White Matter Structure in Autism: Preliminary Evidence from Diffusion Tensor Imaging

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Background: *Individuals with autism have severe difficulties in social communication and relationships. Prior studies have suggested that abnormal connections between brain regions important for social cognition may contribute to the social deficits seen in autism.*

Methods: *In this study, we used diffusion tensor imaging to investigate white matter structure in seven male children and adolescents with autism and nine age-, gender-, and IQ-matched control subjects.*

Results: *Reduced fractional anisotropy (FA) values were observed in white matter adjacent to the ventromedial prefrontal cortices and in the anterior cingulate gyri as well as in the temporoparietal junctions. Additional clusters of reduced FA values were seen adjacent to the superior temporal sulcus bilaterally, in the temporal lobes approaching the amygdala bilaterally, in occipitotemporal tracts, and in the corpus callosum.*

Conclusions: *Disruption of white matter tracts between regions implicated in social functioning may contribute to impaired social cognition in autism.*

Key Words: Autism, social cognition, DTI, white matter, brain, MRI

Impairment in social cognition is a hallmark symptom of autism, a severe developmental disorder also manifested by deficits in communication and by restricted or stereotyped patterns of behavior and motoric activity (American Psychiatric Association 1994).

The ability to engage in social interactions involves diverse cognitive and perceptual abilities and is hypothesized to require the integrated activity of multiple brain regions (Adolphs 1999, 2001; Brothers 1990; Grady and Keightley 2002). One of the most important cognitive skills for social interaction is the ability to attribute mental states to self and others, also referred to as "theory of mind." Converging evidence from functional magnetic resonance imaging (MRI) studies investigating theory of mind tasks in control subjects has shown activation in a network consisting of the amygdala, the medial prefrontal cortex, cingulate cortex, the extrastriate cortex, and the temporoparietal junction (Brunet et al 2000; Castelli et al 2000, 2002; Frith 2001; Gallagher et al 2000). Functional MRI studies investigating social perception in healthy control subjects have shown activation in the fusiform gyrus, the superior temporal gyrus (STG), superior temporal sulcus (STS), and the amygdala (Allison et al 2000; Baron-Cohen et al 1999; Haxby et al 2002; Schultz et al 2003).

Functional brain imaging studies in individuals with autism have found evidence of abnormal brain activation during tasks aimed at eliciting social cognitive responses. For example, compared to control subjects, individuals with autism performing theory of mind tasks show aberrant activation in the ventromedial prefrontal cortex, anterior cingulate cortex, the temporoparietal junction, and the temporal poles adjacent to the amygdala (Baron-Cohen et al 1999; Castelli et al 2002; Happe et al 1996). Subjects with autism also fail to activate the amygdala normally when processing emotional facial and eye expressions (Baron-Cohen et al 1999; Critchley et al 2000). In addition, the fusiform face area is activated less in subjects with autism compared with control subjects during face perception tasks (Critchley et al 2000; Pierce et al 2001; Schultz et al 2000). Finally, subjects with autism show similar levels of activation to those of control subjects in the extrastriate cortex in response to a theory of mind task; however, subjects with autism demonstrate reduced functional connectivity between the extrastriate region and the superior temporal sulcus at the temporoparietal junction, an area associated with theory of mind tasks (Castelli et al 2000, 2002). Consequently, investigators have hypothesized that aberrant connections between such regions comprising a social cognition network may contribute to the social deficits seen in autism (Frith 2001; Grady and Keightley 2002).

The first evidence for potential aberrant connectivity in autism was recently presented in a cross-sectional MRI study (Courchesne et al 2001). This study showed an abnormal developmental trajectory of white matter in autism. Specifically, when compared with control subjects, 2- to 3-year-old boys with autism had increased cerebral and cerebellar white matter volume, whereas adolescent boys had reduced cerebral white matter volume. Such volumetric abnormalities may have resulted from aberrations in axonal density or organization or from myelin abnormalities, either of which could result in aberrant connectivity; however, the volumetric abnormalities seen in autism also could be a result of abnormal glial cell proliferation, which would not necessarily affect white matter connectivity.

To investigate the structural integrity of white matter tracts in individuals with autism, we used diffusion tensor imaging (DTI), a noninvasive magnetic-resonance-based method that enables visualization of white matter tract structure and

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Received May 27, 2003; revised October 28, 2003; accepted October 29, 2003.

