Decreased connectivity and cerebellar activity in autism during motor task performance

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Although motor deficits are common in autism, the neural correlates underlying the disruption of even basic motor execution are unknown. Motor deficits may be some of the earliest identifiable signs of abnormal development and increased understanding of their neural underpinnings may provide insight into autism-associated differences in parallel systems critical for control of more complex behaviour necessary for social and communicative development. Functional magnetic resonance imaging was used to examine neural activation and connectivity during sequential, appositional finger tapping in 13 children, ages 8–12 years, with high-functioning autism (HFA) and 13 typically developing (TD), age- and sex-matched peers. Both groups showed expected primary activations in cortical and subcortical regions associated with motor execution [contralateral primary sensorimotor cortex, contralateral thalamus, ipsilateral cerebellum, supplementary motor area (SMA)]; however, the TD group showed greater activation in the ipsilateral anterior cerebellum, while the HFA group showed greater activation in the SMA. Although activation differences were limited to a subset of regions, children with HFA demonstrated diffusely decreased connectivity across the motor execution network relative to control children. The between-group dissociation of cerebral and cerebellar motor activation represents the first neuroimaging data of motor dysfunction in children with autism, providing insight into potentially abnormal circuits impacting development. Decreased cerebellar activation in the HFA group may reflect difficulty shifting motor execution from cortical regions associated with effortful control to regions associated with habitual execution. Additionally, diffusely decreased connectivity may reflect poor coordination within the circuit necessary for automating patterned motor behaviour. The findings might explain impairments in motor development in autism, as well as abnormal and delayed acquisition of gestures important for socialization and communication.

Keywords: pediatric; movement; neuroimaging; motor learning; development; connections
**Introduction**

Autism is characterized by deficits in social cognition, disordered communication and restricted interests and repetitive behaviours (American Psychiatric Association, 2000). Subsumed within each of these, motor impairments are a common finding in autism spectrum disorders (ASD), noted in even the earliest descriptions (Kanner, 1943; Frith, 1991). Since then a number of studies have revealed impairments in basic motor control (Vilensky et al., 1981; Ghaziuddin and Butler, 1998; Teitelbaum et al., 1998, 2004; Noterdaeme et al., 2002; Nayate et al., 2005; Jansiewicz et al., 2006) and skilled motor gestures (DeMyer et al., 1972; Jones and Prior, 1985; Ohta, 1987; Smith and Bryson, 1994; Rogers et al., 1996; Williams et al., 2001; Mostofsky et al., 2006) as well as impairments in motor learning (Hughes, 1996; Mostofsky et al., 2000; Rinehart et al., 2001).

Greater insight into motor functioning in autism may prove beneficial to understanding its neurological basis and to early identification of children in the autism spectrum. Though autism is most often diagnosed during toddler or preschool years, retrospective studies using video analysis suggest motor signs may be apparent in children with ASD as early as the first year of life (Teitelbaum et al., 1998, 2004). Furthermore, recently reported findings suggest that gross and fine motor delays are among the earliest identifiable signs distinguishing infants with autism from their typically developing (TD) peers (Landa and Garrett-Mayer, 2007; Chawarska et al., 2003, 2007). In the current study, we looked at fMRI activity during self-paced sequential finger tapping in children with high-functioning autism (HFA), as compared with their TD age- and sex-matched peers. We additionally examined functional connectivity within the motor systems critical for motor coordination and execution. There has been increasing speculation that autism may be associated with abnormalities in structural and functional connectivity, with deficits in ‘weak central coherence’ (Shah and Frith, 1993) and ‘complex information processing’ (Minshew et al., 1997) thought to be related to abnormal connectivity between distant brain regions. Consistent with this proposed model, post-mortem analysis has revealed an abundance of short relative to long connective fibres in frontal and temporal regions (Casanova et al., 2002). Similarly, MRI analysis of white matter in autism has revealed increased volume localized to outer zone (‘radiate’) regions principally comprised of localized U-fibre connections (Herbert et al., 2004), with contrasting decreased size of the corpus callosum, comprised of long-range interhemispheric white matter tracts (Chung et al., 2004; Piven et al., 1997). Functional relevance of these findings was recently established using measures of motor function, with increased local white matter volume in primary motor cortex predicting the degree of motor impairment in children with autism (Mostofsky et al., 2007).

