Chapter 10

Diffusion MRI Data Processing and Analysis: A Practical Guide with *ExploreDTI*

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Abstract

This chapter introduces neuroimaging researchers to the concepts and techniques of diffusion magnetic resonance imaging data processing. Using the freely available ExploreDTI software, we provide a step-by-step guide for processing multi-shell High Angular Resolution Diffusion Imaging data and generating tractography based on constrained deconvolution. Brief explanations of the rationale behind each processing step are provided to aid the researcher in understanding the concepts and principles involved. Potential processing pitfalls will be discussed, and tips for troubleshooting common issues will be provided. An additional step-by-step guide for processing DTI data using the open-access AOMIC data set is also provided, demonstrating command-line that can also be applied to process other large neuroimaging datasets.

Key words Diffusion, Tractography, DTI, Multi-shell, HARDI

1 Introduction

White matter refers to the nerve fibers, also known as axons, that interconnect regions of the brain [1]. Healthy development of white matter is essential for neurotypical brain function and cognition [2]. This complex developmental process involves several mechanisms such as axonal growth, myelination, and synaptic pruning [3]. The intricate interplay between these processes is key for the establishment of neural networks for the efficient transmission of information within the brain [2]. Abnormalities in white matter development have been linked to a range of cognitive functions [4] and psychiatric impairments, including autism [5], ADHD [6], and schizophrenia [7].

Supplementary Information The online version contains supplementary material available at https://doi.org/10.1007/978-1-0716-4260-3_10.

Robert Whelan and Hervé Lemaître (eds.), *Methods for Analyzing Large Neuroimaging Datasets*, Neuromethods, vol. 218, https://doi.org/10.1007/978-1-0716-4260-3_10,

Diffusion-weighted magnetic resonance imaging (dMRI) [*see* Glossary] is a powerful neuroimaging technique that allows for the investigation of white matter microstructure through the diffusion measurement of water molecules within biological tissue [8]. In white matter, the diffusion of water molecules is affected by cellular membranes (i.e., myelin sheaths), defining the diffusion-weighted contrast. This diffusion-weighted signal can then be mathematically modeled to estimate the underlying microstructure and reconstruct the organization of white matter tracts [9]. In the early 2000s, the most common dMRI modeling technique was Diffusion Tensor Imaging (DTI) [*see* Glossary] [10, 11]. While DTI remains a key tool for researchers in understanding the impact of white matter microstructure [12–14], DTI has some limitations, such as its inability to accurately model areas in which crossing white matter fibers are present [15–17].

In recent years, advances in dMRI acquisition parameters have enabled higher-order diffusion modeling techniques that increase reconstruction accuracy and can overcome some of the limitations of DTI. With High Angular Resolution Diffusion Imaging (HARDI), an increased number of diffusion direction gradients is acquired, which allows for the estimation of microstructural properties along multiple fiber populations within a single voxel and provides improved reconstruction accuracy of white matter tracts compared to the traditional DTI framework [18]. Another advance in dMRI for tractography is the integration of multiple b-values [19, 20]. Briefly, b-values are a summary measure of the strength, duration, and amplitude of the diffusion-weighting applied during the scan. Different strength b-values elicit altered tissue responses which can be used to increase the reconstruction accuracy of various neurocellular environments. Higher b-values are more sensitive to detecting diffusion of water molecules within brain tissues [21] but are also more susceptible to noise and artifacts compared to lower b-values [22]. As such, multi-shell dMRI data leverages the increased signal of high b-value images with the reduced noise of low b-value images to provide increased anatomical accuracy [19].

In the context of higher-order diffusion modelling, techniques have been developed, such as constrained deconvolution (CSD), Q-ball, and neurite orientation and dispersion density imaging (NODDI) among many others [17, 23–26]. These techniques can describe the distribution of water diffusion within a voxel more accurately compared to DTI (e.g., the fiber orientation distribution function for CSD and diffusion orientation distribution function for DSI and Q-ball) and can be used to model voxels containing crossing white matter fibers. Thus, metrics derived from these higher-order models have increased accuracy, yielding clinically more relevant information that cannot be obtained from the DTI model [9]. Higher-order diffusion models provide more

detailed information about the microstructure and organization of white matter tracts, which can provide important insights into the pathophysiology of neurological and psychiatric disorders.

The aim of this chapter is to introduce neuroimaging researchers to the concepts and techniques of dMRI data processing used in the field, with a focus on providing a practical step-by-step guide for processing multi-shell HARDI data and generating CSD-based tractography using the ExploreDTI software [*see* Resources] [27]. Brief explanations of the rationale behind each processing step will be provided to aid the researcher in understanding the concepts and principles involved. Potential processing pitfalls will be discussed, and tips for troubleshooting common issues will be provided. Overall, this guide aims to provide a comprehensive resource for researchers to gain the skills and knowledge necessary to process dMRI data effectively and efficiently.

2 Methods

2.1 Starting Point for the Data

Advanced fiber orientation distribution modeling techniques, such as CSD, require specific diffusion parameters. Typically for multi-shell HARDI, a minimum of two b-values images (b = 2500–3000 s/mm^2) and 45 diffusion-weighted directions [9, 28] are required for CSD modeling for white matter tractography purposes. The Neuroimaging of the Children's Attention Project (NICAP) [29] study diffusion parameters were used for the processing step-by-step guide provided below. Data from the NICAP cohort are available via Lifecourse (https://lifecourse.melbournechildrens.com/cohorts/cap-and-nicap/).

2.2 Data Storage and Computational Expense

The step-by-step guide provided here was run on a Linux system with an Intel Core i7 processor and 32 GB RAM using MATLAB R2016b. A standalone version of ExploreDTI is also available. Details of the ExploreDTI instalment are provided below. It is recommended to run Steps 9 and 10 using high-performance computers, given the large processing time of these steps. Table 1 shows the estimated processing times per participant for each step, with MATLAB parallel processing enabled.

2.3 Step-by-Step Guide

In this section, we will provide a step-by-step guide for processing multi-shell HARDI data (in BIDS format [*see* Chapter 4]) and generating CSD-based tractography using the ExploreDTI software. As this guide is for those relatively new to neuroimaging, the ExploreDTI graphic user interface (GUI) is used. A step-by-step guide to installing and using ExploreDTI is provided in the user manual. As we are using BIDS format, each subject folder containing the diffusion files should have *.json*, *.bval*, *.bvec*, and *.nii* files (*see* Table 2). Although advanced diffusion modeling is not

Table 1
Approximate processing times for each per participant are provided below (Steps 7 and 8 are optional if multiple b-value data sets were acquired separately)

Processing step	Name of section	Approximate processing time
1	Convert Bval and Bvec files into text files	< 1 min
2	Signal drift	< 1 min
3	Sort Bvals	< 1 min
4	Gibbs ringing	< 1 min
5	Flip permute	< 1 min
6	Generate Mat File	5 min
7	Concatenate all b-value .mat files	2 min
8	Generate Mat File of concatenated .nii files	20 min
9	SM/EC/EPI distortion corrections	360 min
10	Whole brain tractography	70 min

Table 2
Description of dMRI files in BIDS format

File name	Comment
.json	File containing a description of scan acquisition details
.bval	File containing a summary of diffusion-weightings applied during scanning
.bvec	Files containing details on the diffusion gradient vectors of the scan
.nii	The raw diffusion scan in NIfTI format

feasible with the AOMIC datasets (https://nilab-uva.github.io/AOMIC.github.io/), we have also included step-by-step command lines to demonstrate the possibility to preprocess several subjects all at once (Appendix 5.2) [*see* Chapter 2].

