Published in final edited form as:

Psychiatry Res. 2007 December 15; 156(3): 225–245.

Voxel-Wise Comparisons of the Morphology of Diffusion Tensors Across Groups of Experimental Subjects

Ravi Bansal^{a,b}, Lawrence H. Staib^c, Kerstin J. Plessen^{b,d}, Dongrong Xu^{a,b}, Jason Royal^b, and Bradley S. Peterson^{a,b}

^a New York State Psychiatric Institute, New York, NY 10032

^b Department of Psychiatry, Columbia University, New York, NY 10032

^c Departments of Electrical Engineering and Diagnostic Radiology, Yale University, New Haven, CT 06512

^d Center for Child-and Adolescent Mental Health, University of Bergen, Norway

Abstract

Water molecules in the brain diffuse preferentially along the fiber tracts within white matter, which form the anatomical connections across spatially distant brain regions. A diffusion tensor (DT) is a probabilistic ellipsoid composed of 3 orthogonal vectors, each having a direction and an associated scalar magnitude, that represent the probability of water molecules diffusing in each of those directions. The 3D morphologies of DTs can be compared across groups of subjects to reveal disruptions in structural organization and neuroanatomical connectivity of the brains of persons with various neuropsychiatric illnesses. Comparisons of tensor morphology across groups have typically been performed on scalar measures of diffusivity, such as Fractional Anisotropy (FA), rather than directly on the complex 3D morphologies of DTs. Scalar measures, however, are related in nonlinear ways to the eigenvalues and eigenvectors that create the 3D morphologies of DTs. We present a mathematical framework that permits the direct comparison across groups of mean eigenvalues and eigenvectors of individual DTs. We show that group-mean eigenvalues and eigenvectors are multivariate Gaussian distributed, and we use the Delta method to compute their approximate covariance matrices. Our results show that the theoretically computed Mean Tensor (MT) eigenvectors and eigenvalues match well with their respective true values. Furthermore, a comparison of synthetically generated groups of DTs highlights the limitations of using FA to detect group differences. Finally, analyses of in vivo DT data using our method reveal significant between-group differences in diffusivity along fiber tracts within white matter, whereas analyses based on FA values failed to detect some of these differences.

Keywords

Central Limit Theorem; Delta Method; Fisher F-Distribution; Fractional Anisotropy; Multivariate Gaussian Distribution

Corresponding Author: Dr. Ravi Bansal. Contact Address: Room #2410, Unit 74, New York State Psychiatric Institute, 1051 Riverside Dr., New York, NY 10032.

Publisher's Disclaimer: This is a PDF file of an unedited manuscript that has been accepted for publication. As a service to our customers we are providing this early version of the manuscript. The manuscript will undergo copyediting, typesetting, and review of the resulting proof before it is published in its final citable form. Please note that during the production process errors may be discovered which could affect the content, and all legal disclaimers that apply to the journal pertain.

1 Introduction

Diffusion Tensor Imaging (DTI) can detect subtle abnormalities in white matter by assessing the degree to which fiber tracts within that tissue have lost their directional organization (Basser et al., 1994; Basser, 1995). The myelin sheath and cell membranes of axons in white matter fibers of the brain restrict water molecules from diffusing in a direction perpendicular to the fiber tracts. Water molecules therefore diffuse preferentially along the length of axons (Pierpaoli et al., 1996). Disruption of the myelin sheath, loss of the integrity of microstructures with the neurons of the fiber tracts, or an increase in fluid volume can cause water molecules to diffuse more equally in all directions in the areas of disruption. Thus, by quantifying the diffusivity of water molecules, DTI provides an indirect *in vivo* measure of the integrity of fiber tracts in white matter. Statistical analyses of differences in diffusivity between groups have revealed associations of possible disruption or disorganization in neuroanatomical connections with specific neuropsychiatric disorders.

DT data representing the diffusivity of water are encoded as 3×3 symmetrical diffusion tensors, and scalar measures derived from these DTs, such as Fractional Anisotropy (FA), Apparent Diffusion Coefficient (ADC) (Pierpaoli et al., 1996), Voxel Scale Connectivity (VSC) (Parker et al., 2002), and the coherence measure (Klingberg et al., 2000), have been used extensively to compare diffusivity across individuals and thereby to identify disturbances in neuroanatomical connectivity within specific diagnostic groups of subjects (Lim and Helpert, 2002). FA and the coherence measure, for example, have been used to document disruptions in the microarchitecture of white matter in the brains of infants born prematurely (Nagy et al., 2003); an *in vivo* study of the white matter fibers that form temporal-frontal connections showed that patients with schizophrenia lack the left-greater-than-right asymmetry in FA values found in normal controls (Kubicki et al., 2002); and ADC values in infants with overt white matter pathology have been reported to be significantly higher than ADC values in healthy infants (Counsell et al., 2003).

These scalar measures, however, are not ideal for detecting group differences. FA values, for example, are useful as a measure of the degree of anisotropy, but they are related nonlinearly to the eigenvalues of a diffusion tensor (Fig. 1). Therefore, even when sets of eigenvalues differ, the FA values calculated for the sets may be similar (i.e., the mapping of eigenvalues to FA values is non-unique) and thus may conceal between-group differences. Moreover, the sample average FA value calculated from the FA values of individual tensors will not necessarily equal the FA value calculated from the mean of those individual tensors. Similarly, the ADC is a linear combination of eigenvalues that provides information about overall diffusivity, but it too is limited in its ability to detect group differences. Furthermore, the FA and ADC each quantify the magnitude of diffusion but provide little information about the direction of diffusion, and thus comparisons of these scalar indices provide little insight into differences in local fiber trajectory across groups. The coherence measure, in contrast, depends only upon the eigenvectors of diffusion tensors and therefore ignores all differences in their associated eigenvalues; it therefore provides little insight into differences in the rates of diffusion across groups. Each of these measures therefore is limited in its ability to reveal group differences in tensor morphology.

Recently, a number of advanced methods have been proposed to compare tensors across groups of individuals. Between-group differences, for example, have been studied by comparing diffusion along the principal directions (PDs) of tensors (Schwartzman et al., 2005). The accuracy of this methods, however, requires the unambiguous determination of the principal direction of each tensor (the eigenvector of the tensor having the largest associated eigenvalue), which the presence of noise in DTI datasets renders difficult. When estimating tensors, noise in DTI datasets is commonly assumed to follow a Gaussian distribution (Pajevic and Basser,

2003; Basser and Pajevic, 2003); the distribution of noise, however, has finite weight where tensors are not positive definite, because positive-definite tensors are only a subspace of Euclidean space. To ensure that tensors are positive definite, some investigators have suggested using logarithmic transformations to first convert tensors to symmetric matrices, then computing statistics for these symmetric matrices and using exponential transformations to generate positive-definite tensors (Arsigny et al., 2005; Schwartzman, 2006). These methods, however, depend upon the estimation of only a single parameter to capture variations in all three eigenvalues. The use of Riemannian geometry to describe a positive-definite tensor subspace within $\mathbb{R}^6$ Euclidean space has allowed computation of an intrinsic mean tensor and an index of geodesic anisotropy (Fletcher and Joshi, 2004; Batchelor et al., 2005). However, the probability distributions and statistical properties of the eigenvectors and eigenvalues of the mean tensor (MT) cannot be computed using these techniques because the MT is computed using nonlinear optimization methods. Furthermore, other methods that compare mean eigenvalues (Martin et al., 1999) or eigenvectors (Schwartzman et al., 2005) across individuals are prone to errors in determining the corresponding eigenvalues and eigenvectors across tensors in the presence of noise.

We have developed a mathematical framework for directly comparing the eigenvalues and eigenvectors of the MTs across groups of subjects. We first use the Central Limit Theorem (CLT) to compute the statistical properties of the MT for a group of subjects, and then we use the Delta method to derive the probability distribution of the eigenvalues and eigenvectors of this MT. Because the space of positive-definite tensors is dense and closed under addition, our computed MT is positive-definite for each group of subjects being compared. Additionally, our method averages the tensor elements together across individuals, thereby avoiding the need to determine corresponding eigenvalues and eigenvectors across different tensors for a group of subjects. Furthermore, in comparing tensors across groups, we are interested in the statistical properties of the MT only and not those of the individual tensors themselves. Therefore, because the diffusion tensor space is closed under addition, and because our estimate of the sample MT is unbiased, we can apply the CLT to determine the distribution of the MT. Comparing the MTs across different groups of subjects using this method yields the statistical significance, or *P-value*, of the differences in average tensor morphology across groups at each voxel in the imaging volume. Our method thus allows us to specify better the morphological origins (in terms of the directional orientation of fibers through a voxel element, or the magnitude of diffusion along fiber tracts) of the statistically significant, between-group differences in tensor morphology.

In our framework, we use a multivariate Gaussian distribution to represent the variability in diffusion tensors across subjects; therefore, 3×3 symmetric tensors are represented in their equivalent 6×1 vector forms. Furthermore, because correlations among random variables do not depend upon their representation, a covariance matrix within a multivariate Gaussian distribution captures the full range of variability in the tensors that compose that distribution. Additionally, the elements of the covariance matrix are uniquely related on a one-to-one basis to the elements of the positive-definite and symmetric, fourth-order precision tensor $\mathbf{A}$ that is used to describe diffusion tensors (Basser and Pajevic, 2003). Therefore, all information about tensors represented in the precision tensor $\mathbf{A}$ can be deduced from the covariance matrix. Thus, the covariance matrix and the mean tensor uniquely describe the distribution of the group of tensors, under the assumption that tensors are multivariate Gaussian distributed. Finally, representing tensors as 6×1 vectors allows us to use well developed tools for multivariate statistics to compare tensors across groups.

2 Methods

To detect voxel-wise, between-group differences in diffusion tensors, we compute the mean tensor (MT) for a group of tensors, which in turn is used to calculate the eigenvalues and eigenvectors of the MT, as well as their covariance matrices. Under the null hypothesis for this comparison, MT eigenspace components for the groups are equal at each voxel. We test this hypothesis using Hotelling's T^2 -statistic to calculate probability values (P -values) at each voxel in the imaging space, thereby generating a P -value map that depicts where in the volume the DT measure being compared is likely to differ across groups. By thresholding the P -value map at a specified significance level, we can then detect statistically significant differences in the diffusivity of water at each voxel.

In the presence of noise in a DT dataset, mean eigenvalues and eigenvectors and their covariance matrix for a group of subjects cannot be computed from the eigenvalues and eigenvectors of the individual DTs. For example, the principal eigenvector of one tensor could be closely aligned in a second tensor with an eigenvector other than the principal eigenvector. In this instance, we cannot determine whether to average the eigenvalues that correspond to the PDs of these two tensors, or to average the eigenvalues that correspond to the eigenvectors that are most closely aligned across the two tensors. Computing the average of the principal diffusivity across these two tensors could be erroneous using either approach. Our method explicitly addresses these ambiguities by averaging elements y_i , $i = 1, \dots, N$ across N different DTs and then computing their mean values μ_y and covariance matrices Σ_y , for each group of subjects. According to the Central Limit Theorem (CLT), the mean values μ_y of tensor elements are multivariate Gaussian distributed. We then infer the probability distribution of the eigenvectors and eigenvalues (the *eigenspace components*) of the MT from the probability distribution of μ_y , as described in the following subsections.