These findings of white matter abnormalities in autism have further led to investigation into functional connectivity, or how distant brain regions are ‘connected’ based on similarities in their...
profiles of functional activity. Decreased functional connectivity has been observed in autism during performance of cognitive tasks (Just et al., 2004, 2007; Kana et al., 2006, 2007); however, functional connectivity during simple motor coordination and execution has not yet been examined.

Acquisition and execution of motor skills is dependent on coordinated activity across a network of cortical and subcortical regions (Doyon et al., 2002). Given that autism is a developmental disorder with onset of skill deficits (motor, social and communicative) very early in childhood, we hypothesized that, when compared with TD children, children with autism would show decreased activity in regions important for achieving automaticity of motor skills, in particular, the cerebellum, with associated increased activity in cerebral cortical regions, necessary for continued effortful control of movement. We further hypothesized that children with autism would show decreased connectivity between these regions.

Materials and Methods

Participant selection

Twenty-six children, aged 8–12 years, participated in the study: 13 meeting study criteria for HFA (mean age = 10.9 years, SD = 1.5) and 13 age- and sex-matched, TD peers (mean age = 10.5 years, SD = 1.4). Each group consisted of eleven boys and two girls. All subjects were right-handed based upon the Physical and Neurological Exam for Subtle Signs (PANESS) (Denckla, 1985).

Subjects were recruited from several sources, including out-patient clinics at Kennedy Krieger Institute and through advertisements placed within community-wide service groups, schools, and hospitals. Autism diagnoses were based on DSM-IV criteria (American Psychiatric Association, 2000) and were confirmed using the Autism Diagnostic Interview-Revised (ADI-R) (Lord et al., 1994) and Autism Diagnostic Observation Schedule-Generic (ADOS-G) (Lord et al., 2000).

The Diagnostic Interview for Children and Adolescents, Fourth Edition (DICA-IV) (Reich, 2000) was used to determine the presence of additional psychiatric diagnoses. TD children with any diagnosis on the DICA-IV or with any immediate family members with autism were excluded. All TD subjects had no history of seizures or evidence of any other neurological disorder. Within the high-functioning autism group, the following DICA-IV diagnoses were also met: Attention-Deficit/Hyperactivity Disorder (two children), Obsessive-Compulsive Disorder (two), Specific Phobia (three), Generalized Anxiety Disorder (one) and Oppositional Defiant Disorder (five); DICA-IV results were not available for two children in the HFA group. Eight children with HFA were instructed not to move their hands. For all subjects, the hands were comfortably positioned on the torso.

Immediately prior to entering the scanner, participants were taught the proper sequence of movements and asked to demonstrate understanding by performing it with each hand. Care was taken to instruct participants not to count taps or name the fingers aloud during training or testing. Just before the acquisition of images, subjects engaged in a short practice session in which they were shown the same computer screens presented during the actual task (‘Tap your Right Hand’, ‘Tap your Left Hand’ and ‘Rest’). During the practice session, they received verbal feedback about their performance.

fMRI finger-sequencing paradigm

For both right-handed finger sequencing (RHFS) and left-handed finger sequencing (LHFS), subjects were asked to successively tap each finger to the thumb in a fixed sequence (index-middle-ring-little) until they received their next visual instruction. Periods of RHFS and LHFS alternated with periods of rest, during which subjects were instructed not to move their hands. For all subjects, the hands were comfortably positioned on the torso.

The fMRI task consisted of a 30 s block of rest, followed by four cycles of 30 s blocks of RHFS, LHFS and rest for a total scan time of 390 s; starting hand was counterbalanced across subjects. Paradigm programming and display were done using E-Prime (Psychology Software Tools Inc., 2002) on a Windows operating system. Subjects were prompted with visual instructions that remained on the screen throughout each 30 s time period. During scanning, finger movements were video recorded; videotapes were later reviewed by an examiner, blind to diagnosis, to determine the total number of finger taps for each hand.

