Robust, fast and accurate mapping of diffusional mean kurtosis

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Abstract Diffusional kurtosis imaging (DKI) is a methodology for measuring the extent of non-Gaussian diffusion in biological tissue, which has shown great promise in clinical diagnosis, treatment planning, and monitoring of many neurological diseases and disorders. However, robust, fast, and accurate estimation of kurtosis from clinically feasible data acquisitions remains a challenge. In this study, we first outline a new accurate approach of estimating mean kurtosis via the sub-diffusion mathematical framework. Crucially, this extension of the conventional DKI overcomes the limitation on the maximum $b$-value of the latter. Kurtosis and diffusivity can now be simply computed as functions of the sub-diffusion model parameters. Second, we propose a new fast and robust fitting procedure to estimate the sub-diffusion model parameters using two diffusion times without increasing acquisition time as for the conventional DKI. Third, our sub-diffusion-based kurtosis mapping method is evaluated using both simulations and the Connectome 1.0 human brain data. Exquisite tissue contrast is achieved even when the diffusion encoded data is collected in only minutes. In summary, our findings suggest robust, fast, and accurate estimation of mean kurtosis can be realised within a clinically feasible diffusion-weighted magnetic resonance imaging data acquisition time.

eLife assessment

This paper proposes a valuable new method for the assessment of the mean kurtosis for diffusional kurtosis imaging by utilizing a recently introduced sub-diffusion model. The evidence supporting the claims that this technique is robust and accurate in brain imaging is solid; however, there is a need to include a summary of the clear limitations.

Introduction

Diffusion-weighted magnetic resonance imaging (DW-MRI) over a period of more than 30 years has become synonymous with tissue microstructure imaging. Measures of how water diffuses in heterogeneous tissues allow indirect interpretation of changes in tissue microstructure (Le Bihan and Johansen-Berg, 2012). DW-MRI has predominantly been applied in the brain, where properties of white matter connections between brain regions are often studied (Lebel et al., 2019), in addition to mapping tissue microstructural properties (Tournier, 2019). Applications outside of the brain have clinical importance as well, and interest is growing rapidly.

Generally, DW-MRI involves the setting of diffusion weightings and direction over which diffusion is measured. Whilst diffusion tensor imaging (DTI) can be performed using a single diffusion weighting,
a so-called b-shell, and at least six diffusion encoding directions (Le Bihan et al., 2001), other models tend to require multiple b-shells each having multiple diffusion encoding directions. Diffusional kurtosis imaging (DKI) is a primary example of a multiple b-shell, multiple diffusion encoding direction method (Jensen et al., 2005). DKI is considered as an extension of DTI (Jensen and Helpern, 2010; Hansen et al., 2013; Veraart et al., 2011b), where the diffusion process is assumed to deviate away from standard Brownian motion, and the extent of such deviation is measured via the kurtosis metric. Essentially, the increased sampling achieved via DKI data acquisitions allows more complex models to be applied to data (Van Essen et al., 2013; Shafto et al., 2014), in turn resulting in metrics of increased utility for clinical decision making.

Recent clinical benefits of using kurtosis metrics over other DW-MRI-derived measures have been demonstrated for grading hepatocellular carcinoma (Li et al., 2022b), prognosing chronic kidney disease (Liu et al., 2022), differentiating parotid gland tumours (Huang et al., 2022), measuring response to radiotherapy treatment in bone tumour (Guo et al., 2022b) and glioblastoma (Goryawala et al., 2023), identifying tissue abnormalities in temporal lobe epilepsy patients with sleep disorders (Guo et al., 2022a) and brain microstructural changes in mild traumatic brain injury (Wang et al., 2022), monitoring of renal function and interstitial fibrosis (Li et al., 2022a), detecting the invasiveness of bladder cancer into muscle (Li et al., 2022d), aiding management of patients with depression (Maralakunte et al., 2023), delineating acute infarcts with prognostic value (Hu et al., 2022), predicting breast cancer metastasis (Zhou et al., 2023), diagnosing Parkinson’s disease (Li et al., 2022c), amongst others reported prior and not listed here.

The routine use of DKI in the clinic has nonetheless lagged due the inability to robustly estimate the kurtosis metric (Veraart et al., 2011a; Tabesh et al., 2011; Kuder et al., 2012; Henriques et al., 2021). A known requirement for estimating kurtosis in DKI is to restrict the maximum b-value to 2000 – 3000 s/mm$^2$ for brain studies (Jensen et al., 2005; Jensen and Helpern, 2010; Poot et al., 2010), with the optimal maximum b-value found to be dependent on tissue type (Poot et al., 2010). This suggests that the traditional kurtosis model is less accurate at representing the diffusion signal at large b-values. Moreover, multiple b-shell, multiple direction high-quality DW-MRI data can take many minutes to acquire, which poses challenges for clinical imaging protocols involving a multitude of MRI contrasts already taking tens of minutes to execute. Reduction of DKI data acquisition times through parallel imaging, optimisation of b-shells and directions have been investigated (Zong et al., 2021; Heidemann et al., 2010; Zelinski et al., 2008), and DW-MRI data necessary for DKI analysis has been shown to supersede the data required for DTI (Veraart et al., 2011b). Therefore, an optimised DKI protocol can potentially replace clinical DTI data acquisitions without adversely affecting the estimation of DTI metrics.

For DKI to become a routine clinical tool, DW-MRI data acquisition needs to be fast and provides a robust estimation of kurtosis. The ideal protocol should have a minimum number of b-shells and diffusion encoding directions in each b-shell. The powder averaging over diffusion directions improves the signal-to-noise ratio (SNR) of the DW-MRI data used for parameter estimation. Whilst this approach loses out on directionality of the kurtosis, it nonetheless provides a robust method of estimating mean kurtosis (Henriques et al., 2021), a metric of significant clinical value.

Instead of attempting to improve an existing model-based approach for kurtosis estimation, as has been considered by many others, we considered the problem from a different perspective. In view of the recent generalisation of the various models applicable to DW-MRI data (Yang et al., 2022), the sub-diffusion framework provides new, unexplored opportunities, for fast and robust kurtosis mapping. We report on our investigation into the utility of the sub-diffusion model for practically useful mapping of mean kurtosis.

**Results**

Simulation studies were conducted to establish the requirements on the number of diffusion times and the separation between them for accurate estimation of mean kurtosis based on the sub-diffusion model augmented with random Gaussian noise, following (Equation 10). Testing and validation were performed using the human Connectome 1.0 brain dataset (Tian et al., 2022). The $2 \times 2 \times 2$ mm$^3$ resolution data were obtained using two diffusion times ($\Delta = 19.49$ ms) with a pulse duration of $\delta = 8$ ms and $G = 31, 68, 105, 142, 179, 216, 253, \text{ and } 290$ mT/m, respectively, generating $b$-values = 50, 350, 800, 1500, 2400, 3450, 4750, and 6000 s/mm$^2$, and $b$-values = 200, 950, 2300, 4250, 6750,
9850, 13500, and 17800 s/mm², according to \( b \)-value = \((\gamma G)^2(\Delta - \delta/3)\). Up to 64 diffusion encoding directions per b-shell were set. The traditional method for mean kurtosis estimation was implemented (producing \( K_{DKI} \)), which is limited to the use of DW-MRI generated using a single diffusion time (Jensen et al., 2005; Jensen and Helpern, 2010; Veraart et al., 2011a; Poot et al., 2010), alongside our implementation based on the sub-diffusion model (Equation 3), wherein mean kurtosis \( K^* \) is computed as a function of the sub-diffusion model parameter \( \beta \) (refer to (Equation 9)) using either a single or multiple diffusion times.

Figure 1. Simulation results on the effect of the number of diffusion times involved in the fitting of the sub-diffusion model (Equation 3) parameters \( (D_\beta, \beta) \) and computing \( K^* \) following (Equation 9) at various SNR levels. The true values for \( (D_\beta, \beta, K^*) \) are set to \((3 \times 10^{-4}, 0.75, 0.8125)\) to represent white matter (blue) and \((5 \times 10^{-4}, 0.85, 0.4733)\) to represent grey matter (orange). Rows 1–3: Distributions of fitted parameter values using different number of diffusion times. 2’ represents an additional simulation using two diffusion times but set to be the same, so it has the same number of data points in the fitting as for using two different diffusion times. Row 4: Coefficient of variation (CV) of the parameter values fitted using different number of diffusion times.

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Multiple diffusion times for robust and accurate mean kurtosis estimation

In our simulations, we tested up to five distinct diffusion times to generate b-values. Figure 1 illustrates the effects of the number of diffusion times on the parameter estimation at various SNR levels. We draw attention to a number of features in the plots. First, as SNR is increased from 5 to 20 (rows 1–3), the variability in the estimated parameters ($D$, $\beta$, $K^*$) decreases. Second, increasing the number of distinct diffusion times used for parameter estimation decreases estimation variability, with the most significant improvement when increasing from one to two diffusion times (rows 1–3). Third, sampling with two distinct diffusion times provides more robust parameter estimates than sampling twice as many b-values using one diffusion time (compare 2 and 2′ violin plots, rows 1–3). Fourth, the last row (row 4) highlights the improvement in the coefficient of variation (CV) for each parameter estimate with increasing SNR. This result again confirms that the most pronounced decline of CV occurs when increasing from one to two diffusion times, and parameter estimations can be performed more robustly using DW-MRI data with a relatively high SNR.

