Optimization of Macaque Brain DMRI Connectome by Neuron Tracing and Myelin Stain Data

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Abstract

Accurate assessment of connectional anatomy of primate brains can be an important avenue to better understand the structural and functional organization of brains. To this end, numerous connectome projects have been initiated to create a comprehensive map of the connectional anatomy over a large spatial expanse. Tractography based on diffusion MRI (dMRI) data has been used as a tool by many connectome projects in that it is widely used to visualize axonal pathways and reveal microstructural features on living brains. However, the measures obtained from dMRI are indirect inference of microstructures. This intrinsic limitation reduces the reliability of dMRI in constructing connectomes for brains. In this work, we proposed a framework to increase the accuracy of constructing a dMRI-based connectome on macaque brains by integrating meso-scale connective information from tract-tracing data and micro-scale axonal orientation information from myelin stain data. Our results suggest that this integrative framework could advance the mapping accuracy of dMRI based connections and axonal pathways, and demonstrate the prospect of the proposed framework in constructing a large-scale connectome on living primate brains.

Graphical abstract
Joint statistic matrix color-coded by TPs, FPs, FNs and TNs, which were obtained from the comparison between the connective matrices derived from tract-tracing database and dMRI data. The FVE brain map, the optimal brain map (out of the three brain maps in the present work) for global connectome, was used to construct connective matrices. The dMRI tractography connective matrix was constructed under the optimal tractography parameters (QA=0.2, Angle=70°) and binarized by the connective strength threshold corresponding to the maximal Youden index. Those parameters also apply to (b)–(i); (b) dMRI tractography fibers corresponding to the TP connections in (a). Fibers corresponding to the same connection have the same color; (c) The connective matrix color-coded by the average local $C$ values on the dMRI fiber tracts corresponding to the TP connections. Pink block highlights those connecting brain sites in frontal lobe; (d) Top 1% connections in term of their large average local $C$ values. Color blocks highlight some of these connections, and (e)–(i) show the corresponding dMRI fibers as well as the brain sites they connect. Dashed blocks indicate that those connections can find their symmetry counterparts in regions the solid blocks highlight. Interpretations of those blocks are found in the text.

**Keywords**
Tract-tracing; myelin stain; dMRI; multi-scale and multi-modal fusion; connectome

1 **Introduction**

Primate brains are among the most complex organs in the known world because they exhibit and behave as multi-scale networks, which consist of a huge number of structurally or functionally distinct components and a greater number of connections linking these component nodes (Bressler, 1995; McIntosh, 2000; Friston, 2002; Bullmore and Sporns,
Therefore, assessments of connectional anatomy of primate brains are critical to deep understanding of the structural organization of such brain networks and interpretation of their functional aspects (Chédotal and Richards, 2010; Johansen-Berg and Behrens, 2006; Passingham et al., 2002; Schmahmann and Pandya, 2007). To this end, a growing interest has been evoked in creating a map of the connectional anatomy over a large spatial expanse, also known as connectome (Sporns, 2005; Van Essen et al., 2013), accompanied by the initiation of numerous projects, such as the Human Connectome Project (http://www.humanconnectome.org/, Van Essen et al., 2013), the CONNECT project (Assaf et al., 2013), Allen Mouse Brain Connectivity Atlas (AMCA, http://connectivity.brain-map.org/, Oh et al., 2014), BRAIN project (http://www.nih.gov/science/brain/) (NIH, 2014; Tsien et al., 2013), Brainnetome Project (Jiang, 2013; Jiang, 2014) (http://www.brainnetome.org), and the Brain Decoding Project (http://braindecodingproject.org/). All these projects aim to precisely and comprehensively elaborate the brain networks’ structural organization as a whole, as well as the dynamic activities on this substrate.

Although sharing similar goals, these projects emphasize different methodologies and scales that fundamentally determine the ways for connectional anatomy assessments. For example, neuron tracing method provides trustworthy information of locations of afferent or efferent termini of neurons, such that it can be readily used to create a connective map at meso-scale (Jbabdi et al., 2013; Young, 1993; Felleman and Van Essen, 1991; Scannell et al., 1999; Burns and Young, 2000). Histology staining method provides direct in-plane neuronal and axonal morphologies at micro-scale (Budde and Annese, 2013; Leergaard et al., 2010). Along the same vein, connectivity between brain regions can also be assessed using a number of other techniques, such as MR-visible tract tracing methods (Wu et al., 2011) and electrical micro-stimulation methods (Tolias et al., 2005; Axer et al., 2011). However, unlike mouse brain databases such as the Allen Mouse Brain Connectivity Atlas (AMCA, http://connectivity.brain-map.org/), these ex vivo methodologies are impracticable for primate brains (especially human brains) due to ethical concerns (Bakker et al., 2012; Van Essen, 2013).

In addition to those invasive techniques, diffusion MRI (dMRI), a technique measuring restricted water diffusion in brain tissues (axonal bundles for most of the time), is one of the few non-invasive methodologies that are widely applied to primates (Van Essen, 2013; Van Essen et al., 2013; Assaf et al., 2013; Sporns, 2005). To derive a connectome from such a technique, local axonal pathway orientation is estimated for voxels based on diffusion models that are subject to a number of assumptions. Axonal pathways are then inferred by tracking those estimated orientations between predefined origins and targets (Basser et al., 1994; Mori et al., 1999; Tuch et al., 2002; Wedeen et al., 2005; Aganj et al., 2010; Uğurbil et al., 2013; Sotiriospolouls et al., 2013). This technique, also known as dMRI tractography, has been widely applied to reconstruct macro-scale fiber pathways of primate brains (Assaf and Pasternak, 2008; Bassett and Bullmore, 2009; Mori et al., 2008; Rilling et al., 2008; Li et al., 2013) and other mammalian ones (Calamante et al., 2012; Moldrich et al., 2010; Zhang et al., 2002). However, the reliability and validity of dMRI tractography have been questioned since its advent. Taking diffusion tensor imaging (DTI, Basser et al., 1994) for example, a tensor model of local diffusion is used. However, it has only one primary diffusion...
orientation to represent the estimated displacement profiles of water molecules in white matter under the assumption that axonal orientations within a voxel are highly anisotropic. This assumption is problematic at locations where axons cross, kiss and fan in individual voxels, and further result in spurious pathways (Jbabdi and Johansen-Berg, 2011). To enhance the accuracy of orientation estimation at these locations, great efforts have been made to improve data quality during acquisition, such as the image resolution (Uğurbil et al., 2013), and develop more advanced diffusion models and tractography methods, such as high angular resolution diffusion-weighted imaging (HARDI) (Tuch et al., 2002), diffusion spectrum magnetic resonance imaging (DSI) (Wedeen et al., 2005) and multi-shell dMRI (Aganj et al., 2010). In despite of these promising advancements in local diffusion estimation, reliability of dMRI tractography in precisely mapping primate brain connectomes is controversial (Jbabdi and Johansen-Berg, 2011; Jbabdi et al., 2013; Thomas et al., 2014; Leerberg et al., 2010; Donahue et al., 2016). For example, it is difficult to determine the starting or terminating locations of a tractography fiber. It is not clear whether a long pathway is a direct connection between two remote regions or a cascade of shorter ones (Jbabdi and Johansen-Berg, 2011).