Scanning procedure

Scanning was carried out in a 1.5 T ACS-NT Powertrack 6000 MRI scanner (Philips Medical Systems, Inc.) using body coil transmission and quadrature end-capped head coil reception. T$_1$-weighted high resolution fast-field echo (FFE) anatomical images were acquired coronally [flip angle 45°, repetition time (TR) 35 ms, TE 6 ms, matrix size 256 × 256, field-of-view 240 mm, pixel spacing 0.9375 × 0.9375, slice thickness 1.5 mm]; these were used to create a study-specific template in standardized space used for normalization. For the functional images, axially oriented volumes were acquired every 3.0 s using single-shot echo planar imaging (EPI) 64 × 64 voxel matrix,
3.59 x 5.5 mm voxels, TE 64 ms and flip angle 70°. Each volume was composed of 26 5 mm slices (0.5 mm gap).

Post-acquisition processing

All post-acquisition image processing was carried out using MATLAB version 6.1 (The Mathworks, Inc.) and SPM2 (Wellcome Department of Imaging Neuroscience http://www.fil.ion.ucl.ac.uk/spm/software/spm2/). Region of interest (ROI) analyses were run using MarsBar (http://marsbar.sourceforge.net/).

fMRI data preprocessing

Paediatric brains differ from adult brains in both regional and global dimensions; spatial normalization of paediatric brains to a standard adult template is therefore problematic (Casey et al., 2000; Courchesne et al., 2000). In order to achieve the best possible spatial normalization, a study specific template was created from all 26 participating children. To create the template, each subject’s high resolution anatomical Digital Imaging and Communications in Medicine (DICOM) images were converted to Analyze format and segmented in SPM2. The grey-matter images were then normalized to the Montreal Neurological Institute (MNI) grey-matter template using a 12-parameter affine transformation and averaged to create the study-specific template. As anatomical images have superior geometrical fidelity with respect to echo planar images, the parameters of the transformation into standardized space was not determined from functional images alone. Instead, each individual’s grey-matter image was co-registered to the first volume of the functional scan using a six-parameter affine transformation. The functional volumes were time-corrected to adjust for within-volume time of acquisition differences (Calhoun et al., 2000) and spatially realigned to the location of the first image in the time series. Following this, the co-registered grey-matter images were normalized to the study-specific template using a 12-parameter linear transformation and 16 non-linear transformation iterations and then resampled in voxels of (2 mm)³; these parameters were applied to the functional images. The functional images were then smoothed (Friston et al., 1996) using a Gaussian kernel that was half the resolution of the acquisition matrix (7 x 7 x 11 mm³).

fMRI activation data analysis

SPM2 was used to construct and test the fit of the image data to a general linear model (Friston et al., 1995) corresponding to the time-course of RHFS in contrast to rest and LHFS in contrast to rest. Motion regressors were included as regressors of no interest to account for variance associated with head movement during scanning. Voxel-wise t-maps were constructed for each subject as a first level analysis; the amplitude maps were then carried to a second level to test for significant group effects using Gaussian random field theory. The two level strategy described is equivalent to a random effects analysis, in that it provides a representative activation for a given population that is dominated by inter-subject variance rather than inter-scan variance (Holmes and Friston, 1998).

Single-group, whole-brain random effects analyses for both the HFA and TD groups were accomplished in SPM2 by executing one sample t-tests on the individual subject’s right- and left-rest contrast images. These maps were thresholded at P = 0.0001 and 32 voxels, in order to achieve a corrected statistical threshold of P = 0.05, as determined by the program AlphaSim (B. D. Ward; http://afni.nimh.nih.gov/afni/docpdf/ALPHASim.pdf), used to run 1000 Monte Carlo simulations. The location of voxels significantly associated with RHFS (right-rest contrast) and with LHFS (left-rest contrast) were determined for the single group’s contrasts. They were summarized by their local maxima separated by at least 8 mm, and the maxima were converted from MNI to Talairach coordinate space using formulas provided by Matthew Brett (Medical Research Council-Cognition and Brain Sciences Unit, http://www.mrc-cbu.cam.ac.uk/Imaging/Common/mnspace.shtml). These coordinates were assigned neuroanatomic and cytoarchitectonic labels using the Talairach Daemon (Research Imaging Center, University of Texas Health Science Center at San Antonio http://ric.uthscsa.edu/resources/body.html) and were reviewed by a neurologist (SHM).