In Figure 2, we provide simulation results evaluating the choice of the two distinct diffusion times (assuming $\Delta_1 < \Delta_2$) by measuring the goodness-of-fit of the model. The smaller of the two diffusion times is stated along the abscissa, and the difference, i.e., $\Delta_2 - \Delta_1$, is plotted along the ordinate. The quality of fitting was measured using the coefficient of determination (the larger the $R^2$ value, the better the goodness-of-fit of the model) for each combination of abscissa and ordinate values. The conclusion from this figure is that $\Delta_1$ should be small, whilst $\Delta_2$ should be as large as practically plausible. Note, DW-MRI echo time was not considered in this simulation, but as $\Delta_2$ increases, the echo time has to proportionally increase. Because of the inherent consequence of decreasing SNR with echo time, special attention should be paid to the level of SNR achievable with a specific $\Delta_2$. Nonetheless, our findings suggest that when $\Delta_1 = 8$ ms, $\Delta_2$ can be set as small as 21 ms to achieve an $R^2 > 0.90$ with an SNR as low as 5. If $\Delta_1$ is increased past 15 ms, then the separation between $\Delta_1$ and $\Delta_2$ has to increase as well, and such choices benefit from an increased SNR in the DW-MRI data. The Connectome 1.0 DW-MRI data was obtained using $\Delta_1 = 19$ ms and $\Delta_2 = 49$ ms, leading to a separation of 30 ms. For this data, it is expected that with SNR = 5 an $R^2$ around 0.9 is feasible, and by increasing SNR to 20, the $R^2$ can increase to a value above 0.99.

In Figure 3, we present the scatter plots of simulated $K$ values versus fitted $K$ values using simulated data with different number of diffusion times at various SNR levels. Four cases are provided, including fitting simulated data generated with $\Delta_1 = 19$ ms (row 1) or $\Delta_2 = 49$ ms (row 2), fitting data with both diffusion times (row 3), and fitting data with three diffusion times (row 4). $R^2$ values for each case at each SNR level are provided. This result verifies that sub-diffusion based kurtosis estimation (blue dots) improves using multiple diffusion times. The improvement in $R^2$ is prominent when moving from fitting single diffusion time data to two diffusion times data, especially when the data is noisy.
The improvement gained by moving from two to three distinct diffusion times is marginal (less than 0.01 improvement in $R^2$ value at SNR = 5 and 10, and no improvements for SNR = 20 data). Moreover, our simulation findings highlight the deviation away from the true kurtosis $K$ by using the traditional DKI method (orange dots), especially with kurtosis values larger than 1. Overall, fitting sub-diffusion model (Equation 3) to data with two adequately separated diffusion times can lead to robust estimation of mean kurtosis, via (Equation 9).

**Time-dependence in DKI metrics**

In Figure 4, we show the time-dependence effect of the DKI metrics ($\mu_{DKI}$ and $\kappa_{DKI}$) after fitting the standard DKI model to our simulated data. In this fitting, we consider $b$-values of 0, 1000, 1400,
and 2500 s/mm², and vary the diffusion time, as in Jelescu et al., 2022. We depict the parameter estimates, $D_{DKI}$ and $K_{DKI}$, from simulated data with added noise (SNR = 20) to estimate the parameters $D_{DKI}$ and $K_{DKI}$. In (C) and (D), using data with no added noise, we illustrate the long-term fitting results and trends in the parameter estimates. In both (A) and (C), as diffusion time increases, $D_{DKI}$ decreases as expected for an effective diffusion coefficient. The mean $D_{DKI}$ (averaged over 1000 simulations) agrees with the true diffusivity $D^*$. When it comes to the kurtosis $K_{DKI}$, in the noiseless data setting (D), we see $K_{DKI}$ is converging to the true kurtosis value $K^*$ at large diffusion time, whilst in the noisy data setting (B), this trend is not obvious within the experimental diffusion time window. More explanations of the observed time-dependence of diffusivity and kurtosis are provided in the Discussion.

**Towards fast DKI data acquisitions**

Next, we sought to identify the minimum number and combination of $b$-values to use for mean kurtosis estimation based on the sub-diffusion model (Equation 3). The simulation results were generated using the Connectome 1.0 DW-MRI data $b$-value setting, which has 16 $b$-values, 8 from $\Delta_1 = 19$ ms and 8 from $\Delta_2 = 49$ ms.

In Figure 5, we computed $R^2$ values for all combinations of choices when the number of non-zero $b$-values is 2, 3, and 4. We then plotted the $R^2$ values sorted in ascending order for each considered SNR level. Notably, as the number of non-zero $b$-values increase, the achievable combinations increase as well (i.e., 120, 560, and 1820 for the three different non-zero $b$-value cases). The colours illustrate the proportion of $b$-values used in the fitting based on $\Delta_1$ and $\Delta_2$. For example, 1:0 means only $\Delta_1$ $b$-values were used, and 2:1 means two $\Delta_1$ and one $\Delta_2$ $b$-values were used to generate the
result. The b-value combinations achieving highest $R^2$ values are displayed in the inset pictures and the b-values are provided in Table 1.

Our simulation results in Table 1 suggest that increasing the number of non-zero b-values from two to four improves the quality of the parameter estimation, also achievable by fitting the sub-diffusion model (Equation 3) to higher SNR data. The gain is larger by using higher SNR data than by using more b-values. For example, going from two to four non-zero b-values with SNR = 5 data approximately doubles the $R^2$, whereas the $R^2$ almost triples when SNR = 5 data is substituted by SNR = 20 data. Additionally, the use of $\Delta_1$ or $\Delta_2$ alone is not preferred (also see Figure 5), and preference is towards first using $\Delta_1$ and then supplementing with b-values generated using $\Delta_2$. At all SNR levels when only two non-zero b-values are used, one b-value should be chosen based on the $\Delta_1$ set, and the other based on $\Delta_2$. Moving to three non-zero b-values requires the addition of another $\Delta_1$ b-value, and when four non-zero b-values are used then two from each diffusion time are required. If we consider an $R^2 = 0.90$ to be a reasonable goodness-of-fit for the sub-diffusion model, then at least three or four non-zero b-values are needed with an SNR = 20. If SNR = 10, then three non-zero
b-values will not suffice. Interestingly, an $R^2$ of 0.85 can still be achieved when SNR = 20 and two optimally chosen non-zero b-values are used. Table 1 summarises findings based on having different number of non-zero b-values with $R^2$ values deduced from the SNR = 5, 10, and 20 simulations. We have chosen to depict five b-value combinations producing the largest $R^2$ values for the two, three, and four non-zero b-value sampling cases. We found consistency in b-value combinations across SNR levels. Thus, we can conclude that a range of b-values can be used to achieve a large $R^2$ value, which is a positive finding, since parameter estimation does not stringently rely on b-value sampling. For example, using three non-zero b-values an $R^2 \geq 0.90$ is achievable based on different b-value sampling. Importantly, two distinct diffusion times are required, and preference is towards including a smaller diffusion time b-value first. Hence, for three non-zero b-values we find two b-values based on $\Delta_1$ and one based on $\Delta_2$. This finding suggests one of the $\Delta_1$ b-values can be chosen in the range 50 to 350 s/mm^2, and the other in the range 1500 to 4750 s/mm^2. Additionally, the $\Delta_2$ b-value can also be chosen in a range, considering between 2300 to 4250 s/mm^2 based on the Connectome 1.0 b-value settings. The b-value sampling choices made should nonetheless be in view of the required $R^2$ value. Overall, sampling using two distinct diffusion times appears to provide quite a range of options for the DW-MRI data to be used to fit the sub-diffusion model parameters. The suggested optimal b-value sampling in the last row of Table 1, primarily chosen to minimise b-value size whilst maintaining a large $R^2$ value, may be of use for specific neuroimaging studies, which will be used to inform our discussion on feasibility for clinical practice.

Table 1. A selection of the best b-value sampling regimes to achieve the highest $R^2$ value in the three cases considered.

