

# Enhancing Measured Diffusion Anisotropy in Gray Matter by Eliminating CSF Contamination With FLAIR

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**In this work, the effect of fluid-attenuated inversion recovery (FLAIR) on measured diffusion anisotropy was investigated in gray matter. DTI data were obtained with and without FLAIR in six normal volunteers. The application of FLAIR was experimentally demonstrated to lead to a consistent increase in fractional anisotropy (FA) in gray-matter regions, which was attributed to suppressed partial volume effects from CSF. In addition to these experimental results, Monte Carlo simulations were performed to ascertain the effect of noise on the measured FA under the experimental conditions of this study. The experimentally observed effect of noise was corroborated by the simulation, indicating that the increase in the measured FA was not due to a noise-related bias but to an actual increase in diffusion anisotropy. This enhanced measurement of diffusion anisotropy can be potentially used to differentiate directionally dependent structure and tracking fibers in gray matter. Magn Reson Med 51:423–427, 2004. © 2004 Wiley-Liss, Inc.**

**Key words:** diffusion tensor imaging; FLAIR; fractional anisotropy; SNR; gray matter

By measuring the diffusion characteristics of water in tissue along at least six noncolinear directions in space, diffusion tensor imaging (DTI) maps the directional dependence of water diffusion and is useful for visualizing microstructural tissue organization in the human brain (1). The advent of DTI in the last decade allows investigators to noninvasively visualize neural fiber tracts in vivo (2,3), which facilitates the study of brain development and the pathology of diseases associated with white-matter damage and disruption. Recent advances in tract-tracking techniques have further advanced the exploration of neural connectivity and neurological pathways in the human brain (4–7).

The application of DTI assumes a parallel relationship between the direction of maximum diffusion (i.e., the principal eigenvector of the diffusion tensor) and the direction of fiber fascicles traversing the imaged voxel. The measurement of diffusion-weighted MR signals, however, suffers from several limitations and is prone to artifacts that compromise accuracy in the derived diffusion tensor. For instance, cerebrospinal fluid (CSF) can degrade diffusion

mapping, resulting in an overestimate of the trace of the diffusion tensor (8,9). Therefore, a technique that reduced the inaccuracy caused by CSF contamination in diffusion tensor measurements would allow better tract-tracking in both white and gray matter, especially in gray-matter areas such as the hippocampus, thalamus, and cortex. This would increase our understanding of the brain anatomy of neural fibers, neurological pathways, and disruptions in disease states.

Recent studies have suggested that applying fluid-attenuated inversion recovery (FLAIR) to suppress CSF (10) before the diffusion weighting could reduce CSF contamination in the measured diffusion tensor (11,12). Although FLAIR has been used to address CSF contamination in ADC measurements in acute stroke (12) and epilepsy (13), and diffusion anisotropy measurements in white matter (11), the effect of CSF contamination on the measurement of diffusion anisotropy in gray matter has not been investigated. Therefore, the main goal of this study was to extend previous studies of FLAIR-DTI in white matter (11) to examine the effect of suppressing CSF, using FLAIR, on the measurement of diffusion anisotropy in gray matter. In this study, DTI was conducted with and without FLAIR in normal volunteers to investigate whether suppression of the CSF signal improves the measured diffusion anisotropy in gray matter in the human brain.

It has been demonstrated that noise in DTI data can bias the resultant diffusion tensor and lead to an overestimate of diffusion anisotropy when both the SNR and actual anisotropy are low (2,14). This overestimation may overwhelm fractional anisotropy (FA) in areas with relatively low diffusion anisotropy, such as gray matter. Because FLAIR reduces the signal-to-noise ratio (SNR) of the DTI data, the noise-related bias may contribute significantly to the FLAIR results. To rule out this possibility, we performed experiments and numerical simulations to evaluate the influence of noise at a level corresponding to our experimental data.

## MATERIALS AND METHODS

Diffusion-tensor data were acquired using a diffusion-weighted, single-shot, spin-echo, echo-planar imaging (EPI) sequence. A dual spin-echo technique combined with bipolar gradients (15) was employed to minimize the geometric distortion induced by eddy currents. Diffusion-weighting gradients were applied in 12 directions. In FLAIR-DTI, the DTI sequence was preceded with an inversion recovery pulse (TI = 2250 ms) to suppress the CSF signal.