Region of interest (ROI) analyses were run to determine if there were significant differences in activation between the HFA and TD groups within regions seen in the single-group, whole-brain analyses. This approach has precedence in the literature (Durston et al., 2006; Suskauer et al., 2008), as it restricts between-group analyses to task-relevant regions and allows for more robust statistical comparisons within regions relative to whole-brain analyses. ROI were defined as the union (‘OR’) between the HFA and TD single-group maps; separate sets of ROIs were created for right- and left-rest contrasts. Within ROIs, two-sample t-tests were run comparing activation between groups for RHFS and LHFS. Results were adjusted for multiple comparisons using a Bonferroni correction based upon the number of ROIs in each analysis (RHFS: 10 ROIs, LHFS: eight ROIs), and significant group differences are reported at a Bonferroni-corrected level of P < 0.05, with trends reported at a Bonferroni-corrected level of P < 0.1.

Additionally, between-group whole-brain analyses were run via a two-sample t-test. These results are shown in uncorrected maps, thresholded at P = 0.05, in order to further explore regions that differ between groups.

fMRI connectivity data analysis

Functional connectivity analyses were run using approaches similar to those previously employed in autism (Just et al., 2004, 2007; Kana et al., 2006, 2007). Motor network ROIs were selected based on one-sample t-tests of RHFS- and LHFS-rest contrasts across the entire sample (HFA and TD, n = 26), for a total of seven regions (bilateral primary motor, bilateral anterior cerebellum, bilateral thalamus and supplementary motor area (SMA)). These ROIs were then used as a mask on each individual subject and only voxels active at a threshold of P = 0.001 in each subject were considered for further analysis. If a subject had less than thirteen active voxels in a ROI, they were excluded from further analyses of that ROI (Kana et al., 2006; Kana et al., 2007). Additionally, control ROIs in the right primary auditory cortex and the brainstem were used to verify the specificity of differences in connectivity.

Time courses for each ROI were extracted using MarsBar, were subsequently high-pass filtered (128 s) and extreme outlier volumes (variance >5 SDs from mean) were excluded (Mazaika et al., 2007). Partial correlations were then run between each ROI pair (21 motor pairs and 15 control pairs), covarying for motion (six regressors, derived from spatial realignment during preprocessing). Of the 21 motor pairs, six were classified as right hand circuits (both ROIs derived from RHFS map), six were classified as left hand circuits (both ROIs derived from LHFS map) and nine were classified as neutral circuits (one ROI derived from each map). Further analyses of these ROIs covaried for activity in the control ROIs (two time course regressors); results of these analyses were qualitatively the same as those that did not covary for activity in control ROIs, and hence only the
analyses covarying for both motion and control ROI activity are reported.

In addition to correlations across the entire time course, connectivity within each condition (rest, RHFS, LHFS) was examined by dividing the time course into the separate conditions. For each 30 s block, composed of 10 3 s TR’s, the first three volumes (6 s) were discarded and one volume (3 s) of the next block was included so as to minimize the effects of hemodynamic delay from previous conditions on within-condition connectivity analyses; this resulted in eight time points per block. Connectivity analyses were run in two ways: (i) by calculating connectivity within each block and averaging correlation coefficients across blocks in the same condition; and (ii) by concatenating blocks in the same condition and correlating the resulting time courses. Partial correlations covarying for motion and control activation could not be carried out due to a lack of degrees of freedom (8 time points versus 10 regressors); however, correlations without covariates demonstrated that the results of the two methods were qualitatively the same, and hence the concatenation method was used for further analyses. After concatenation, partial correlation analyses were run as described above.

After correlations were calculated, these values were normalized using a Fisher’s z transformation. Repeated measures MANOVA analyses were then run examining for effects of diagnosis and condition. For analyses examining individual region-pairs (i.e. correlation between two specific regions), Bonferroni-corrections for multiple comparisons were made separately for each circuit ($P = 0.0083$ for right- and left-hand circuits, $P = 0.0056$ for neutral circuits).

**Results**

**Task performance**

On the motor assessment prior to scanning, children with HFA had significantly higher total PANESS scores than TD children, indicating poorer performance on this composite measure of subtle motor abnormalities [HFA mean = 40.1 ± 12.8, TD mean = 18.8 ± 8.7; $t(23) = 4.93$, $P = 0.0001$; data missing for 1 HFA child]. However, there were no significant between-group differences in time to complete five finger-tapping sequences during either RHFS [HFA mean = 10.5 s ± 3.0, TD mean = 9.5 s ± 3.7; $t(23) = 0.72$, $P = 0.48$] or LHFS [HFA mean = 11.0 s ± 2.3, TD mean = 9.5 s ± 3.3; $t(23) = 1.29$, $P = 0.21$], consistent with published PANESS findings in children with HFA (Jansiewicz et al., 2006).