The various categories correspond with two, three and four non-zero b-value sampling schemes, with $\Delta_1$ and $\Delta_2$ denoting the diffusion time setting used to generate the b-values. Note, entries are b-values in unit of s/mm^2, and $\Delta_1 = 19$ ms and $\Delta_2 = 49$ ms were used to match the Connectome 1.0 DW-MRI data collection protocol. The entries listed at the bottom row are suggested optimal non-zero b-values for clinical practice.

| Two b-values | Three b-values | Four b-values |
|--------------|---------------|---------------|
| $\Delta_1$  | $\Delta_2$   | $R^2$ | $\Delta_1$  | $\Delta_2$ | $R^2$ | $\Delta_1$  | $\Delta_2$ | $R^2$ |
| 350          | 6750          | 0.01  | 350, 800    | 2300        | 0.46  | 350, 4750   | 950, 4250   | 0.61  |
| 800          | 2300          | 0.11  | 350, 950, 4250 | 0.47  | 350, 6000   | 950, 6750   | 0.62  |
| SNR = 5      | 350, 4250    | 0.18  | 350, 2300, 4250 | 0.47  | 350, 4750   | 950, 6750   | 0.62  |
| 350, 950     | 0.26          | 50, 800 | 2300        | 0.48  | 350, 2400   | 950, 6750   | 0.63  |
| 350, 2300    | 0.30          | 350, 950 | 6750        | 0.49  | 350, 2400   | 950, 4250   | 0.63  |
| 350, 4250    | 0.63          | 350, 4750 | 2300        | 0.83  | 350, 2400   | 950, 9850   | 0.91  |
| 800          | 4250          | 0.64  | 50, 800     | 2300        | 0.84  | 350, 3450   | 950, 6750   | 0.91  |
| SNR = 10     | 350, 950     | 0.67  | 350, 800    | 2300        | 0.84  | 350, 6000   | 950, 9850   | 0.91  |
| 350, 2300    | 0.72          | 350, 1500 | 2300       | 0.84  | 350, 4750   | 950, 9850   | 0.91  |
| 800          | 2300          | 0.73  | 350, 2400   | 2300        | 0.85  | 350, 3450   | 950, 9850   | 0.91  |
| SNR = 20     | 1500, 2300   | 0.77  | 350, 6000   | 2300        | 0.89  | 350, 4750   | 2300, 13,500 | 0.96  |
| 350, 950     | 0.78          | 50, 1500 | 4250        | 0.90  | 350, 4750   | 2300, 6750  | 0.96  |
| SNR = 10     | 350, 2300    | 0.80  | 350, 1500   | 2300        | 0.90  | 350, 6000   | 2300, 9850  | 0.96  |
| 800          | 4250          | 0.82  | 350, 4750   | 2300        | 0.90  | 350, 4750   | 2300, 9850  | 0.96  |
| 800          | 2300          | 0.85  | 50, 2400    | 4250        | 0.90  | 350, 4750   | 950, 6750   | 0.96  |
| 800, 2300    | 0.85          | 350, 1500 | 2300        | 0.90  | 350, 1500   | 950, 4250   | 0.92  |
Benchmark mean kurtosis in the brain

The benchmark mean kurtosis estimation in the brain is established using the entire b-value range with all diffusion encoding directions available in the Connectome 1.0 DW-MRI data. For two subjects in different slices, Figure 6 provides the spatially resolved maps of mean kurtosis computed using the sub-diffusion method (i.e., $K^*$) with one or two diffusion times, and using the standard method (i.e., $K_{DKI}$). The diffusion times, $\Delta$, used to generate each plot are provided for each case. We consider the mean kurtosis maps using two diffusion times ($\Delta = 19.49$ ms) as the benchmarks.
KDKI considering the two distinct diffusion times. First, we notice a degradation in the KDKI image with an increase in diffusion time. Second, the use of a single diffusion time with the sub-diffusion model leads to \( K^* \) values which are larger than either the KDKI values or \( K^* \) values generated using two diffusion times. Third, the quality of the mean kurtosis map appears to visually be best when two diffusion times are used to estimate \( K^* \). Superior grey-white matter tissue contrast (TC) was found for the \( K^* \) map (TC = 1.73), compared to the KDKI maps (TC = 0.80 for the \( \Delta = 19 \) ms dataset and TC = 1.01 for the \( \Delta = 49 \) ms dataset).

In Figure 7, an error map (measured by root-mean-square-error, RMSE) from Subject 5 slice 74 was presented for fitting the sub-diffusion model to the DW-MRI data with two diffusion times. Sample parameter fittings in both \( b \)-space (Equation 3) and \( q \)-space (Equation 4) were provided for four representative white and grey matter voxels.

Quantitative findings for kurtosis are provided in Table 2. The analysis was performed for subcortical grey matter (scGM), cortical grey mater (cGM) and white matter (WM) brain regions. For specifics we refer the reader to the appropriate methods section. The table entries highlight the differences in mean kurtosis when computed using the two different approaches. The trend for the traditional single diffusion time approach is that an increase in \( \Delta \) results in a slight decrease in the mean KDKI, and an increase in the coefficient of variation (CV) for any region. For example, the mean KDKI in the thalamus reduces from 0.65 to 0.58, whilst the CV increases from 30% to 39%. As much as 30% increase in CV is common for scGM and cGM regions, and around 10% for WM regions. The CV based on the \( K^* \) value for each region is less than the CV for KDKI with either \( \Delta = 19 \) ms or \( \Delta = 49 \) ms.

Figure 8 presents the distributions of the fitted parameters (\( D \) and \( K \)) in specific brain regions, based on the sub-diffusion model (Panel A) and the standard DKI model with \( \Delta = 19 \) ms (Panel B). The distributions are coloured by the probability density. Yellow indicates high probability density, light blue indicates low probability density. In each subplot, the diffusivity is plotted along the abscissa axis and the kurtosis is along the ordinate axis. Results for the standard DKI model with \( \Delta = 49 \) ms are qualitatively similar, so are not shown here. From Panel (B), we see an unknown nonlinear relationship between the DKI pair, \( D_{\text{DKI}} \) and \( K_{\text{DKI}} \), in all regions considered. By comparison, the sub-diffusion based \( K^* \) and \( D^* \) (Panel A) are less correlated with each other, indicating \( D^* \)...
and $K^*$ carry distinct information, which will be very valuable for characterising tissue microstructure. Tables 3 and Table 4 present the $D_\beta$ and $\beta$ values used to compute $D^*$ and $K^*$ in Table 2 and Figure 8.

**Reduction in DKI data acquisition**

Results for reductions in diffusion encoding directions to achieve different levels of SNR with the purpose of shortening acquisition times will be benchmarked against the $K^*$ maps in Figure 6 and the $K^*$ values reported in Table 2.

Figure 9 presents the qualitative findings for two subjects generated using all, optimal and sub-optimal $b$-value samplings with SNR = 6 (3 non-collinear directions, 6 measurements), 10 (8 non-collinear directions, 16 measurements), and 20 (32 non-collinear directions, 64 measurements) DW-MRI data. The quality of the mean kurtosis map improves with increasing SNR, and also by optimising $b$-value sampling. Optimal sampling at SNR = 10 is qualitatively comparable to the SNR = 20 optimal sampling result, and to the benchmark sub-diffusion results in Figure 6.

Quantitative verification of the qualitative observations are provided in Table 5. Significant differences in brain region-specific mean kurtosis values occur at the SNR = 6 level, which are not apparent when SNR = 10 or 20 data with optimal $b$-value sampling were used. The average errors are relative errors compared to the benchmark kurtosis values reported in Table 2. When using optimal $b$-values, average errors range from 22% to 43% at SNR = 6, from 13% to 43% at SNR = 10, and from 8% to 20% at SNR = 20, across brain regions. When using sub-optimal $b$-values, average errors range from 47% to 57% at SNR = 6, from 24% to 102% at SNR = 10, and from 27% to 72% at SNR = 20. Also, the brain region-specific CV for mean kurtosis was not found to change markedly when SNR = 10 or 20 data were used to compute $K^*$. The result of reducing the SNR to 6 leads to an approximate doubling of the CV for each brain region. These findings confirm that with optimal $b$-value sampling, the data quality can be reduced to around the SNR = 10 level, without a significant impact on the region-specific mean kurtosis estimates derived using the sub-diffusion model.

**Table 2.** Benchmark kurtosis values estimated using the Connectome 1.0 DW-MRI data for different regions of the human brain.

Results are provided for the traditional mean kurtosis ($K_{DKI}$) at two distinct diffusion times, and values ($K^*$) obtained based on fitting the sub-diffusion model across both diffusion times. Results are for grey matter (GM) and white matter (WM) brain regions, in categories of sub-cortical (sc) and cortical (c), and CC stands for corpus callosum. The pooled means and standard deviations across participants have been tabulated, along with the coefficient of variation in parentheses.

|          | $K_{DKI}$ | $K^*$  |
|----------|-----------|--------|
|          | $\Delta = 19$ ms | $\Delta = 49$ ms | $\Delta = 19,49$ ms |
| scGM     | 0.57 ± 0.23 (40%) | 0.50 ± 0.24 (48%) | 0.60 ± 0.21 (35%) |
| Thalamus | 0.65 ± 0.19 (30%) | 0.58 ± 0.22 (39%) | 0.70 ± 0.17 (25%) |
| Caudate  | 0.41 ± 0.24 (58%) | 0.37 ± 0.23 (64%) | 0.39 ± 0.14 (35%) |
| Putamen  | 0.54 ± 0.21 (40%) | 0.45 ± 0.22 (49%) | 0.49 ± 0.13 (27%) |
| Pallidum | 0.68 ± 0.25 (37%) | 0.64 ± 0.26 (41%) | 0.93 ± 0.18 (19%) |
| cGM      | 0.53 ± 0.24 (46%) | 0.46 ± 0.23 (51%) | 0.40 ± 0.16 (39%) |
| Fusiform | 0.55 ± 0.22 (40%) | 0.44 ± 0.22 (49%) | 0.40 ± 0.15 (37%) |
| Lingual  | 0.59 ± 0.21 (35%) | 0.53 ± 0.22 (42%) | 0.47 ± 0.16 (34%) |
| WM       | 0.78 ± 0.20 (25%) | 0.76 ± 0.19 (26%) | 0.87 ± 0.22 (25%) |
| Cerebral WM | 0.77 ± 0.19 (25%) | 0.75 ± 0.19 (26%) | 0.85 ± 0.22 (26%) |
| Cerebellum WM | 0.99 ± 0.18 (18%) | 0.95 ± 0.19 (20%) | 1.07 ± 0.22 (21%) |
| CC       | 0.65 ± 0.22 (35%) | 0.65 ± 0.22 (34%) | 0.95 ± 0.25 (26%) |
Scan–rescan reproducibility of mean kurtosis