We assume that DT images have been normalized (registered and reoriented) into a common coordinate space to permit voxel-wise statistical analysis of tensors across subjects. Errors in normalizing these images (e.g., from motion, cardiac pulsation, or thermal noise) will increase noise in statistical analyses of the tensors and thereby reduce our power to detect significant between-group differences. Furthermore, using many degrees of freedom in the spatial transformation of tensor maps could diminish the real differences in tensor morphology across groups. DTI datasets, however, can be normalized accurately using any one of several excellent existing methods (Ashburner and Friston, 1999; Alexander et al., 2001; Ruiz-Alzola et al., 2000, 2002; Xu et al., 2003) (Netsch and Muiswinkel, 2003; Park et al., 2003; Westin and Knutsson, 2003; Wang et al., 2004) (Bansal et al., 2005b), thereby avoiding this inappropriate elimination of real group differences. We should note that although the accurate spatial normalization of DTI datasets is essential for the valid comparison of tensor morphologies across groups, our method for the statistical comparison of tensor morphologies is independent of the method used to normalize DT images.

2.1 Distribution of Mean Tensor Elements

Let D_i , $i = 1, \dots, N$ be N diffusion tensors at a corresponding voxel in the spatially normalized and reoriented DT images of N subjects, and let $y_i = (d_{11}^i \ d_{12}^i \ d_{23}^i \ d_{22}^i \ d_{23}^i \ d_{33}^i)^T \in \mathbb{R}^6$, be a 6×1 random vector of the elements of a diffusion tensor D_i . Because of the positive-definite constrain on diffusion tensors, y_i is restricted to only a subspace of $\mathbb{R}^6$. Consider a sequence of random vectors $\bar{y}_n$, $n = 1, \dots, N$, in which $\bar{y}_n$ is computed by averaging the first n random vectors y_i : $\bar{y}_n = \frac{1}{n} \sum_{i=1}^n y_i$. If y_i are independent and identically distributed (i.i.d.) and $E\|y_i\| < \infty$ then by the strong law of large numbers, $\bar{y}_n$ converges *almost surely* (a.s.) to the

expected value of the random vector $\mathbf{y}_i$, i.e., $\bar{\mathbf{y}}_n \xrightarrow{a.s.} E[\mathbf{y}_i]$. Under the stronger assumption that $E\|\mathbf{y}_i\|^2 < \infty$, we obtain by the CLT (van der Vaart, 2000)

$$\sqrt{n}\bar{\mathbf{y}}_n \rightsquigarrow N(E[\mathbf{y}_i], \Sigma_{\mathbf{y}_i}),$$

where $\Sigma_{\mathbf{y}_i}$ is the covariance matrix of the random vector $\mathbf{y}_i$; that is, the sequence of random vectors $\bar{\mathbf{y}}_n$ converge in distribution to a multivariate Gaussian distribution, irrespective of the distributions of the tensors $\mathbf{y}_i$.

Because the positive-definite subspace of diffusion tensors in $\mathbb{R}^6$ is dense and closed under the mathematical operation of addition, the MT for a group of subjects is well defined and positive-definite. Additionally, the variance of the MT will be much smaller than is the distribution of the tensors used to compute the MT itself, with its variance-covariance reducing by a factor of n for an increasing number n of observations. With 20 subjects in each group, for example, the variance of the MT will be 20 times smaller than the overall variance for the tensors across all 20 subjects in the group. Asymptotically, the distribution of the MT $\bar{\mathbf{y}}_n$ is degenerate at $E[\mathbf{y}_i]$ (i.e. will have nonzero probability mass at only $E[\mathbf{y}_i]$). Therefore, although the multivariate Gaussian distribution describing the mean random vector $\bar{\mathbf{y}}_n$ will have nonzero probability mass in the portion of Euclidean space $\mathbb{R}^6$ where the tensor is not positive-definite, this mass is zero asymptotically, and we can apply the CLT to determine the multivariate distribution of the MT.

For sufficiently large n , the random vectors $\mathbf{y}_i$ need not be identically distributed to apply the CLT. If $E(\mathbf{y}_i) = \mu_i$ and $\text{Cov } \mathbf{y}_i = \Sigma_i$, and if we define

$$\mu_{\mathbf{y}} = \frac{1}{n} \sum_{j=1}^n \mu_j \quad \text{and} \quad \Sigma = \frac{1}{n} \sum_{j=1}^n \Sigma_j,$$

then $\sqrt{n}\bar{\mathbf{y}}_n \rightsquigarrow N(\mu_{\mathbf{y}}, \Sigma)$ as $n \rightarrow \infty$.

2.2 Transformation of Tensor Elements into Eigenspace Components

We derive herein a closed form analytic transformation $\phi(\cdot)$ between tensor elements and eigenspace components. Because the eigenvectors of a symmetric 3×3 matrix form an orthonormal coordinate system (Golub and Loan, 1996), we represent eigenvectors of a diffusion tensor by three angles $(\alpha \beta \gamma)$ that rotate the x-, y-, and z-coordinate axes into the eigenvectors. We therefore represent eigenspace components by a 6×1 random vector $\mathbf{x} = (\lambda_1 \lambda_2 \lambda_3 \alpha \beta \gamma)^T$ whose elements are three eigenvalues $(\lambda_1 \lambda_2 \lambda_3)$ and three rotation angles $(\alpha \beta \gamma)$.

2.2.1 Transformation of Eigenvalues—Eigenvalues are estimated by setting the determinant of the matrix $(D - \lambda I_3)$ equal to zero, where I_3 is an identity 3×3 matrix. Thus, we have

$$\det(D - \lambda I_3) = 0 = -\lambda^3 + c_2 \lambda^2 - c_1 \lambda + c_0,$$

where (Schneider and Eberly, 2002)

$$\begin{aligned}
c_0 &= d_{11}d_{22}d_{33} + 2d_{12}d_{13}d_{23} - d_{11}d_{23}^2 - d_{22}d_{13}^2 - d_{33}d_{12}^2, \\
c_1 &= d_{11}d_{22} - d_{12}^2 + d_{11}d_{33} - d_{13}^2 + d_{22}d_{33} - d_{23}^2, \text{ and} \\
c_2 &= d_{11} + d_{22} + d_{33}.
\end{aligned}$$

Note that c_0 is the determinant of the matrix D , and c_2 is its trace. To evaluate the transformation for eigenvalues, define $a = (3c_1 - c_2^2)/3$, $b = \left(\frac{-2c_2^3 + 9c_1c_2 - 27c_0}{27} \right)$, and $Q = \left(\frac{b^2}{4} + \frac{a^3}{27} \right)$; then eigenvalues depend upon the value of Q .

Case I: $Q > 0$: This is possible only if the tensor D is a multiple of the identity matrix (Schneider and Eberly, 2002), i.e., $D = \lambda I_3$. In this case, all eigenvalues are equal.

Case II: $Q = 0$: Here, only two distinct eigenvalues exist:

$$\lambda_1 = \frac{c_2}{3} + \left(\frac{b}{2}\right)^{1/3}, \quad \lambda_2 = \lambda_1, \quad \lambda_3 = \frac{c_2}{3} - 2\left(\frac{b}{2}\right)^{1/3}. \quad (1)$$

Case III: $Q < 0$: In this instance, each of the three eigenvalues is distinct. If we define

$$\theta = \arctan(\sqrt{-Q} - b/2), \quad \rho = \sqrt{(b/2)^2 - Q};$$

then the eigenvalues are computed as (Hasan et al., 2001; Shen et al., 2006)

$$\begin{aligned}
\lambda_1 &= \frac{c_2}{3} + 2\rho^{1/3} \cos(\theta/3) \\
\lambda_2 &= \frac{c_2}{3} - \rho^{1/3} [\cos(\theta/3) + \sqrt{3} \sin(\theta/3)] \\
\lambda_3 &= \frac{c_2}{3} - \rho^{1/3} [\cos(\theta/3) - \sqrt{3} \sin(\theta/3)].
\end{aligned} \quad (2)$$

2.2.2 Transformation of Eigenvectors—Eigenvectors are specified by the columns of the rotation matrix R that rotates the x-, y-, and z- coordinate axes. If α , β , and γ denote rotations about the x-, y-, and z-axes, respectively, then the rotation matrix R is

$$R = \begin{pmatrix} \cos \beta \cos \gamma & \cos \beta \sin \gamma & -\sin \beta \\ -\cos \alpha \sin \gamma + \sin \alpha \sin \beta \cos \gamma & (\cos \alpha \cos \gamma + \sin \alpha \sin \beta \sin \gamma) & \sin \alpha \cos \beta \\ (\sin \alpha \sin \gamma + \cos \alpha \sin \beta \cos \gamma) & (-\sin \alpha \cos \gamma + \cos \alpha \sin \beta \sin \gamma) & \cos \alpha \cos \beta \end{pmatrix}^T.$$

Eigenvectors, v_i ; $i = 1, 2, 3$, are estimated by solving the equations $(D - \lambda_i I_3)v_i = 0$. Because eigenvectors of a symmetric matrix are orthogonal, columns of the rotation matrix R represent the eigenvectors themselves. Thus, to estimate eigenvectors, we estimate the rotation angles α , β , and γ . We discuss the degenerate case in the next subsection.

Let $M = (D - \lambda_3 I_3) = [m_{ij}]$. Then we solve the equation

$$0 = M \begin{pmatrix} \cos \beta \cos \gamma \\ \cos \beta \sin \gamma \\ -\sin \beta \end{pmatrix} = \begin{pmatrix} m_{11} & m_{12} & m_{13} \\ m_{12} & m_{22} & m_{23} \\ m_{13} & m_{23} & m_{33} \end{pmatrix} \begin{pmatrix} \cos \beta \cos \gamma \\ \cos \beta \sin \gamma \\ -\sin \beta \end{pmatrix}.$$

Assuming that $\cos \beta \neq 0$, that is, the rotations are less than 90° ($-\frac{\pi}{2} < \beta < \frac{\pi}{2}$), we obtain

$$\tan \gamma = \frac{m_{12}m_{33} - m_{13}m_{23}}{m_{23}^2 - m_{22}m_{33}}, \text{ and } \tan \beta = \cos \gamma \left(\frac{m_{13} + m_{23} \tan \gamma}{m_{33}} \right). \quad (3)$$

To estimate α , consider a different eigenvalue and let the eigenvector that corresponds to this eigenvalue be $((-\cos \alpha \sin \gamma + \sin \alpha \sin \beta \cos \gamma) (\cos \alpha \cos \gamma + \sin \alpha \sin \beta \sin \gamma) \sin \alpha \cos \beta)^T$. Solving for α and assuming that $\cos \alpha \neq 0$ and $\cos \gamma \neq 0$, we obtain

$$\tan \alpha = \left[\frac{m_{11}m_{23} - m_{13}m_{12}}{\sin \beta \tan \gamma (m_{13}m_{12} - m_{11}m_{23}) + (m_{13}^2 - m_{11}m_{33}) \frac{\cos \beta}{\cos \gamma}} \right]. \quad (4)$$

These relations are used to compute the Jacobian of the transformation $\phi(\cdot)$ in the analysis below. To compute the Jacobian using Eqns. (3) and (4) requires a correspondence between eigenvalues and eigenvectors; we determine this correspondence using numerical methods (Press et al., 1992) that compute eigenvalues and eigenvectors of matrices.