2.3.1 Convert Bval and Bvec Files into Text Files (Step 1)

The first processing step is to generate *.txt* file(s) from the *.bval* and *.bvec* files for the images you are processing. The *.txt* file is a summary file of the b-values and diffusion-weighting directions used during image acquisition and is required for image processing.

In ExploreDTI:

1. Plugins → Convert → *.bval/*.bvec to B-matrix *.txt files (s) (*see* Fig. 1)
   (a) Select folder containing *.bval and *.bval file(s)
   (b) Select output folder for *.txt file(s)
2. The output folder now includes the converted .txt file(s)

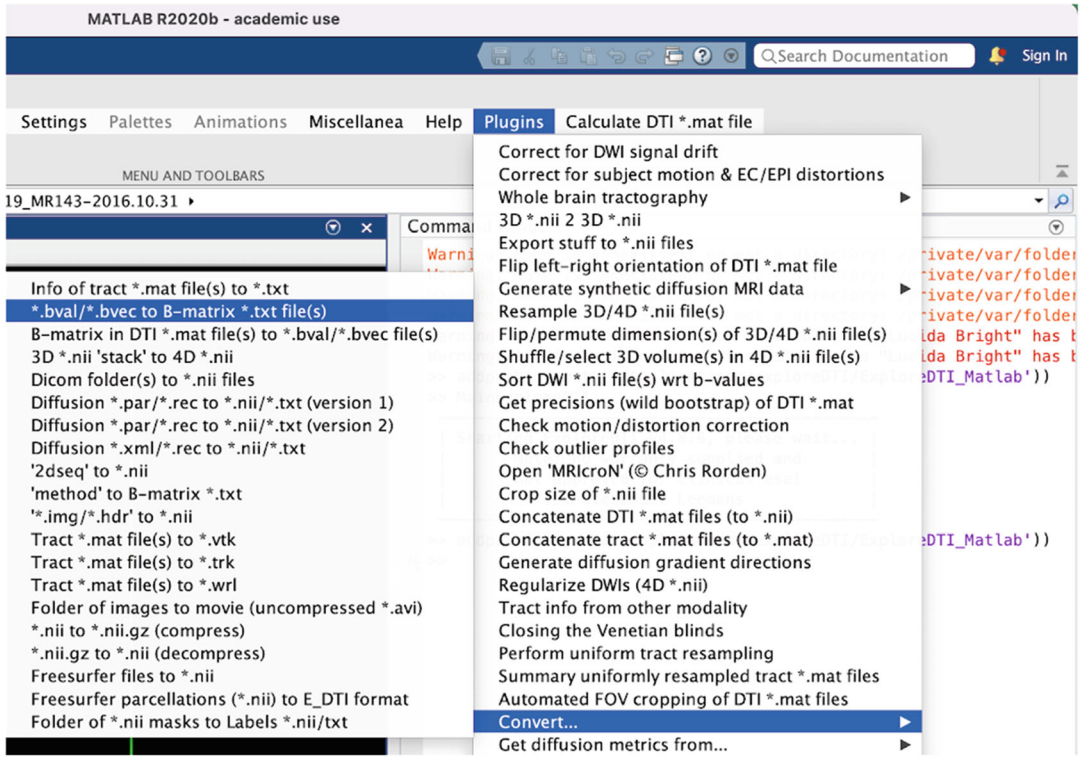

Fig. 1 Step 1 using the ExploreDTI GUI

2.3.2 Signal Drift Correction (Step 2)

Signal drift is a phenomenon caused by scanner imperfections, which leads to an adverse alteration of the acquired signal and a bias in the estimation of diffusion measures if not corrected [30].

> **Note**
> We recommend the use of "quadratic fit," but a signal drift fitting guide is provided in the Appendix 5.1 for users who want to investigate the impact of different fitting approaches (*see* Fig. A1).

In ExploreDTI:

1. Plugins → Correct for DWI signal drift (*see* Fig. 2).
   (a) Single or multiple data sets: multiple.
   (b) Select the folder of .nii file(s).
   (c) Select output folder.

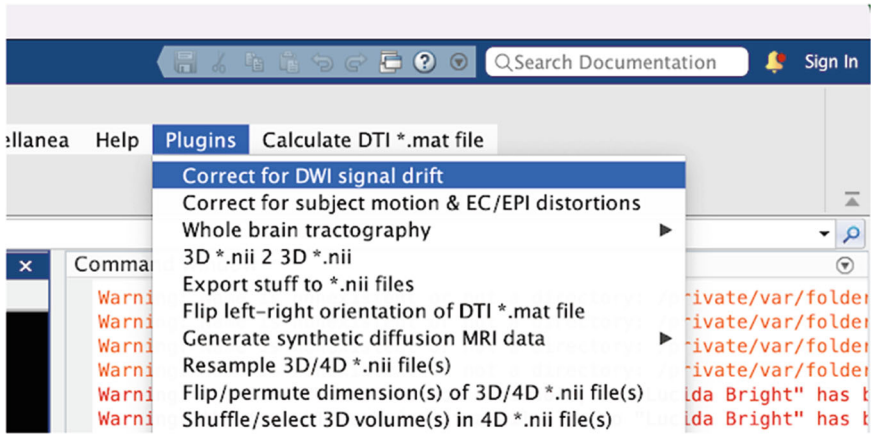

Fig. 2 Step 2 using the ExploreDTI GUI

2. The output folder now includes:
   (a) *_sdc.txt file(s)
   (b) *_sdc.nii file(s)
   (c) *_sdc.png file(s)

> **Note**
> As the signal drift correction uses the non-diffusion weighted (b-0) files acquired to correct for signal drift, it is crucial that signal drift correction is completed before sorting b values, which may change the order of the acquired dMRI volumes (**step 3**).

2.3.3 Sort B-Values, Organize and Remove Excess b-0 Files (Step 3)

For the remaining processing steps ExploreDTI requires that all b-0 files are sorted to the beginning of the diffusion files. This step quickly organizes the files to have all the b-0 files at the beginning of the *.nii* and *.txt* files.

In ExploreDTI:

1. Plugins → Sort DWI *.nii file(s) wrt b-values (*see* Fig. 3)
   (a) File name suffix: *_sorted.nii.
   (b) Single or multiple data sets: multiple.
   (c) Select the folder of *.nii file(s) and *.txt file(s).
   (d) Select output folder.
2. The output folder now includes:
   (a) *_sdc_sorted.txt file(s)
   (b) *_sdc_sorted.nii file(s)

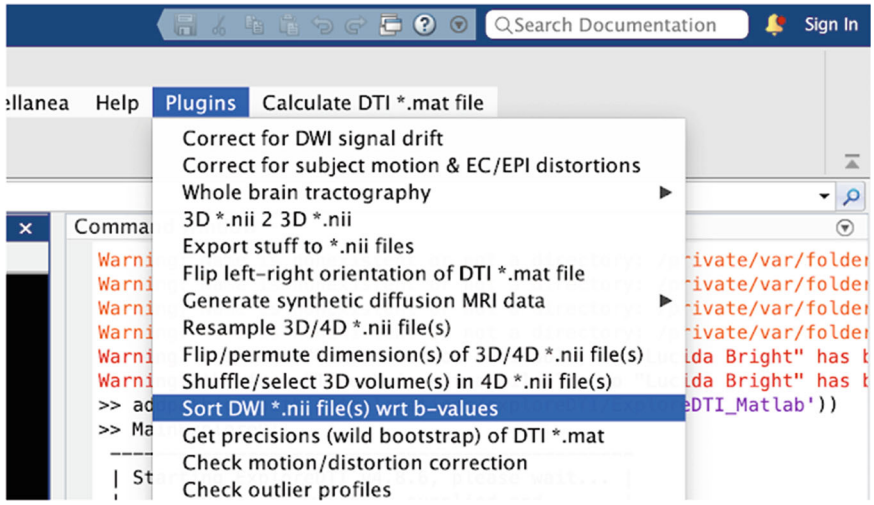

Fig. 3 Step 3 using the ExploreDTI GUI

> **Note**
> Rarely, additional b-0 files are collected during scanning. To investigate if extra b-0 files are present, open the newly sorted .txt file and investigate (*see* Fig. 4). If excess b-0 files are present (see red box in Fig. 4), these can be removed in ExploreDTI.