Figure 10 summarises the intraclass correlation coefficient (ICC) distribution results ($\mu$ for mean; $\sigma$ for standard deviation) for the specific brain regions analysed. The two sets of ICC values were computed based on all DW-MRI (i.e., SNR = 20; subscript A) and the SNR = 10 optimal $b$-value sampling scheme (subscript O). As the value of $\mu$ approaches 1, the inter-subject variation in mean kurtosis is expected to greatly outweigh the intra-subject scan–rescan error. The value of $\mu$ should always be above 0.5, otherwise parameter estimation cannot be performed robustly and accurately, and values above 0.75 are generally accepted as good. The $\mu_A$ values for all regions were in the range 0.76 (thalamus) to 0.87 (caudate), and reduced to the range 0.57 (thalamus) to 0.80 (CC) when optimal sampling with SNR = 10 was used to estimate the $K^*$ value. Irrespective of which of the two DW-MRI data were used for $K^*$ estimation, the value of $\mu$ was greater than or equal to 0.70 in 20 out of 24 cases. The $\mu_O$ was less than 0.70 for only the thalamus, putamen, and pallidum. The loss in ICC by going to SNR = 10 data with optimal $b$-value sampling went hand-in-hand with an increase in $\sigma$, which is not unexpected, since the uncertainty associated with using less data should be measurable. Overall, $\mu_A$, $\mu_O$, and $\sigma_A$, $\sigma_O$, were fairly consistent across the brain regions, suggesting the DW-MRI data with SNR = 10 is sufficient for mean kurtosis estimation based on the sub-diffusion framework.
Discussion

DW-MRI allows the measurement of mean kurtosis, a metric for the deviation away from standard Brownian motion of water in tissue, which has been used to infer variations in tissue microstructure. Research on mean kurtosis has shown benefits in specific applications over other diffusion related measures derived from DW-MRI data (Li et al., 2022b; Liu et al., 2022; Huang et al., 2022; Guo et al., 2022b; Goryawala et al., 2023; Guo et al., 2022a; Wang et al., 2022; Li et al., 2022a; Maral-akunte et al., 2023; Hu et al., 2022; Zhou et al., 2023; Li et al., 2022c). Whilst many efforts have been made to optimise mean kurtosis imaging for clinical use, the limitations have been associated with lack of robustness and the time needed to acquire the DW-MRI data for mean kurtosis estimation. The choice of the biophysical model and how diffusion encoding is applied are critical to how well kurtosis in the brain is mapped. Here, we evaluated the mapping of mean kurtosis based on the sub-diffusion model, which allows different diffusion times to be incorporated into the data acquisition. Using simulations and the Connectome 1.0 public DW-MRI dataset, involving a range of diffusion encodings, we showed that mean kurtosis can be mapped robustly and rapidly provided at least two different diffusion times are used and care is taken towards how $b$-values are chosen given differences in the SNR level of different DW-MRI acquisitions.

Reduction in scan time

Previous attempts have been made in optimising the DW-MRI acquisition protocol for mean kurtosis estimation based on the traditional, single diffusion time kurtosis model (Hansen et al., 2013; Hu et al., 2022; Poot et al., 2010; Hansen et al., 2016). Considerations have been made towards reducing the number of b-shells, directions per shell, and sub-sampling of DW-MRI for each direction in each shell. Our findings suggest that robust estimation of kurtosis cannot be achieved using the classical model for mean kurtosis, as highlighted previously (Yang et al., 2022; Ingo et al., 2014; Ingo et al., 2015; Barrick et al., 2020). A primary limitation of the traditional method is the use of the cumulant expansion resulting in sampling below a $b$-value of around 2500 s/mm² (Jensen et al., 2005;
Jensen and Helpern, 2010), and using the sub-diffusion model this limitation is removed (Yang et al., 2022). Our simulation findings and experiments using the Connectome 1.0 data confirm that mean kurtosis can be mapped robustly and rapidly using the sub-diffusion model applied to an optimised DW-MRI protocol. Optimisation of the data acquisition with the use of the sub-diffusion model has not been considered previously.

Mean kurtosis values can be generated based on having limited number of diffusion encoding directions (refer to Figure 9 and Table 5). Given that each direction for each b-shell takes a fixed amount of time, then a four b-shell acquisition with six directions per shell will take 25 times longer than a single diffusion encoding data acquisition (assuming a single $b$-value=0 data is collected). The total acquisition time for the diffusion MRI protocol for the Connectome 1.0 data was 55 min, including 50 $b=0$ s/mm² scans plus seven $b$-values with 32 and nine with 64 diffusion encoding directions (Tian et al., 2022). This gives a total of 850 scans per dataset. As such, a single 3D image volume took 3.88 s to acquire. Conservatively allowing 4 s per scan, and considering SNR = 20 data (i.e., 64 directions) over four $b$-values and a single $b$-value = 0 scan, DW-MRI data for mean kurtosis estimation can be completed in 17 min 8 s ($R^2 = 0.96$). At SNR = 10 (i.e., 32 directions), DW-MRI data with the same number of $b$-values can be acquired in 4 min 20 s ($R^2 = 0.91$). If an $R^2 = 0.85$ (SNR = 10) is deemed adequate, then one less b-shell is required, saving an additional 64 s. We should point out that even though 2-shell optimised protocols can achieve $R^2 = 0.85$ with SNR = 20, this is not equivalent in time to using 3-shells with SNR = 10 (also $R^2 = 0.85$). This is because 4×4× additional data are required to double the SNR (equivalent to acquiring an additional 4-shells). However, only one extra b-shell is required to convert 2-shell data to 3-shells with SNR = 10. Our expected DW-MRI data acquisition times are highly feasible clinically, where generally neuroimaging scans take around 15 min involving numerous different MRI contrasts and often a DTI acquisition.

An early study on estimating mean kurtosis demonstrated the mapping of a related metric in less than 1 min over the brain (Hansen et al., 2013). Clinical adoption of the protocol lacked, possibly since $b$-values are a function $\delta$, $G$, and $\Delta$. Hence, different $b$-values can be obtained using different DW-MRI protocol settings, leading to differences in the mapping of mean kurtosis based on the data.
(we showed the $\Delta$ effect in Figure 6 and Table 2). Our findings suggest this impediment is removed by sampling and fitting data with $b$-values across two distinct diffusion times. Nonetheless, we should consider what might be an acceptable DW-MRI data acquisition time.

A recent study on nasopharyngeal carcinoma investigated reducing the number of $b$-shell signals based on fixing diffusion encoding directions to the three Cartesian orientations (He et al., 2022). The 3-shell acquisitions took 3 min 2 s to acquire, whilst the 5-shell data required 5 min 13 s. They investigated as well the impact of using partial Fourier sampling, i.e., reducing the amount of data needed for image reconstruction by reducing $k$-space line acquisitions for each diffusion encoded image. Their benchmark used 5-shells (200, 400, 800, 1500, 2000 s/mm$^2$), and found partial Fourier sampling with omission of the 1500 s/mm$^2$ $b$-shell produced acceptable results. With this acquisition the scan could be completed in 3 min 46 s, more than 2 times faster than the benchmark 8 min 31 s.

