30	L	40	Inferior parietal lobule
	736	0.11	−34, 50, 24	L	10	Superior frontal gyrus
	734	0.13	−48, 2, 36	L	6	Precentral gyrus
	586	0.08	46, 46, 12	R	10	Middle frontal gyrus
	493	0.08	16, −64, 64	R	7	Superior parietal lobule
	453	0.10	62, −32, 36	R	40	Inferior parietal lobule
PCC-R						
TD > ASD	1,988	−0.21	−4, −48, 18	L	30	Posterior cingulate
	1,031	−0.14	48, −70, 40	R	39	Angular gyrus
	1,802	−0.12	−2, 54, −14	L	11	Medial frontal gyrus
	471	−0.12	−54, −64, 36	L	39	Angular gyrus
	534	−0.08	66, −14, −16	R	21	Inferior temporal gyrus
ASD > TD	740	0.13	−48, 2, 36	L	6	Precentral gyrus
	831	0.13	−62, −34, 30	L	40	Inferior parietal lobule
	738	0.11	−34, 50, 24	L	10	Superior frontal gyrus
	954	0.09	62, −32, 36	R	40	Inferior parietal lobule
	588	0.08	46, 46, 12	R	10	Middle frontal gyrus

Brain regions across the entire brain that show significant differences in functional connectivity with the left and right PCC, between typically developing controls and individuals with ASD (all: $p < 0.01$, cluster: $p < 0.05$). Mean *z* score differences are included. Coordinates are given in Montreal Neurological Institute space. Medial–lateral (X), anterior–posterior (Y), and superior–inferior (Z) positive values are right, anterior, and superior.
ASD = autism spectrum disorders; BA = Brodmann area; L = left; PCC = posterior cingulate cortex; R = right; TD = typically developing controls.

right inferior temporal gyrus (BA 21). Significantly higher connectivity in typically developing participants compared to those with ASD was also found between the right PCC and the left medial frontal gyrus (BA 11), the left PCC (BA 30), the left and right angular gyrus (BA 39), and the right inferior temporal gyrus (BA 21; Table 3).

In contrast, individuals with ASD, relative to controls, had higher connectivity between the left PCC, and the left inferior parietal lobule (BA 40), the left superior frontal gyrus (BA 10), the left precentral gyrus (BA 6), the right middle frontal gyrus (BA 10), the right superior parietal lobule (BA 7), and the right inferior parietal lobule (BA 40). The ASD group also showed higher connectivity, relative to controls, between the right PCC and the left and right inferior parietal lobule (BA 40), the right middle frontal gyrus (BA 10), the left precentral gyrus (BA 6), and the left superior frontal gyrus (BA 10; see Table 3). See Figure 2 for voxelwise maps.

No significant group differences were found in connectivity between any region of the brain and the occipital poles (left and right; all p values > 0.05).