2.3 Special Case Mean Tensors

For an isotropic MT, Eqns. (3) and (4) cannot be used to determine the eigenvectors. Because the eigenvectors of an isotropic MT can be rotated to match those of the other group, in this case we compare only the eigenvalues of the MT across the two groups. When the MT is either prolate (i.e. the smaller two eigenvalues are equal) or oblate (i.e. the larger two eigenvalues are equal), we statistically compare only the eigenvalues and the one angle that can be computed uniquely. However, exactly isotropic, prolate, or oblate MTs are rare because eigenvalues are continuous-valued random variables, and the precise equality of eigenvalues within a tensor is unlikely. Therefore, we do not discuss these cases further in the paper.

2.4 Linearized Distribution of Mean Eigenspace Components

We estimate the linearized probability distribution of the mean values $\bar{\mathbf{x}}$ of the eigenspace components for a group of subjects and show that it is multivariate Gaussian distributed. Eqns. (2), (3), and (4) define the transformation $\phi(\cdot)$ between $\bar{\mathbf{y}}$ and $\bar{\mathbf{x}}$. Because eigenspace components $\mathbf{x} = \phi(\mathbf{y})$ and tensor elements $\mathbf{y}$ are related through a nonlinear transformation $\phi(\cdot)$, the *change of variables* method cannot yield the covariance matrix of the eigenspace components. From the continuous-mapping theorem (van der Vaart, 2000; Carew et al., 2006; Anderson, 2001), if the sequence $\bar{\mathbf{y}}_n$ converges in probability to $\mu_{\mathbf{y}}$, and $\phi(\cdot)$ is continuous at $\mu_{\mathbf{y}}$, then $\phi(\bar{\mathbf{y}}_n)$ converges in probability to $\phi(\mu_{\mathbf{y}})$. To obtain the asymptotic distribution of $\phi(\bar{\mathbf{y}}_n)$, we therefore use the Delta method (van der Vaart, 2000) from the theory of large samples to linearize $\phi(\cdot)$. The Delta method uses Taylor's expansion of $\phi(\cdot)$ around $\mu_{\mathbf{y}}$ to linearize the transformation, and then estimates an asymptotic distribution of $\mu_{\mathbf{x}}$. The theorem states that if $\bar{\mathbf{y}}_n$ is a random 6×1 vector with an asymptotic distribution $\sqrt{n}(\bar{\mathbf{y}}_n - \mu_{\mathbf{y}}) \rightsquigarrow N(\mathbf{0}, \Sigma)$, and if $\phi(\bar{\mathbf{y}}_n)$ is a differentiable function, then the asymptotic distribution of $\phi(\bar{\mathbf{y}}_n)$ is

$$\sqrt{n}(\phi(\bar{\mathbf{y}}_n) - \phi(\mu_{\mathbf{y}})) \rightsquigarrow N(\mathbf{0}, G(\mu_{\mathbf{y}}) \Sigma G(\mu_{\mathbf{y}})^T), \quad (5)$$

$$\text{where the Jacobian } G \text{ is evaluated as } G(\mu_{\mathbf{y}}) = \left. \frac{\partial \phi(\bar{\mathbf{y}}_n)}{\partial \bar{\mathbf{y}}_n^T} \right|_{\bar{\mathbf{y}}_n = \mu_{\mathbf{y}}}.$$

In the Delta method, the linear term $(\bar{\mathbf{y}}_n - \mu_{\mathbf{y}})^T \cdot G(\mu_{\mathbf{y}})$ determines the order at which $(\phi(\bar{\mathbf{y}}_n) - \phi(\mu_{\mathbf{y}}))$ converges to zero (van der Vaart, 2000). The quadratic term determines the asymptotic

behavior of $(\phi(\bar{y}_n) - \phi(\mu_y))$ only when $G(\mu_y) = 0$. Furthermore, calculation of the quadratic term is analytically more challenging and computationally unstable in the presence of noise. In our experiments, the linear term was nonzero and thus we used only the linear term to determine the limiting behavior of the sequence $\sqrt{n} \phi(\bar{y}_n)$.

2.5 Hypothesis Testing for Group Differences

The null hypothesis for comparing tensors across two groups of subjects is that the mean values μ_x^1 and μ_x^2 of the eigenspace components will be equal each voxel of the imaging space. Thus, we test the null hypothesis

$$H_0 : \mu_x^1 = \mu_x^2, H_1 : \mu_x^1 \neq \mu_x^2, \text{ with } \Sigma_x^1 = \Sigma_x^2.$$

To test this hypothesis, we compute sample mean values $\hat{\mu}_x^1 = \phi(\mu_y^1)$ and $\hat{\mu}_x^2 = \phi(\mu_y^2)$. We calculate sample covariance matrices, $\hat{\Sigma}_x^1$ and $\hat{\Sigma}_x^2$, for each group of subjects using equation (5) and then compute the pooled covariance matrix $S_{pl} = \frac{(N_1 - 1)\hat{\Sigma}_x^1 + (N_2 - 1)\hat{\Sigma}_x^2}{N_1 + N_2 - 2}$. Using the sample mean values and the pooled covariance matrix, we compute Hotelling's T^2 -statistic (Stevens, 2002) as $T^2 = \frac{N_1 N_2}{N_1 + N_2} (\hat{\mu}_x^1 - \hat{\mu}_x^2)^T S_{pl}^{-1} (\hat{\mu}_x^1 - \hat{\mu}_x^2)$. From Hotelling's T^2 -statistic, we calculate the F-statistic (Stevens, 2002)

$$F(6, N_1 + N_2 - 6 - 1) = \frac{N_1 + N_2 - 6 - 1}{6} \frac{T^2}{N_1 + N_2 - 2}, \quad (6)$$

which follows the *Fisher* F-distribution with 6 and $N_1 + N_2 - 6 - 1$ degrees of freedom. We also compute $F_{\alpha(6, N_1 + N_2 - 6 - 1)}$ for a specified significance level, α . If the computed statistic is greater than this value, then the null hypothesis can be rejected, and we conclude that the average tensor morphologies of the two groups do in fact differ significantly at that particular voxel. We then perform a *post hoc* analysis to determine which particular elements of the mean tensors contribute most to the overall multivariate significance by conducting univariate *t*-tests for each eigenspace component at the $\alpha/6$ level of significance. The Bonferroni inequality ensures that the overall type I error rate for this set of *t* tests is less than α (Stevens, 2002). (Equality of covariance matrices could be tested using likelihood ratio statistic (Gupta and Tang, 1984). If the covariance matrices of the two populations are not equal, i.e. *in the generalized Behrens-Fisher problem*, other advanced procedures must be used to test for equality of the mean vectors (Kim, 1992; Bennett, 1951; Yao, 1965; Anderson, 1984). However, these advanced procedures are unnecessary when comparing tensors across groups containing a sufficiently large number of subjects (Anderson, 1984).)

2.6 Testing Our Formulation

2.6.1 Definitions—We first define notations and terms used subsequently in this paper. The mean tensor used to generate a set of random tensors will be called the *Mean Specified Tensor* (MST). The eigenspace components of the MST will be called the *MST eigenspace components* and the eigenvalues *MST eigenvalues*. From the set of tensors generated randomly, we compute the *Mean Tensor* (MT) by calculating the average value of each tensor element. The eigenvalue, eigenspace components, FA, and PD of the MT will be called the *MT eigenvalues*, *MT eigenspace components*, *Mean FA* (MT-FA), and *MT principal diffusivity*, respectively.

In addition to calculating these values from the MT, we also calculate average FA and diffusivity along the PD for each tensor in a set of sampled tensors. These will be referred to as the *Sample Average FA (SA-FA)* and the *SA principal diffusivity*. In the synthetically generated sets of tensors, where we know correspondence across eigenspace components, we also compute average eigenspace components, which are referred to as *SA eigenspace components* and *SA eigenvalues*.

2.6.2 Generating Random Tensors—To generate random tensors, we first specified elements of the MST $\bar{\gamma} = (23.99, -3.75, 0.0, 11.004, 0.0, 50.0)$, that represented a tensor with eigenvalues 50.0, 25.0, and 5.0, and rotation angles $\alpha = 0.0$, $\beta = 0.0$, and $\gamma = 15^\circ$. The FA equals 0.7 for the MST, which is representative of the average FA values of white matter in the brain of an adult human (Madden et al., 2004). Furthermore, the MST can always be aligned with the coordinate axes. However, we elected $\gamma = 15^\circ$ to study the effect of nonzero angle on the generated random tensors. Note that even if MST α and β equal zero, these angles will differ significantly from zero in tensors generated randomly. Groups of tensors were generated assuming a multivariate Gaussian distribution for the tensor elements. We considered two different covariance matrices for the tensor elements: (1) a diagonal matrix $\text{diag}(\sigma^2, \sigma^2, \sigma^2, \sigma^2, \sigma^2, \sigma^2)$; and (2) an upper block-diagonal matrix (Basser and Pajevic, 2003; Batchelor et al., 2003) of the form

$$\begin{pmatrix} 0.75\sigma^2 & 0.25\sigma^2 & 0.25\sigma^2 & 0 & 0 & 0 \\ 0.25\sigma^2 & 0.75\sigma^2 & 0.25\sigma^2 & 0 & 0 & 0 \\ 0.25\sigma^2 & 0.25\sigma^2 & 0.75\sigma^2 & 0 & 0 & 0 \\ 0 & 0 & 0 & \sigma^2 & 0 & 0 \\ 0 & 0 & 0 & 0 & \sigma^2 & 0 \\ 0 & 0 & 0 & 0 & 0 & \sigma^2 \end{pmatrix}.$$

The diagonal form of the covariance matrix captures the situation in which the tensor elements vary independently in the presence of noise. However, the block-diagonal covariance matrix is richer and captures more realistic variations in tensor elements (Basser and Pajevic, 2003; Batchelor et al., 2003). We increased σ^2 from 1.0 to 10.0 in increments of 1.0. For the specified mean values and their covariance matrix, we used the Cholesky method of decomposition (Press et al., 1992) to factor the correlation matrix and to generate 10,000 positive-definite, random tensors from independently generated zero-mean, Gaussian random variables by keeping only those tensors that were positive-definite. For each value of variance σ^2 , we computed values of the MT eigenspace components and their covariance matrices, both theoretically and empirically. To test the validity of our formulation, we plotted graphs of the MT eigenspace components and SA eigenspace components for varying σ . We expected that the theoretical and empirical values of the covariance matrices would be equal, and that the MT eigenspace components would remain constant.