Our proposed 3-shells acquisition with an $R^2 = 0.85$ (see SNR = 10 results in Table 1) executable under 4 min is therefore inline with current expectations. Note, at the $R^2 = 0.85$ level the ICC for the different brain regions were in the range 0.60–0.69, and these were not formally reported in Figure 10. This level of reproducibility is still acceptable for routine use. We should additionally point out that we used the Subject 1 segmentation labels, after having registered each DW-MRI data to the Subject 1 first scan. This approach results in slight mismatch of the region-specific segmentations when carried across subjects, inherently resulting in an underestimation of ICC values.
Less than 4 min DW-MRI data acquisitions can potentially replace existing data acquisitions used to obtain DTI metrics, since even the estimation of the apparent diffusion coefficient improves by using DW-MRI data relevant to DKI (Veraart et al., 2011b; Wu and Cheung, 2010). Additionally, it is increasingly clear that in certain applications the DKI analysis offers a more comprehensive approach for tissue microstructure analysis (Li et al., 2022b; Liu et al., 2022; Huang et al., 2022; Guo et al., 2022b; Goryawala et al., 2023; Guo et al., 2022a; Wang et al., 2022; Li et al., 2022a; Maralakunte

| Table 5. Kurtosis values ($K^*$) under the optimal and sub-optimal b-value sampling regimes for specific brain regions. $K^*$ was estimated based on fitting the sub-diffusion model to the Connectome 1.0 DW-MRI data with two diffusion times and selected four b-shells. Optimal b-value sampling is considered to have $R^2 = 0.63, 0.91$ and $0.96$ for the SNR = 6, 10, and 20 columns, according to Table 1. Sub-optimal b-values are chosen to have $R^2 = 0.3, 0.45$, and $0.5$, respectively, as reported in Figure 9. Individual entries are for grey matter (GM) and white matter (WM) brain regions, in categories of sub-cortical (sc) and cortical (c), and CC stands for corpus callosum. A reduction in SNR level was achieved by reducing the number of diffusion encoding directions in each b-shell of the DW-MRI data. The pooled means and standard deviations across participants have been tabulated, along with the coefficient of variation in parentheses. The entries identified in italic under the optimal b-value heading were found to be significantly different from the benchmark mean $K^*$ reported in Table 2. Sub-optimal result population means were mostly significantly different from the benchmark mean $K^*$, and they are not italicised. The average errors (last column) are relative errors compared to the benchmark kurtosis values reported in Table 2. |

|                  | SNR = 6 | SNR = 10 | SNR = 20 | Average error for SNR = 6/10/20 |
|------------------|---------|----------|----------|-------------------------------|
| **Optimal b-values** |         |          |          |                               |
| scGM             | 0.47 ± 0.27(57%) | 0.65 ± 0.22(33%) | 0.61 ± 0.21(34%) | 37/23/12% |
| Thalamus         | 0.57 ± 0.26(46%) | 0.75 ± 0.20(26%) | 0.71 ± 0.17(24%) | 33/20/11% |
| Caudate          | 0.30 ± 0.20(68%) | 0.46 ± 0.17(38%) | 0.40 ± 0.15(38%) | 43/30/15% |
| Putamen          | 0.38 ± 0.21(56%) | 0.58 ± 0.16(28%) | 0.52 ± 0.14(28%) | 40/28/13% |
| Pallidum         | 0.66 ± 0.33(49%) | 0.90 ± 0.24(26%) | 0.89 ± 0.20(22%) | 39/19/12% |
| cGM              | 0.40 ± 0.22(54%) | 0.51 ± 0.20(39%) | 0.45 ± 0.18(40%) | 30/36/19% |
| Fusiform         | 0.37 ± 0.21(57%) | 0.54 ± 0.20(37%) | 0.45 ± 0.17(38%) | 35/43/20% |
| Lingual          | 0.45 ± 0.22(50%) | 0.59 ± 0.19(33%) | 0.52 ± 0.18(34%) | 28/34/18% |
| WM               | 0.72 ± 0.26(36%) | 0.91 ± 0.23(26%) | 0.89 ± 0.22(25%) | 23/13/8% |
| Cerebral WM      | 0.71 ± 0.25(35%) | 0.89 ± 0.23(25%) | 0.88 ± 0.22(25%) | 22/13/8% |
| **Sub-optimal b-values** |         |          |          |                               |
| scGM             | 0.31 ± 0.22(71%) | 0.82 ± 0.29(36) | 0.58 ± 0.25(43%) | 55/45/34% |
| Thalamus         | 0.36 ± 0.23(65%) | 0.95 ± 0.25(27%) | 0.72 ± 0.19(26%) | 54/43/31% |
| Caudate          | 0.23 ± 0.19(81%) | 0.57 ± 0.17(31%) | 0.40 ± 0.20(52%) | 56/57/40% |
| Putamen          | 0.25 ± 0.16(67%) | 0.65 ± 0.17(26%) | 0.44 ± 0.20(46%) | 54/42/36% |
| Pallidum         | 0.44 ± 0.29(66%) | 1.27 ± 0.32(25%) | 0.76 ± 0.25(33%) | 56/42/32% |
| cGM              | 0.25 ± 0.17(67%) | 0.44 ± 0.14(33%) | 0.37 ± 0.17(47%) | 47/30/36% |
| Fusiform         | 0.22 ± 0.15(68%) | 0.47 ± 0.16(35%) | 0.33 ± 0.17(52%) | 51/32/38% |
| Lingual          | 0.29 ± 0.19(66%) | 0.53 ± 0.16(30%) | 0.39 ± 0.20(51%) | 48/29/40% |
| WM               | 0.42 ± 0.18(44%) | 1.16 ± 0.36(31%) | 0.72 ± 0.22(31%) | 53/38/28% |
| Cerebral WM      | 0.41 ± 0.18(43%) | 1.15 ± 0.35(31%) | 0.74 ± 0.20(28%) | 53/38/27% |
| Cerebellum WM    | 0.47 ± 0.21(45%) | 1.27 ± 0.33(26%) | 0.35 ± 0.26(74%) | 57/24/72% |
| CC               | 0.69 ± 0.42(61%) | 1.94 ± 0.44(23%) | 0.92 ± 0.19(20%) | 47/102/28% |
| Cerebellum WM    | 0.83 ± 0.29(35%) | 1.17 ± 0.25(21%) | 1.15 ± 0.23(20%) | 27/15/11% |
| CC               | 0.88 ± 0.36(41%) | 0.98 ± 0.31(31%) | 0.86 ± 0.25(29%) | 26/17/13% |
Farquhar et al., 2023; Hu et al., 2022; Zhou et al., 2023; Li et al., 2022c). As such, multiple b-shell, multiple diffusion encoding direction DW-MRI acquisitions should be used for the calculation of both DTI and DKI metrics.

**Figure 10.** Interclass correlation coefficient (ICC) results for mean kurtosis are depicted for the 12 brain regions analysed. The mean ($\mu$) and standard deviation ($\sigma$) computed based on all the Connectome 1.0 DW-MRI data (subscript $A$), and the reduced data achieving an SNR = 10 with optimal four non-zero $b$-value sampling (subscript $O$), are provided for each brain region. Histograms were generated using all data. Mean kurtosis based on the optimised protocol was computed using the sub-diffusion framework using DW-MRI data with the four non-zero $b$-values suggested in Table 1 and diffusion encoding directions down sampled to achieve an SNR = 10.

**DW-MRI data acquisition considerations**

To achieve $R^2 > 0.92$ for estimating kurtosis $K^*$, it is necessary to have four $b$-values, e.g., two relatively small $b$-values (350 s/mm$^2$ using $\Delta = 19$ ms, and 950 s/mm$^2$ using $\Delta = 49$ ms, both with...
Farquhar et al., eLife 2023;12:RP90465. DOI: https://doi.org/10.7554/eLife.90465  18 of 29

Computationally increasing \( \Delta \) values are considered sufficient (with \( R^2 = 0.85 \) or 0.90), then the larger \( \Delta \) needs to be used to set the largest \( b \)-value to be 2300 s/mm\(^2\), and the other(s) should be set using the smaller \( \Delta \). For the two non-zero \( b \)-values case, the \( b \)-value from the smaller \( \Delta \) should be around 800 s/mm\(^2\). For the three non-zero \( b \)-values case, the \( b \)-values from the smaller \( \Delta \) would then need to be 350 s/mm\(^2\) and 1500 s/mm\(^2\). Interestingly, \( b \)-value = 800 s/mm\(^2\) lies around the middle of the 350 s/mm\(^2\) to 1500 s/mm\(^2\) range. The additional gain to \( R^2 = 0.92 \) can be achieved by splitting the \( b \)-value with the larger \( \Delta \) into two, again with 2300 s/mm\(^2\) near the middle of the two new \( b \)-values set. In addition, the separation between \( \Delta_1 \) and \( \Delta_2 \) needs to be as large as plausible, as can be deduced from the simulation result in Figure 2, but attention should be paid to signal-to-noise ratio decreases with increased echo times (He et al., 2022).

A recent study on optimising quasi-diffusion imaging (QDI) considered \( b \)-values up to 5000 s/mm\(^2\) (Spilling et al., 2022). Whilst QDI is a non-Gaussian approach, it is different to the sub-diffusion model, but still uses the Mittag-Leffler function and involves the same number of model parameters. The DW-MRI data used in their study was acquired with a single diffusion time. Nevertheless, particular points are worth noting. Their primary finding was parameter dependence on the maximum \( b \)-value used to create the DW-MRI data. They also showed the accuracy, precision, and reliability of parameter estimation are improved with increased number of \( b \)-shells. They suggested a maximum \( b \)-value of 3960 s/mm\(^2\) for the 4-shell parameter estimation. Our results do not suggest a dependence of the parameter estimates on maximum \( b \)-value (note, if a maximum \( b \)-value dependence is present, benchmark versus optimal region specific results in Tables 2 and 5 should show some systematic difference; and we used the real part of the DW-MRI data and not the magnitude as commonly used). These findings potentially confirm that \( \Delta \) separation is an important component of obtaining a quality parameter fit from which mean kurtosis is deduced (Figure 2).