2.3.4 Gibbs Ringing Correction (Step 4)

A phenomenon known as Gibbs ringing may occur due to the shortening/truncation of Fourier transforms to reconstruct the MRI signal. If uncorrected, Gibbs ringing leads to artifacts that appear as multiple fine parallel lines in the image.

In ExploreDTI:

1. *Plugins → TV for Gibbs ringing in non-DWI's (4D *.nii)* (*see* Fig. 5)
   (a) *Select Gibbs Ringing Correction settings* (*see* Table 3).
   (b) *Single or multiple data sets:* Multiple.
   (c) Select the folder of *.nii files.
   (d) Select output folder.
2. The output folder now includes:
   (a) *_sdc_sorted_GR_TV.nii files.
3. Create and move *_sdc_sorted_GR_TV.nii files into a new folder.

sub-0751_ses-wave2_acq-mB1000_dwi_sorted.txt

0.00000000	0.00000000	0.00000000	0.00000000	0.00000000	0.00000000
0.00000000	0.00000000	0.00000000	0.00000000	0.00000000	0.00000000
0.00000000	0.00000000	0.00000000	0.00000000	0.00000000	0.00000000
0.00000000	0.00000000	0.00000000	0.00000000	0.00000000	0.00000000
0.00000000	0.00000000	0.00000000	0.00000000	0.00000000	0.00000000
0.00000000	0.00000000	0.00000000	0.00000000	0.00000000	0.00000000
0.00000000	-0.00000000	-0.00000000	0.03680566	12.01135738	979.96276035
186.72698939	-193.76339460	747.45690612	50.26623790	-387.81160643	748.00625811
227.47005810	92.36385555	-825.06390529	9.37604919	-167.50794369	748.15390374
88.76529892	235.05331942	514.53197590	155.60715631	681.24847413	745.62683125
506.25574580	234.06808929	961.66793922	27.05543141	222.31429365	456.68875514
18.04569042	128.08936946	-231.84363532	227.29646507	-822.81986367	744.65800407
436.56855645	-449.19630631	-874.45195565	115.54732849	449.87274361	437.88438921
44.67902543	-187.45677693	-365.79262985	196.62494243	767.36574655	748.69721727
573.37732659	-588.96640527	787.47242635	151.24483061	-404.44117921	270.37728626
87.73072406	-397.98017735	399.99161671	451.34764150	-907.25761280	455.92150058
551.61256248	565.77163321	-811.30467735	145.07353653	-416.06482843	298.31412674
885.45216734	-204.10868928	-588.50553422	11.76245273	67.82923892	97.78584789
924.06067726	-13.57524705	511.88749951	0.04985802	-3.76003408	70.89058614
410.36323522	-899.08545434	-399.38085739	492.46288705	437.51180516	97.17309908
158.56403640	597.41777469	-420.44928627	562.71902132	-792.05815731	278.71642010
130.77264062	555.72794200	381.90420675	590.40167738	811.46498933	278.82518552
791.40951054	-810.10025866	63.70298025	207.30810672	-32.60372796	1.28191209
19.73772108	-232.12653573	-153.32934856	682.48416788	901.61904613	297.77866748
868.56062516	658.49921662	-151.77103589	124.81029123	-57.53260356	6.63006320
592.55509018	831.09173750	524.42683507	291.41318993	367.76901994	116.03288450
326.92905733	-891.89171102	302.08120397	608.29009103	-412.05227225	69.78047052
0.86791375	-55.51647338	20.09778370	887.78372985	-642.78165694	116.34822891
414.26095255	983.25904440	-109.92057572	583.44766895	-130.44965930	7.29161950
76.37513962	-532.07105731	-46.07865938	926.67460715	160.50459048	6.95004572
51.71877864	443.55065292	38.82648649	950.99491373	166.49188060	7.28698595

Fig. 4 Example of a sorted *.txt* file containing 6 b-0 images

2.3.5 Flip Permute (Step 5)

Permutations and flips in spatial configuration and/or mismatches between spatial and diffusion coordinate systems can accidentally occur during processing and analyses across different software packages, potentially resulting in errors. The "flip/permute" tool in ExploreDTI can reorientate images and also avoid further unexpected axis flips and permutations in any following image processing step. Use default ExploreDTI settings as orientations will be inspected at the next step.

In ExploreDTI:

1. Plugins → Flip/permute dimension(s) of 3D/4D *.nii files (*see* Fig. 6)
   - (a) Use default setting:

     File name suffix: _FP

     Permute dimensions: 1 2 3

     Flip dimensions: 0 0 0.

     Force voxel size: leave empty
   - (b) Single or multiple data sets: multiple.
   - (c) Select the folder of *.nii file(s).
   - (d) Select output folder.

The output folder now includes *_sdc_sorted_GR_TV_FP.nii files.