2.6.3 Single Group Analysis—We tested the validity of our formulation for calculating descriptive statistics for tensors in a group of individuals by generating random diffusion tensors whose elements were multivariate Gaussian distributed with a known covariance matrix. From these randomly generated tensors, we computed mean values $\bar{\gamma}$ of the tensor elements and then calculated the MT eigenspace components $\bar{\kappa}$ using the transformation $\phi(\cdot)$. We also empirically computed the covariance matrix of the randomly generated tensor elements; this covariance matrix, along with the Jacobian G of the transformation $\phi(\cdot)$, was used to compute the covariance matrix of the MT eigenspace components.

In addition, we computed eigenvalues and eigenvectors for each randomly generated tensor. Because the tensors were generated synthetically, we knew which eigenvalues corresponded across groups. This allowed us to compute empirically the values of the SA eigenspace components and their covariance matrix. We then compared the values of the SA eigenspace components with the MT eigenspace components, as well as their covariance matrices to assess the accuracy of our proposed formulation.

We computed the MT for a group of subjects by averaging the tensor elements across each tensor in the group. Intuitively, averaging different tensors should reduce the MT-FA. We therefore plotted a graph to track the effect on MT-FA values of varying amounts of noise in the randomly generated tensors and with a varying number of subjects. Additionally, we plotted graphs of SA-FA for varying amounts of noise and numbers of subjects. Under ideal conditions, the values of MT-FA and SA-FA should equal the value of FA for the MST.

2.6.4 Between-Group Analysis—To evaluate the performance of our method in comparing tensor morphologies across groups, we generated different sets of 100 diffusion tensors for five differing cases of interest (see below). We compared the results of between-group analyses using our method with results from the method that uses values of SA-FAs. The null hypotheses to be tested for between-group comparisons were that the values of the various statistics (computed using the MT eigenspace components and SA-FA) would be equal for the tensors from the two groups of subjects. We should note that a failure to reject the null hypothesis for a given statistic does not imply that the values of the statistic were equal for the two groups of tensors.

For the first group of tensors, the MST $\bar{\gamma} = (6.0598, -5.075, 0.0, 23.6402, 0.0, 50.0)$ represented eigenvalues 50.0, 25.0, 4.7, and rotation angles $\alpha = 0.0$, $\beta = 0.0$, and $\gamma = 15^\circ$. The FA of the MST was 0.7. For the second group of tensors, we specified MSTs uniquely for the five differing cases of interest. In each case, we assumed a diagonal covariance matrix between the MST elements, with a variance that ranged from 1.0 to 10.0.

Case I: Equal MSTs: In this case, the MSTs for the two groups are equal. Therefore, neither the analyses based on our method nor those based on SA-FA should reject the null hypotheses.

Case II: Different Mean Specified Rotations: In this case, the second set of tensors was generated for MST $\bar{\gamma} = (4.7, 0.0, 0.0, 25.0, 0.0, 50.0)$ with eigenvalues 50.0, 25.0, 4.7, and rotation angles $\alpha = 0.0$, $\beta = 0.0$, and $\gamma = 0^\circ$. Therefore, the two groups have equal MST eigenvalues but differing values of γ . We expect that our method will reject the null hypothesis, i.e., that the MT eigenspace components will be equal for the groups and will localize the origin of the difference to the angle γ . We expect that the method based on SA-FA will not reject the null hypothesis.

Case III: Differing MST Eigenvalues and MST-FA Values: We generated the second set of tensors with MST $\bar{\gamma} = (18.189, -1.825, 0.0, 24.511, 0.0, 50.0)$ to represent a tensor having eigenvalues 50.0, 25.0, 17.7, rotation angles $\alpha = 0.0$, $\beta = 0.0$, and $\gamma = 15^\circ$, and FA of 0.5. The MST eigenspace components and an FA differed between the two groups of tensors. Therefore, both our method and the method based on SA-FA should reject the null hypothesis.

Case IV: Small Differences in MST Eigenvalues: Here we generated the second set of diffusion tensors with MST $\bar{\gamma} = (6.8063, -4.875, 0.0, 23.6937, 0.0, 50.0)$ that represented a tensor having eigenvalues 50.0, 25.0, and 5.5, rotation angles $\alpha = 0.0$, $\beta = 0.0$, and $\gamma = 15^\circ$, and an FA of 0.687. We provide this example to illustrate that a relatively small change in the eigenvalue (from 4.7 to 5.5) leads to a relatively large change in FA values (from 0.7 to 0.687, respectively). Small differences in eigenvalues can be caused by noise in the DT images, which

in turn produces large perturbations in FA. In this instance, it is preferable that the null hypothesis should not be rejected. In the presence of large noise values, we expect that our method will not reject the null hypothesis, whereas the analysis based on SA-FA will tend to reject it.

Case V: Differing MST Eigenvalues but Equal MST-FAs: Tensors with differing values for their tensor elements could have equal FA values; therefore, even if the MSTs in two groups differ, their difference cannot be detected using SA-FAs. To illustrate this, we generated a second set of diffusion tensors with MST $\bar{\gamma} = (134.4424 \ 29.325 \ 0.0 \ 32.8576 \ 0.0 \ 50.0)$, representing a tensor with eigenvalues 50.0, 25.0, 142.2, rotation angles $\alpha = 0.0$, $\beta = 0.0$, and $\gamma = 15^\circ$, and an FA of 0.7. Whereas our method should reject the null hypothesis and localize the origin of the between-group difference to the eigenvalues 142.2 and 4.7, we expect that the method based on SA-FA will not be able to reject the null hypothesis.

2.6.5 Effects of Calculation of the MT from Errors in Reorienting Tensors—Errors in registering and reorienting subject DT images into a common coordinate space will cause errors in the computed MTs, and therefore errors in the between-group analyses. To study the effects of error in reorienting tensors on calculation of MTs, we considered an MST with eigenvalues 10, 1, and 1, and eigenvectors aligned along the X, Y, and Z axes. The principal eigenvector was aligned along the X-axis. We then generated sets of simulated tensors by rotating the MST about the Z-axis by varying degrees that were sampled uniformly at random from a specified range of angle. The simulated tensors therefore were randomly rotated but had the same eigenvalues as the MST. We generated 7 sets of 50 simulated tensors for a differing range of angle: $\pm 1^\circ$, $\pm 2^\circ$, $\pm 4^\circ$, $\pm 8^\circ$, $\pm 16^\circ$, $\pm 32^\circ$, and $\pm 64^\circ$. The last set of tensors, generated for the rotation angle γ sampled uniformly $\pm 64^\circ$, represented the situation in which the DT images were poorly normalized. (Note that a range of $\pm 90^\circ$ in fact covers the entire 360° because tensors are symmetric along the eigenvectors. In this case the MT cannot be determined.) To study the effect of errors in registering and reorienting large number of tensors on the computed MT, we generated 7 other sets of 5000 simulated tensors for the same ranges of angle. For each of these sets, we computed the MT eigenvalues, eigenvectors and MT-FA and compared these values with those of the MST. We expected that the MT-FA would decrease for an increasing range of rotation angles. Furthermore, for an increasing number of subjects, we expected that the MT-FA would decrease but that the MT eigenvectors would align better with the X-, Y-, and Z-axes.

2.6.6 A Real-World Example—We used our method to compare DT images from a group of 24 healthy subjects and a group of 22 individuals with Tourette's Syndrome (TS). The DT images were acquired along 6 directions (8 averaged measurements per slice) on a Siemens 1.5-Tesla MRI scanner, with Repetition Time (TR) = 4000ms, Echo Time (TE) = 96 ms, $b = 1000 \text{ s/mm}^2$, an image matrix of size 128×128 , and a Field of View (FOV) of 240 millimeters. Nineteen slices, each 4mm thick, were acquired in the sagittal orientation such that the 10th slice was located at the interhemispheric fissure in the brain. Therefore, the DTI data covered only the midportion of the brain in each subject. Furthermore, to ensure that the reconstructed tensors were positive-definite, we masked out the background in the images and reconstructed tensors only in the brain region. Rarely, a reconstructed tensor within the brain was not be positive-definite. For these tensors, we imposed the positivity constraint by inverting the sign of the negative eigenvalue and then recomputing the tensor. Such an approach reduced computation time without compromising the imaging data or findings. More advanced methods, including constrained nonlinear least-squares estimation (Koay et al., 2006), could be used to ensure that the reconstructed tensors are positive-definite; however, these approaches are computationally expensive.

To compare DT images across groups of subjects, images were coregistered and reoriented into a common coordinate space. We defined this common coordinate space using an anatomical MR image of a normal subject as the *reference image*. DT images from every other subject were then coregistered to this reference image, and their tensors were reoriented into this common coordinate space using a two-step procedure (Bansal et al., 2005b; Xu et al., 2005). First, we used a similarity transformation to register the FA image of each subject to the corresponding anatomical MR image, such that the mutual information across images was maximized (Viola and Wells, 1997). Because DTI data spanned only the sagittal and parasagittal portions of the brain of each subject, we cropped the anatomical MR image in the reference space to match the total extent of the FA image. Then, using a fluid dynamical model of spatial transformation, we warped the FA image from each subject to the corresponding cropped anatomical MR image for that person, such that mutual information across the FA and anatomical images was maximized (Christensen et al., 1994; Bansal et al., 2005a). Second, the uncropped anatomical image of each subject was warped to the reference anatomical image, such that the differences in pixel intensities between images were minimized (Christensen et al., 1994). Thus, each step estimated a deformation field that satisfied the smoothness and topology constraints imposed by fluid dynamics. We then concatenated these two deformation fields and used Procrustean estimation (Xu et al., 2003) to coregister and reorient the DT images of each subject into the common reference space.

After coregistering and reorienting the DT images of each subject to the template space, we computed the MTs and their covariance matrices for the two groups of subjects, as detailed above. We used these MTs and covariance matrices to compare DTs across diagnostic groups. To validate visually the accuracy of the computed MTs, we computed and compared two FA images: (1) an image of values of MT-FA at each voxel and (2) an image of the values of SA-FA for the group of healthy subjects. Additionally, we compared numerically the values of MT-FA and SA-FA at six corresponding locations selected manually (two locations were selected within the Corpus Callosum (CC) region and four locations were selected from the surface of white matter in the reference image).