For task performance during scanning, videotape recordings for 24 (12 TD and 12 HFA) subjects were available for visual count of individual taps. Finger-sequencing speed measured outside the scanner correlated with that inside the scanner for both RHFS [$r^2 = 0.26$; $t(21) = 2.70$, $P = 0.01$] and LHFS [$r^2 = 0.34$; $t(21) = 3.31$, $P = 0.03$]. There was no significant difference in individual taps per 30 s block between groups during RHFS [HFA mean = 58 ± 9, TD mean = 65 ± 15; $t(22) = 1.36$, $P = 0.19$], although there was during LHFS [HFA mean = 56 ± 9, TD mean = 65 ± 11; $t(22) = 2.38$, $P = 0.03$].

**Within-group whole brain activation analyses**

First, separate random effects analyses were computed with right-rest (RHFS) and left-rest (LHFS) contrasts for the HFA and TD groups. During RHFS and LHFS (Fig. 1), both groups demonstrated the expected finding of predominant activation in the contralateral pre/postcentral gyrus (BA3/4) and ipsilateral anterior cerebellum (lobules IV/V). Additionally, both groups showed bilateral activation (left>right) in the superior medial wall (BA6) and contralateral activation in the thalamus. Full details of within-group results can be seen in Table 1.

![Figure 1](http://brain.oxfordjournals.org/) Glass brain and sectional maps showing regions where fMRI activation was significantly associated with LHFS (left images) and RHFS (right images), each contrasted with rest, for TD children (upper images) and children with autism (lower images). All maps were thresholded at $P = 0.05$ corrected for multiple comparisons. Neurologic convention is used (i.e. right = right hemisphere; projections looking rightward or into the page).
Between-group activation analyses

The results of between-group ROI analyses can be seen in Table 2 and Fig. 2. For both RHFS and LHFS, the TD group showed greater activation in the ipsilateral anterior cerebellum (lobules IV/V). Additionally, for RHFS, the TD group showed greater activation in the left (contralateral) anterior cerebellum (lobules IV/V), right (ipsilateral) posterior/inferior cerebellum (lobule VIII A/B) and the left lingual/fusiform gyrus (BA18/19).

As group differences in finger-sequencing speed were noted (significant only for LHFS), ROI analyses were recalculated covarying for sequencing speed in the scanner. This data was not available for two children (1 TD, 1 HFA); however, because finger-sequencing speed inside and outside the scanner were significantly correlated, the regression between the two was calculated and values were imputed for the two children with missing data. After covarying for finger-sequencing speed, all ROIs with greater TD than HFA activation remained significant. Additionally, for LHFS, the right posterior cerebellum (lobule VI) ROI reached significance with Bonferroni correction (Table 2).

Because there was a significant difference in FSIQ, an additional analysis was run covarying for both finger-sequencing speed and FSIQ. Again, results were qualitatively identical, except for two regions that no longer met Bonferroni correction: left lingual gyrus (BA18/19) for RHFS and right posterior cerebellum (lobule VI) for LHFS (Table 2).

Whole brain analyses were also run to determine regions differing between groups, although not at a statistically corrected threshold (Fig. 3). The findings, which should be viewed as exploratory, largely confirmed ROI analyses, highlighting greater activation for both RHFS and LHFS in the TD group in several regions spanning both hemispheres of the cerebellum, as well as several left-lateralized posterior regions, including the left inferior parietal lobule (BA40) and left lingual/fusiform gyrus (BA18/19).

In contrast, greater activation for both RHFS and LHFS in the HFA group was seen bilaterally in the posterior portion of the SMA (BA6).