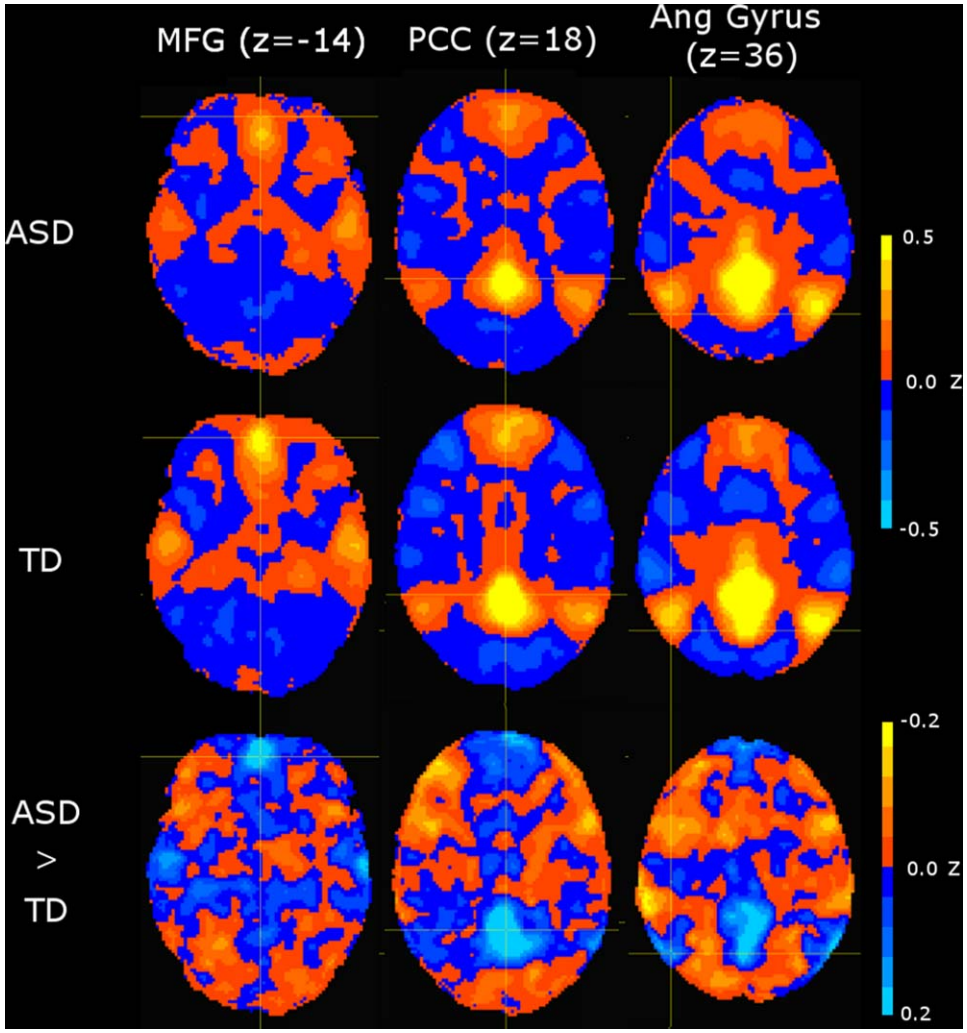

FIGURE 2: Voxelwise maps illustrating the mean autism spectrum disorders (ASD), typically developing (TD), and ASD > typically developing differences for the left posterior cingulate cortex (PCC) versus the middle frontal gyrus (MFG), the PCC, and the angular gyrus.

Secondary Analysis: A Priori ROI Analysis—Effect of Age and IQ

A significant difference was found in functional connectivity between the left PCC and right MPFC (slope difference typically developing, ASD: 0.0115 ± 0.0045; $p = 0.01$) and the right PCC and right MPFC (slope difference typically developing, ASD: 0.0108 ± 0.0042; $p = 0.01$) as a function of age (Fig 3, Table 4). Functional connectivity strengthened with age in the typically developing group but was weaker in older children in the ASD group. No other regions showed significant differences with age (see Table 3). Functional connectivity also did not differ as a function of IQ, within the IQ range reported in our sample. No group differences were found in connectivity between the occipital pole and regions of the visual system (all p values > 0.05).

The inclusion and exclusion of motion as a covariate in the mixed-effect analyses yielded similar results. Significant age effects remained in both conditions. Results presented were obtained when motion was included as a covariate.

Exploratory Analysis: Association between DMN Connectivity and Empathic Accuracy

Significant group differences were found in functional connectivity between the left PCC and the right MPFC; and the right PCC and the left precuneus, the right MPFC, the right MTG, and the right precuneus, as a function of RMET scores. In all significant cases except the right PCC–right MTG correlation, better empathic accuracy was associated with stronger correlations in controls, but weaker correlations in individuals with ASD. In comparison, as the right PCC–right MTG correlation got stronger, empathic accuracy in the ASD group also improved, whereas controls showed poorer accuracy with