To compare tensor morphologies statistically across groups of subjects, we computed the *P-values* for between-group differences in the values of the various DTI statistics. *P-values* were computed at each voxel in the common coordinate space without correcting for correlations between tensors at neighboring voxels. First, we computed the values of all the MT eigenspace components and their covariance matrices for both groups of subjects. The vectors corresponding to the MT eigenvalues (which were arranged in order of decreasing magnitude) and their covariance matrices for the two groups were used to compute Hotelling's T^2 statistics and their corresponding *P-values*. Second, we statistically compared the MT principal diffusivities for the two groups using Student's *t*-statistics and *P-values*. Third, we compared the SA principal diffusivity between groups using Student's *t*-statistics. We expected that the MT principal diffusivities and SA principal diffusivities for the two groups would match well, and that therefore *P-values* of between-group differences would also match well. Computation of SA principal diffusivity assumes, however, that the largest eigenvalues always correspond with the principal directions of a tensor, but the presence of noise in the DT dataset can invalidate this assumption. Therefore, *P-values* based on these two statistics may differ. Finally, we computed at each voxel the values of SA-FA and their variances for the two groups; these values were used to compute the *t*-statistic and *P-value* for between-group differences (Rosner, 1995). *P-values* were color encoded and displayed on the reference image.

3 Results

3.1 Generation of Random Tensors

Fig. 2 shows the range of values for the eigenspace components of the 10,000 DTs that were generated at random (for a diagonal covariance matrix between tensor elements). The range of values for each component increased with increasing variance in the covariance matrix. While DTs were generated randomly, values of the MT eigenspace components converged to their respective MST eigenspace components (see Section 3.2 below).

3.2 Single Group Analyses

Single group analyses show that the values for the MT eigenspace components and their covariance matrices match well with the corresponding values for the SA eigenspace components. We present results for two sets of 10,000 diffusion tensors generated randomly. The first set of diffusion tensors was generated assuming a diagonal covariance matrix between the elements of the MST. The second set was generated assuming a block-diagonal covariance matrix.

3.2.1 Diagonal Covariance Matrix—The graphs in Fig. 3 show that the theoretically and empirically computed values of the covariance matrix of eigenspace components match closely, even for a large covariance in the tensor elements. (We did not plot the values of the off-diagonal elements in the covariance matrix because they were small.)

The graphs in Fig. 4 show that the values for the SA eigenspace components depend substantially on variability in the tensor elements and diverge away from their respective MST eigenspace components. On the other hand, the values for the MT eigenspace components remain close to their respective MST eigenspace components.

3.2.2 Upper Block-Diagonal Covariance Matrix—The graphs of the values for the SA eigenspace and MT eigenspace components match well for a block-diagonal covariance matrix (Fig. 5). Additionally, the graphs in Fig. 6 show that the MT eigenspace components are closer to the corresponding MST eigenspace components than to the SA eigenspace components. A close match of the MT and SA eigenspace components, together with their respective covariance matrices (for both the diagonal and the block-diagonal covariance matrices) shows the accuracy of our mathematical formulation. Because our method linearizes the relation between the tensor elements and the eigenspace components, the theoretical values of the covariance matrix will not match exactly with those computed empirically.

Fig. 7 shows that the MT-FA decreases with increasing variance in the tensor elements, even though the MT eigenspace components are similar to their respective specified values (Fig. 6). Therefore, even though the MT eigenvalues differ only slightly from their respective MST eigenvalues (Fig. 6), the cumulative effect of the difference is to decrease the anisotropy of the MT, consistent with findings from an earlier report (Jones et al., 2002). Figs. 6 and 7 suggest that the increased values of the smallest eigenvalue (labeled as Eigenvalue1 in Fig. 6) decrease the MT-FA.

Decreasing values of the MT-FA with increasing noise is of concern when comparing DT images across groups of subjects because errors in coregistration and reorientation of the images of each subject will increase noise in the corresponding DTs. Our results, however, suggest that reduced values of MT-FA may not necessarily imply that the values of the MT eigenspace components differ greatly from their true (though unknown in a real-world setting) values.

3.3 Between-Group Analyses

When the MSTs were equal for the two groups of tensors, the null hypothesis could not be rejected for any statistic; therefore, as expected, the two groups of tensors did not differ significantly in any statistic at the significance level of 0.05. When only one rotation angle differed between the MSTs of the two groups (Fig. 8, **Case II**), our method determined that the null hypothesis for the Hotelling's T^2 statistic could be rejected; therefore, our method detected that the average morphology of the two groups of tensors differed significantly. Additionally, we were able to localize the differences between the MSTs to the difference in the rotation angle γ based on its associated P -value. The SA-FA method could not reject the null hypothesis because the MST eigenvalues were equal for the two groups. This case example demonstrates the superiority of our approach in detecting differences between groups that would otherwise be overlooked by the method based on SA-FA.

When the MST eigenvalues differ between groups (Fig. 8, **Case III**), the null hypothesis can be rejected based both by our method and by the values of SA-FA. Additionally, however, our method localizes the cause of the between-group difference to the correct eigenvalue (Eigenvalue1 in this case). When comparing two sets of diffusion tensors that differed only slightly in the MST eigenvalues (Fig. 8, **Case IV**), our method did not reject the null hypotheses for small amounts of variances in the tensor elements; however, the method based on the values of SA-FA rejected the null hypothesis even in the presence of large variances. A small between-group differences in eigenvalues can cause large differences in the SA-FAs (fig. 1), leading to the conclusion that when using this statistic group differences are statistically significant. This example demonstrates that an analyses based on SA-FA may erroneously detect differences between groups when in fact none exist, thereby increasing the rate of false-positives in the presence of noise. Finally, when the MST eigenvalues of the two groups differed but their FA values were equal (Fig. 8, **Case V**), our method was able correctly to reject the null hypothesis and to localize the origin of between-group differences to the correct eigenvalues; the method based on SA-FA, however, could not reject the null hypothesis even though the eigenvalues differed significantly between groups.

These cases show that the method using SA-FA will sometimes erroneously detect nonexistent group differences (Fig. 8, **Case IV**) or fail to detect true group differences (Fig. 8, **Case V**). In contrast, our method correctly detected between-group differences in both examples. Note that the P -values of the statistics, such as FA, changed wildly when the null hypothesis could not be rejected. This is because under the null hypothesis, the distributions of these indices are equal for the two groups of subjects, and therefore the between-group differences in the index and its associated P -value will vary randomly.

3.4 The Effects of Errors in the Reorienting of Tensors

The MT-FA declined significantly for the sets of tensors generated with rotation angle γ sampled uniformly in the range $\pm 32^\circ$ or greater (Fig. 9). However, for smaller ranges of γ , MT eigenvalues and eigenvectors, and MT-FA, matched closely those of the MST (Fig. 9). In contrast, even though a larger number of subjects further reduced the MT-FA only slightly, the MT eigenvectors were better aligned with those of the MST (Fig. 9). Therefore, the arithmetical averaging of tensors yielded an MT that approximated closely the MST. In the presence of large noise levels, however, the MT differed substantially from the MST, indicating that precise registration and reorientation of DT images during spatial normalization is essential for detecting between-group differences. Furthermore, in the absence of systematic bias in the registration of subjects in a group to the reference brain, errors in the registration will contribute to Type II (false negative) error by increasing variance in our DTI measures.

3.5 Analyses Using a Real-World Data Set

Visually, fiber pathways in the two FA images (one computed from the values of MT-FA and the other from the values of SA-FA at each voxel) match well (Fig. 10). However, the values of MT-FA are smaller than those of SA-FA, and therefore the MTs are slightly less anisotropic; this does not necessarily imply, however, that the MT eigenvalues will differ greatly from their true (although unknown) values (see Figs. 6 and 7).

Fig. 11 displays the P -values of the differences across groups in various statistics. The computed P -values for the entire 3D volume are color-coded and displayed on 4 representative sagittal slices from the reference MR image. Images in Fig. 11 *Column (a)* show statistically significant between-group differences in the SA-FA of the two groups, especially in the Corpus Collosum (CC). However, images in Fig. 11 *Column (b)* show that the SA principal diffusivities in the CC do not differ statistically across groups, implying that the differences in the values of SA-FA are a function of differences in the diffusivities in directions orthogonal to the PDs.

We compared the vectors of the MT eigenvalues for the two groups using the Hotelling's T^2 statistic. Images in Fig. 11 *Column (c)* show the differences in the Hotelling's T^2 detected across groups; significant differences included the CC, where SA-FA values also differed statistically. Because Hotelling's T^2 statistic (which compares all three eigenvalues simultaneously) differed significantly across groups but the values of SA principal diffusivity did not, we conclude that the observed group differences in SA-FA were attributable to differences in diffusivity across groups in directions orthogonal to the PDs. Note that while we have been very careful in registering and reorienting subject DTI datasets into a common coordinate space, errors in normalization will effect these analysis by reducing our statistical power to detect between-group differences. Therefore, in the absence of grossly obvious morphological deformities in the brain of one group of subjects as compared to the other, errors in the registration will contribute to Type II (false negative) error by increasing variance in our DTI measures.

Images in the Fig. 11 *Column (d)* depict between-group differences in MT principal diffusivities. Their good correspondence with the location of group differences in SA principal diffusivities (Fig. 11 *Column (b)*) further attested to the accuracy of the mean values and variances of the principal diffusivities that were computed using our method. In addition, our method allows for an unambiguous computation and comparison of diffusivities along PDs because these diffusivities were computed from the MTs. Our results show that the differences in SA-FA are because of differences in diffusion in directions orthogonal to PD (Fig. 12).

The relatively lower values of SA-FA in the CC of TS subjects, combined with the absence of significant differences between principal diffusivities across groups, suggest that fiber tracts are likely intact in the CCs of persons with TS (because principal diffusivities are normal in this region), but that either the numbers of nerve fibers or their degree of myelination in the CC may be reduced compared to normal controls (because diffusivity was significantly increased in directions orthogonal to the PDs). This interpretation is consistent with our prior findings of reduced size of the CC in a large sample of TS children (Plessen et al., 2004).

3.5.1 Summary—Across these two groups of real subjects, while SA-FA differed significantly, SA-PD was not significantly different in the CC region: i.e., while eigenvalues were different, the eigenvalue along the principal direction (PD) was not significantly different across groups. Therefore, while the tensors were equally elongated along the PD, tensors in one group were either fatter or thinner as compared to those in the other group. This was further confirmed by our method that tested differences in the eigenvalues using T^2 statistics. Differing eigenvalues (and therefore diffusion of water molecules) in directions perpendicular to PD may suggest fewer fibers in individuals with TS.