In this work, we propose an integrative framework aiming to investigate whether the accuracy of dMRI based macro-scale connectome can be enhanced by adjusting tractography parameters according to evaluations from micro- and meso-scale structural measurements of the aforementioned neuron tracing data and histology staining data (Figure 1).

To achieve this goal, we construct large-scale connectome with a predefined cortical parcellation scheme based on dMRI data from normal adult macaque brains in vivo. Neuron tracing based connectome derived from the CoCoMac database (Bakker et al., 2012) with the same cortical parcellation scheme (the left branch in Figure 1) and axonal orientations estimated from myelin stain data (Mikula et al., 2007) (the right branch in Figure 1) are used as references to provide evaluations to dMRI derived connections and pathways (Figure 1). These quantitative evaluations from the two modalities constitute integrative scores to tractography parameters such that connectomes based on dMRI are optimized by selecting the parameters with the best score (the middle branch in Figure 1). It is worth noting that many dMRI tractography methods are available (e.g., Jiang et al., 2006; Behrens et al., 2007; Yeh et al., 2010), but comparison between these state-of-the-art dMRI tractography methods is not our major concern. This work only focuses on proposing a framework, and we thus select one of these methods (Yeh et al., 2010) as an example to present how to evaluate and optimize its tractography parameters. It is easy to apply this framework in a similar way to other dMRI tractography methods. We envision that by means of micro- and meso-scale measurements, the assessment of the macro-scale connectome based on dMRI tractography could be refined.

2 Materials and Preprocessing

2.1 Database

**MRI dataset:** Both T1-weighted MRI and DTI scans were conducted on twenty macaques under institutional animal care and use committee (IACUC) approval. Imaging
parameters for T1-weighted MRI data used were as follows: repetition time/inversion time/echo time of 2500/950/3.49 msec, a flip angle of 8°, a matrix of 256×256×192, and resolution of 0.5×0.5×0.5 mm³, with 3 averages. DTI data: A dual-spin-echo technique combined with bipolar gradients was used to minimize eddy current effects. Diffusion-weighted gradients were applied in 60 directions with a \( b \) value of 1000 sec/mm²; repetition time/echo time of 7000/108 msec, field of view of 141×132 mm², matrix size of 128×120, resolution of 1.1×1.1×1.1 mm³, 43 slices covering the whole brain. Diffusion-weighted images were acquired with phase-encoding directions of opposite polarity (left–right), each with 4 averages, to correct for susceptibility-induced distortion. For each average of diffusion-weighted images, 5 images without diffusion weighting \( (b = 0 \text{ s/mm}^2) \) were also acquired with matching imaging parameters.

**Caret dataset:** This dataset includes the macaque ‘F99’ atlas (F99 space was introduced in Van Essen, 2002) and template surface and volume image (T1-weighted, 0.5 mm resolution) in the atlas space to which parcellation schemes, also termed brain maps, from 15 published studies have been mapped (Van Essen, 2004). Of these published studies, BA (Brodmann, 1909), LVE (Lewis and Van Essen, 2000), and FVE (Felleman and Van Essen, 1991) were used in this paper, because they have different spatial resolutions that allow us to study the impact of parcellation resolution on dMRI connectome creation.

**CoCoMac database:** This database is established based on 40,000 experimental findings on macaque anatomical connections (Stephan, 2013; Bakker et al., 2013). We retrieved wiring information from those collated reports and constructed a meso-scale tract-tracing connective matrix based on the abovementioned parcellation schemes.

**Myelin stain database:** 36 coronal slices from one adult macaque brain are available in http://brainmaps.org (Mikula et al., 2007). Those slices cover the whole brain and the Weil’s stain method has been applied to those slices. In this database, the myelin sheaths are stained dark blue and blood cells are stained brown. The in-plane resolution is 0.46 micron/pixel. The section thickness is 40 microns. Each slice has a stack of images with different resolutions. The original images are at the bottom level with the image size of 80,000×100,000 in pixel. The images in an upper level is a down sampled version of those in the neighboring lower level, with 2 as the down-sampling factor.

### 2.2 Preprocessing

**MRI Data:** DMRI data was skull stripped using FSL-BET (Smith, 2002) and corrected for eddy current distortions using FSL-FDT toolkit (http://fsl.fmrib.ox.ac.uk). On the corrected data, an advanced diffusion model, ball-and-sticks (B&S) model, was adopted to estimate local diffusion displacement profiles for individual voxels (Behrens et al., 2003). Currently, two fibers were modelled per voxel, because it is suggested that \( b \)-values upwards of 4000 are required to resolve a three-fiber-orthogonal system robustly (Behrens et al., 2007). Based on this voxel-wise diffusion model, DSI studio was used for streamline tractography (Yeh et al., 2010). Of all tractography parameters, quantitative anisotropy (QA) and angular threshold served as critical termination criteria. QA is similar to fractional anisotropy (FA) which is used as the index for the filter to determine the fiber threshold in diffusion.
tensor model where only one diffusion orientation is estimated. QA in DSI Studio works for models of multiple diffusion orientations. It is defined as the amount of anisotropic spins of multiple orientations that diffuse along the fiber orientation (Yeh et al., 2010). We manipulated these two parameters while using default configurations for the others. For T1-weighted MRI data, we conducted skull removal via FSL-BET (Smith, 2002) and tissue segmentation via FSL-FAST (Zhang, et al., 2001). DMRI data was used as intra-subject standard space. Co-registration was performed to align T1-weighted MRI data to dMRI space. We used FSL-FNIRT (Andersson et al., 2010) to nonlinearly register T1-weighted MRI data to FA map to mitigate the misalignment between the two modalities. White matter surfaces were reconstructed on the registered T1-weighted MRI data via the methods in Liu et al., 2008.