**Diffusion gradient pulse amplitudes**

Commonly available human clinical MRI scanners are capable of 40 mT/m gradient amplitudes. Recently, the increased need to deduce tissue microstructure metrics from DW-MRI measurements has led to hardware developments resulting in 80 mT/m gradient strength MRI scanners. These were initially sought by research centres. The Connectome 1.0 scanners achieve 300 mT/m gradient amplitudes (Tian et al., 2022), which in turn allow large \( b \)-values within reasonable echo times. The Connectome 2.0 scanner is planned to achieve 500 mT/m gradient amplitudes (Huang et al., 2021), providing data for further exploration of the \((q, \Delta)\) space by providing a mechanism for increasing our knowledge of the relationships between the micro-, meso-, and macro-scales. Whilst there are three Connectome 1.0 scanners available, only one Connectome 2.0 scanner is planned for production. Hence, robust and fast methods utilising existing 80 mT/m gradient systems are needed, and the Connectome scanners provide opportunities for testing and validating methods.

The \( b \)-value is an intricate combination of \( \delta \), \( G \), and \( \Delta \). An increase in any of these three parameters increases the \( b \)-value (note, \( \delta \) and \( G \) increase \( b \)-value quadratically, and \( \Delta \) linearly). An increase in \( \delta \) can likely require an increase in \( \Delta \), the consequence of which is a reduction in signal-to-noise ratio as the echo time has to be adjusted proportionally. Partial Fourier sampling methods aim to counteract the need to increase the echo time by sub-sampling the DW-MRI data used to generate an image for each diffusion encoding direction (Zong et al., 2021; Heidemann et al., 2010). The ideal scenario, therefore, is to increase \( G \), as for the Connectome 1.0 and 2.0 scanners.

Based on our suggested \( b \)-value settings in Table 1, a maximum \( b \)-value of around 4250 s/mm\(^2\) is required for robust mean kurtosis estimation (assume \( R^2 > 0.90 \) is adequate and achieved via four non-zero \( b \)-values and two distinct \( \Delta \)s, and we should highlight that \( b \)-value = 1500 s/mm\(^2\) (\( \Delta = 19 \) ms) and \( b \)-value = 4250 s/mm\(^2\) (\( \Delta = 49 \) ms) were achieved using \( G = 142 \) mT/m). Considering 80 m/T/m gradient sets are the new standard for MRI scanners, an adjustment to \( \delta \) to compensate for \( G \) is needed (recall, \( q = \gamma \delta G \)). Hence, for a constant \( q \), \( G \) can be reduced at the consequence of proportionally increasing \( \delta \). This then allows MRI systems with lower gradient amplitudes to generate the relevant \( b \)-values. For example, changing the \( \delta \) from 8 ms to 16 ms would result in halving of \( G \) (using a maximum \( G \) of 142 m/T/M from the Connectome 1.0 can achieve the optimal \( b \)-values; hence halving would require around 71 m/T/M gradient pulse amplitudes). Increasing \( \Delta \) above 49 ms to create larger
b-value data is unlikely to be a viable solution due to longer echo times leading to a loss in SNR. SNR increases afforded by moving from 3 T to 7 T MRI are most likely counteracted by an almost proportional decrease in $T_2$ times (Pohmann et al., 2016), in addition to 7 T MRI bringing new challenges in terms of increased transmit and static field inhomogeneities leading to signal inconsistencies across an image (Kraff and Quick, 2017).

**Relationship between sub-diffusion based mean kurtosis $K^*$ and histology**

Following Table 2 (last column), white matter regions showed high kurtosis (0.87 ± 0.22), consistent with a structured heterogeneous environment comprising parallel neuronal fibres, as shown in Maiter et al., 2021. Cortical grey matter showed low kurtosis (0.40 ± 0.16). Subcortical grey matter regions showed intermediate kurtosis (0.60 ± 0.21). In particular, caudate and putamen showed similar kurtosis to grey matter, whilst thalamus and pallidum showed similar kurtosis properties to white matter. Histological staining results of the sub-cortical nuclei (Maiter et al., 2021) showed the subcortical grey matter was permeated by small white matter bundles, which could account for the similar kurtosis in thalamus and pallidum to white matter. These results confirmed that our proposed sub-diffusion based mean kurtosis $K^*$ is consistent with published histology of normal human brain.

**Time-dependence of diffusivity and kurtosis**

The time-dependence of diffusivity and kurtosis has attracted much interest in the field of tissue microstructure imaging. Although our motivation here is not to map time-dependent diffusion, we can nonetheless point out that the assumption of a sub-diffusion model provides an explanation of the observed time-dependence of diffusivity and kurtosis. From (Equation 4), the diffusivity that arises from the sub-diffusion model is of the form $D_{SUB} = D_β\bar{\Delta}^{\beta-1}$, where $\bar{\Delta}$ is the effective diffusion time, $D_{SUB}$ has the standard units for a diffusion coefficient mm$^2$/s, and $D_β$ is the anomalous diffusion coefficient (with units mm$^2$/s$^{\beta}$) associated with sub-diffusion. In the sub-diffusion framework (Equation 1), $D_β$ and $\beta$ are assumed to be constant, and hence $D_{SUB}$ exhibits a fractional power-law dependence on diffusion time. Then, following (Equation 8), $D^*$ is obtained simply by scaling $D_{SUB}$ by a constant $1/Γ(1+\beta)$, and hence also follows a fractional power-law dependence on diffusion time. This time-dependence effect of diffusivity was illustrated in our simulation results, Figure 4(A) and (C).

When it comes to kurtosis, the literature on the time-dependence is mixed. Some work showed kurtosis to be increasing with diffusion time in both white and grey matter (Aggarwal et al., 2020), and in grey matter (Ianus et al., 2021), whilst others showed kurtosis to be decreasing with diffusion time in grey matter (Lee et al., 2020; Olesen et al., 2022; Jelescu et al., 2022). In this study, we provide an explanation of these mixed results. We construct a simulation of diffusion MRI signal data based on the sub-diffusion model (Equation 3) augmented with random Gaussian noise. Then we fit the conventional DKI model to the synthetic data. As shown in Figure 4(D), when there is no noise, $KDKI$ increases with diffusion time in white matter, whilst decreasing with diffusion time in grey matter. When there is added noise, as shown in Figure 4(B), the time-dependency of kurtosis within the timescale of a usual MR experiment is not clear. This goes some way to explaining why the results in the literature on the time-dependence of kurtosis are quite mixed.

Furthermore, we summarise the benefits of using the sub-diffusion based mean kurtosis measurement $K^*$. First, as shown in (Equation 9), sub-diffusion based mean kurtosis $K^*$ is not time dependent, and hence has the potential to become a tissue-specific imaging biomarker. Second, the fitting of the sub-diffusion model is straightforward, fast and robust, from which the kurtosis $K^*$ is simply computed as a function of the sub-diffusion model parameter $\beta$. Third, the kurtosis $K^*$ is not subject to any restriction on the maximum b-value, as in standard DKI. Hence, its value truly reflects the information contained in the full range of b-values.

**Extension to directional kurtosis**

The direct link between the sub-diffusion model parameter $\beta$ and mean kurtosis is well established (Yang et al., 2022; Ingo et al., 2014; Ingo et al., 2015). An important aspect to consider is whether mean $\beta$ used to compute the mean kurtosis is alone sufficient for clinical decision making. Whilst benefits of using kurtosis metrics over other DW-MRI data-derived metrics in certain applications are clear, the adequacy of mean kurtosis over axial and radial kurtosis is less apparent. Most studies perform
the mapping of mean kurtosis, probably because the DW-MRI data can be acquired in practically feasible times. Nonetheless, we can point to a few recent examples where the measurement of directional kurtosis has clear advantages. A study on mapping tumour response to radiotherapy treatment found axial kurtosis to provide the best sensitivity to treatment response (Goryawala et al., 2023).

In a different study, a correlation was found between glomerular filtration rate and axial kurtosis assessing renal function and interstitial fibrosis (Li et al., 2022a). Unipolar depression subjects have been shown to have brain region specific increases in mean and radial kurtosis, whilst for bipolar depression subjects axial kurtosis decreased in specific brain regions and decreases in radial kurtosis were found in other regions (Maralakunte et al., 2023). This selection of studies highlights future opportunities for extending the methods to additionally map axial and radial kurtosis.

Notably, estimates for axial and radial kurtosis require directionality of kurtosis to be resolved, resulting in DW-MRI sampling over a large number of diffusion encoding directions within each b-shell (Jensen and Helpern, 2010; Poot et al., 2010). As such, extension to directional kurtosis requires a larger DW-MRI dataset acquired using an increased number of diffusion encoding directions. The number of b-shells and directions therein necessary for robust and accurate mapping of directional kurtosis based on the sub-diffusion model is an open question.