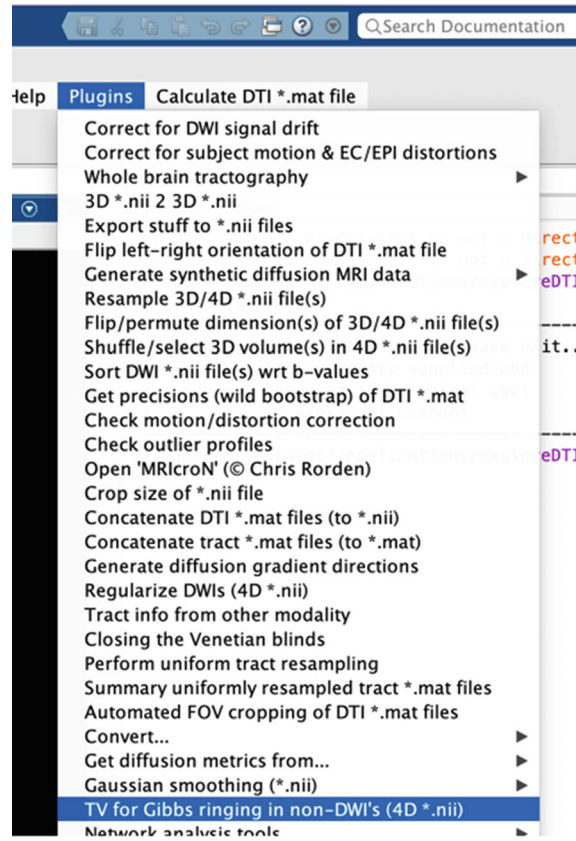

Fig. 5 Step 4 using the ExploreDTI GUI

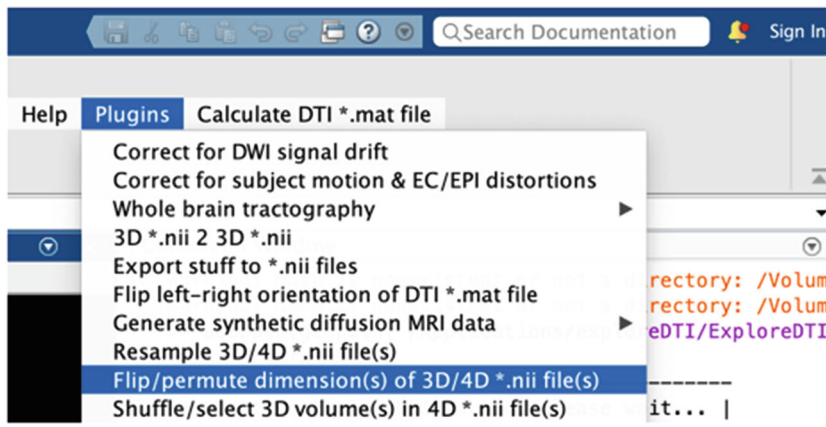

Fig. 6 Step 5 using the ExploreDTI GUI

Table 3
Setting Gibbs ringing parameters

Parameter	Comment
Number of non-DWIs	This information is provided in the *.txt file
Lambda ([1 200])	Lambda is a parameter that can be used to control the degree of Gibbs ringing in image reconstruction algorithms. A higher value of lambda will suppress Gibbs ringing more. However, it should be noted that a high value of lambda will also reduce the level of high-frequency information in the processed image, and therefore it is important to find a balance between reducing Gibbs ringing and preserving image quality *Recommendation*: 100 (Default setting)
Number of iterations ([1200])	The number of iterations is another parameter that controls the degree of Gibbs ringing correction *Recommendation*: 100 (Default setting)
Step size ([0.001–0.1])	The step size determines the magnitude of the update applied to the image estimate at each iteration of the algorithm. A smaller step size will result in a slower convergence of the algorithm and less Gibbs ringing, while a larger step size will result in a faster convergence but more Gibbs ringing. The optimal value is a desired trade-off between Gibbs ringing reduction and computational time *Recommendation*: 0.01 (Default setting)
Imaging plane (coronal:1, sagittal:2, axial:3)	The Gibbs ringing correction algorithm takes into account the imaging plane in which the image was acquired. This information is found in the subject specific *.json file, Phase Encoding Directions **i** left-right (sagittal) **i**- right-left (sagittal) **j** anterior–posterior (axial) **j**- posterior–anterior (axial) **k** inferior-superior (coronal) **k**- superior–inferior (coronal)

The next processing step requires each individual image to have matching .nii and .txt file names. Thus, rename *****_sdc_sorted.txt_ files in the previous folder to match the current .nii file (e.g. *****_sdc_sorted_GR_TV_FP.txt_) and create a new folder containing matching .txt and .nii files (*see* Appendix 5.2, **steps 3–7** for an example of how to batch rename files).

2.3.6 Generate .mat File (Step 6)

It is required to generate a .mat file from the processed .nii and corresponding .txt files before tractography or other analysis tools can be applied. The DTI .mat file is a MATLAB format file and can be loaded into ExploreDTI for further processing and analysis.

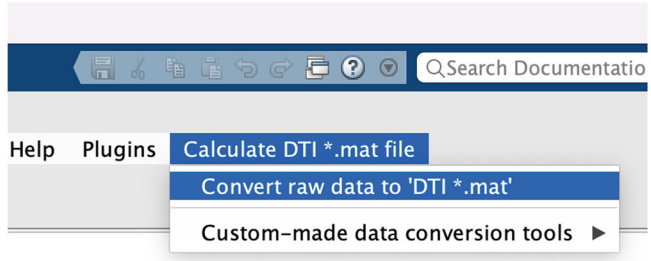

Fig. 7 Step 6 using the ExploreDTI GUI

In ExploreDTI:

1. Calculate DTI *.mat file → Convert raw data to 'DTI *.mat' (*see* Fig. 7)
   (a) Select settings (*see* Table 4).
   (b) Select the folder of *.nii.
   (c) Select folder .txt files: Press cancel if each .nii has its associated .txt file.
   (d) Select output folder.

The output folder now includes *_sdc_sorted_GR_TV_FP.mat files.

Note
A common pitfall of dMRI processing is orientation issues. During the .mat generation step you should investigate the flip/permutations to ensure appropriate orientations were selected. It is advised that you first use the default settings as ExploreDTI is able to automatically provide the correct orientation settings [31]. ExploreDTI deploys the widely used color convention to ensure the orientations ("Permute gradient components") are correct (left-right: Red, top-bottom: Blue, and front-back: Green). Good tracts to investigate when checking orientations are the corpus callosum—a white matter tract that is orientated left-right (Red) and the corticospinal tracts—white matter tracts that are orientated top-bottom (Blue). To see an example of orientation checks *see* Figs. 8 and 9.

Before-and-after correct flipping of the "Permute gradient components." As you can see in Fig. 8a, while the corticospinal tract is the correct orientation (blue arrow) the corpus callosum (red arrow)—a white matter tract that is orientated left-right—is

Table 4
Selecting .mat generation parameters

Parameter	Comment
Format diffusion weighted data	4D Nifti (*.nii)
Permute spatial dimensions	This allows you to flip spatial dimensions of image. *Recommendation*: Use default settings (AP RL IS) if there are no issues with spatial dimensions
Flip spatial orientations	This step allows you flip the direction of the dimensions. This is important if your data was collected in Neurological dimensions rather than Radiological conventions[*see* Glossary]. In this instance, you may need to flip dimensions from Right—Left to Left—Right. To flip, change the parameter from "AP RL IS" to "AP LR IS." If your data were collected in radiological dimensions the default setting of "AP RL IS" should be appropriate
Perform visual data check	This allows you to quickly visualize the orientation of the image
Diffusion tensor estimation	The robust tensor estimation algorithms aim to minimize the impact of outliers on the final diffusion tensor estimate, leading to more reliable results *Recommendation*: Robust (exclude outliers)
Format diffusion information	Text file (*.txt)
Background masking approach	Automatic
Permute gradient components	Permute gradient components should correspond to data and may require some investigation (*see* Note below this table)
Flip sign of gradient components	The sign of gradient components should correspond to data and may require some investigation (*see* Note below this table)
Data processing mode	Single or multiple data sets
b-value in units s/mm^2	E.g., 1000
Voxel size [AP RL IS] (in mm)	E.g., 2 2 2
Number of non-DW images	E.g., 3
Number of DW images	E.g., 30
Matrix size [AP RL IS]	E.g., 128 128 60

green. This indicates that the x and y axis need to be flipped. To do so, change the "Permute gradient components" from $x\,y\,z \rightarrow y\,x\,z$ and generate a new correctly orientated .mat file (Fig. 8b).