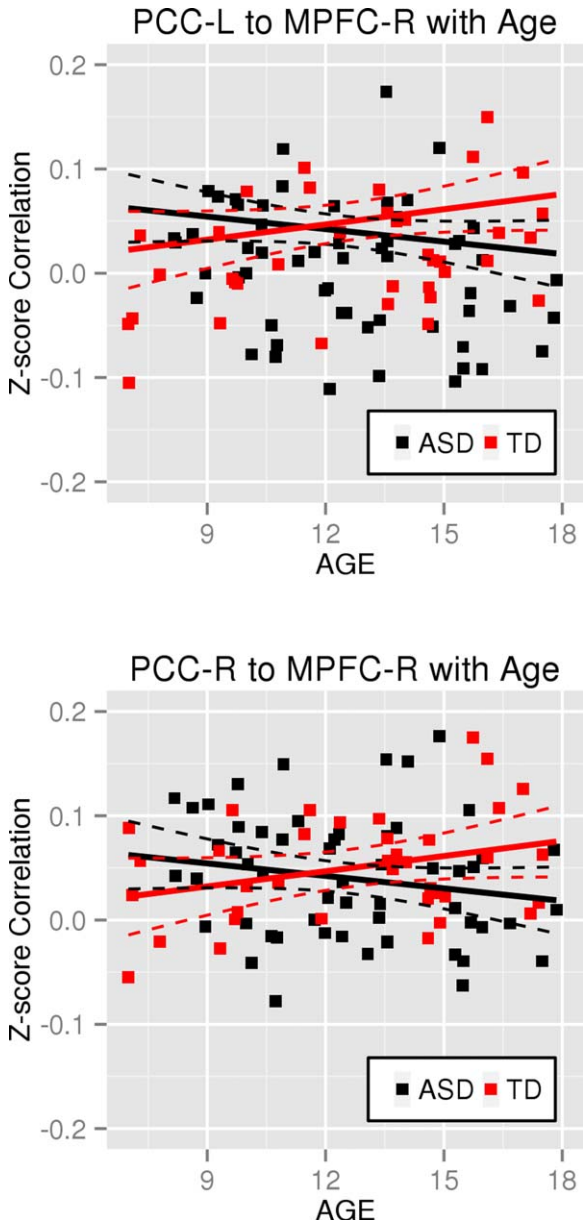

FIGURE 3: Scatter plot depicting a significant age-by-group interaction in functional connectivity between the left (L) and right (R) posterior cingulate cortex (PCC), and right medial prefrontal cortex (MPFC; $p < 0.05$). Typically developing (TD) participants are indicated using red squares; line of best fit is shown in red. Participants with autism spectrum disorders (ASD) are indicated using black squares; line of best fit is shown in black.

stronger correlations. No other significant correlations were found (Table 5).

Discussion

The current study is the largest study to date to examine DMN connectivity in children and adolescents with ASD. We compared functional connectivity between the PCC, a hub of the DMN, and regions across the brain in children/adolescents with ASD and typically developing controls. Additionally we examined the effect of age, IQ, and empathic accuracy on DMN connectivity in these groups. Individuals with ASD showed atypical, both strong and weak, correlations between the PCC and regions across the brain that were significantly different from those observed in controls. Groups also differed in connectivity between the PCC and the MPFC as a function of age. However, no connectivity differences were found as a function of IQ. Some correlations between the PCC and other DMN regions were also correlated with RMET scores, and showed a very different relation in individuals with ASD compared to typically developing controls. In general, stronger connectivity between the PCC and regions across the DMN was associated was better empathic accuracy in controls, but poorer empathic accuracy in individuals with ASD. Connectivity between the right PCC and right MTG was the only case where the opposite was true. The lack of significant correlation between connectivity and participants' motion suggests that the reported connectivity differences were not a result of motion differences between groups.

The DMN connectivity differences found between groups further expand on previous findings of aberrant and inefficient connectivity reported in smaller samples of participants with ASD. Functional connectivity atypicalities in children and youth included both reduced and increased connectivity between the PCC and the rest of the brain.

The age-related findings in the current study are modest; however, they are in agreement with previous findings on DMN development in typically developing individuals, and the long-range atypical connectivity theory in individuals with ASD. Studies in typically developing individuals have showed an poorly connected DMN in children younger than 9 years that matures into a more integrated system in late adolescence/early adulthood, after controlling for motion.[46–48] Nearly all developmental changes among regions of the DMN in typically developing individuals were increases in connection strength, particularly between longer range structures (eg, the MPFC and PCC).[46–48] The typically developing findings of the current study were consistent with these. However, functional connectivity between the PCC and MPFC in individuals with ASD, relative to controls, decreased with age. This may suggest the presence of a transition between childhood and adolescence, when unknown mechanisms facilitate a changeover to long-range underconnectivity.[37] Because connectivity within the visual system was intact, aberrant connectivity between the PCC and MPFC is unlikely to be a result of whole brain connectivity atypicalities.