Six normal volunteers, who provided informed consent, participated in this study. All experiments were carried out on a 3 T Siemens Trio system (Siemens Medical Sys-

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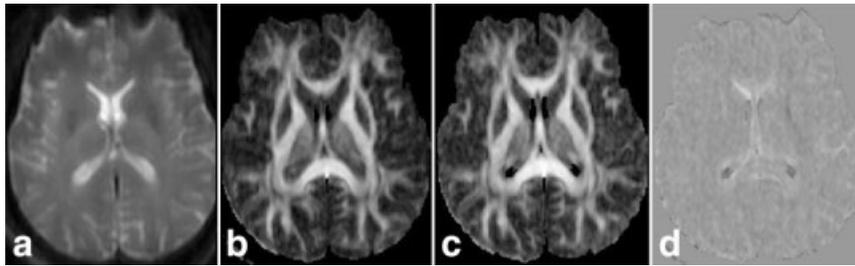


FIG. 1. DTI images in a normal volunteer: (a)  $T_2$ , (b) FA map from conventional DTI, (c) FA map from FLAIR-DTI, and (d) difference map obtained by subtracting **b** from **c**.

tems, Iselin, NJ). The following parameters were used: TR/TI/TE = 6462/2250/85 ms for FLAIR-DTI, and TR/TE = 2692/85 ms for conventional DTI; FOV = 22 cm  $\times$  22 cm; slice thickness = 5 mm; slice gap = 2.5 mm; number of slices = 5;  $b$ -values = 0 and 1000 s/mm<sup>2</sup>; and six averages. The total imaging times for FLAIR-DTI and conventional DTI were 8:25 min and 3:30 min, respectively. Images (128  $\times$  96 matrix size) were acquired in the axial orientation. The diffusion tensor was calculated for each voxel, and FA maps were generated using DtiMap (7). To study the effects of experimental noise on FA, we also performed the experiments in one subject using different numbers of averages (i.e., 1, 2, 4, or 6), while the rest of the parameters were the same as those described previously.

We conducted Monte Carlo simulations to ascertain the effects of noise on our experimentally derived FA calculations. Starting from an assumed diffusion tensor, diffusion-weighted measurements were simulated for two  $b$ -values ( $b = 0$  and 1000 s/mm<sup>2</sup>) in the same 12 diffusion gradient directions used for the experimental data acquisition. Two diffusion tensors, representing values of gray matter measured with and without FLAIR, respectively, were assumed in the simulation. White Gaussian noise was added to individually simulated measurements for both  $b$ -values. Different levels of noise were used to cover an SNR range of 5–50 dB. Since the image SNR depends on the  $b$ -value, the SNR was defined operationally as measured in the  $b = 0$  measurement. The simulated data sets were used to derive the diffusion tensor and FA in the same way as used for the experimental data. The simulations were repeated 100 times for each case, and the mean and standard deviation (SD) of the obtained FA values were computed.

## RESULTS

FA images obtained with and without FLAIR from a representative subject are shown in Figs. 1 and 2. While the

average FA over the entire brain was not significantly increased by FLAIR, the FA in the gray matter increased by 80–100% in the FLAIR-DTI result. FA values in gray matter of the frontal, parietal, and occipital regions, as well as white matter in the splenium (CCs) and genu (CCg) of the corpus callosum, obtained using both techniques, were calculated with the results listed in Table 1. A paired  $t$ -test revealed that the increase in gray matter FA was statistically significant ( $P < 0.001$ ). The increase in FA in white matter was consistent with the results reported previously (11).

To further examine the effect of FLAIR on the gray-matter DTI measurement, we identified pixels within the gray matter by thresholding the FA map obtained with conventional DTI using an upper threshold of 0.15, as proposed by Stieltjes et al. (16). Figure 3a–c illustrate the gray-matter FA maps obtained with the two methods, and their difference; their corresponding histograms are shown in Fig. 3d–f. An increase of FA in gray matter is evident in Fig. 3a–c. This increase is also evident in the histograms shown in Fig. 3d–f. The overall enhancement of FA using FLAIR in gray matter is >30%. The small number of pixels exhibiting negative values in difference map is likely due to noise and/or motion between the scans.