There are three primary ways of determining mean kurtosis. These include the powder averaging over diffusion encoding directions in each shell, and then fitting the model, as in our approach. A different approach is to ensure each b-shell in the DW-MRI data contains the same diffusion encoding directions, and then kurtosis can be estimated for each diffusion encoding direction, after which the average over directions is used to state mean kurtosis. Lastly, the rank-4 kurtosis tensor is estimated from the DW-MRI data, from which mean kurtosis is computed directly. The latter two approaches are potential candidates for extending to axial and radial kurtosis mapping. Note, in DTI a rank-2 diffusion tensor with six unique tensor entries is needed to be estimated, whilst in DKI in addition to the rank-2 diffusion tensor, the kurtosis tensor is rank-4 with 15 unknowns, resulting in 21 unknowns altogether (Hansen et al., 2016). As such, DKI analysis for directional kurtosis requires much greater number of diffusion encoding directions to be sampled than DTI. This automatically means that at least 22 DW-MRI data (including b-value = 0) with different diffusion encoding properties have to be acquired (Jensen and Helpern, 2010). The traditional approach has been to set five distinct b-values with 30 diffusion encoding directions within each b-shell (Poot et al., 2010). Hence, to obtain the entries of the rank-4 kurtosis tensor, much more DW-MRI data is needed in comparison to what is proposed for mean kurtosis estimation in this study. Estimation of the tensor entries from this much data is prone to noise, motion and image artifacts in general (Tabesh et al., 2011), posing challenges on top of long DW-MRI data acquisition times.

A kurtosis tensor framework based on the sub-diffusion model where separate diffusion encoding directions are used to fit a direction specific β is potentially an interesting line of investigation for the future, since it can be used to establish a rank-2 β tensor with only six unknowns, requiring at least six distinct diffusion encoding directions. This type of approach can reduce the amount of DW-MRI data to be acquired, and potentially serve as a viable way forward for the combined estimation of mean, axial, and radial kurtosis.

### Kurtosis estimation outside of the brain

Although our study has been focusing on mean kurtosis imaging in the human brain, it is clear that DKI has wide application outside of the brain (Li et al., 2022b; Liu et al., 2022; Huang et al., 2022; Guo et al., 2022b; Li et al., 2022a; Zhou et al., 2023). Without having conducted experiments elsewhere, we cannot provide specific guidelines for mean kurtosis imaging in the breast, kidney, liver, and other human body regions. We can, however, point the reader in a specific direction.

The classical mono-exponential model can be recovered from the sub-diffusion equation by setting \( \beta = 1 \). For this case, the product between the b-value and fitted diffusivity has been reported to be insightful for b-value sampling (Yablonskiy and Sukstanskii, 2010), in accordance with a theoretical perspective (Istratov and Vyvenko, 1999). It was suggested the product should approximately span the (0, 1) range. Considering our case based on the sub-diffusion equation, we can investigate the size of \( hD_{\text{SUB}} \) by analysing the four non-zero b-value optimal sampling regime (\( \Delta_1 \): 350 s/mm\(^2\) and 1500 s/mm\(^2\); \( \Delta_2 \): 950 s/mm\(^2\) and 4250 s/mm\(^2\) from Table 1). Considering scGM, cGM, and WM brain regions alone, the rounded and dimensionless \( hD_{\text{SUB}} \) values are (0.09, 0.38, 0.20, 0.88), (0.13, 0.55, 0.79, 2.79) for scGM, cGM, and WM brain regions alone.
0.30, 1.34), and (0.05, 0.23, 0.11, 0.48), respectively, and note that in each case the first two effective sampling values are based on \( \Delta_1 \), and the other two are derived using \( \Delta_2 \). Interestingly, the log-linear sampling proposed in Istratov and Vyvenko, 1999 is closely mimicked by the effective sampling regime (scGM: \(-2.42, -1.62, -0.97, -0.13\); cGM: \(-2.06, -1.20, -0.60, 0.29\); WM: \(-2.94, -2.23, -1.48, -0.73\); by sorting and taking the natural logarithm). This analysis also informs on why it may be difficult to obtain a generally large \( R^2 \) across the entire brain, since \( \beta \) and \( D_{SUB} \) are brain region specific and the most optimal sampling strategy should be \( \beta \) and \( D_{SUB} \) specific. Whilst region specific sampling may provide further gains in the \( R^2 \) value, and improve ICC values for specific brain regions, such data would take a long time to acquire and require extensive post-processing and in-depth analyses.

**Methods**

**Theory**

**Sub-diffusion modelling framework**

In biological tissue, the motion of water molecules is hindered by various microstructures, and hence the diffusion can be considerably slower than unhindered, unrestricted, free diffusion of water. The continuous time random walk formalism provides a convenient mathematical framework to model this sub-diffusive behaviour using fractional calculus (Metzler and Klafter, 2000). The resulting probability density function \( P(x, t) \) of water molecules at location \( x \) (in units of mm) at time \( t \) (in units of s) satisfies the time fractional diffusion equation:

\[
\frac{\partial^\beta}{\partial t^\beta} P(x, t) = D_\beta \nabla^2 P(x, t), \quad 0 < \beta \leq 1,
\]

where \( \frac{\partial^\beta}{\partial t^\beta} \) is the time fractional derivative of order \( \beta \) \((0 < \beta \leq 1)\) in the Caputo sense, \( D_\beta \) is the generalised anomalous diffusion coefficient with unit of \( \text{mm}^2/\text{s}^\beta \), and the parameter \( \beta \) characterises the distribution of waiting times between two consecutive steps in the continuous time random walk interpretation. When \( \beta = 1 \), the waiting times have finite mean; when \( 0 < \beta < 1 \), the waiting times have infinite mean, leading to sub-diffusion behaviour. The solution to the time fractional diffusion Equation 1 in Fourier space is:

\[
p(k, t) = E_\beta \left( D_\beta |k|^2 \bar{\Delta}^\beta \right),
\]

where \( E_\beta(z) = \sum_{n=0}^{\infty} \frac{z^n}{\Gamma(1+n\beta)} \) is the single-parameter Mittag-Leffler function, \( \Gamma \) is the standard Gamma function and by definition \( E_1(z) = \exp(z) \). In the context of diffusion DW-MRI, \( k \) in Equation 2 represents the \( q \)-space parameter \( q = \gamma G \delta \), \( t \) represents the effective diffusion time \( \bar{\Delta} = \Delta - \delta/3 \) and \( p(k, t) \) represents the signal intensity \( S(q, \bar{\Delta}) \), leading to the diffusion signal equation (Magin et al., 2020):

\[
S(q, \bar{\Delta}) = S_0 E_\beta \left( -D_\beta q^2 \bar{\Delta}^{\beta} \right).
\]

Defining \( b = q^2 \bar{\Delta} \), the DW-MRI signal then can be expressed in terms of \( b \)-values:

\[
S(b) = S_0 E_\beta(-bD_{SUB}).
\]

where

\[
D_{SUB} = D_\beta \bar{\Delta}^{\beta-1}
\]

has the standard unit for a diffusion coefficient, \( \text{s/mm}^2 \).

**Diffusional kurtosis imaging**

The traditional DKI approach was proposed by Jensen and colleagues (Jensen et al., 2005; Jensen and Helpern, 2010) to measure the extent of non-Gaussian diffusion in biological tissues using DW-MRI data:
\[ S(b) \approx S_0 \exp \left( -bD_{DKI} + \frac{1}{6} b^2 D_{DKI}^2 K_{DKI} \right). \]  

where \( S \) is the signal for a given diffusion weighting \( b \) (i.e., b-value), \( S_0 \) is the signal when \( b = 0 \), \( D_{DKI} \) and \( K_{DKI} \) are the apparent diffusivity and kurtosis. A major limitation of (Equation 6) is that it was developed based on the Taylor expansion of the logarithm of the signal at \( b = 0 \), as such \( b \)-values should be chosen in the neighbourhood of \( b = 0 \) (Yang et al., 2022; Kiselev, 2010). Hence, to estimate diffusivity and kurtosis, Jensen and Helpern, 2010 suggested the use of three different \( b \)-values (such as 0, 1000, 2000 s/mm\(^2\)) and the maximum \( b \)-value should be in the range 2000 s/mm\(^2\) to 3000 s/mm\(^2\) for brain studies. Subsequently, the optimal maximum \( b \)-value was found to be dependent on the tissue types and specific pathologies, which makes the experimental design optimal for a whole brain challenging (Chuhutin et al., 2017). The procedure for fitting kurtosis and diffusivity tensors is also not trivial, and a variety of fitting procedures are currently in use. We refer readers to the descriptive and comparative studies for detail on the implementation and comparison of methods (Veraart et al., 2011b; Chuhutin et al., 2017).

Mean kurtosis from the sub-diffusion model

Yang et al., 2022 established that the traditional DKI model corresponds to the first two terms in the expansion of the sub-diffusion model:

\[ S(b) = S_0 E_\beta(-bD_{SUB}) = S_0 \exp \left( -bD^* + \frac{1}{6} b^2 D^* K^* + O(b^3) \right). \]  

where diffusivity, \( D^* \), and kurtosis, \( K^* \), are computed directly via sub-diffusion parameters \( D_{SUB} \) and \( \beta \):

\[ D^* = \frac{D_{SUB}}{\Gamma(1+\beta)}. \]  

\[ K^* = \frac{\Gamma^2(1+\beta)}{\Gamma(1+2\beta)} - 3, \]  

where \( D_{SUB} \) is defined in (Equation 5). Note the mean kurtosis expression in (Equation 9) was also derived by Ingo et al., 2015 using a different method. Their derivation follows the definition of kurtosis, \( K = \langle x^4 \rangle / \langle x^2 \rangle^2 - 3 \), i.e., by computing the fourth moment \( \langle x^4 \rangle \) and the second moment \( \langle x^2 \rangle \) based on the sub-diffusion Equation 1.