TABLE 4. Secondary Analysis: Effect of Age on Default Mode Network and Control Network Functional Connectivity

TD–ASD Comparison	Region	Side	Slope Difference	*p*
PCC-L	MPFC	L	0.0067 ± 0.0051	n.s.
	MPFC	R	0.0115 ± 0.0045	0.01[a]
	ACC	L	0.0046 ± 0.0085	n.s.
	ACC	R	0.0082 ± 0.0078	n.s.
	MTG	L	0.0028 ± 0.0095	n.s.
	MTG	R	0.0038 ± 0.0089	n.s.
	Precuneus	L	−0.0003 ± 0.0077	n.s.
	Precuneus	R	−0.0034 ± 0.0074	n.s.
	PCC	L	0.0072 ± 0.0078	n.s.
	PCC	R	0.0013 ± 0.0062	n.s.
PCC-R	MPFC	L	0.0055 ± 0.005	n.s.
	MPFC	R	0.0108 ± 0.0042	0.01[a]
	ACC	L	0.0067 ± 0.0092	n.s.
	ACC	R	0.0067 ± 0.0081	n.s.
	MTG	L	0.011 ± 0.0116	n.s.
	MTG	R	0.0123 ± 0.0111	n.s.
	Precuneus	L	−0.0036 ± 0.0073	n.s.
	Precuneus	R	−0.0048 ± 0.0074	n.s.
	PCC	L	0.0059 ± 0.0082	n.s.
	PCC	R	0.0013 ± 0.0062	n.s.
OP-L	Cuneal	L	−0.0081 ± 0.0188	n.s.
	Cuneal	R	−0.0118 ± 0.0195	n.s.
	Lingual	L	−0.0231 ± 0.0206	n.s.
	Lingual	R	−0.0173 ± 0.0191	n.s.
	Calcarine	L	−0.0026 ± 0.014	n.s.
	Calcarine	R	−0.0012 ± 0.0143	n.s.
	Occipital pole	L	0.0094 ± 0.008	n.s.
	Occipital pole	R	0.0148 ± 0.0086	n.s.
OP-R	Cuneal	L	−0.0096 ± 0.0197	n.s.
	Cuneal	R	−0.0057 ± 0.0209	n.s.
	Lingual	L	−0.0201 ± 0.0206	n.s.
	Lingual	R	−0.0175 ± 0.019	n.s.
	Calcarine	L	−0.0012 ± 0.0151	n.s.
	Calcarine	R	−0.0025 ± 0.0152	n.s.
	Occipital pole	L	0.009 ± 0.0089	n.s.
	Occipital pole	R	0.0132 ± 0.0084	n.s.

Brain regions that show significant differences in functional connectivity with the left and right PCC, between typically developing controls and individuals with ASD as a function of age.
[a]Significant results.
ACC = anterior cingulate cortex; ASD = autism spectrum disorders; L = left; MPFC = medial prefrontal cortex; MTG = middle temporal gyrus; n.s. = not significant; OP = occipital pole; PCC = posterior cingulate cortex; R = right; TD = typically developing controls.

TABLE 5. Exploratory Analysis: Association between Empathic Accuracy and Default Mode Network Functional Connectivity