The influence of noise on experimentally obtained FA values in gray matter is illustrated in Fig. 4. The four data points in Fig. 4 correspond to the average of 1, 2, 4, and 6 measurements, respectively. As expected, the SNR increased proportionally to the square root of the number of averages in both cases. The results also show that the application of FLAIR reduced the SNR by approximately 20%. For both methods, a slight increase in FA was observed when SNR was decreased. However, at any given SNR, the FA value obtained with FLAIR was always larger than for conventional DTI.

Figure 5 presents the mean and SD of FA values derived from simulated data as a function of SNR. The results in Fig. 5 are in good agreement with the experimental data

FIG. 2. FA maps in a volunteer obtained using (a) conventional DTI and (b) FLAIR-DTI. **c** and **d**: Magnifications of the left frontal areas of **a** and **b**, respectively.

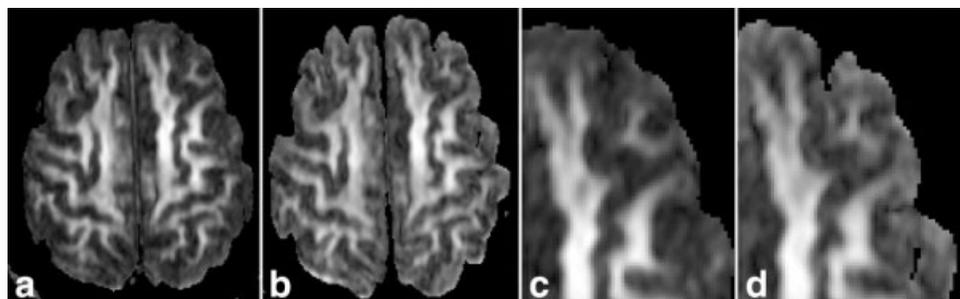


Table 1  
FA Values in Gray and White Matter for Six Subjects Obtained With and Without FLAIR

	Frontal lobe	Gray matter parietal lobe	Occipital lobe	White matter	
				CCs	CCg
Without FLAIR	0.105 ± 0.017	0.090 ± 0.020	0.128 ± 0.047	0.706 ± 0.037	0.611 ± 0.030
With FLAIR	0.197 ± 0.029	0.197 ± 0.028	0.205 ± 0.045	0.761 ± 0.037	0.683 ± 0.032
Increase	0.092 ± 0.028	0.107 ± 0.039	0.078 ± 0.040	0.053 ± 0.024	0.072 ± 0.022

shown in Fig. 4. The Monte Carlo simulations indicate that the SD of the calculated FA increases with decreasing SNR, and noise leads to a bias in the derived FA. However, the increase in FA in our experimental data from FLAIR is significantly larger than the noise-related bias in the range of SNR of our experimental data at 3 T. The significance of this FLAIR-induced FA increase in gray matter (with six averages) was assessed by calculating the z-score corresponding to the increase according to

$$z = \frac{(FA_{FLAIR-DTI} - bias_{FLAIR-DTI}) - (FA_{DTI} - bias_{DTI})}{\sqrt{SD_{FLAIR-DTI}^2 + SD_{DTI}^2}},$$

where the biases and SDs were derived based on simulations at SNRs of the DTI data with and without FLAIR, respectively. The resultant z-score was 3.76, which corresponds to  $P < 0.001$ , and confirms that the increase in measured gray-matter FA was not primarily a result of noise.

## DISCUSSION

While FLAIR was previously shown to increase measured diffusion anisotropy in white matter (11), the present work demonstrates that FLAIR also leads to a statistically significant increase in FA in gray matter. The effect of FLAIR on DTI can be intuitively understood based on the following argument: Diffusion in CSF is likely isotropic, and the partial volume effect between brain tissue and CSF would lead to a reduction of FA in the measured data. It follows, therefore, that the elimination of the CSF signal increases the observed FA. This increase is more prominent in the gray matter, where the CSF contamination is likely more significant. The increase of measured FA in the gray matter may make it possible to track fibers in the gray matter, a possibility that will significantly expand the utility of DTI in neuroscience. It is also interesting to note that if the partial volume effect explains the observed increase in FA with FLAIR, such an increase may depend on voxel size,