**Myelin stain data:** Because the staining was performed on each slice individually, we first reconstructed a 3D image stack from the staining slices via a rigid body 2D image co-registration method (Lu et al., 2008) to minimize the zig-zag effect across sections (Figure S4). The alignment was performed on the top-level images with the lowest resolution. Image of slice #22 was selected as a starting reference. Images from the neighboring slices (#21 and #20) were registered to the reference via the method in (Lu, et al., 2008). Next, we registered slice #19 and #22 to the warped slice #21 and #20, respectively and repeated this step for all the other slices. For each slice, the transformation matrix resulted from the top-level image registration was applied to the other levels with finer resolutions accordingly, so that the local axonal orientations estimated in the bottom-level images were also aligned. As dMRI data was used as the intra-subject standard space, we adopted FSL-FNIRT (Andersson et al., 2010) to register myelin stain images to the FA map (Figure S5). The local axonal orientations estimated in the bottom-level images were transposed to dMRI space, accordingly.

**Caret template data:** ‘F99’ atlas in Caret dataset was transposed to individual space by registering T1-weighted MRI data of the atlas to individual T1-weighted MRI data via FSL-FNIRT (Andersson et al., 2010). All the brain maps, BA (Brodmann, 1909), LVE (Lewis and Van Essen, 2000) and FVE (Felleman and Van Essen, 1991) in ‘F99’ atlas (Van Essen, 2004) were transposed to individual spaces, accordingly. As a result, connective matrices derived from CoCoMac database and dMRI data could be compared in the same space.

It is noted that the relatively stereotyped macaque brain structure has very little inter-individual variability in the connective pattern (Saleem et al., 2014; Webster et al., 1994) making it feasible to warp the atlas to individual spaces. The misalignment errors among data modalities (myelin stain data and T1-weighted MRI data) were mitigated via the aforementioned registration strategies (see Figure S4 and Figure S5 for investigation of registration errors). Finally, it is also important to note that most records in CoCoMac database are within a hemisphere and it is usually ambiguous on which hemisphere those experiments are performed. Therefore, we only used ipsilateral connections and streamlines obtained from the dMRI tractography in this work. Left hemisphere was used to present the results in the main text. Comparisons between the two hemispheres are presented in the Supplemental Materials.
3 Methods

3.1 Local Axonal Orientations on Micro-scale Myelin Stain Data

Micro-scale myelin stain axonal orientations were estimated on the bottom level images with the highest granularity. In practice, we divided the original image into image blocks of 256×256 in size. We identified myelin structures and estimated local axonal orientations for every block via methods as follows:

1) Color channel (step 1 in Figure 2):

The blood cells, myelin structures and background are brown, dark blue and white, respectively. We used support vector machine (SVM) method (Chang and Lin, 2011) to decompose an original myelin stain image block into three probabilistic color maps. In the training stage, we randomly selected 500 pixels as training samples and manually labeled them as ‘myelin’, ‘blood cell’ and ‘background’. RGB values were used as features to learn a classification model. In the testing stage, the model was applied to all pixels in image blocks to classify them into three color classes. Figure 2(e1) and (e2) illustrate the probabilistic maps of ‘blood cell’ class and ‘myelin’ class based on the color features. Cyan dots and red arrow heads highlight the locations of some typical blood cells and myelin structures. Mathematically, we used \( m(x) \) to denote the myelin probability for pixel \( x \) based on the color features in image block \( I(x) \).

2) Texture channel (step 2 in Figure 2):

Because stained myelin usually has a thin line shape, we adopted texture shape analysis to estimate the probability of myelin structure. We converted the color image block into a gray-scale one and applied Hessian matrix to filter the image to obtain the shape information:

\[
H(x) = \begin{bmatrix}
L_{xx}(x) & L_{xy}(x) \\
L_{yx}(x) & L_{yy}(x)
\end{bmatrix}
\]  

where \( H(x) \) is the Hessian matrix for pixel \( x \). We computed the eigen values of \( H(x) \) and used \( l_1(x) \) and \( l_2(x) \) (\(|l_1(x)| > |l_2(x)|\)) to denote them. Line shape descriptor was defined as follows:

\[
f(x) = e^{-\frac{(l_2(x)/l_1(x))^2}{2\sigma_r^2}} \cdot (1 - e^{-\frac{(l_2(x))^2}{2\sigma_s^2}})
\]  

where the first term describes the line shape. The texture of a pixel has a thin line shape when the ratio \( l_2(x)/l_1(x) \) is small. The second term describes the contrast of line structures to the image background.

The Hessian matrices \( H(x) \) s, the eigen values \( l_1(x) \) s and \( l_2(x) \) s, and shape descriptor \( f(x) \) s were computed on a series of smoothed images for the same pixel \( x \). The derivatives, \( L_{xx}, L_{xy}, L_{yx}, L_{yy} \),
$L_{yy}$ and $L_{xy}$ in Eq. (1), associated with the $i$th smoothing scale were obtained by convolving image with a Gaussian kernel of scale $\sigma_i$

$$L_i \ast o(x) = g(\sigma_i) \otimes I(x) \quad (3)$$

Therefore, we obtained a series of $f_i(x)$ values at different smoothing scales for each pixel $x$. From those $f_i(x)$s, the extrema $f_{\text{max}}(x)$ was used to represent the line shape probability of the pixel. An example of the probabilistic map of line shape is shown in Figure 2(d).

Finally, the product of the probabilistic maps of the two channels was used to yield the final myelin structure probabilistic map:

$$p(x) = f_{\text{max}}(x) \cdot m(x) \quad (4)$$

After thresholding the map $(x)$ by 0.05, we estimated the myelin structure orientations on this binary map (Figure 2(f)). The isolated components were extracted from the binary map and their orientations were computed. The orientation $\alpha$ measures the angle (in degrees ranging from $-90^\circ$ to $90^\circ$) between the $x$-axis and the major axis of the ellipse which has the same second-moments as the isolated component. The components having less than 20 pixels were discarded. The distribution of isolated components’ orientations for image block of Figure 2(b) is shown in (h). We used unimodal Gaussian model to fit the distribution. The expectation (e.g., 50 degrees in Figure 2(h)) of the distribution was defined as the primary orientation of myelin structures for the image block.

The reasons for identifying myelin structures based on the combination of the two probabilistic maps are two-folds: 1) dense blood cells might form a line shape. Therefore, using texture shape analysis alone might erroneously recognize those blood cells as myelin structures, leading to false positives; 2) it is difficult to infer myelin shape information on the color based probabilistic map alone.