TD–ASD Comparison	Region	Side	Slope Difference	*p*
PCC-L	MPFC	L	0.0007 ±0.0056	n.s.
	MPFC	R	0.0135 ± 0.0049	0.01[a]
	ACC	L	−0.0045 ± 0.0095	n.s.
	ACC	R	0.004 ± 0.0082	n.s.
	MTG	L	−0.0069 ± 0.0087	n.s.
	MTG	R	−0.0019 ± 0.0092	n.s.
	Precuneus	L	0.0123 ± 0.0079	n.s.
	Precuneus	R	0.0182 ± 0.0079	0.05[a]
	PCC	L	0.0022 ± 0.0076	n.s.
	PCC	R	0.0054 ± 0.007	n.s.
PCC-R	MPFC	L	0.0095 ± 0.0052	n.s.
	MPFC	R	0.0128 ± 0.0047	0.01[a]
	ACC	L	−0.0045 ± 0.0095	n.s.
	ACC	R	−0.0019 ± 0.0092	n.s.
	MTG	L	−0.0007 ± 0.0114	n.s.
	MTG	R	−0.0289 ± 0.0117	0.05[a]
	Precuneus	L	0.0204 ± 0.0074	0.01[a]
	Precuneus	R	0.0225 ± 0.0078	0.05[a]
	PCC	L	−0.0055 ± 0.008	n.s.
	PCC	R	0.0005 ± 0.0061	n.s.

Brain regions that show significant differences in functional connectivity with the left and right PCC, between typically developing controls and individuals with ASD as a function of Reading the Mind in the Eyes Test scores.
[a]Significant results.
ACC = anterior cingulate cortex; ASD = autism spectrum disorders; L = left; MPFC = medial prefrontal cortex; MTG = middle temporal gyrus; n.s. = not significant; PCC = posterior cingulate cortex; R = right; TD = typically developing controls.

In the present study, we found that empathic accuracy was associated with functional connectivity differently in individuals with ASD than it was in controls. All but 1 correlation showed that increased functional connectivity was associated with poorer emotion perception in ASD, whereas it was associated with better empathic accuracy in controls, suggesting an atypical relation between functional connectivity and social cognition processes in ASD.

There are several limitations to this study. First, the current study has a cross-sectional design, which is only a proxy for a longitudinal design, where developmental changes are more directly assessed. Second, our sample was limited to individuals with ASD and controls who were able to complete the scan while awake. Third, we did not monitor cardiac or respiratory rhythms during data acquisition, which may have contributed to motion. However, we addressed motion effects by excluding participants with excessive motion, omitting problematic volumes, and including average maximum displacement as a covariate in our mixed effects analyses. There was no significant difference in either volumes omitted due to motion, or average maximum displacement in ASD compared to typically developing controls. Fourth, our resting state scan was relatively short; however, it allowed us to study participants with ASD regardless of age and functioning. In the future, it would be beneficial to attempt longer scans in this population to allow for more data. Lastly, we observed substantial variability in mean correlations, both within participants and within groups. Variability may reflect the heterogeneity within the ASD population and also within the typically developing population, and suggests largely overlapping distributions of

functional connectivity between the 2 groups. Although this is the largest study to date, the current sample might still be too small to fully describe the magnitude of variance within these groups.

In the future, longitudinal data or at least large replications of cross-sectional data are needed to confirm and extend our results. Samples need to be expanded to include resting state data in lower functioning and younger individuals to expand our knowledge of DMN connectivity across the spectrum and development. The resting state scan is particularly accommodating to these individuals, because participants can be scanned while asleep or sedated. Larger sample sizes will also give us the opportunity to examine the biology of the large variance we observed, by combining imaging data sets with phenomics and genomics.

In summary, atypical functional connectivity was found in children and adolescents with ASD relative to typically developing age-matched controls within the DMN, and between the PCC and the rest of the brain. An atypical developmental trajectory was found in functional connectivity between the PCC and MPFC in ASD. Whereas typically developing individuals showed increased correlation across age, correlation was reduced with age in individuals with ASD. No association was found between functional connectivity and IQ. Empathic accuracy showed significantly different associations with functional connectivity between groups. These results may suggest atypical structural and/or functional properties in the ASD brain, which may impact the development of typical DMN connections.

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Authorship

L.Z., K.L.H., A.C.E., J.L., E.A., and the NeuroDevNet ASD Imaging group conceptualized the project. Data collection was carried out by K.A.R.D.-T., N.E.V.F., A.T., and T.O. Data analysis and manuscript preparation were completed by W.L. and K.A.R.D.-T. under the supervision of E.A.

Potential Conflicts of Interest

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