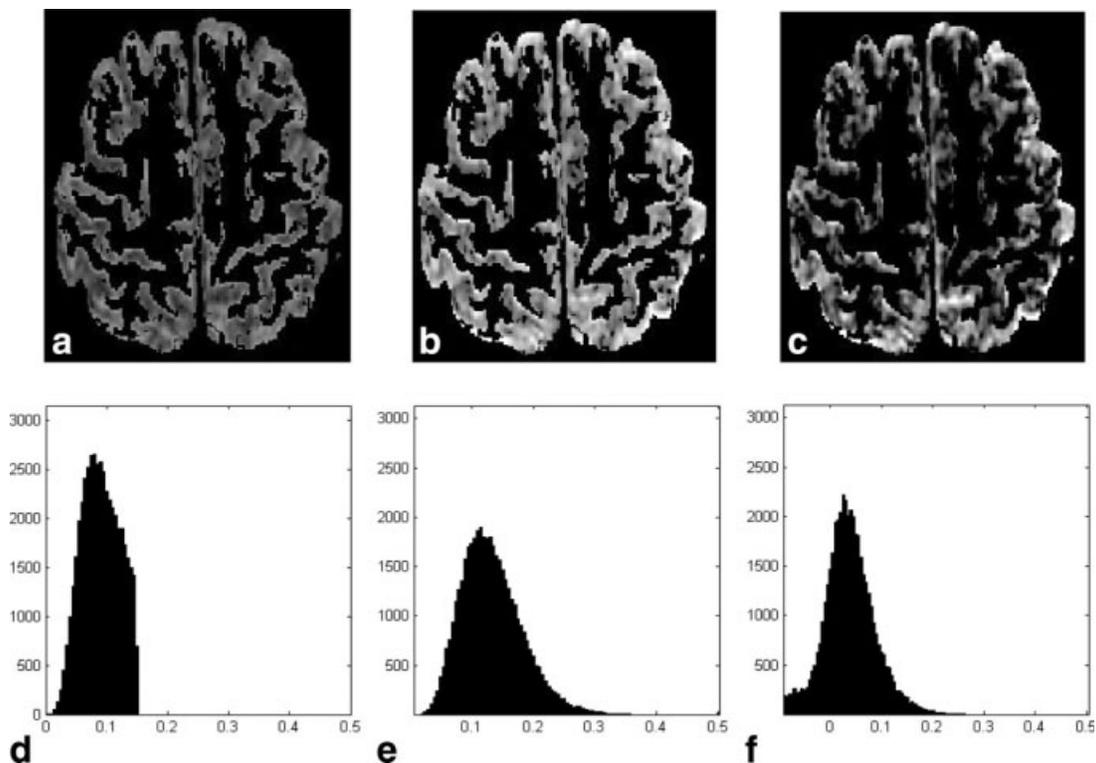


FIG. 3. Comparison of the gray-matter FA maps between (a) conventional DTI and (b) FLAIR-DTI. c: The difference map produced by subtracting a from b. d-f: The histograms corresponding to a-c. The image slice is the same as that shown in Fig. 2. Notice that the gray-matter mask was derived from an upper limit of 0.15 in FA from a.

with the amount of enhancement in FA by FLAIR diminishing with increasing spatial resolution.

An alternative explanation for the observed increase in FA may be that it is due to a reduction of signal arising from long- $T_1$  compartments if diffusion in these compartments is less anisotropic. At 3 Tesla, these compartments may have a sufficiently long  $T_1$  such that their signal is significantly attenuated by the inversion recovery with the 2.25 TI used in this work. This possibility remains to be further studied.

Although FLAIR increases the derived FA according to our experimental data, it also suppresses the raw image signal, reducing the SNR. Noise is known to bias the resultant FA, particularly if the true FA is low (2,14). To rule out the possibility that this bias explains the gray-matter FA increase, experimental and simulated SNR dependence studies were conducted. They indicated that the decrease in SNR due to FLAIR increased the measured FA at a much lower level compared to the observed increase. In fact, the simulations showed that for SNR corresponding to our data, the increase in calculated gray-matter FA due to noise was about 0.01, which is substantially less than the increase of FA that we observed in the experiments (0.04–0.08). In our experiments at 3 T, the reduction in SNR using FLAIR-DTI was typically about 20% compared to DTI without FLAIR and with the same number of averages. With noise taken into account, the measured increase in gray-matter FA using FLAIR is statistically significant. Therefore, the enhancement of FA in gray matter using FLAIR-DTI at 3 T primarily represents an improvement from CSF suppression, rather than a biased result from the reduction in SNR. Of note is that the incorporation of FLAIR lengthens the TR and thus reduces the SNR achievable per unit time. As a result, the efficiency of FLAIR-DTI will be reduced, offsetting to some extent the enhancement by FLAIR. The full impact of this aspect remains to be investigated.