4 Discussion

We have presented a voxel-wise method for accurately detecting between-group differences in the diffusivity of water that likely reflect group differences in anatomical connectivity or white matter integrity (Pfefferbaum et al., 2000; Plessen et al., 2004). Because white matter fiber bundles are not visible using anatomical MRI, DTI is currently the only viable means to identify within- and between-group differences in the diffusion of water when studying the integrity of white matter fibers *in vivo*. Differences in connectivity or white matter integrity typically have been assessed by comparing mean FAs across diagnostic groups, which does not require identification of corresponding eigenspace components because FA is a simple scalar index. This method may not provide an optimal or complete comparison of tensors across groups, however, because FA is related nonlinearly to the underlying eigenspace components and it does not permit comparison of the directionality of tensors across groups. Other methods have therefore been developed that first establish a correspondence of the various eigenspace components and then compares statistically the numerical values of corresponding components across tensors from differing subjects. These approaches are limited by the difficulty of establishing a correct correspondence of eigenspace components across individuals in the presence of the noise that are inherent in DTI datasets.

We have developed a method for the comparison of tensors across individuals, or groups of individuals, that circumvents the difficulty of correctly identifying eigenspace components that correspond across individuals. We first compute the MT for each group of subjects, and then we use this MT to compute the various eigenspace components of the MT. We have shown that MT eigenspace components are multivariate Gaussian distributed, irrespective of the distribution from which the tensors at each voxel are sampled. Once the MT eigenspace components are calculated, we then use the Delta method to compute their covariance matrix. Once the distribution of the MT eigenspace components is determined, standard multivariate statistical procedures can be used to detect between-group differences in tensor morphology at each voxel. Analyses of simulated data demonstrated that the MT eigenspace components computed using this approach matched well with the corresponding MST eigenspace components, whereas the values of SA eigenspace components diverged quickly as the noise increased. Therefore, arithmetic averaging of the tensors yielded a valid MT for a group of subjects, which can be then used for detecting between-group differences.

Because diffusion of water is nonnegative in any particular direction, a tensor must be positive definite to describe quantitatively the diffusion of water in the brain. For detecting between-group differences in diffusion of water, we computed MT as an arithmetic average of the DTs across individuals in a group. For an increasing number of subjects, the distribution of our MT is increasingly restricted to the subspace portion of $\mathbb{R}^6$ in which tensors are positive definite. Another interesting way to ensure that the positive-definite constraint on the MT is satisfied is to first decompose individual tensors into the lower triangular factor ($\mathbf{L}_i$) using Cholesky decomposition (Press et al., 1992; Koay et al., 2006), then compute the Mean Lower (ML) factor and its distribution by taking the arithmetic average of $\mathbf{L}_i$ s across subjects in a group, and then finally calculate the MT and its distribution from ML. Using this method, the distribution of $\mathbf{ML}$ does not need to be restricted to a particular subspace of $\mathbb{R}^6$ because the MT computed from any ML is always positive-definite. However, the comparison of tensor morphologies across groups require determination of the distributions of the mean eigenvalues and eigenvectors in each group, and in this respect the method of tensor decomposition suffers from at least two serious limitations. First, the elements of the ML are nonlinearly related to those of the MT, and therefore when the number of subjects is small, estimation of the distribution of the MT using linear methods will have nonzero probability mass in subregions of $\mathbb{R}^6$ for tensors that are not positive-definite. Second, the nonlinear relation of the elements

of ML to those of MT will increase the errors in linear approximations of the distributions of eigenvectors and eigenvalues in the Delta method. Thus, in comparison, our method satisfies the positivity constraint while also producing relatively fewer errors when determining the distributions of the MT eigenspace components.

In comparing the performance of our method with that based on SA-FA, we found that both methods were equally likely to correctly reject null hypotheses in trivial cases. However, when the MST eigenvalues differed from one another only minimally (Fig. 8, **Case IV**), the method using SA-FA rejected the null hypothesis even in the presence of high noise levels, whereas our method (appropriately) did not reject the null hypothesis, even in the presence of low noise levels. These results suggest that, in the presence of noise, comparing tensor morphologies across groups using the SA-FA method can detect between-group differences when in fact none exist. Finally, Fig. 8, **Case V** showed that real group differences may not be detected using the SA-FA method (i.e., the method is prone to generating false negative findings), whereas our method correctly identified real group differences in this particular case.

We evaluated the performances of the two methods in a real-world setting by comparing the DT images of a group of 24 normal subjects with those of a group of 22 subjects with TS. In analyses of SA-FA within the corpus callosum, we found that the SA-FA for the TS group was significantly lower than that for the healthy subjects. These analyses, however, could not determine whether SA-FAs differed because of variations in mean diffusivities along the PDs or because of differences in diffusivities in directions orthogonal to PDs. Furthermore, SA principal diffusivities did not differ statistically between groups within the CC, indicating that between-group differences in values of the SA-FA in this region arose from differences in the diffusivity of water in directions orthogonal to the PD. This interpretation was supported by findings of significant between-group differences in the remaining MT eigenvalues (Song et al., 2005). These between-group differences in diffusivities could be caused by any of several differences in tissue characteristics across groups, including a reduction in the numbers of fibers connecting the left and right cerebral hemispheres, and reduced myelination of axons within the CCs of TS children (Song et al., 2002; Mori and Zhang, 2006). By combining statistical analyses based on MT principal diffusivity, MT eigenvalues, and SA-FA, we can therefore describe better the between-group differences in DT images.

Between-group differences could also be detected by comparing corresponding eigenvalues that were ordered according to their magnitude and coherence in local fiber direction in each subject brain (Martin et al., 1999; Basser and Pajevic, 2000). These approaches are attractive because they are simple to implement and because when the data are free of noise these methods should yield results comparable to those obtained using our approach. In the ubiquitous presence of noise, corresponding eigenvalues across different tensors cannot be determined uniquely, therefore causing bias in the ordered eigenvalues. Thus, our method will yield accurate results even in the presence of noise because it does not have to identify the corresponding eigenvectors across individuals.

In the presence of noise, the FA of the mean tensor for a group of subjects in a normalized reference space may be lower than the corresponding SA-FA. In our simulation data, we found that increasing the number of tensors used to compute the MT did not correct this reduction in anisotropy. Furthermore, in our real subject datasets, MT-FA was smaller than the SA-FA for the groups of 24 healthy subjects. We attribute the lower anisotropy of the MTs to the introduction of errors in tensor alignment that accompany errors when normalizing DT images into the common coordinate space. Better normalization of the DT images will reduce differences between the values of MT-FA computed using our method and the values of SA-FA, leading to accurate detection of between-group differences.

Selection of the appropriate index or measure of tensor morphology to detect between-group differences in tissue organization depends upon the scientific context and aims of a particular study. The best index will presumably be the one that is most sensitive to the particular morphological feature of the tensor that is of interest to the investigator, and using one that is not sufficiently sensitive will fail to detect significant differences in morphology across groups. Whereas a multivariate index, such as the vector of eigenvalues, is sensitive to differences in a wide range of linear combinations of the component eigenvalues, this index will be less sensitive to overall differences in tensor morphology than will other scalar measures, such as FA, for those tensors in which the index value changes rapidly for a small change in the tensor's eigenvalues. Our results (Figs. 11 and 12) suggest that the index that is most sensitive in detecting between-group differences in tensor morphology may actually vary according to brain region and tissue type. Thus, assessing group differences in tensor morphology using both a scalar index (such as FA) and a multivariate measure (such as the vector of eigenvalues) may yield a more complete understanding of the morphological origin of between-group differences than will either measure alone.

Comparing tensor morphologies across groups at each voxel within the entire brain entails many statistical tests and a proportionately increased probability of detecting between-group differences by chance (i.e., of generating false positive, or type I, errors). Standard multiple-comparison procedures can be used to limit the false positive rate to a desired level. The simple method of *Bonferroni adjustment* to control for false positive rate is generally considered to be excessively conservative because test statistics on neighboring voxels are correlated, and therefore the actual number of independent comparisons is less than the number used for correction. Additional methods, including those based on false discovery rate (Benjamini and Hochberg, 1995; Genovese et al., 2002; Schwartzman et al., 2005) and permutations testing (Bullmore et al., 1999; Golland and Fischl, 2003), can be used to compute corrected *P*-values of voxels and clusters of voxels. These procedures can be applied to the test statistic *F* (Eqn. (6)) within our method to control for multiple comparisons and to compute corrected *P*-value.

We used the Central Limit Theorem to justify our estimation of the multivariate Gaussian distribution for the elements of the mean tensor. Whereas the MT itself will be positive-definite, the Gaussian distribution in Euclidean space, $\mathbb{R}^6$, will have nonzero probability mass for vectors that do not form positive-definite tensors. Because the mean tensor of the sample is an unbiased, consistent estimator of the mean, however, the mean tensor of the sample asymptotically equals the true mean in probability, and the distribution of the mean tensor is degenerate. Therefore, the mass of the distribution that describes the MT, will equal zero asymptotically in $\mathbb{R}^6$ for tensors that are not positive-definite. Despite this potential limitation, however, our method yields a valid and complete description of the mean eigenvalues and eigenvectors by determining their multivariate distribution. This multivariate Gaussian distribution of the MTs can be used to detect between-group differences in tensor morphologies.

We have presented a mathematical framework for computing the voxel-wise mean values of the eigenspace components and their covariance matrices for tensors within groups of subjects. Because we compute the mean eigenvalues and eigenvectors from the mean tensor, which is computed by the element-wise averaging of the tensors across individuals, we need not define the correct correspondence of eigenvalues and eigenvectors across those tensors. Furthermore, we have shown that the mean values are multivariate Gaussian distributed. Once the distribution of the mean eigenspace components has been estimated, we can compare these mean values statistically across groups. This method allows for a comparison of mean DTs across groups of subjects that is more direct and accurate than are methods that are based on calculation of scalar indices alone. Our method also permits determination of the morphological origins of group differences in tensor morphology, in that it indicates whether

between-group differences derive either from differences in diffusivity along PDs, or from differences in diffusivity in directions perpendicular to the PDs.

Acknowledgements

This work was supported in part by NIMH grants MH068318, MH36197, and K02-74677, NIDA grant DA017820, a NARSAD Independent Investigator Award, the Thomas D. Klingenstein & Nancy D. Perlman Family Fund, and the Suzanne Crosby Murphy Endowment at Columbia University.