In summary, for an image block of a myelin stain slice, we estimated one orientation. A myelin orientation map (Figure 2(i)) was finally obtained for the image slice that typically has around 100x100 – 300x400 blocks. Each block is analogous to a voxel in dMRI data, which contains estimated axonal pathway orientations. Therefore, this myelin orientation map slice has a comparable matrix size to that of dMRI coronal slice (128x120).

### 3.2 Connectivity Matrices on Meso-scale Tract-tracing Data and Macro-scale DMRI Data

We used $E$ to denote the tract-tracing derived meso-scale connective matrix. The nodes of the connective matrix were defined as the cortical ROIs, termed BrainSites, on each hemisphere under a parcellation scheme (or brain map). We used the online search wizard of the second edition of CoCoMac (Bakker et al., 2012, http://cocomac.g-node.org), which provides a full access to all tables and their nested dependencies of the database, to yield a connectivity matrix based on a predefined brain map. The element of the matrix is
connection strength pre-defined as ‘0’ (absent), ‘1’ (weak), ‘2’ (medium), ‘3’ (strong), ‘x’ (existing connection of unknown strength) and ‘–’ (no data). We took the ‘maximum’ option for the strength, such that the ‘largest’ connection strength found in multiple reports was used in the matrix. Details of nomenclature used to query CoCoMac can be found in Supplemental Materials.

For dMRI data, we used $\mathbf{A}$ to denote the macro-scale connective matrix derived from it. Similarly, the nodes were defined as the BrainSites in a brain map. Connection strength $a_{ij}$ was defined as the number of streamlines that pass through BrainSite $i$ and $j$ simultaneously. Within each subject, $\mathbf{A}$ was normalized by dividing by the total streamline number. An average connective matrix $\overline{\mathbf{A}}$ of all subjects was used to represent the group-wise information.

### 3.3 Investigation and Selection of Tractography Parameters

The tractography parameters were investigated and selected based on the agreement between tract-tracing connective matrix $\mathbf{E}$ and tractography connective matrix $\overline{\mathbf{A}}$ as well as the agreement between axonal orientations inferred from myelin stain and tractography streamlines across the entire brain.

#### 3.3.1 Agreement between dMRI tractography based and tract-tracing based connective matrices

The agreement between the two matrices, $\mathbf{E}$ and $\overline{\mathbf{A}}$ was estimated in terms of sensitivity and specificity (Seehaus et al., 2012; Iturria-Medina et al., 2011; Thomas et al., 2014). The motivation of using sensitivity and specificity in evaluating and estimating tractography parameters is to answer two questions: 1) to what extent are DTI tractography pathways coincide with stained axons? 2) to what extent can non-exist DTI tractography paths be trusted when there are no corresponding stained axons (Seehaus et al., 2012)? Because $\overline{\mathbf{A}}$ was regulated by tractography parameters such as angular value and quantitative anisotropy (QA), we performed comparisons between $\mathbf{E}$ and a group of $\overline{\mathbf{A}}$ Equation with different combinations of tractography parameters, and selected the one producing the best tradeoff between specificity and sensitivity (ideally, the maximum sensitivity without any decrease in specificity). Because $\mathbf{E}$ was used as a reference to compute the statistic measurements, we binarized it by assigning ‘1’ to a connection with its strength being 1 (weak), 2 (medium), 3 (strong) or $x$ (existing connection of unknown strength). Otherwise, we assigned ‘0’ to it. (The strength was 0 (absent) and – (no data)). Notably, unlike $\overline{\mathbf{A}}$s which are unable to distinguish afferent projections from efferent ones, $\mathbf{E}$ is not a symmetric matrix. For an $\overline{\mathbf{A}}$ we used thresholds ranging from 0 to the maximal connective strength to convert it to a series of binary ones by assigning zeros to elements if they were below the thresholds. Then, we computed the true positives (TPs), true negatives (TNs), false positives (FPs) and false negatives (FNs) (see the example in Figure 3(a)) and generated the receiver operating characteristic (ROC) curve, and finally used the maximal Youden’s index (sensitivity+specificity−1, denoted by $\gamma$) (Youden, 1950) to evaluate the performance of an $\overline{\mathbf{A}}$ given a tractography parameter combination.
3.3.2 Agreement between dMRI tractography streamlines and axonal orientation from myelin stain data—In brief, the comparisons between myelin stain data and dMRI data were performed with a dMRI tractography streamline as a basic unit. We scored points on a streamline by measuring to which degree their local orientations were in agreement with axonal orientations inferred from myelin data at the same locations. We used \((x)\) to denote a streamline at spatial location \(x\). As the orientation maps derived from myelin stain images and streamlines have been registered in the same space (see Figure 3(a)), we could find the locations where \((x)\) penetrated the slices. In Figure 3(c), we show a streamline that penetrates two adjacent slices at locations highlighted by black dots. At a penetrating location, as illustrated in Figure 3(d), we used the angle \(\beta(0° ≤ \beta ≤ 90°)\) between axonal orientation from myelin stain data and streamline orientation to measure the agreement between them. A measurement \(C\) was defined for all streamlines on a subject as follows:

\[
C = \sum_{m=1}^{M} \sum_{n}^{N_m} \frac{(90 - \beta_m(x_n))}{90} \quad (5)
\]

where \(M\) denotes the number of streamlines. We used \(N_m\) to denote the number of points on the \(m\)th streamline as different streamlines have different number of points. Since the myelin stain slices only contain in-plane information (\(x\)-\(y\) plane in Figure 3(b)), we also measured the angle between local streamline orientation and the slice plane for each point \(x\) (Figure 3(c)), denoted by \(\gamma(0° ≤ \gamma ≤ 90°)\), which measures the confidence level for \(\beta\). If \(\gamma = 90°\), that is the streamline perpendicularly penetrated a slice, the primary orientation of the axonal pathway will be perpendicular to this plane and \(\beta\) will have very low confidence level. Therefore, the whole brain measurement \(C\) was updated as follows:

\[
C = \sum_{m=1}^{M} \sum_{n}^{N_m} \frac{(90 - \beta_m(x_n))}{90} \cdot \frac{(90 - \gamma_m(x_n))}{90} \quad (6)
\]

where \(\frac{(90 - \beta_m(x_n))}{90}\) and \(\frac{(90 - \gamma_m(x_n))}{90}\) is the local measurement for point \(x\). Finally, an average measurement \(\bar{C}\) across subjects was computed to incorporate group-wise information. For the sake of convenience, we use \(C\) hereafter to denote the group-wise measurement. Similar to measurement \(Y\) in section 3.3.1, we measured \(C\) for every combination of angular thresholds and QAs.