Functional connectivity analyses

Within both groups, connectivity was seen within motor circuits (HFA: mean z = 9.4, P < 0.0001; TD: mean z = 14.9, P < 0.0001) that was not seen within control circuits (HFA: mean z = 1.3, P = 0.19; TD: mean z = 0.4, P = 0.72), and between-group analyses revealed no significant main effects of diagnosis or condition on connectivity within control circuits and no significant interactions. However, within motor circuits, connectivity in the HFA group was significantly less than in the TD group across the entire time-course [F(1,18) = 13.6, P = 0.0017]; when examined separately by condition, robust differences were seen for RHFS [mean difference = 0.47, F(1,18) = 26.9, P < 0.0001] and LHFS [mean difference = 0.37, F(1,18) = 27.0, P < 0.0001], whereas only marginally significant differences were seen for rest [mean difference = 0.19, F(1,18) = 3.1, P = 0.096] (for details, see Figs 4 and 5). In examining region-pairs individually, a main effect of diagnosis was seen for all the region-pairs (all P < 0.0056) except for the
right and left cerebellum ($P = 0.081$). Significant interaction between diagnosis and condition was seen between the left and right thalamus ($P = 0.004$), left thalamus and SMA ($P = 0.0004$) and right thalamus and SMA ($P = 0.0006$).

### Discussion

To our knowledge, this is the first fMRI study to explore the neural activation and connectivity associated with simple motor execution in children with autism. The results are remarkable for a relative dissociation of cerebral and cerebellar motor regions between children with HFA and their TD peers. While both groups displayed the expected predominant activations in cortical and subcortical regions critical to motor execution (e.g., contralateral pericentral gyrus and ipsilateral cerebellum), children in the TD group showed greater activation in the ipsilateral anterior lobe of the contralateral cerebellum (lobules IV/V) that was absent in the HFA group. The TD findings are consistent with prior imaging studies of basic finger movements in normal adults (Rijntjes et al., 1999; Grodd et al., 2001; Nitschke et al., 2003; Thickbroom et al., 2003; Habas et al., 2004), including the finding of significant, though lesser, activation in the posterior lobe. In contrast, exploratory whole brain analyses revealed greater cerebral activation in the HFA group, located in the SMA proper, which is consistent with findings of increased premotor activation during cued finger sequencing in adults with autism (Muller et al., 2003). This observed pattern of decreased cerebellar activation and increased premotor activation in the HFA group is both distinctive and robust, with interesting implications for further exploration.

There are several potential explanations for the observed cerebral/cerebellar dissociation. First, it is possible that the HFA group’s increased frontal activation and failure to recruit cerebellar regions results directly from anatomical or functional abnormalities in those regions. Consistent with this, adults with ASD were found to show increased frontal activation compared with controls during executive tasks, with structural analyses revealing associated decreases in grey-matter density in those areas (Schmitz et al., 2001; Nitschke et al., 2003; Akshoomoff et al., 2004). Moreover, cerebellar abnormalities are a common finding in post-mortem studies (Williams et al., 1980; Ritvo et al., 1986; Bailey et al., 1998; Kemper and Bauman, 2002), with several studies demonstrating direct associations between behavioural functioning and cerebellar integrity (Pierce and Courchesne, 2001; Pessier and Courchesne, 2001; Akshoomoff et al., 2004; Kates et al., 2004).

The lesser anterior cerebellar activation in our HFA group may also reflect those subjects’ relative inability to shift responsibility of continued motor execution from premotor regions associated with effortful movement to those associated with over-learned or habitual movement. Several studies in normal adults have demonstrated stage-dependent activation in various motor regions, leading to the suggestion that the cerebellum may be preferentially involved in automatic or ‘learned’ motor execution (Seitz et al., 1990; Burnod and Duffose, 1991; Doyon et al., 1996; Shadmehr and Holcomb, 1997; Krebs et al., 1998; Muller et al., 2002).