References

- Alexander D, Pierpaoli C, Basser P, Gee J. Spatial transformations of diffusion tensor magnetic resonance images. *IEEE Transactions on Medical Imaging* 2001;20(11):1131–1139. [PubMed: 11700739]
- Anderson AW. Theoretical analysis of the effects of noise on diffusion tensor imaging. *Magnetic Resonance in Medicine* 2001;46:1174–1188. [PubMed: 11746585]
- Anderson, T. An Introduction to the Multivariate Statistical Analysis. John Wiley & Sons; 1984.
- Arsigny, V.; Fillard, P.; Pennec, X.; Ayache, N. Fast and simple calculus on tensors in the log-Euclidean framework. In: Duncan, J.; Gerig, G., editors. *Proceedings of MICCAI 2005, Eighth International Conference on Medical Image Computing and Computer-Assisted Intervention (LNCS 3749)*; Palm Springs, CA, USA. 2005. p. 115-122.
- Ashburner J, Friston K. Nonlinear spatial normalization using basis functions. *Human Brain Mapping* 1999;7(4):254–266. [PubMed: 10408769]
- Bansal R, Staib L, Peterson B. Mutual information based non-rigid image registration: A stochastic formulation. *International Society for Magnetic Resonance in Medicine*. 2005a
- Bansal R, Xu D, Peterson B. Eigen function based coregistration of diffusion tensor images to anatomical magnetic resonance images. *International Society for Magnetic Resonance in Medicine*. 2005b
- Basser P. Inferring microstructural features and the physiological state of tissues from diffusion-weighted images. *NMR in biomedicine* 1995;8(7):333–344. [PubMed: 8739270]
- Basser P, Mattiello J, LeBihan D. MR diffusion tensor spectroscopy and imaging. *Biophysical Journal* 1994;66:259–267. [PubMed: 8130344]
- Basser P, Pajevic S. Statistical artifacts in diffusion tensor MRI (DT-MRI) caused by background noise. *Magnetic Resonance in Medicine* 2000;44(1):41–50. [PubMed: 10893520]
- Basser P, Pajevic S. A normal distribution for tensor-valued random variables: applications to diffusion tensor MRI. *IEEE Transactions on Medical Imaging* 2003;22(7):785–794. [PubMed: 12906233]
- Basser P, Pierpaoli C. Microstructural and physiological features of tissues elucidated by quantitative-diffusion-tensor MRI. *Journal of Magnetic Resonance* 1996:209–219. [PubMed: 8661285]B III
- Batchelor P, Atkinson D, Hill D, Calamante F, Connelly A. Anisotropic noise propagation in diffusion tensor MRI sampling schemes. *Magnetic Resonance in Medicine* 2003;49:1143–1151. [PubMed: 12768593]
- Batchelor P, Moakher M, Atkinson D, Calamante F, Connelly A. A rigorous framework for diffusion tensor calculus. *Magnetic Resonance in Medicine* 2005;53(1):221–225. [PubMed: 15690523]
- Benjamini Y, Hochberg Y. Controlling the false discovery rate: a practical and powerful approach to multiple testing. *Journal of the Royal Statistical Society* 1995;57(1):289–300.B
- Bennett B. Note on a solution of the generalized behrens-fisher problem. *Annals of the Institute of Statistical Mathematics* 1951;2:87–90.
- Bullmore E, Suckling J, Overmeyer S, Rabe-Hesketh S, Taylor E, Brammer M. Global, voxel, and cluster tests, by theory and permutation, for a difference between two groups of structural MR images of the brain. *IEEE Transactions on Medical Imaging* 1999;18(1):32–42. [PubMed: 10193695]
- Carew J, Koay CG, Wabha G, Alexander AL, Basser PJ, Meyerand ME. The asymptotic distribution of diffusion tensor and fractional anisotropy estimates. *Proceedings of the International Society for Magnetic Resonance in Medicine* 2006;14
- Christensen G, Rabbitt R, Miller M. 3D brain mapping using a deformable neuroanatomy. *Physics in medicine and biology* 1994;39(3):609–618. [PubMed: 15551602]

- Counsell S, Allsop J, Harrison M, Larkman D, Kennea N, Kapellou O, Cowan F, Hajnal J, Edwards A, Rutherford M. Diffusion-weighted imaging of the brain in preterm infants with focal and diffuse white matter abnormality. *Pediatrics* 2003;112(1):1–7. [PubMed: 12837859]
- Fletcher, P.; Joshi, S. Principal geodesic analysis on symmetric spaces: Statistics of diffusion tensors. Workshop on Computer Vision Approaches to Medical Image Analysis (CVAMIA) LNCS 3117; 2004. p. 87–98.
- Genovese C, Lazar N, Nichols T. Thresholding of statistical maps in functional neuroimaging using the false discovery rate. *NeuroImage* 2002;15:870–878. [PubMed: 11906227]
- Golland, P.; Fischl, B. Permutation tests for classification: Towards statistical significance in image-based studies. In: Taylor, C.; Noble, J., editors. *Proceedings of Information Processing in Medical Imaging (LNCS 2732)*; Berlin: Springer-Verlag; 2003. p. 330–341.
- Golub, G.; Loan, C. *Matrix Computation*. Johns Hopkins University Press; 1996.
- Gupta A, Tang J. Distribution of likelihood ratio statistic for testing equality of covariance matrices of multivariate gaussian models. *Biometrika* 1984;71(3):555–9.
- Hasan K, Basser P, Parker D, Alexander A. Analytical computation of the eigenvalues and eigenvectors in DT-MRI. *Journal of Magnetic Resonance* 2001;152:41–47. [PubMed: 11531362]
- Jones D, Griffin L, Alexander D, Catani M, Horsfield M, Howard R, Williams S. Spatial normalization and averaging of diffusion tensor MRI data sets. *NeuroImage* 2002;17:592–617. [PubMed: 12377137]
- Kim SJ. A practical solution to the multivariate Behrens-Fisher problem. *Biometrika* 1992;79(1):171–176.
- Klingberg T, Hedehus M, Temple E, Salz T, Gabrieli J, Moseley M, Poldrack R. Microstructure of temporo-parietal white matter as a basis for reading ability: Evidence from diffusion tensor magnetic resonance imaging. *Neuron* 2000;25(2):493–500. [PubMed: 10719902]
- Koay C, Carew J, Alexander A, Basser P, Meyerand M. Investigation of anomalous estimates of tensor-derived quantities in diffusion tensor imaging. *Magnetic Resonance in Medicine* 2006;55:930–936. [PubMed: 16526013]
- Kubicki M, Westin CF, Maier S, Frumin M, Nestor P, Salisbury D, Kikinis R, Jolesz F, McCarley R, Shenton M. Uncinate fasciculus findings in schizophrenia: A magnetic resonance diffusion tensor imaging study. *American Journal of Psychiatry* 2002;159(5):813–820. [PubMed: 11986136]
- Lim K, Helpert J. Neuropsychiatric applications of DTI - a review. *NMR in biomedicine* 2002;15(2):587–593. [PubMed: 12489105]
- Madden D, Whiting W, Huettel S, White L, MacFall J, Provenzale J. Diffusion tensor imaging of adult age differences in cerebral white matter: relation to response time. *NeuroImage* 2004;21:1174–1181. [PubMed: 15006684]
- Martin K, Papadakis N, Huang C, Hall LD, Carpenter T. The reduction of the sorting bias in the eigenvalues of the diffusion tensor. *Magnetic Resonance in Imaging* 1999;17(6):893–901.
- Mori S, Zhang J. Principles of diffusion tensor imaging and its applications to basic neuroscience research. *Neuron* 2006;51:527–539. [PubMed: 16950152]
- Nagy Z, Westerberg H, Skare S, Andersson JL, Anders L, Flodmark O, Elisabeth F, Kirsten H, Birgitta B, Hans F, Hugo L, Torkel K. Preterm children have disturbances of white matter at 11 years of age as shown by diffusion tensor imaging. *Pediatric Research* 2003;54(5):672–679. [PubMed: 12904607]
- Netsch T, Muiswinkel A. Image registration for distortion correction in diffusion tensor imaging. *Biomedical Image Registration LNCS* 2003;2717:171–180.
- Pajevic S, Basser P. Parametric and non-parametric statistical analysis of DT-MRI data. *Journal of magnetic resonance* 2003;161(1):1–14. [PubMed: 12660106]
- Park H, Kubicki M, Shenton M, Guimond A, McCarley R, Maier S, Kikinis R, Jolesz F, Westin CF. Spatial normalization of diffusion tensor MRI using multiple channels. *NeuroImage* 2003;20(4):1995–2009. [PubMed: 14683705]
- Parker G, Wheeler-Kingshott C, Barker G. Estimating distributed anatomical connectivity using fast marching methods and diffusion tensor imaging. *IEEE Transactions on Medical Imaging* 2002;21(5):505–512. [PubMed: 12071621]

- Pfefferbaum A, Sullivan E, Hedehus M, Lim K, Adalsteinsson E, Moseley M. Age-related decline in brain white matter anisotropy measured with spatially corrected echo-planar diffusion tensor imaging. *Magnetic resonance in medicine* 2000;44(2):259–268. [PubMed: 10918325]
- Pierpaoli C, Jezzard P, Basser P, Barnett A, Chiro G. Diffusion tensor MR imaging of human brain. *Radiology* 1996;201(3):637–648. [PubMed: 8939209]
- Plessen K, Larsen T, Hugdahl K, Feineigle P, Klein J, Staib L, Bansal R, Peterson B. Altered interhemispheric connectivity in individuals with tourette syndrome. *American Journal of Psychiatry* 2004;161:2028–2037. [PubMed: 15514403]
- Press, W.; Teukolsky, S.; Vetterling, W.; Flannery, B. *Numerical Recipes in C. The Art of Scientific Computing*. Cambridge University Press; 1992.
- Rosner, B. *Fundamentals of Biostatistics*. Duxbury; 1995.
- Ruiz-Alzola J, Westin CF, Warfield S, Alberola C, Maier S, Kikinis R. Nonrigid registration of 3D tensor medical data. *Medical Image Analysis* 2002;6(2):143–161. [PubMed: 12045001]
- Ruiz-Alzola, J.; Westin, CF.; Warfield, S.; Nabavi, A.; Kikinis, R. Nonrigid registration of 3d scalar vector and tensor medical data. In: DiGioia, A.; Delp, S., editors. *Proceedings of MICCAI 2000, Third International Conference on Medical Image Computing and Computer-Assisted Intervention*; Pittsburgh. 2000. p. 541-550.
- Schneider, P.; Eberly, D. *Geometric Tools for Computer Graphics*. Morgan Kaufmann Pub; 2002.
- Schwartzman, A. PhD thesis. Stanford University; 2006. Random Ellipsoids and False Discovery Rates: Statistics for Diffusion Tensor Imaging Data.
- Schwartzman A, Dougherty R, Taylor J. Cross-subject comparison of principal diffusion direction maps. *Magnetic Resonance in Medicine* 2005;53:1423–1431. [PubMed: 15906307]
- Shen Y, Pu I, Clark C. Analytic expressions for noise propagation in diffusion tensor imaging. *Proceedings of the International Society for Magnetic Resonance in Medicine* 2006;14
- Song S, Sun S, Ramsbottom M, Chang C, Russell J, Cross A. Dysmyelination revealed through MRI as an increased radial (but unchanged axial) diffusion of water. *Neuroimage* 2002;17:1429–1436. [PubMed: 12414282]
- Song S, Yoshino J, Le T, Lin S, Sun S, Cross A, Armstrong R. Demyelination increases radial diffusivity in corpus callosum of mouse brain. *NeuroImage* 2005;26(1):132–140. [PubMed: 15862213]
- Stevens, J. *Applied Multivariate Statistics for the Social Sciences*. Lawrence Erlbaum Associates; London: 2002.
- van der Vaart, A. *Asymptotic Statistics*. Cambridge University Press; 2000.
- Viola P, Wells W. Alignment by maximization of mutual information. *International Journal of Computer Vision* 1997;24(2):137–154.
- Wang Z, Vemuri B, Chen Y, Mareci T. A constrained variational principal for direct estimation and smoothing of the diffusion tensor field from complex DWI. *IEEE Transactions on Medical Imaging* 2004;23(8):930–939. [PubMed: 15338727]
- Westin, C.; Knutsson, H. Tensor field regularization using normalized convolution. In: Diaz, RM.; Arencibia, AQ., editors. *Computer Aided Systems Theory (EUROCAST'03)*, LNCS 2809. Las Palmas de Gran Canaria, Spain: Springer Verlag; 2003. p. 564-572.
- Xu, D.; Bansal, R.; Plessen, K.; Martin, L.; Peterson, B. A DTI study of Tourette's Syndrome. *Human Brain Mapping*; 2005.
- Xu D, Mori S, Shen D, van Zijl P, Davatzikos C. Spatial normalization of diffusion tensor fields. *Magnetic Resonance in Medicine* 2003;50:175–182. [PubMed: 12815692]
- Yao Y. An approximate degrees of freedom solution to the multivariate Behrens-Fisher problem. *Biometrika* 1965;52(1):139–147.