Figure 9 illustrates an investigation into the flip sign gradients. While the orientations of the images of both images are correct, the gradient sign may be flipped. Use Glyphs [*see* Glossary]

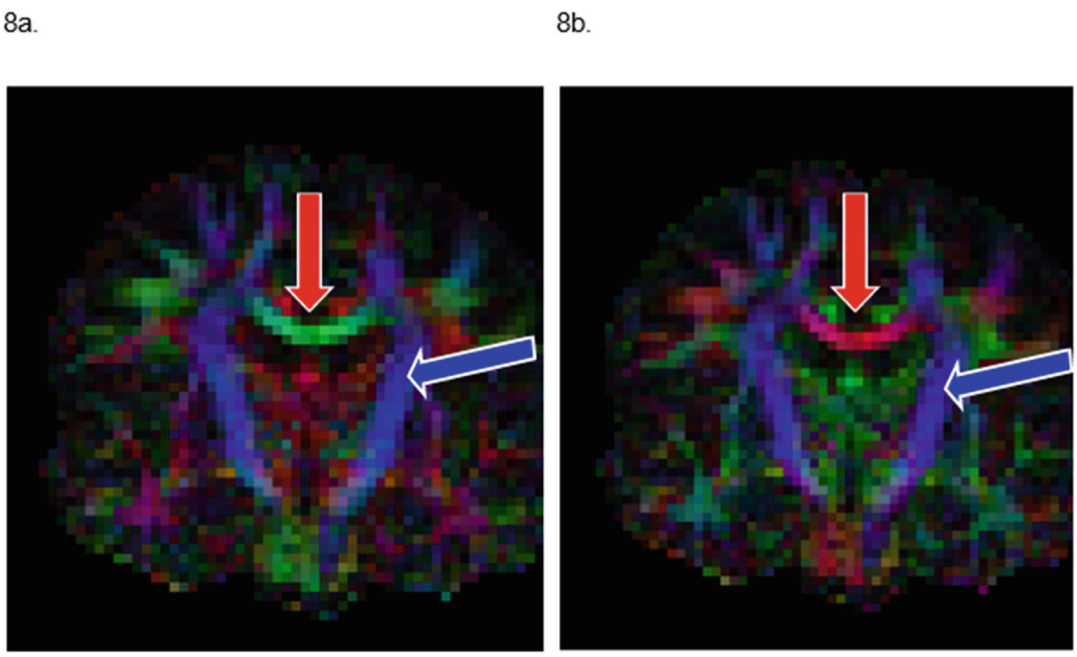

Fig. 8 Checking Permute Gradient Components

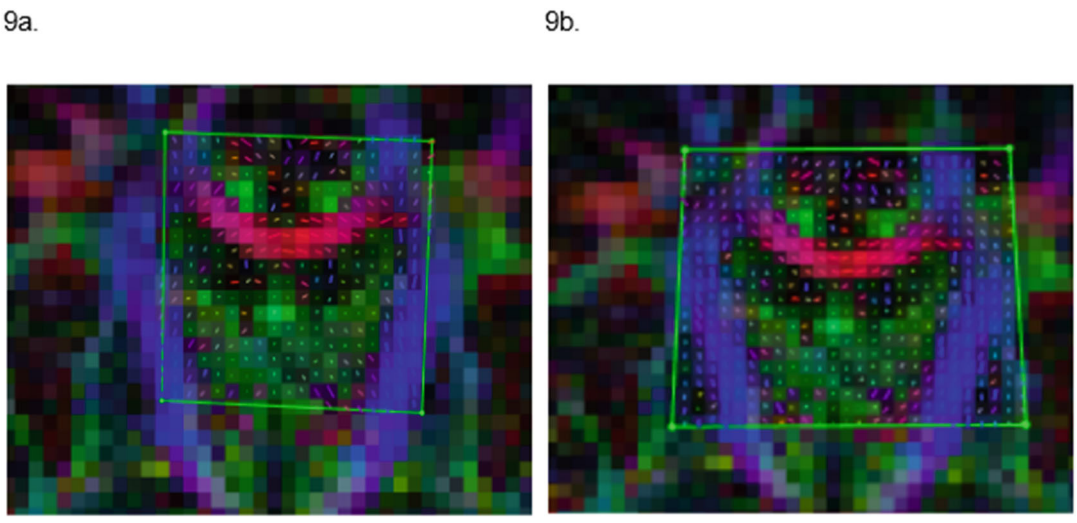

Fig. 9 Checking Flip Sign Components

(in ExploreDTI: *Draw ROI* → *Draw Glyphs*) to inspect the signs and investigate the "Flip sign of gradient components." Figure 9a shows an incorrect flip sign gradient as the glyphs are not following the curvature of the Corpus Callosum. To fix this, the z component must be flipped. To do so, change the "flip sign gradients" from *x y z* → *x y -z* and generate the correct .mat file (Fig. 9b).

Fig. 10 b-value folders

2.3.7 Concatenate All b-Value .mat Files (Step 7)

This step concatenates all the single b-values (shells) .mat files together to create a multi-shell .nii file. This enables a major benefit of multi-shell imaging; namely, leveraging the increased signal of high b-value images with the reduced noise of low b-value images to produce an image with increased anatomical accuracy.

Firstly, you should organize all your b-value .mat files into scan-specific folders (*see* Fig. 10).

In ExploreDTI:

1. Plugins → Concatenate DTI *.mat files (to *.nii) (*see* Fig. 11)
   (a) Select folder of folders: select the folder containing all the scan-specific folders.
2. The output folder now includes:
   (a) *_concatenated.txt file(s).
   (b) *_concatenated.nii file(s).

2.3.8 Generate Concatenated .mat File (Step 8)

It is now required that you convert the concatenated .nii files into .mat files. As any orientation issues should have been resolved at **step 5** (*see* Subheading 2.3.5), default orientation settings will be used. For processing efficiency, it is advised that you move *_concatenated.nii and *_concatenated.txt files into a folder.

In ExploreDTI:

1. Calculate DTI *.mat file → Convert raw data to 'DTI *.mat' (*see* Fig. 7)
   (a) Select settings (*see* Table 5).
   (b) Select the folder of *_concatenated.nii files.
   (c) Select folder *_concatenated.txt files: Press cancel.
   (d) Select output folder.
2. The output folder now includes:
   (a) *_concatenated.mat file(s).

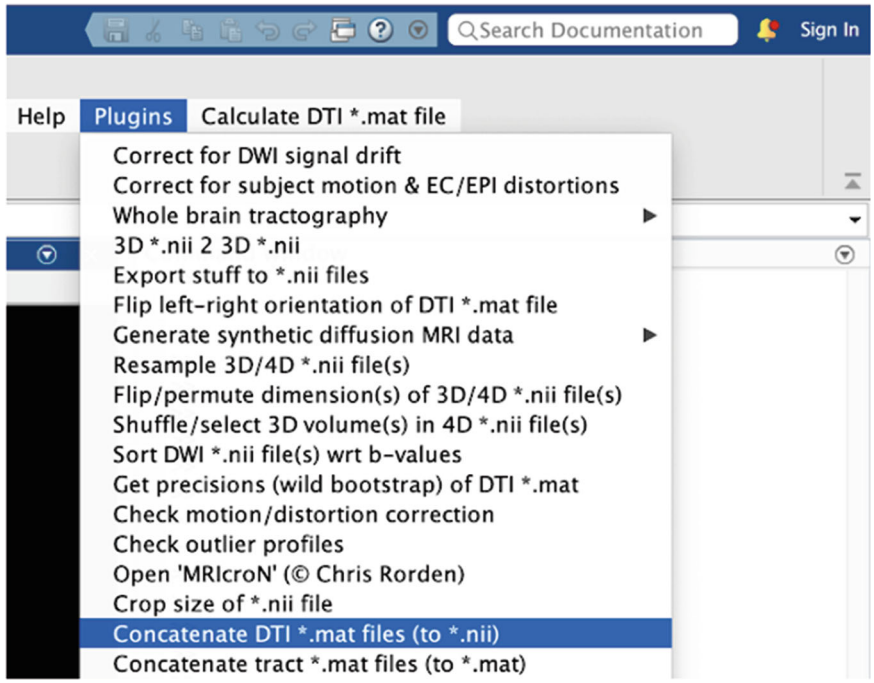

Fig. 11 Step 7 using the ExploreDTI GUI

2.3.9 Correcting Subject Motion, Eddy Currents, and EPI-Induced Distortions (Step 9)

This step corrects subject motion (SM), eddy currents (EC), and EPI-induced distortions (EPI). This is a crucial processing step, as such distortions can lead to significant changes in diffusion metric estimates. Additionally, you can use "undistorted" structural MRI (T1 or T2) images to unwrap the deformations in the diffusion data (For more details see ExploreDTI manual). If you do not have a structural MRI, this step can be conducted in native space. To process this step in ExploreDTI without a structural MRI file ensure the following setting is selected, *Settings → SM/EC/EPI correction → Also register to other data → No thanks (stay in native space)*. Before beginning this step move *_concatenated.mat files (and *_nu.nii and *_mask.nii files if necessary) to a folder.