### Table 2 ROI results for between-group analyses for RHFS and LHFS

<table>
<thead>
<tr>
<th>Hem</th>
<th>Region</th>
<th>BA</th>
<th>t*</th>
<th>Uncorrected P</th>
<th>Uncorrected P covarying for FS speed</th>
<th>Uncorrected P covarying for FS speed and FSIQ</th>
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<tbody>
<tr>
<td>RHFS</td>
<td>L Primary sensorimotor cortex</td>
<td>3/4</td>
<td>0.08</td>
<td>0.467</td>
<td>0.400</td>
<td>0.451</td>
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<tr>
<td></td>
<td>R Anterior cerebellum (Lobules IV/V)</td>
<td></td>
<td>2.55</td>
<td>0.009†</td>
<td>0.008†</td>
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<tr>
<td></td>
<td>L Thalamus</td>
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<td>-0.72</td>
<td>0.238</td>
<td>0.809</td>
<td>0.775</td>
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<td></td>
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<td>0.46</td>
<td>0.324</td>
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</tr>
<tr>
<td></td>
<td>B Medial frontal wall</td>
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<td>1.03</td>
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<td>0.154</td>
<td>0.401</td>
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<tr>
<td></td>
<td>R Posterior/Inferior cerebellum (Lobule VIII A/B)</td>
<td></td>
<td>3.90</td>
<td>&lt;0.001‡</td>
<td>&lt;0.001‡</td>
<td>&lt;0.001‡</td>
</tr>
<tr>
<td></td>
<td>L Anterior cerebellum (Lobules IV/V)</td>
<td></td>
<td>3.39</td>
<td>&lt;0.001‡</td>
<td>&lt;0.001‡</td>
<td>&lt;0.001‡</td>
</tr>
<tr>
<td></td>
<td>R Lingual/Fusiform gyrus</td>
<td>18/19</td>
<td>0.91</td>
<td>0.186</td>
<td>0.211</td>
<td>0.505</td>
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<td>R Postcentral gyrus</td>
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<td>0.409</td>
<td>0.424</td>
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<td>18/19</td>
<td>2.49</td>
<td>0.010‡</td>
<td>0.004‡</td>
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<tr>
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<td>R Primary sensorimotor cortex</td>
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<td>0.374</td>
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<tr>
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<td>L Anterior cerebellum (Lobules IV/VI)</td>
<td></td>
<td>3.33</td>
<td>0.001‡</td>
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<tr>
<td></td>
<td>B Medial frontal wall</td>
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<td>0.311</td>
<td>0.317</td>
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<td>L Precentral gyrus</td>
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<tr>
<td></td>
<td>L Inferior parietal lobe</td>
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<td>1.35</td>
<td>0.096</td>
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<td>0.063</td>
</tr>
</tbody>
</table>

*Positive t-values show regions where TD > HFA, negative t-values show regions where HFA > TD.

$t^*P<0.1$, Bonferroni-corrected for multiple comparisons.

$t^*P<0.05$, Bonferroni-corrected for multiple comparisons.

Cond = Condition, Hem = Hemisphere, BA = Brodmann Area.
In fact, for simple, repeated single digit tapping that requires much less in-scanner learning, adults with autism show the opposite pattern of fMRI activation with decreased premotor (Muller et al., 2001) and increased ipsilateral cerebellar (Allen et al., 2004) activation.

Efficient neuro-functioning is predicated upon the automatization of learned or habitual outputs. From a developmental perspective, deficits in automatization and motor sequence learning might explain impairments in motor coordination commonly reported in autism (Jansiewicz et al., 2006), as well as abnormal and delayed acquisition of motor gestures important for social communication (Gidley Larson and Mostofsky, 2006).

Learning-dependent shifts in motor control depend on the integrity of connections between cortical and subcortical regions. Inefficient, or less organized, neural activation (Muller et al., 2001, 2003; Turner et al., 2006). As such, aberrant neural organization may be a broad neurofunctional or neuroanatomic characteristic of the disorder. This functional disorganization has been attributed to a ‘local overconnectivity’ and ‘long-distance underconnectivity’ of neural circuits in individuals with autism, leading to lesser integration of remote cortical areas (Minshew et al., 1997; Herbert et al., 2004; Happe and Frith, 2006). Anatomic imaging studies reveal increased volume of outer ‘radiate’ white matter volumes comprising localized connections to be the primary contributor to overall brain volume increases in boys with autism (Herbert et al., 2003) and recent findings revealed increased volume of primary motor cortex white matter to be a highly robust predictor of impaired motor function in children with HFA (Mostofsky et al., 2007).