3.3.3 Integrated evaluations of the data modalities and decision on tractography parameters—Given a pair of tractography parameters \((qa, angle)\), \(Y\) measures the overall agreement between dMRI tractography based and tract-tracing based connective matrices, while \(C\) measures the agreement between dMRI tractography streamlines and axonal orientations from myelin stain data. \(Y\) was defined in section 3.3.1 and \(C\) was defined in section 3.3.2. Therefore, we found the best combination of tractography parameters by taking both of the evaluations in our consideration.
\[
\arg\max_{\text{qa, angle}} (\lambda Y(\text{qa, angle}) + (1 - \lambda) C(\text{qa, angle})) 
\]

\(\lambda\) was used to balance the weights for the two measurements. \(\lambda = 0.5\) was used in this paper to give equal weights to them. The optimal parameters identified by Eq. 7 allow the dMRI induced macro-scale global connective matrix to maximally match the tract-tracing induced meso-scale one, and the streamlines to be locally coherent to micro-scale axonal orientations derived from myelin stain data.

It is noted that \(Y\) is susceptible to the selection of brain parcellation scheme whereas \(C\) is independent of it. Therefore, Eq. 7 is for one parcellation scheme. Comparison results among different brain maps are also reported in the result section.

4 Experimental Results

4.1 Evaluating Axon Identification Performance on Myelin Stain Data

We used the production of color channel and texture channel to identify myelin structures. To justify the performance, we manually selected 200 pixels and labeled them as ‘myelin’ or ‘non-myelin’, as the accuracy of myelin structure classification is of our major interest. We used probabilistic threshold 0 (we used 0 as the threshold in the following steps for the purpose of fair comparison) for ‘myelin’ class obtained based on SVM to separate the 200 pixels as ‘myelin’ and ‘non-myelin’ as well. The sensitivity and specificity of the classification are 0.83 and 0.81, respectively. It is seen that the values are not high and balanced. Similarly, after thresholding the probabilistic texture map obtained based on Gaussian model, the sensitivity and specificity for the texture channel alone are 0.90 and 0.71, respectively. It is seen that the specificity is relatively low as many ‘blood cell’ pixels were identified as ‘myelin’. The sensitivity and specificity are 0.92 and 0.86, which are improved by the production of the two maps, demonstrating that the effectiveness of the production which outperforms the result of either of the two channels alone.

4.2 Evaluating dMRI Tractography Parameters by Tract-tracing Connectome

Two key tractography parameters, QA and angular threshold, used to terminate fiber tracking were within the ranges of \(\{0.1, 0.2, 0.3, 0.4\}\) and \(\{10^\circ, 20^\circ, 30^\circ, 40^\circ, 50^\circ, 60^\circ, 70^\circ\}\), respectively. Their effects on the specificity and sensitivity of dMRI tractography were studied on a connectome base. Also, we used three brain maps, BA (29 BrainSites), FVE (58 BrainSites) and LVE (100 BrainSites) with different granularities, to study their impacts on sensitivity and specificity.

Instead of using ROC curves to illustrate the sensitivity and specificity for all combinations of tractography parameters, we used the maximal Youden’s indices (\(Y_s\)) of the ROC curves (Figure 4(a)–(c)). All \(Y_s\) values are reported in Table 1. It is found that for all brain maps, the optimal combinations of tractography parameters yielding the maximal (black arrow heads), are the ones having small QA values and large angular thresholds (QAs = 0.1 and
angle thresholds above 60° for the three brain maps). Therefore, there is no competition between parcellation resolution and tractography parameters.

We first studied the effects of QA and angular thresholds. Generally, a decreasing QA value results in an incremental growth in the true positive rate (TPR) and false positive rate (FPR), especially for FVE and LVE brain maps (Figure 4(e)). The $Y$ increases with a decreasing QA as well (Table 1). For angular threshold, it is found that larger angular threshold yields higher TPR. For example, the symbols for 70° (black dots in Figure 4(f)) of all brain maps are diagonally further from the origin than those for other angular thresholds, e.g., 10° represented by red diamonds. The statistics in the angular threshold panel in Table 1 clearly depicts this trend.

An intuitive interpretation of these findings is that lower QA value and larger angular threshold allow fiber tracking to proceed to locations where homogeneity of voxel-wise axonal orientations is low, and where two consecutive axonal orientations have large crossing angle. They increase the chance that a fiber connects two BrainSites and consequently give a rise to increasing sensitivity. However, they also increase the spurious fibers, and thus increase the false positives and consequently lead to decreasing specificity.

Taking the optimal $Y$ for FVE brain map for example (Figure 4(b)), it has relatively high sensitivity (TPR was about 0.6), whereas its specificity is low (FPR was around 0.4).

When studying the effect of brain map, we found that the disparity between the sensitivity and specificity was not improved when either coarser or granular brain maps were used. By superimposing the $Y$s of the three brain maps (Figure 4(d)) on each other, we can observe that the $Y$s of the brain maps having the most (LVE) and fewest (BA) BrainSites are both further away from the chance line than those of FVE. This result suggests that parcellation scheme with finer grid might not necessarily lead to more accurate dMRI tractography connectome in terms of sensitivity and specificity.

In summary, we investigated the effects of tractography parameters and brain maps on dMRI tractography connectome creation in reference to tract-tracing connectome. The implication is that a brain map with median granularity and tractography parameters of small QA threshold and large angular threshold tend to yield more accurate global dMRI tractography connectome.

### 4.3 Evaluating Tractography Parameters of dMRI Connections by Myelin Stain Data

Myelin stain data is more suitable for evaluating the performance of tractography parameters on a local scale. To convert local measurements to a global one, we devised $C$, a summation of all local evaluations. Similar to evaluations from tract-tracing data, we had one $C$ value for each combination of tractography parameters. It allows us to conduct a comparative study among tractography parameters.

Generally, $C$ value increases with QA and angular threshold (the middle matrix in Figure 5(a)). To intuitively interpret this finding, we compared the fiber bundles near posterior limb of the internal capsule (PLIC) derived from parameter combination of {QA=0.4, angular threshold=10°} (in Figure 5(d)), and the one of {QA=0.4, angular threshold=70°} (in Figure 5(d)).
5(e). **Loose** angular constraint (70°) allows this bundle to slightly curve (pink arrow head). Therefore, the local orientations along it match the myelin stain ones slightly better than those in Figure 5(d). This improvement is not significant because the fiber bundles are restricted within regions with high QAs but do **not** turn into regions with lower QAs, although sharp turns are allowed by the loose angular constraint. In contrast, although lower QA threshold in Figure 5(c) allows a few fiber bundles to enter more regions where their orientations are consistent with the myelin stain data (red arrow heads), relaxing QA constraint does tractography more harm than good because more spurious fibers (green arrow head) are also generated.