> **Note**
> Due to the large computational demand of **step 9**, it is recommended to use multi-core computing support for this tool with a minimum of 32 GB RAM for dMRI data if you have more than 100 DW images.

Table 5
Selecting .mat generation parameters

Parameter	Comment
Format diffusion weighted data	4D Nifti (*.nii)
Permute spatial dimensions	AP RL IS
Flip spatial orientations	AP RL IS
Perform visual data check	No.
Diffusion tensor estimation	Robust (exclude outliers)
Format diffusion information	Text file (*.txt)
Background masking approach	Automatic
Permute gradient components	x y z
Flip sign of gradient components	x y z
Data processing mode	Multiple data sets
b-value in units s/mm^2	NaN (for multi-shell data) or any integer for DTI
Voxel size [AP RL IS] (in mm)	E.g., 2 2 2
Number of non-DW images	The total number of b-0 images in the concatenated images (using the parameters in NICAP study, e.g., number of non-DW images = 16)
Number of DW images	The total number of b-value images in the concatenated images (using the parameters in NICAP study, e.g., number of DW images = 130)
Matrix size [AP RL IS]	E.g., 128 128 60

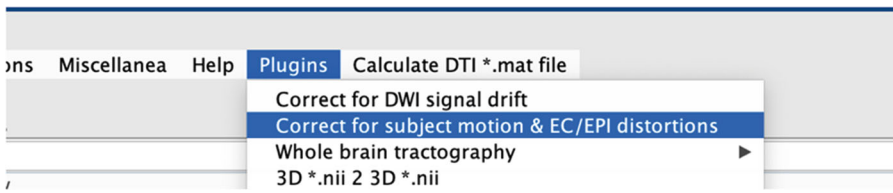

Fig. 12 Step 9 using the ExploreDTI GUI

Table 6
Selecting SM/EC/EPI distortion parameters

Parameter	Comment
Settings → SM/EC/EPI correction → masking stuff	This setting allows you to use a mask generated from a structural MRI scan. If you do not have a structural MRI mask, do not select the "masking stuff" setting
Settings → SM/EC/EPI correction → also register to other data → yes, to do EPI correction (non-rigid)	This setting allows you to register your diffusion image to a structural MRI image during the EPI correction, enabling increased distortion correction. *Recommendation*: select "orig_nu" from Freesurfer processed structural MRI files
Settings → SM/EC/EPI correction → also register to other data → registration details → Deformation axes	By default, the non-linear deformations are allowed along any orientation. *Recommendation*: correction will likely improve if the registration is constrained to model deformations only along the phase encoding direction. To do this (example A-P orientation), change "Deformation axes" to [1 0 0]
Settings → SM/EC/EPI correction → registration details for SM/EC corrections → interpolation method	*Recommendation*: Linear or cubic spline

In ExploreDTI:

1. Select settings (*see* Table 6).
2. Start MATLAB parallel pooling.
3. Plugins → Correct for subject motion & EC/EP distortions (*see* Fig. 12)
   (a) Single or multiple data sets: Multiple.
   (b) Select the folder of *_concatenated.mat files.
   Include *_nu.nii and *_mask.nii for using structural MRI files for registration.
   (c) Select output folder.
   The output folder now includes *_concatenated_trafo.mat files.

2.3.10 Whole Brain Tractography (Step 10)

Whole brain tractography in ExploreDTI generates white matter tracts using a deterministic approach. Other software packages, such as FSL and MRtrix are available if you would like to do probabilistic tractography. It is recommended that you complete whole brain tractography before reconstructing specific white matter tracts for analysis.

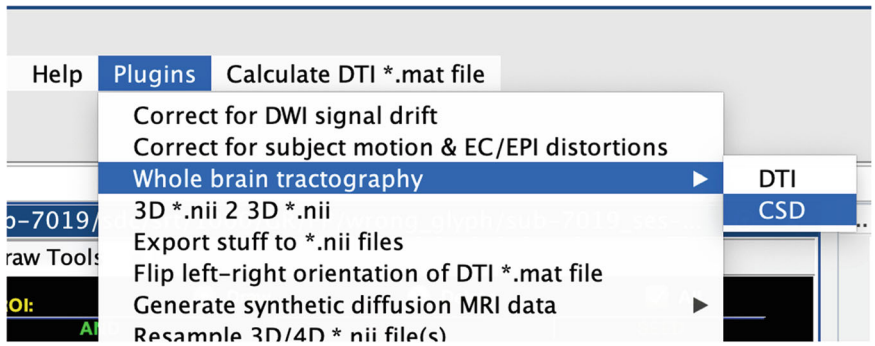

Fig. 13 Step 10 using the ExploreDTI GUI

Table 7
Whole brain tractography parameters

Parameter	Comment
Seedpoint resolution (mm)	Seed point resolution is a measure of how close together the seed points are placed in the brain. A higher seed point resolution will result in a higher number of seed points used in the tractography algorithm, and therefore a higher number of the reconstructed tracts. However, this also increases the computation time. *Recommendation:* 2 2 2
Step size (mm)	The step size is a parameter that determines the distance between each point in the reconstructed tracts. A smaller step size will result in a higher accuracy of the reconstructed tracts, but it will also increase the computation time. *Recommendation*: 1
Angle threshold	The angle threshold is a parameter that controls the angular deviation of consecutive steps during pathway reconstruction. A higher angular threshold will result in more or longer tracts, but it will also increase the risk of false positive tracts. If you are planning to exact tracts with high curvature (such as the fornix) it is advised to set this threshold higher (e.g., 60°)
Fiber length range	This step allows you to set the upper and lower bound of the length of the reconstructed fibers. Change this setting if you are investigating particularly long or short white matter tracts. If you are investigating both long and short fiber, it is recommended to set this setting to, 10–500
Random permutations of seed points	*0 = no/1 = yes (setting to get rid of rectilinear grid-pattern artifacts)* *Recommendation*: 0

In ExploreDTI:

1. *Plugins → whole brain tractography → CSD* (*see* Fig. 13)
   (a) *Select settings (see* Table 7*).*
   (b) *Single or multiple data sets:* multiple.
   (c) Select the folder of *_trafo.mat files.