0006-3223/04/$30.00
doi:10.1016/j.biopsych.2003.10.022

Table 1. Subject Demographics

Variable	Control Group	Autism Group[a]	p^b	t
Number of Subjects	9	7	.5	
Age (years)	13.4 ± 2.8	14.6 ± 3.4	.5	t (10.5) = 0.6
FSIQ[c]	107 ± 8.5	101 ± 12.2	.28	t(10.2) = 1.14
VIQ	105.2 ± 11.4	84 ± 17	.018	t(10) = 2.8
PIQ	107 ± 10	121.5 ± 8	.008	t(13) = 3.1

FSIQ, full-scale IQ; PIQ, performance IQ; VIQ, verbal IQ.

[a]Inclusion criteria were DSM-IV-based diagnosis of autism and Autism Diagnostic Observational Schedule, Generic.

[b]Statistically significant was $p < .05$.

[c]Using Wechsler Abbreviated Scale of Intelligence, only subjects with FSIQ ≥70 were included.

coherence in vivo. We hypothesized that white matter located between brain regions known to be involved in social cognition would be morphologically abnormal in autism.

Methods and Materials

Subjects were 16 male children and adolescents, 7 diagnosed with high-functioning autism and 9 typically developing control subjects (Table 1). The diagnosis of autism was based on DSM-IV criteria (American Psychiatric Association 1994), the Autism Diagnostic Inventory—Revised (Rutter et al 1995), and the Autism Diagnostic Observational Schedule, Generic (Lord et al 1999). In addition, a history of delayed phrase speech development (i.e., after 36 months) was an inclusion criterion for the autism group.

Standardized cognitive testing using the Wechsler Abbreviated Scale of Intelligence (Zhu 1999) was administered to all subjects. Only subjects with full-scale IQ ≥ 70 were included in our study.

All subjects with autism were screened to exclude major medical, genetic (e.g., fragile X syndrome), or psychiatric conditions. All control subjects were in good physical health and were screened to exclude neurologic, developmental, or psychiatric disorders.

All participating subjects and their parents signed a written informed consent approved by the institutional review board of Stanford University.

Magnetic resonance images were acquired using a GE-Signa 3-Tesla scanner (General Electric, Milwaukee, Wisconsin). A DTI sequence was based on a single-shot spin-echo, echo-planar imaging (EPI) sequence with diffusion sensitizing gradients applied on either side of the 180° refocusing pulse (Basser et al 1994; Moseley et al 1991). Imaging parameters for the diffusion-weighted sequence were as follows: field of view (FOV) = 24 cm, matrix size 128 × 128, echo time/response time = 106/6000 msec, 19 axial-oblique slices, slice thickness 5 mm/skip 1.5 mm. The scan was prescribed from the top of the brain and included only the most superior part of the cerebellum. Diffusion gradient duration was δ = 32 msec, diffusion weighting was b = 900 sec/mm^2. In addition, T2-weighted images were acquired by removing the diffusion sensitizing gradients. Diffusion was measured along six noncollinear directions: XY, XZ, YZ, −XY, −XZ, and −YZ. This pattern was repeated four times for each slice with the sign of all diffusion gradients inverted for odd repetitions.

The variable of interest, fractional anisotropy (FA), is a measure that reflects the degree of diffusion anisotropy within a voxel, which is determined by microstructural features of the tissue, including fiber diameter and density, degree of myelination (Basser 1995), and macrostructural features such as intravoxel fiber-tract coherence (Pierpaoli and Basser 1996).

The FA was calculated for each voxel according to Basser and Pierpaoli (1996) to produce a fractional anisotropy image. The FA images were further processed using Statistical Parametric Mapping software (SPM99; Wellcome, London, United Kingdom). The T2-weighted image map was used to determine normalizing parameters subsequently applied to the FA images using SPM99. The normalized FA images were smoothed with a 4-mm kernel to increase the signal-to-noise ratio. A white matter mask was applied to the images to eliminate noise and edge effects. Because the brain stem and cerebellum were only partially scanned, they were excluded from the mask. These smoothed images for controls and subjects with autism were compared using voxelwise two-tailed t tests. Results were normalized to Z scores to provide a statistical measure of differences

Table 2. Brain Regions Showing Significant Differences between Subjects with Autism and Control Subjects