Further study of the issue of CSF contamination and its effect on the measurement of diffusion anisotropy could be

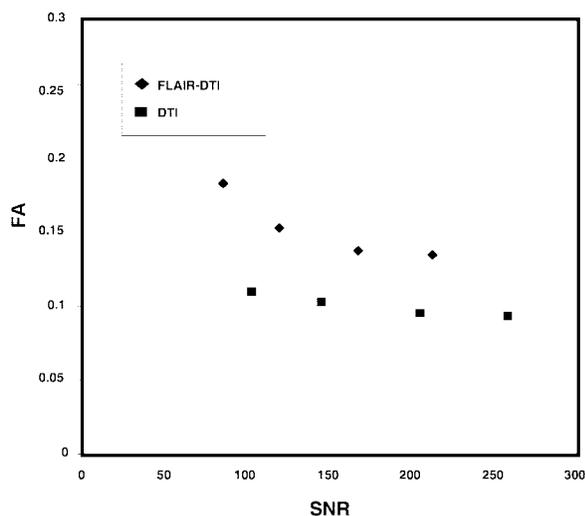


FIG. 4. ROI measurements of FA as a function of SNR in gray matter in an axial slice. Different numbers of averages (1, 2, 4, or 6) were used to achieve different SNRs experimentally.

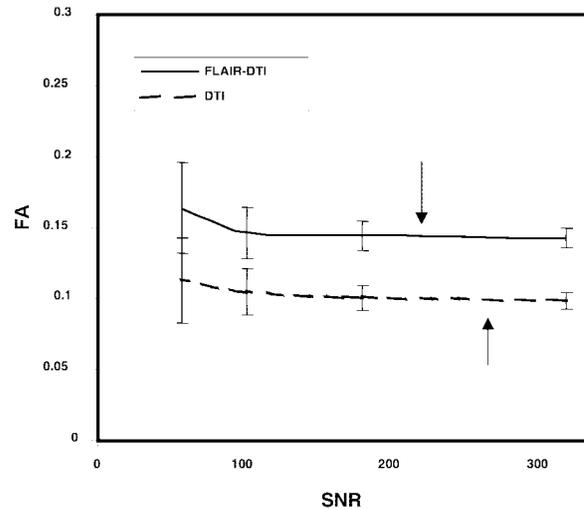


FIG. 5. Monte Carlo simulation results: FA and SD as a function of SNR, with and without FLAIR, simulated for gray matter. The assumed diffusion tensors were chosen based on the experimental results shown in Fig. 4. The arrows indicate FA values corresponding to the SNR measured in the experiments when the number of averages equals 6.

facilitated by improving the slice profile of the inversion pulse. For example, an adiabatic frequency offset-corrected inversion (FOCI) inversion pulse (17), which has been used for multislice perfusion imaging (18), can be incorporated into FLAIR-DTI to generate a sharper slice profile of the inversion pulse compared to that from the hyperbolic secant pulse used in our current FLAIR-DTI.

A preliminary version of this work was presented at the 11th annual meeting of the International Society of Magnetic Resonance in Medicine, in Toronto (19). At the same meeting, another group also reported an increase in FA in gray matter when FLAIR was used, which they attributed to partial volume averaging (20). However, their results showed a smaller increase (5–14%) in gray matter, in contrast to our result. This discrepancy probably reflects the facts that 1) the slice thickness used in our study was larger than theirs, and hence we observed a larger CSF partial voluming effect; and 2) their gray-matter ROIs contained mostly pixels next to the white matter. The latter point is confirmed by the high FA in their gray-matter ROI obtained without FLAIR.

In summary, FLAIR-DTI suppresses CSF signal in raw DTI images and improves the resultant diffusion anisotropy in the brain, particularly in gray matter. At the SNR level for our data, the observed enhancement was not due to noise. The enhanced FA in gray matter reveals the potential of this technique for assessing directionally-dependent structures in gray matter, which could significantly expand the clinical utility of DTI.

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