In summary, it is concluded that tractography benefit **more** from relaxing the angular constraint than the QA constraint.

### 4.4 Optimal Tractography Parameters for dMRI Connectome

In the previous two sections, we evaluated the performance of tractography parameters from the viewpoint of global connectome as well as the one of local coherence. Importantly, for each combination of parameters, these evaluations are respectively summarized by two global measurements ($\mathbf{Y}$ and $\mathbf{C}$), and integrated in one evaluation framework. As illustrated in Figure 5(a), the integrative evaluation is a summation of the two measurements after normalization (subtracted the mean and divided by the standard deviation) within each evaluation matrix to remove the bias. Finally, the decision on the optimal tractography parameters (QA=0.2 and angular threshold=70°) is made by finding the maximum in the combined scores. It is noteworthy that we use the $\mathbf{Y}$ matrix of FVE brain map in Figure 5(a) because it outperforms the other ones. Because the $\mathbf{C}$ matrix is independent of the effects of brain maps, the summation of the $\mathbf{Y}$ matrix of FVE brain map and the $\mathbf{C}$ matrix still outperform the summations based on the other brain maps (the combined scores for all three brain maps are in Table S2–S4). This result was obtained on the left hemispheres because contralateral connection reports were hardly found in CoCoMac database and it is often ambiguous about which hemisphere these reports were made. **The results on the right hemispheres are** presented in Figure S6. Although there are a few differences between the final score matrices on two hemispheres, the final decisions on the optimal tractography parameters are the same. This is attributed to the similarity between the two hemispheres in terms of structural connections on a macro-scale (see the comparisons between connective matrices based on two hemispheres in Table S5).

### 4.5 Trustworthy Connections and dMRI Axonal Pathways

Due to the intrinsic limitations of the two abovementioned *ex vivo* data modalities, it is difficult to derive 3D axonal pathways from them. Alternatively, we can easily extract dMRI derived axonal pathways from the living brains. In the previous sections, we made the decision on the best brain map (out of the three brain maps in the present work) and the optimal tractography parameters for dMRI fiber bundles reconstruction and connectome creation. Therefore, we can identify the validated connections and the corresponding dMRI tractography pathways based on these results.
Specifically, we first reconstructed the whole brain dMRI tractography fiber bundles using QA = 0.2 and angular threshold = 70°, the optimal combination in Figure 5(a). Then, FVE brain map, the best brain map, was used to construct the dMRI connective matrix, which was comparatively studied with the tract-tracing one by analyzing the sensitivity and specificity, as introduced in section 3.3.1. A binary version of the dMRI connective matrix was obtained by applying the connective strength threshold associated with the maximal Youden index. This binary matrix is jointly presented with the tract-tracing one to give the joint statistic matrix in Figure 6(a). The true positive (TP) connections (white elements in Figure 6(a)) of this matrix as well as the corresponding dMRI fiber bundles (Figure 6(b), termed as TP connection pathways), are those validated by the tract-tracing datasets. For each TP connection, the local C measurements on its corresponding dMRI fibers’ points were averaged and superimposed, such that the evaluations from the myelin data were integrated to the TP connections (Figure 6(c)). The local C measurement was defined as \( \frac{90 - \beta_m(s_n)}{90} \times \frac{90 - \gamma_m(s_n)}{90} \) in section 3.3.2. We used local C to distinguish it from the global measurement C 90 in Eq. (6). Therefore, this matrix in Figure 6(c) can be used as a hybrid reference. Applying a threshold to the local C measurement yields the trustworthy connections and axonal pathways at the confidential level desired. We show an example of such usage in Figure 6(d) where the top 1% TP connections with the largest local C values (above the average plus three standard deviations) are preserved. In fact, those top connections and the fiber bundles are the most trustworthy ones because they are both TP connections (validated by tract-tracing database) and those with high consistency with myelin stain data in terms of local axonal orientations. In this work, four major fiber streams were identified and supported by the tract-tracing reports: 1) pathways connected to occipital regions (Figure 6(e)) (Anderson et al., 1998; Rockland and Van Hoesen, 1994; Sincich and Horton, 2003; Yukie and Iwai, 1985); 2) pathways connected to somatosensory and motor cortices (Figure 6(f) and (g)) (Burton et al., 1995; Kunzle, 1978; Disbrow et al., 2003 for Figure 6(f); Tokuno and Tanji, 1993; Tokuno et al., 1997 for Figure 6(g)); 3) pathways connecting occipital lobe and temporal lobe (Figure 6(h) and (i)) (Nakamura et al., 2001; Webster et al., 1991; Webster et al., 1994 for Figure 6(h); Neal et al., 1990 for Figure 6(i)); and 4) pathways connecting inferior parietal lobe and temporal lobe (Figure 6(i)) (Barnes and Pandya, 1992). It is noted that Figure 6(d)–(i) only illustrate an example of the usage of the hybrid connective matrix in Figure 6(c). More trustworthy connections and fibers, such as those in prefrontal lobe (the pink block in Figure 6(c)), can be extracted if we lower the threshold for local C values. It is also noticed that those TP connections have been reported and validated in literatures collected and collated by CoCoMac database (according to the citation policy, the complete list of those reports were found in supplemental materials). However, in those reports, the 3D pathways of those connections were rarely available. Therefore, the presentation of those validated dMRI axonal pathways is one of the major contributions of this work.

The validity of the trustworthy connections can also be cross-validated by comparing the average local C measurements of the TP connections with the FP and FN ones (Figure 6(a)). Because the TP, FP and FN connections were obtained by the comparative study between dMRI data and tract-tracing dataset, using myelin data derived measurements to evaluate...
those connections is a cross-validation between the two non-macro-scale data modalities. On average, the local $C$ measurement for the TP, FP and FN connections are $1.44 \times 10^{-2}$, $0.91 \times 10^{-2}$ and $0.68 \times 10^{-2}$, respectively, indicating that TP connections have the highest local $C$ values. Results of the left-tail $t$-test between TP-FP and TP-FN pairs, under the null hypothesis that TP connections have lower local $C$ measurements than FP or FN ones at the 5% significance level, demonstrate the significance of the discrepancy ($p$-values for TP-FP and TP-FN pairs are $1.19 \times 10^{-7}$ and $1.03 \times 10^{-17}$, respectively). This result suggests that the ‘trustworthy’ connections validated by tract-tracing dataset are still ‘trustworthy’ in terms of local orientations on their pathways.