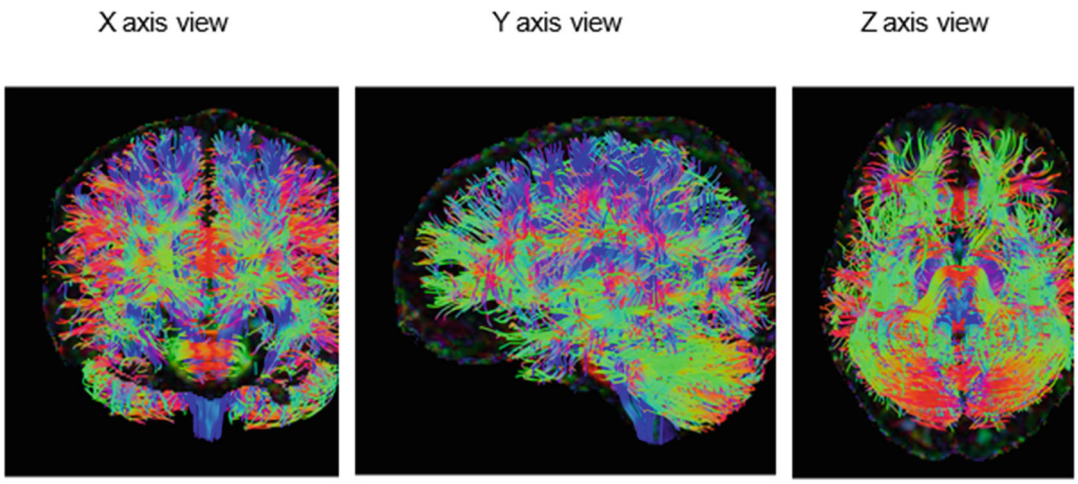

Fig. 14 Complete CSD tractography (subsampled: 50)

(d) Select output folder.

(e) The output folder now includes the *_trafo_Tracts_CSD.mat files (*see* Fig. 14).

2.3.11 Extracting Diffusion MRI Metrics (Step 11)

At this step, you should have already extracted the white matter tracts you want to analyze. A step-by-step guide of conducting manual tractography is provided in the ExploreDTI manual. This step allows you to export diffusion metrics for the analyzed tract pathway of interest (see Supplementary Material Table 2). If you wish to also obtain kurtosis measures, please see Appendix 5.3.

In ExploreDTI:

1. *Plugins → convert → info. of tract *.mat file(s) to .txt.* (*see* Fig. 15)
   (a) Select the folder of .nii.
   (b) Select output folder.
2. The output folder now includes:
   (a) *.txt files.
3. Export *.txt file to Excel.

3 Conclusion

This chapter offers a comprehensive resource that equips researchers with the necessary skills and knowledge to effectively and efficiently process large diffusion MRI data sets. By providing practical, step-by-step guides, researchers can process both DTI and multi-shell HARDI data using the ExploreDTI software. When choosing a diffusion MRI modeling technique, it is important to consider the

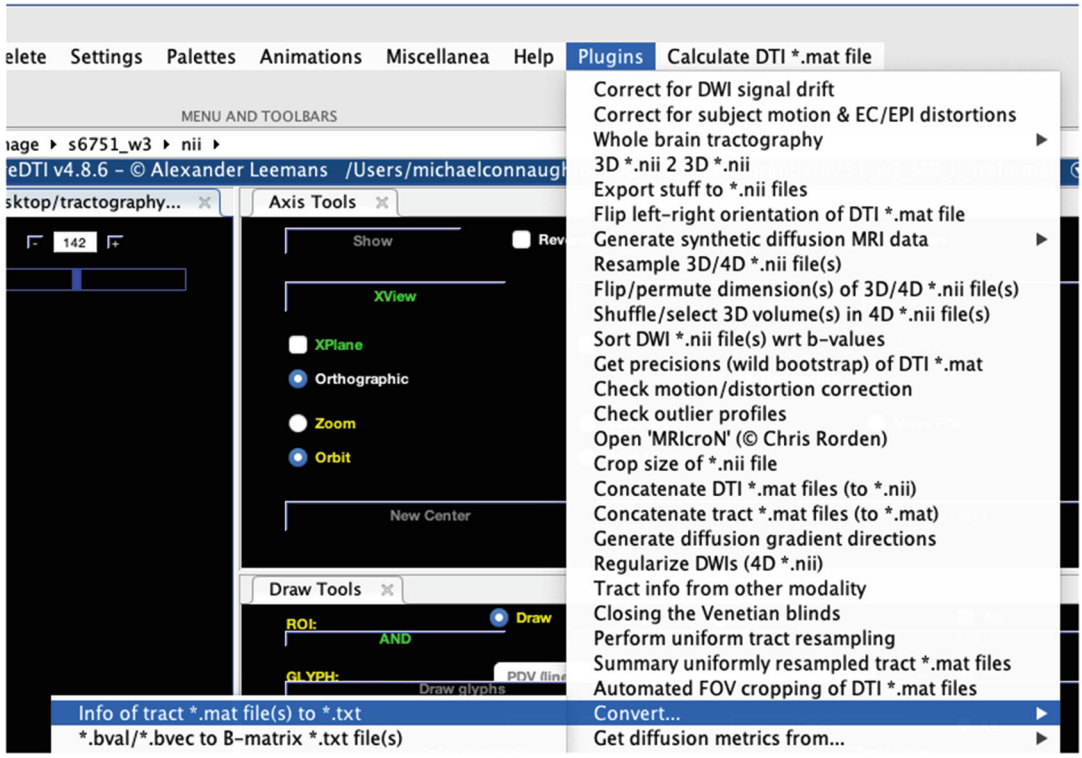

Fig. 15 Step 11 using the ExploreDTI GUI

pros and cons of both DTI and multi-shell HARDI approaches. Multi-shell HARDI offers several advantages over DTI, including increased anatomical accuracy and the ability to model crossing fibers. However, it also comes with certain disadvantages compared to DTI that warrant careful evaluation. A significant drawback of multi-shell HARDI is that it is a highly computationally expensive technique, which results in significantly longer processing times compared to DTI. Nevertheless, steps can be taken to reduce processing time through resource optimization. Researchers can optimize the utilization of computational resources by fine-tuning the processing parameters described in this book chapter. When processing large data sets, it is advised to experiment with different settings parameters to find the optimal balance between reconstruction accuracy and processing time. Overall, it is crucial to assess the accessible resources, both available time and computational resources, before deciding which diffusion MRI modeling technique to employ.

Appendix

4.1 Checking the Impact of Different Signal Drift Fit Approaches

The primary parameter to be considered during this step is the signal drift fit approach (Linear, Quadratic, or Cubic). The ExploreDTI default setting is "Quadratic" however, all three approaches should be investigated to find the best fit for your data set. The signal drift fit approach can be changed by in ExploreDTI, *Settings → Signal drift correction → Fit Approach* (1. Linear, 2. Quadratic, or 3. Cubic). Initially it is advised to run all three approaches (using the instructions below). After you have run all three different signal drift approaches, the *.png* files you be used to evaluate their performance. The approach that is most appropriate for your data set is the one with the least amount of signal loss (see Appendix Figure).

4.2 Data Storage and Computational Expense: Open Neuro Diffusion Data

The code provided here can be run on a single computer using MATLAB. The code was written using MATLAB version R2020a. While earlier versions of MATLAB may also execute the code successfully on a MacBook Pro with an Intel Core i7 processor and 16 GB RAM, it takes 10 min to execute the code for 10 participants with MATLAB parallel processing enabled (*see* Table 1).