Location of Significant FA Differences	Cluster Size in Voxels	Talairach Coordinates of Most Significant Voxel			Z Score
		x	y	z	
Right Motor and Premotor Areas, Extending into the Centrum Semiovale	328	38	5	26	3.97
Adjacent to Right Striate and Extrastriate Cortex, Extending into the Optic Radiations, the Temporoparietal Junction, the Superior Temporal Gyrus, and the Middle Temporal Gyrus	1064	28	−81	11	3.58
Along the Right Middle Frontal Sulcus	366	36	28	17	3.28
Left Superior and Middle Temporal Gyrus Extending into the Temporoparietal Junction	648	−38	−25	10	3.26
Genu and Body of the Corpus Callosum Extending into the Subgenual Ventromedial Prefrontal Regions Bilaterally and the Anterior Cingulate Gyri	1043	0	31	6	3.11
Left Optic Radiations Extending into the Medial Temporal Gyrus, Adjacent to the Fusiform Gyrus, Extending into the Parahippocampal Gyrus	477	−30	−62	3	2.72

Clusters in which controls had higher fractional anisotropy than subjects with autism are shown with their peak coordinates in Talairach space and the associated Z-scores, along with their size in voxels.

FA, fractional anistropy.

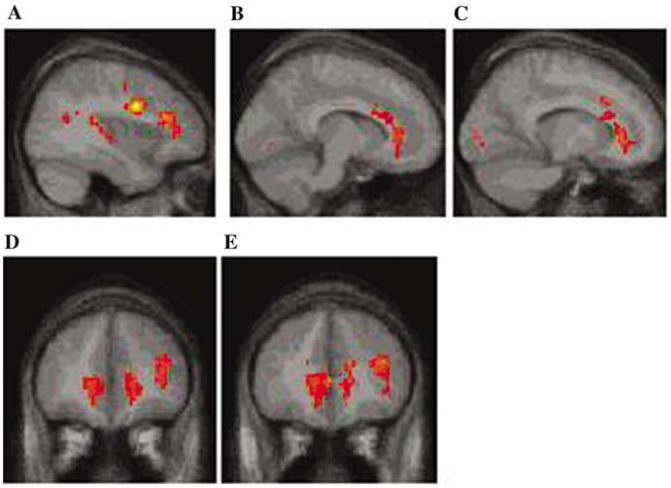

Figure 1. Voxels that showed significant reduction in white matter fractional anisotropy in patients with autism compared with control subjects, mapped onto an average T1-weighted image of control and autism brains. (**A, B, C**) Sagittal view, significant clusters shown in the ventromedial prefrontal region (**A, B**), the anterior cingulate and subgenual area (**C**), the corpus callosum (**B, C**), the superior temporal gyrus (**A**), centrum semiovale (**A**), and temporoparietal junction (**A**). (**D**) Coronal view, significant clusters shown in the ventromedial prefrontal region bilaterally and along the middle frontal sulcus (**D, E**).

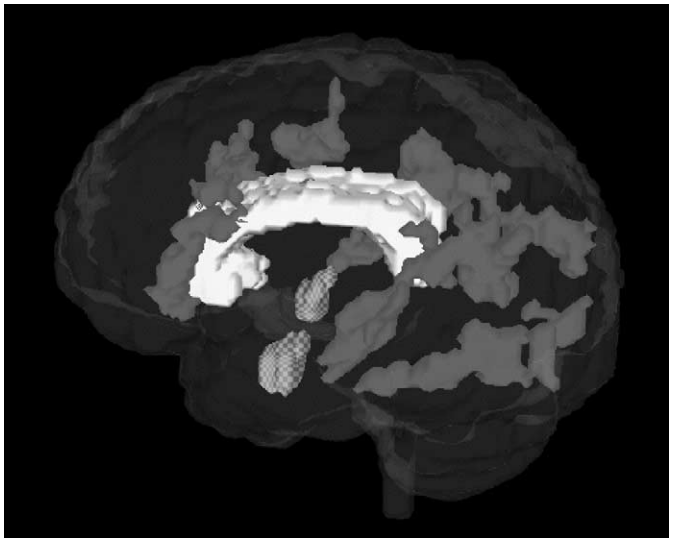

Figure 2. A three-dimensional representation of the aberrant white matter (dark gray) in relation to the corpus callosum (white), and the amygdala (checkered gray).

between voxels that are independent of sample size. Finally, the joint expected probability distribution of the height and extent of Z scores, with height ($Z > 1.67$; $p < .05$) and extent ($p < .05$) thresholds, was used to determine the presence of significant clusters of difference and correct for spatial correlation in the data.