Consistent with this, we found that children with HFA show decreased functional connectivity across nearly the entire network of regions activated during both RHFS and LHFS. The failure of our HFA group to recruit cerebellar regions and their greater reliance instead on premotor cortical regions during finger sequencing, combined with the observed decreased functional connectivity within motor networks that include cerebellar and premotor regions, suggests that autism-associated deficits in motor execution may result from anomalous long-tract connections within the fronto–cerebello–thalamo–frontal network. Further, HFA-associated reductions in functional connectivity within this motor control network were more robust during finger sequencing than during rest, suggesting that decreased connectivity is particularly evident while the network is active (during

Figure 2 Charts showing results of RHFS (upper chart) and LHFS (lower chart) with bar graphs representing mean percent signal change (±SEM) compared with rest for children with autism (blue) and TD controls (red) in ROIs derived from the individual group maps (Fig. 1). **P < 0.05, Bonferroni-corrected for multiple comparisons; *P < 0.1, Bonferroni-corrected for multiple comparisons.

Figure 3 Sectional maps showing localization of differences in fMRI activation between children with autism and TD children during RHFS (red), LHFS (blue), and the overlap between RHFS and LHFS (pink). The upper maps show regions where TD children showed greater activation than did those with autism; the lower maps show regions where children with autism showed greater activation than did TD children. The results are based on a Gaussian random effects analysis of each group of 13 participants; all maps were thresholded at P = 0.05 uncorrected for multiple comparisons. Representative slices are shown in the sagittal (left hemisphere), coronal and axial planes. Neurologic convention is used (i.e. right = right hemisphere; projections looking rightward or into the page).
Indeed, it is worth noting that, as shown in Fig. 4, motor task performance often drives inter-regional correlations in autism below what they are during rest. This might suggest that, given the relative underconnectivity between these distant brain regions, with increasing task demand, it may be more efficient for children with autism to utilize these regions as independent processors, rather than to have them work in concert.

A limitation of the current study is the potential for the behavioural differences between groups to have driven the observed neural activation. While the pre-scanning motor exam suggested no difference in rate of finger apposition, the TD group did have a higher number of taps during imaging of RHFS and LHFS (significant only for LHFS). To address this, the data were reanalysed covarying for number of finger taps; the results did not change, suggesting that the fMRI findings could not be accounted for by differences in finger-sequencing speed.

Some of the children with HFA were taking psychoactive medications, and the potential impact of this cannot be discounted. Future investigations might benefit from exclusion of children taking medications, though this would have a detrimental impact on recruitment of numbers sufficient to examine group differences using BOLD fMRI. With sufficient numbers, comparisons of subjects with autism on/off medications could be applied to future studies.

One particular strength of this study is the use of a study-specific template to normalize the functional data into standardized space. Spatial normalization of paediatric brains to a standard adult template is problematic, since paediatric brains differ from adult brains in both regional and global size and composition (Casey et al., 2000; Courchesne et al., 2000). Additionally, the use of a standard template is especially problematic in disorders such as autism, which have been associated with differences in cerebral volume and composition (Courchesne et al., 2001). As it has been shown that utilizing custom paediatric templates for normalization in paediatric populations improves the quality of normalization (Winkle et al., 2002), our use of a customized study-specific template allowed us to minimize artefacts due to poor normalization.

Though the aetiology of autism is yet unknown, the pervasive-ness of symptoms across modalities suggests that impairments are likely not limited to a single system and that neurological onset is likely quite early. As such, careful examination of the neurologic underpinnings of motor dysfunction in autism may provide insight into mechanisms within parallel systems important for cognitive and behavioural control (Gidley Larson and Mostofsky, 2006). Further, as one of the earliest identifiable traits, motor impairment may serve a principal role in the behavioural phenotype of the disorder, with broad downstream effects across other domains; i.e. early deficits in basic motor abilities may impede the development of compound motor skills and social gestures, contributing to the defining behavioural features of the disorder. Receptive language far outpaces expressive language in many children.
with autism (Gernsbacher et al., 2008) and motor dysfunction might also contribute to delays in productive speech. Indeed, neural systems important for procedural acquisition of motor skills appear to also be critical for language and social development. It follows that abnormalities in these systems may contribute not only to impaired motor skill acquisition in children with autism, but also to impaired communicative and social development (Mostofsky et al., 2000; Walenski et al., 2006). As evidence, recent findings reveal the clearest predictor of optimal outcome in toddlers diagnosed with an autism spectrum disorder is motor skills at age 2 years (Sutera et al., 2007). As such, continued investigation of the neural mechanisms underlying motor development in children with autism is critical to our ongoing understanding of the disorder, as well as the design of effective early interventions. The current study reflects the initial attempts to do so, beginning with a targeted exploration of a simple form of motor execution.

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