4.2.1 Step-by-step Guide of DTI Processing the Open Neuro Data

Step 1: *Convert Bval and Bvec Files into Text Files*

```
d = pwd;


dic_folder =
'/Users/michaelconnaughton/Desktop/Neuroimaging_Book_Chapter/OpenN
euro/ds002790-master/dwi_files/';


if ~ischar(dic_folder)
    return;
end
```

```
files = E_DTI_Get_files_from_folder(dic_folder, '.bval');


if isempty(files)
    uiwait(my_msgbox('No *.bval files found...','Converting
*.bval/*.bvec file(s) B-matrix *.txt file(s)','modal'));
    return;
end
```

Step 3–7: *Signal Drift, Gibbs Ringing, Flip Permute, and Generate Mat File*

```
h_w = my_waitbar(0,'Converting *.bval/*.bvec file(s) B-matrix
*.txt file(s)');pause(0.01)


for i=1:length(files)

    E_DTI_convert_nii_dic_2_txt_exe(files{i});


    my_waitbar(i/length(files))
end


close(h_w);
```

```
d = pwd;


dic_folder =

'/Users/michaelconnaughton/Desktop/Neuroimaging_Book_Chapter/OpenN

euro/ds002790-master/dwi_files/';


if ~ischar(dic_folder)

    return;

end


files = E_DTI_Get_files_from_folder(dic_folder, '.bval');


if isempty(files)

    uiwait(my_msgbox('No *.bval files found...','Converting

*.bval/*.bvec file(s) B-matrix *.txt file(s)','modal'));

    return;

end


h_w = my_waitbar(0,'Converting *.bval/*.bvec file(s) B-matrix

*.txt file(s)');pause(0.01)
```

```
for i=1:length(files)

    E_DTI_convert_nii_dic_2_txt_exe(files{i});

    my_waitbar(i/length(files))
end
```

```
d = pwd;
folder_in =
'/Users/michaelconnaughton/Desktop/Neuroimaging_Book_Chapter/OpenN
euro/ds002790-master/dwi_files/';
if ~ischar(folder_in)
    return;
end

files_in = E_DTI_Get_files_from_folder(folder_in, '.nii');
if isempty(files_in)
    uiwait(my_msgbox('No DWI nii files found...','Signal drift
correction','modal'));
    return;
end
```

```
folder_out_nii = [folder_in filesep 'sdc'];
```

```
if ~isdir(folder_out_nii),mkdir(folder_out_nii);end

folder_out_txt = [folder_in filesep 'sdc'];

if ~isdir(folder_out_txt),mkdir(folder_out_txt);end

h_w = my_waitbar(0,'Processing');pause(0.01)

for i=1:length(files_in)

    [~,n,~] = fileparts(files_in{i});

    par.f_in_nii = files_in{i};

    par.f_in_txt = [folder_in filesep n '.txt'];

    par.f_out_nii = [folder_out_nii filesep n '_sdc.nii'];

    par.f_out_txt = [folder_out_txt filesep n '_sdc_GR_FP.txt'];

    par.bvalC = 1000;

    par.bv_thresh = 10;

    par.method = 2;

    par.masking.do_it = 0;

    par.masking.p1 = 5;

    par.masking.p2 = 1;

    par.show_summ_plot = 1;

    E_DTI_signal_drift_correction(par);

    my_waitbar(i/length(files_in))
```

```
    % Apply Gibbs ringing correction
    f_in = [folder_out_nii filesep n '_sdc.nii'];
    f_out = [folder_out_nii filesep n '_sdc_GR.nii'];
    p.NrB0 = 1;
    p.lambda = 100;
    p.iter = 100;
    p.ss = 0.01;
    p.ip = 3;
    E_DTI_Gibbs_Ringing_removal_with_TV_exe(f_in,f_out,p);

    % Apply flip/permute correction
    File_name_of_DWIs_input = [folder_out_nii filesep n
'_sdc_GR.nii'];
    File_name_of_permuted_flipped_DWIs = [folder_out_nii filesep n
'_sdc_GR_FP.nii'];
    p = [];
    p.suff = '_FP';
    p.permute = [1 2 3];
    p.flip = [0 0 0];
    p.force_voxel_size = [];
    E_DTI_flip_permute_nii_file_exe(File_name_of_DWIs_input, p,
File_name_of_permuted_flipped_DWIs);
    % Generate Mat File
    f_DWI= [folder_out_nii filesep n '_sdc_GR_FP.nii']; %file name
```

```
of the DWIs
    f_BM= [folder_out_txt filesep n '_sdc_GR_FP.txt']; %name of
the B-matrix
    f_mat= [folder_out_nii filesep n '_sdc_GR_FP.mat']; %file name
of the DTI output
    Mask_par.tune_NDWI = 0.7; % (rough range: [0.3 1.5])
    Mask_par.tune_DWI = 0.7; % (rough range: [0.3 1.5])
    Mask_par.mfs = 5; % (uneven integer)
    NrB0= 1;
    perm = 2;
    flip = 4;
    E_DTI_quick_and_dirty_DTI_convert_from_nii_txt_to_mat(f_DWI,
f_BM, f_mat, Mask_par, NrB0, perm, flip)



end


my_waitbar(1);close(h_w);pause(0.01);
% As per the main manuscript, it is recommended to run a quality
check on the orientation of the mat files.
```

Step 7: *SM/EC/EPI Distortion*

```
% Download Parameters_SM_EC_EPI.txt on OpenNeuro
% Note: edit path folder_in and foulder_out of
Parameters_SM_EC_EPI.txt
% Name of the text file containing the parameters for the
SM/EC/EPI correction. The file is as saved in    the GUI using
Settings > SM/EC/EPI correction > Export parameter file.

parameter_filename =
'/Users/michaelconnaughton/Desktop/Neuroimaging_Book_Chapter/OpenN
euro/ds002790-master/dwi_files/sdc/Parameters_SM_EC_EPI.txt';

E_DTI_SMECEPI_Main(parameter_filename);
```

```
folder_in =
'/Users/michaelconnaughton/Desktop/Neuroimaging_Book_Chapter/OpenN
euro/ds002790-master/dwi_files/sdc/epi';
folder_out =
'/Users/michaelconnaughton/Desktop/Neuroimaging_Book_Chapter/OpenN
euro/ds002790-master/dwi_files/sdc/epi/wbt';
```

```
% Get a list of all files in the folder
file_list = dir(fullfile(folder_in, '*.mat'));

% Loop through each file and apply the code block
for i = 1:numel(file_list)
    n = file_list(i).name(1:end-10);
    filename_in = [folder_in filesep n 'native.mat'];
    filename_out = [folder_out filesep n '.mat'];

    parameters.SeedPointRes = [3 3 3];
    parameters.StepSize = 1;
    parameters.FAThresh = 0.2000;
    parameters.AngleThresh = 45;
    parameters.FiberLengthRange = [50 500];
    WholeBrainTrackingDTI_fast(filename_in, filename_out,
parameters);
end
```

4.3 Extracting Diffusion Kurtosis Metrics

```
% Define the path to the DTI and tract files

path_dMRI = 'path to diffusion file';

path_tract = 'path to tract file';

path_tract_new = 'Path to new output file';


% Get a list of all the dMRI files in the folder

dMRI_files = dir(fullfile(path_dti, '*_trafo.mat'));


% Loop through the dMRI files

for i = 1:length(dti_files)

    % Get the subject name from the file name

    subject = dti_files(i).name(1:end-8);


    % Define the input and output file names for the current
subject

    f_in_1 = [path_dMRI, dMRI_files(i).name];

    f_in_2 = [path_tract, subject,'_old_tract.mat'];

    f_out = [path_tract_new, subject,'_new_tract.mat'];


    % Call the script

    E_DTI_Add_DKI_metrics_to_tract_file(f_in_1, f_in_2, f_out);

end
```

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