Figure 1. Graph of Fractional Anisotropy (FA) Values for Increasing Eigenvalues

Two eigenvalues of the tensor were set to 20 and 30, and the third was varied from 0 to 200. Note differing eigenvalues can yield the same FA value and that small changes in eigenvalues can cause large changes in the FAs. The FA values were computed using the formula in Bassar and Pierpaoli (1996). Both FA and the eigenvalues are dimensionless, continuous variables.

Figure 2. Maximum and Minimum Values of Eigenspace Components for Increasing Variance in Tensor Elements

We generated simulated groups of 10,000 random tensors for increasing variance and a diagonal covariance matrix between the tensor elements. *Eigenvalue1*, *Eigenvalue2*, and *Eigenvalue3* are the three eigenvalues of the DTs corresponding to the 5, 25, and 50 eigenvalues of the Mean Specified Tensor (MST), respectively. Blue: Maximum values for eigenspace components; Purple: Minimum values for eigenspace components. The MST eigenspace components are shown in yellow. Therefore, eigenvalues and eigenvectors of the simulated tensors varied in large intervals around the corresponding values of the MST.

Figure 3. Theoretical Versus Sample Average (SA) Values of the Covariance Matrix Between Eigenspace Components

When generating the groups of 10,000 random diffusion tensors (DTs), we assumed the presence of a diagonal covariance matrix between the tensor elements with specified mean values of $\bar{y} = (6.0598, -5.075, 0.0, 23.6402, 0.0, 50.0)$. We increased σ^2 from 1.0 to 10.0 in the diagonal covariance matrix. Ideally (purple), the SA values should equal the theoretical values that were calculated using our method. Theoretical values computed using our method were close (blue) to the SA values, even for large amounts of noise in the tensors. *Covar* (1,1) = variance in Eigenvalue1; *Covar*(2,2) = variance in Eigenvalue2; *Covar*(3,3) = variance in Eigenvalue3; *Covar*(4,4) = variance in Alpha; *Covar*(5,5) = variance in Beta; *Covar*(6,6) = variance in Gamma.

Figure 4. Group-Average Values of Eigenspace Components Computed Using Differing Methods
 Values of mean tensor (MT) eigenspace components (blue); sample average (SA) eigenspace components (purple); mean specified tensor (MST) eigenspace components (yellow). The groups of 10,000 random tensors were generated for increasing variance and assuming a diagonal covariance matrix between the tensor elements. MT eigenspace components matched well with those of the MST, thereby validating our method of computing the MT for a group of subjects.

Figure 5. Theoretical Versus Sample Average (SA) Values of the Diagonal Elements in the Covariance Matrix for Increasing Variance

Groups of 10,000 random tensors were generated using upper block-diagonal covariance matrix across tensor elements. The σ^2 of the covariance matrix was increased from 1.0 to 10.0. Ideally (purple), the SA values equal the theoretical values. SA values (blue) closely matched the theoretical values computed using our method. The off-diagonal covariance values are not plotted because they were all small. Theoretically computed covariance matrix match well with the covariance matrix computed from simulated sample of tensors, in which correspondences across eigenvectors were known. Therefore, the Delta method computed the covariance matrix of the eigenspace components with high accuracy. Definitions for Covar(x,x) are as indicated in the legend for Figure 3.

Figure 6. Group-Average Values of Eigenspace Components Computed By Various Methods
 Values of mean tensor (MT) eigenspace components (blue), sample average (SA) eigenspace components (purple), and MST eigenspace components (yellow). We computed the group average values for each group of 10,000 random tensors generated for increasing σ^2 in the block-diagonal covariance matrix between mean specified tensor (MST) elements.

Figure 7. Group-Average Fractional Anisotropy (FA) Values Computed for Increasing Variance and Increasing Numbers of Subjects

Mean tensor FA (MT-FA) values (blue); sample average FA (SA-FA) values (purple). The top row of figures (a, b, c) plot the graphs for varying number of subjects at three differing levels of noise in the generated tensor elements. The bottom row of figures (d, e, f) plots the graphs for varying amounts of noise for three differing numbers of subjects. The FA of the mean specified tensor (MST) was 0.7.

Figure 8. Graphs of the P -values for Differences When Comparing Two Groups of Synthetically Generated Diffusion Tensors (DTs)

The MST elements for the first group of tensors were $\bar{y} = (6.0598, -5.075, 0.0, 23.6402, 0.0, 50.0)$. The MST elements for the second group of tensors were varied for each of the four differing cases. P -values of fractional anisotropy (FA) and various statistics that were significant are plotted. In each case, our method correctly identified the eigenspace component that differed significantly across the two groups. The graphs and their labels were color encoded: Ht = Hotelling's T^2 statistics (dark blue); FA = FA values (blue); $E1, E2, E3$ = Three eigenvalues ($E1$ = purple); A = Alpha; B = Beta; G = Gamma (green).

Case I: Because the MSTs were equal for the two groups in this case, between-group differences in tensor morphology were not statistically significant using any statistic. Therefore, a graph for this case is not shown.

Case II: Only γ differed between the two groups. Our method correctly identified γ as the source of the between-group difference in tensor morphology.

Case III: Only one eigenvalue ($E1$) differed between the two groups. Both our method and the method using FA determined that the two groups differed significantly in tensor morphology. Our method, in addition, correctly identified differences in morphology due to $E1$.

Case IV: $E1$ differed only minimally (0.8) between the two groups.

Case V: Although the mean specified tensor FAs (MST-FAs) were equal for the two groups, their $E1$ s differed. Our method correctly detected that the between-group difference derived from difference across groups in $E1$. However, the method using FA values failed to detect significant difference between the two groups in tensor morphology.

Figure 9. The Effects of Errors in Reorientation on Mean Tensor (MT) Morphology

We generated two sets of simulated tensors (one with 50 and the other with 5000) by rotating the Mean Specified Tensor (MST) about the Z-axis by randomly varying degrees but sampled uniformly within specified ranges ($\pm 1^\circ$, $\pm 2^\circ$, $\pm 4^\circ$, $\pm 8^\circ$, $\pm 16^\circ$, $\pm 32^\circ$, and $\pm 64^\circ$) of the angle γ . The MST eigenvalues were $E_1 = 10$, $E_2 = 1$, and $E_3 = 1$, and the eigenvectors were aligned along the X, Y, and Z axes, respectively. We computed MT eigenvalues and eigenvectors for each set of tensors. In the figure, the horizontal axis plots the logarithm of the absolute value of the range in which γ was sampled uniformly. The vertical axis plots the eigenvalues and γ computed from the MT. The eigenvalue E_3 , and the angles α and β of the MT equaled those of the MST in each experiment. Even for large amounts of errors in correctly reorienting tensors, the MT eigenvalues and eigenvectors matched closely to those of the MST. Left Column: E_1 = purple; E_2 = yellow.

Figure 10. Fractional Anisotropy (FA) Images Computed for the Group of 24 Healthy Individuals
 The top row shows the values of the mean tensor FA (MT-FA); the bottom row shows the values of the sample average FA (SA-FA). The values of FA at 6 locations in the upper row of images were 0.7528, 0.75524, 0.4861, 0.5787, 0.4725, and 0.5301; values at the corresponding locations in the lower row of images were 0.7963, 0.7781, 0.5382, 0.6044, 0.5323, and 0.5765. We set all three eigenvalues of the tensors in the background (i.e. voxels containing no brain tissue) equal to zero. Then in the common coordinate space, we computed mean FA values at only those locations where every tensor for each subject has nonzero eigenvalues.

Figure 11. Comparing Tensor Morphologies in 24 Healthy and 22 TS Subjects

Diffusion tensor (DT) image of each subject was coregistered and reoriented into a common reference space. The P -values of differences in mean values of the four statistics of tensor morphology for the two groups were computed, color encoded, and displayed on 4 parasagittal slices of the reference brain. Shown in each image are the P -values for the group differences in (1) sample average FA (SA-FA) values, (2) SA principal diffusivities, (3) vectors of mean tensor (MT) eigenvalues, and (4) MT principal diffusivities. For images in the 1st, 2nd, and the 4th columns, *red* shows that the statistic is significantly greater (*violet* shows statistic is smaller) in the TS group as compared to its values in healthy individuals, with a P -value of 0.001 or less. See color bar on the right. For images in the 3rd column, *red* shows statistically significant (P -value ≤ 0.001), between-group differences in MT eigenvalues, and *violet* shows regions of differences with P -values of 0.2. See color bar on the right in Fig. 12. **A** = Anterior; **B** = Posterior; **CC** = Corpus Callosum.

Figure 12. Comparing the TS and Healthy Subjects in the Axial Orientation

The results were resliced, color encoded, and displayed on four slices in the axial orientation. Shown in each image are the P -values for the between-group differences in (1) sample average FA (SA-FA) values, (2) SA principal diffusivities, (3) vectors of mean tensor (MT) eigenvalues, and (4) MT principal diffusivities. Color encoding for the P -values of the various statistics is the same as that in Fig. 11. Note that diffusion tensor data was acquired in only 19 slices around the interhemispheric fissure for each subject. **A** = Anterior; **P** = Posterior.