5 Discussion

In this work, we present a framework to enhance the accuracy of in vivo dMRI in inferring macro-scale connectome on macaque brains by means of meso- and micro-scale data modalities. In this framework, comparisons between dMRI and the two data modalities were quantified and integrated to provide integrative evaluations to dMRI tractography parameters. Because tract-tracing reports in CoCoMac database provide the meso-scale distribution of axonal termini while myelin stain data provides local axonal orientations, the optimal tractography parameters obtained are a tradeoff between the accuracies in global connective patterns and local pathway morphologies. Therefore, with these optimal parameters, the true positive (TP) connections with high evaluation scores from myelin stain data yield the trustworthy dMRI tractography fibers, which are validated by the two non-macro-scale data modalities simultaneously. These abovementioned results underscore the fact that the choice of tractography parameters is crucial to accurate estimation of brain connectome from dMRI. This statement is in agreement with the conclusions from other studies (Dauguet et al., 2007; Bastiani et al., 2012; Seehaus et al., 2013; Girard et al., 2014). However, in those works, only tract-tracing data on a few areas of interest was used as a reference to evaluate the performance of dMRI tractography. The conclusions may not be applicable to in vivo dMRI data (1–3 mm resolution for primate brains) that is more widely used by connectome projects (Van Essen et al., 2013; Assaf et al., 2013; Oh et al., 2014; NIH, 2014; Tsien et al., 2013; Jiang, 2013; Jiang, 2014). Therefore, the present work advances the investigation in three folds: 1) it evaluates the performance of dMRI in constructing a large-scale connectome and provides a better way to select tractography parameters and parcellation schemes; 2) it incorporates evaluations for one dMRI tractography method (Behrens et al., 2003; Yeh et al., 2010) from two non-macro-scale data modalities and is open for more data modalities and tractography models to be easily integrated in this framework; 3) it evaluates the performance of in vivo dMRI and the conclusions are applicable to large-scale data bases. Notably, the present work and the abovementioned related works used animal models. Inferences from these models could be limited due to the anatomical differences across mammal species. From the technique perspective, however, the dMRI and associated tractography are almost the same for these species (Rilling et al., 2008; Li et al., 2013; Hutchinson et al., 2017; Young et al., 2017).

Moreover, these mammal species share similar connectional anatomy and functional organization at a certain level (Song et al., 2014; Taso et al., 2008; Saleem et al., 2007). Therefore, studies on animal brains, especially those having ex vivo data, provide a unique
avenue and a valuable proxy to understand general principles of brain organization and make inferences for human brains, for which in vivo imaging technique is currently the only widely adopted approach.

The results from the present work suggest that the integration of meso- and micro-scale data would guide the selection of dMRI tractography parameters, and dMRI could serve as a more reliable tool for brain connectome estimation with the optimal parameters.

However, in the comparison between dMRI connective matrix and tract-tracing one, even the optimal tractography parameters still yield false positives and false negatives (Figure 4). The contradiction between the sensitivity and specificity are irreconcilable, because high sensitivity is always accompanied by low specificity (Figure 4). This agrees well with the aforementioned studies (Thomas et al., 2014; Chen et al., 2015), underlining the intrinsic limitations of dMRI. Advances in dMRI data acquisition, diffusion model and tractography algorithm design are not within the scope of the present work. We emphasize parameter selection based on the state-of-the-art techniques with the guidance of non-macro-scale data modalities. In this respect, our results suggest that diffusion anisotropy measure has a stronger impact on dMRI tractography accuracy than angular threshold. A lower diffusion anisotropy threshold tends to yield a higher sensitivity and specificity. As for brain maps, dMRI tractography performance favors neither coarse-grained parcellation nor fine-grained one. These suggestions on parameter selection are in line with the findings on mouse brain (Chen et al., 2015). As for the comparisons between dMRI and myelin stain in terms of agreement on local axonal pathway orientations, dMRI tractography accuracy gains more from relaxing angular threshold than doing diffusion anisotropy (Figure 5). Therefore, the final suggestion on parameter selection in the present work is a compromise among global connection based sensitivity and specificity and local orientation based accuracy. The performance of dMRI tractography could be underestimated due to the limitation of the two non-macro-scale data modalities used in the present work. The tract-tracing connective matrix is based on the reports available in CoCoMac database. It is difficult to assess if the connections in these reports completely cover the entire brain. To overcome this limitation, we set those undiscovered connections in tract-tracing based connective matrix as negative identity, such that only a few more false positives could be illusively introduced to dMRI tractography performance evaluation. The efficacy of the true positives and the associated axonal pathways, the major interest of this work, is not negated by this limitation. This type of false positives can be reduced when CoCoMac database is developed to perfection. A few false positives in the present analysis could also result from the fact that dMRI tractography does not differentiate afferent fibers from efferent ones. Therefore, the dMRI connective matrix is symmetric while the tract-tracing one is not. However, these asymmetric connections in tract-tracing matrix only take up around 4% of all connections and they hence lead to no large bias in the global connectome evaluation. Another issue is that, as mentioned in section 4.4, most connections in CoCoMac database are within a hemisphere. Although experiments on the two hemispheres yield similar results (Figure S6), the accuracy of contralateral connections derived from dMRI, such as commissure fibers, is not investigated in this work.
Currently, the myelin stain data used in the present work only includes coronal slices. The information of orientations orthogonal to the slices is missing. To overcome this limitation, Eq. (6) was carefully designed to minimize this impact. Sagittal and axial sections, and an algorithm integrating axonal information from myelin sections of multiple directions would provide a more accurate reference to evaluate dMRI tractography. In addition, only one axonal orientation was estimated from a myelin stain image block while the tractography fibers were based on the model that estimates multiple number of crossing fibers. Crossings of myelin structures were observable in some image blocks in spite of their ultra-high resolution. In fact, more than one axonal orientations could be estimated from the orientation histology of stained data blocks by means of Gaussian mixture model. However, the key limitation is the missing information of stained data on z-axis. Although we have introduced a confidence factor to reduce the impact (Eq. (6)), estimating multiple orientations from the stained data will complicate the confidence factor model and introduce more uncertainty and inaccuracy. Therefore, it will be a better future solution to develop a method that integrates stained slices from multiple orientations and estimates probabilistic 3D axonal orientation maps. On this basis, we could develop advanced approaches similar to those for diffusion MRI (Tuch et al., 2002; Wedeen et al., 2005) in the future to model the anisotropic local orientation profiles, and estimate more trustworthy multiple orientations from the integrated stained data, which would provide more solid guidance to tractography parameter tuning.