Results

As shown in Table 1, there were no significant differences in full-scale IQ between the two groups; however, subjects with autism had a significantly lower verbal IQ score, as expected. In this sample, the performance IQ was significantly higher in subjects with autism compared with control subjects.

Subjects with autism showed significantly reduced FA values in, and adjacent to, the anterior cingulate bilaterally, extending into the body and genu of the corpus callosum, the ventromedial prefrontal areas, and the subgenual prefrontal regions (Figure 1 and Figure 2). In addition, reduced FA values were observed in the temporoparietal junctions bilaterally and adjacent to the superior temporal sulci. In the right hemisphere, this cluster extended into the optic radiation as well as into the middle temporal gyrus approaching the right amygdala. Additional clusters of reduced FA were seen in the left optic radiation extending into the left fusiform gyrus, in the left middle temporal gyrus approaching the left amygdala (shown in Figure 2), and in white matter located between the right middle and inferior frontal gyri and in the right centrum semiovale, as well as motor and premotor areas.

There were no significant increases in FA values in the autism group when compared with the control group.

Discussion

Our preliminary results suggest that white matter structure is disrupted in subjects with autism. Specifically, reduced FA values were seen adjacent to brain regions that have been implicated in social cognition. Among these are regions important for face and gaze processing (fusiform gyrus and the superior temporal sulcus; Kanwisher et al 1997; Puce et al 1995, 1998) and for awareness of mental states and emotional processing (anterior cingulate, amygdala, ventromedial prefrontal cortex; Adolphs et al 2002; Bush et al 2000; Damasio 1994). In addition, in our results, areas of aberrant white matter were found between those gray matter regions activated in theory of mind tasks, including the ventromedial prefrontal cortex, anterior cingulate, temporoparietal junction, superior temporal sulcus, and the amygdala (Brunet et al 2000; Castelli et al 2000, 2002; Frith 2001; Gallagher et al 2000). We also found aberrant white matter structure between the extrastriate region and temporoparietal regions. The latter regions are also activated during theory of mind tasks and were previously shown to have reduced functional connectivity in autism (Castelli et al 2002). Disruptions in these pathways could impair the integration of information necessary for processing socially relevant stimuli.

Reduced FA values were also observed within callosal fibers important for connecting prefrontal (genu) and motor, sensory, and auditory cortices (body) (Pandya 1986), as well as in white matter adjacent to prefrontal regions. Structural changes in the corpus callosum may disrupt executive functioning and sensorimotor processing requiring effective bihemispheric transfer. Taken together, these impairments could lead to disruption of effective neural connectivity necessary for children with autism to develop adaptive social skills. In addition, these white matter disruptions could underlie impairments in cognitive functioning and the abnormal responses to sensory stimuli seen in autism (Rapin 1991).

Previous research has suggested two possible explanations for white matter disruption in autism. First, abnormal cerebrospinal fluid levels of insulinlike growth-factor-1 (IGF-1; Vanhala et al 2001), as well as abnormal blood levels and synthesis capacity of serotonin, have been described in autism (Anderson et al 1990; Chugani et al 1999; Warren and Singh 1996). Both are important neurotrophic factors for brain development (Vanhala et al 2001; Whitaker-Azmitia 2001). In addition, elevated levels of

other brain growth factors important for neuronal and glial development and for circuit organization were found in neonates who later developed autism or mental retardation (Nelson et al 2001). Abnormal levels of neurotrophic factors during critical periods of brain development could result in disrupted white matter development (Dreyfus 1998). Second, data of early overgrowth in brain tissue volumes led to the hypothesis that neuronal pruning in autism is impaired (Courchesne et al 2001; Frith 2003). Such primary dysfunction in brain regions important for social cognition may contribute to abnormalities in the developmental trajectory of white matter tracts to those areas.

Further research with larger samples is needed to replicate our findings. In addition, future studies with fiber-tracking techniques are necessary to better delineate the exact white matter pathways affected in autism.

This is the first report of disrupted white matter tracts using DTI in autism. These findings may help improve our understanding of the neuroanatomic basis underlying the deficits in social information processing that are seen in autism and related disorders.

The research presented in this manuscript was supported by National Institutes of Health Grant Nos. MH01142, HD31715, MH064708, and HD40761, and the Sinclair Fund. We thank Christopher Dant and Amy Garrett for their help in manuscript preparation.

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