These abovementioned limitations are due to the imperfection of the available non-macro-scale data modalities, and have already been taken into consideration and taken care of as discussed above. Therefore, they do not invalidate the effectiveness of the framework proposed in the present work as well as the strength of dMRI, which is to date the only technique to visualize axonal pathways in living brains. Under the guidance of the non-macro-scale data, our framework shows prospect in advancing dMRI tractography usage for connectome assessment and a comprehensive connectional map construction as illustrated in Figure 6.

**Supplementary Material**

Refer to Web version on PubMed Central for supplementary material.

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### Highlights

- Performance of dMRI in constructing a large-scale connectome was evaluated by gold-standard data: neuron tracing data and myelin stain data.
- The presented framework incorporates evaluations from two gold standard data modalities and is open for more data modalities and tractography models to be easily integrated in this framework.
- With optimized tractography parameters and scales, we could substantially trust dMRI with more confidence.
Figure 1.
Flowchart of the present framework. The dashed arrow indicates our goal in the present work. The solid arrows, including those in left/right branch and the one in the middle branch, indicate the solution proposed to achieve the goal.
Figure 2.
Methods of identifying myelin structures and estimating myelin orientations. (a) A myelin stain image slice with the image block grid (green frames) superimposed on it; (b) an image block (256×256); (c) an enlarged view of the region highlighted by the blue frame in (b); (d) the probability map of myelin structure in terms of its texture; (e1) & (e2) the probability maps of blood cell and myelin structure in terms of their colors; (f) identified myelin structures based on the combination of myelin structure color map in (e2) and texture map in (d); (g) Identified myelin structure in the image block of (b); (h) a histogram of the distribution of myelin structure orientations estimated in image block of (b); (i) the image slice with estimated myelin structure orientations for its image blocks. Cyan dots in (c) and (e1): locations of blood cells; Red arrow heads in (c), (d), (e2) and (f): locations of myelin structures.
Figure 3.
(a) An example matrix that comprises connective information from CoCoMac tract-tracing database and dMRI tractography. White elements are the connections identified by both dMRI and tracer data, the true positives (TP). Yellow ones are the connections identified only by dMRI data, the false positives (FP). Red ones are the connections identified only by tracer data, the false negatives (FN). Black ones are the connections found neither in dMRI nor in tracer data, the true negatives (TN). FVE brain map was used to construct the matrix.

Both injection sites and labeled sites are brain sites in FVE brain map. Injection sites refer to the brain sites where anterograde/retrograde tracer is injected. Labeled sites refer to the brain sites where axonal projection source/target are found. In this example, we used a random connective strength to threshold the dMRI tractography connective matrix. The ROC curve was yielded based on a series of such matrices with different thresholds; (b) joint visualization of dMRI streamlines (white curves) and myelin stain slices which is color-coded by FA values. **X-axis: left-right. Y-axis: top-bottom. Z-axis: front-rear**; (c) a streamline is used as an example to illustrate how to measure the angle between its orientation and the myelin stain slice it penetrates; (d) the same streamline is used to
illustrate how to measure the agreement between its local orientation and myelin orientation. The short cylinders represent the orientation estimated from myelin stain image blocks, the same as the one in Figure 2(i). Blue arrow represents myelin structure orientation while black one represents the dMRI fiber orientation at this location.
The specificities and sensitivities of dMRI tractography parameter combinations based on different brain maps. (a)–(c) for each parameter combination, we used a unique symbol (the symbol map is in the top-right corner of (a)) to show the location of the maximal Youden index ($Y$) value, which was used as the representative of a ROC curve and the performance of the corresponding parameter combination. This experiment was executed on three brain maps, BA, FVE and LVE, differing by the growing BrainSite numbers, 29, 58 and 100. For each brain map, the black arrow head highlights the symbol of the parameter combination which yielded the largest $Y$. The tractography parameters corresponding to these largest $Y$s are shown beside them. (d) $Y$s from the three maps are superimposed on each other for comparison. The maximal $Y$s for the three maps are noted. Chance line (true positive rate = false positive rate) is gray. Other color lines, as references, are trend lines for the distribution of $Y$s of the three maps; (e) this column shows the effect of QA on the specificity and sensitivity of results from all dMRI tractography parameter combinations across the three brain maps. We use the same symbol for $Y$s who have the same QA value. For the sake of comparison, we use the elliptic shade to incorporate those symbols having the same QA value; (f) the effect of angular threshold on the specificity and sensitivity of results from all dMRI tractography parameter combinations across the three brain maps. For each brain map, we use the same symbol for those who have the same angular threshold. For
the sake of comparison, we use the elliptic shades to highlight the symbols having the largest and smallest angular values.
Figure 5.
(a) The final decision (right) on the optimal tractography parameters was made based on the integration of meso-scale measurements (left) and micro-scale measurements (middle). Original Y values and C values are shown in the left and middle matrices, respectively. They were normalized before the summation was made. Those matrices are color-coded by their normalized values. White boxes highlight the maximum values in each matrix. Yellow boxes highlight the combinations of tractography parameters which are used to derive fibers shown in (c)–(e); (b) the cylinders represent the axonal orientations estimated from one myelin stain slice. FA map of this slice was used as background; (c)–(e) fibers (yellow curves) were derived from the combinations of tractography parameters highlighted by yellow boxes in (a). Red and green arrow heads highlight the differences of fibers between (c) and (e). Pink arrow heads highlight the difference of fibers between (d) and (e). Those arrow heads are also shown in (b).
Figure 6.
(a) Joint statistic matrix color-coded by TPs, FPs, FNs and TNs, which were obtained from the comparison between the connective matrices derived from tract-tracing database and dMRI data. The FVE brain map, the optimal brain map (out of the three brain maps in the present work) for global connectome, was used to construct connective matrices. The dMRI tractography connective matrix was constructed under the optimal tractography parameters (QA=0.2, Angle=70°) and binarized by the connective strength threshold corresponding to the maximal Youden index. Those parameters are also applied to (b)–(i); (b) dMRI tractography fibers corresponding to the TP connections in (a). Fibers corresponding to the same connection have the same color; (c) The connective matrix color-coded by the average local C values on the dMRI fiber tracts corresponding to the TP connections. Pink block highlights those connecting brain sites in frontal lobe; (d) Top 1% connections in term of their large average local C values. Color blocks highlight some of these connections, and
(e)–(i) show the corresponding dMRI fibers as well as the brain sites they connect. Dashed blocks indicate the symmetric counterparts of the solid blocks. Interpretations of those blocks are found in the text.
Table 1.
Average $Y_s \times 10^{-2}$ for QAs, angular thresholds and brain maps.

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