Connectome 1.0 human brain DW-MRI data

The DW-MRI dataset provided by Tian et al., 2022 was used in this study. The publicly available data were collected using the Connectome 1.0 scanner for 26 healthy participants. The first seven subjects had a scan–rescan available. We evaluated qualitatively the seven datasets, and chose the six which had consistent diffusion encoding directions. Subject 2 had 60 instead of 64 diffusion encoding directions, and hence, was omitted from this study. The \( 2 \times 2 \times 2 \) mm\(^3\) resolution data were obtained using two diffusion times (\( \Delta = 19.49 \) ms) with a pulse duration of \( \delta = 8 \) ms and \( G = 31, 68, 105, 142, 179, 216, 253, 290 \) mT/m, respectively, generating \( b \)-values = 50, 350, 800, 1500, 2400, 3450, 4750, 6000 s/mm\(^2\) for \( \Delta = 19 \) ms, and \( b \)-values = 200, 950, 2300, 4250, 6750, 9850, 13500, 17800 s/mm\(^2\) for \( \Delta = 49 \) ms, according to \( b \)-value = \( (\gamma \delta G)^2 (\Delta - \delta / 3) \). 32 diffusion encoding directions were uniformly distributed on a sphere for \( b < 2400 \) s/mm\(^2\) and 64 uniform directions for \( b \geq 2400 \) s/mm\(^2\).

The FreeSurfer’s segmentation labels as part of the dataset were used for brain-region-specific analyses. Tian et al., 2022 provided the white matter averaged group SNR (23.10 ± 2.46), computed from 50 interspersed \( b \)-value = 0 s/mm\(^2\) images for each subject. Both magnitude and the real part of the DW-MRI were provided. Based on an in-depth analysis, the use of the real part of the DW-MRI data was recommended, wherein physiological noise, by nature, follows a Gaussian distribution (Gudbjartsson and Patz, 1995).
Simulated DW-MRI data at specific b-values

DW-MRI data were simulated to establish (i) the correspondence between actual versus fitted mean kurtosis using the traditional DKI and sub-diffusion models based on various choices for $\Delta$, and (ii) to investigate the impact of SNR levels and sub-sampling of b-values on the mean kurtosis estimate. The DW-MRI signal was simulated using the sub-diffusion model (Equation 3) with random Gaussian noise added to every normalised DW-MRI signal instance:

$$S(D_\beta, \beta, q, \Delta) = E_\beta(-D_\beta q^2 \Delta^\beta) + N(0, \sigma^2),$$  \hspace{1cm} (10)

where $N(0, \sigma^2)$ is white noise with mean of zero and standard deviation of $\sigma$ according to the normal distribution.

Two aspects influence $\sigma$ in the case of real-valued DW-MRI data. These include the SNR achieved with a single diffusion encoding direction (i.e., Connectome 1.0 DW-MRI data SNR was derived using only $b$-value $= 0 \text{ s/mm}^2$ data), and the number of diffusion encoding directions in each b-shell across which the powder average is computed:

$$\sigma = \frac{1}{\text{SNR} \sqrt{N_{\text{DIR}}}}.$$

where $N_{\text{DIR}}$ is the number of diffusion encoding directions for each b-shell and assuming it is consistent across b-shells. The $\sigma$ for the Connectome 1.0 data is approximately $1/(23.10 \times 8) = 0.0054$ based on 64 diffusion encoding directions. Achieving of SNR $= 5$, $10$, and $20$ for the simulation study can therefore be accomplished by changing only the $\sigma$ and keeping $N_{\text{DIR}} = 64$. As such, $\sigma_{\text{SNR}=5} = 0.0250$, $\sigma_{\text{SNR}=10} = 0.0125$, and $\sigma_{\text{SNR}=20} = 0.0063$.

Three simulation experiments were carried out at various SNR levels. The first simulation experiment was to examine the effect of the number of diffusion times on the accuracy of the parameter fitting for idealised grey matter and white matter cases. In (Equation 10) the choices of $D_\beta = 3 \times 10^{-4} \text{ mm}^2/\text{s}^\beta$, $\beta = 0.75$, and $D_\beta = 5 \times 10^{-4} \text{ mm}^2/\text{s}^\beta$, $\beta = 0.85$, were made for white matter and grey matter, respectively. These two distinct $\beta$s led to $K^*$ of 0.8125 and 0.4733 using (Equation 9). Diffusion times were chosen from the range $\Delta_t \in [\delta, \delta + 50]$, where diffusion pulse length was set to $\delta = 8$ ms to match the Connectome 1.0 data. A minimum required separation between any two $\Delta$s was enforced, i.e., $30(n - 1)$, where $30$ corresponds with the $30$ ms difference between the $\Delta$s for the Connectome 1.0 DW-MRI data, and $n$ is the number of distinct diffusion times simulated. We considered as many as five distinct diffusion times. Simulations were conducted by randomly selecting sets of $\Delta$s for 1000 instances, and then generating individual DW-MRI simulated signals using (Equation 10), before fitting for $D_\beta$ and $\beta$, from which $K^*$ was computed using (Equation 9). For the case of two diffusion times, suggestions on the separation between them was given based on the goodness-of-fit of the model.

The second simulation experiment was to investigate the effect of the number of diffusion times on the accuracy of parameter fitting. To generate simulated data reflecting observations in human data, $D_\beta$ and $\beta$ were restricted to $D_\beta \in [10^{-4}, 10^{-3}]$ in the unit of $\text{mm}^2/\text{s}^\beta$ and $\beta \in [0.5, 1]$, corresponding to $K^* \in [0, 1.7124]$. For the case of two diffusion times, suggestions on the separation between them were given based on the goodness-of-fit of the model.

The third simulation experiment is to study b-value sub-sampling under various SNR levels (SNR $= 5$, $10$, and $20$). We set the b-values in the simulated data the same as those used to acquire the Connectome 1.0 dataset. At each SNR level, we selected combinations of two, three, and four b-values, irrespective of the difference between them and the diffusion time set to generate the b-value. Essentially, we explored the entire possible sets of b-values for the three regimes, resulting in $120$, $560$, and $1820$ combinations, respectively. A goodness-of-fit measure for model fitting was used to make comparisons between the different b-value combinations.

**SNR reduction by downsampling diffusion encoding directions**

We performed SNR reduction of the Connectome 1.0 DW-MRI data by downsampling of diffusion directions in each b-shell. The method of multiple subsets from multiple sets (P-D-MM) subsampling algorithm provided in DMRITool (Cheng et al., 2018) was applied to the b-vectors provided with the Connectome 1.0 DW-MRI data. Note, the b-shells contained 32 directions if $b < 2400 \text{ s/mm}^2$, and 64
directions if \( b \geq 2400 \text{s/mm}^2 \). We consider SNR = 20 to be the full dataset. The SNR = 10 data was constructed by downsampling to eight non-collinear diffusion encoding directions in each b-shell, and three were required for the SNR = 6 data. In the downsampled data, each diffusion encoding direction was coupled with the direction of opposite polarity (i.e., SNR = 10 had sixteen measurements for each b-value, and SNR = 6 had six).

**Parameter estimation**

**Standard DKI model**

The maximum b-value used to acquire the DW-MRI for standard DKI model fitting is limited to the range 2000 s/mm\(^2\) to 3000 s/mm\(^2\) due to the quadratic form of (Equation 6). We opted to use DW-MRI data generated with b-values = 50, 350, 800, 1500, 2400 s/mm\(^2\) using \( \Delta = 19 \text{ ms} \), and b-values = 200, 950, 2300 s/mm\(^2\) using \( \Delta = 49 \text{ ms} \). Note, the apparent diffusion coefficient, \( D_{DDKI} \) in (Equation 6) is time dependent, as can be deduced from (Equation 5) and (Equation 8). Thereby, standard DKI fitting can only be applied to DW-MRI data generated using a single diffusion time. The model in (Equation 6) was fitted in a voxelwise manner to the powder averaged (i.e., geometric mean over diffusion encoding directions, often referred to as trace-weighted) DW-MRI data using the lsqcurvefit function in MATLAB (Mathworks, Version 2022a) using the trust-region reflective algorithm. Optimisation function specific parameters were set to TolFun = \( 10^{-4} \) and TolX = \( 10^{-6} \). Parameters were bounded to the ranges of \( D_{DKI} > 0 \) and \( 0 < K_{DKI} \leq 3 \).

**Sub-diffusion model**

For the single diffusion time case, the sub-diffusion model in (Equation 3) was fitted to the powder averaged DW-MRI data in a voxelwise manner using the same MATLAB functions as in the previous section. For each subject, spatially resolved maps of \( D_β \) and \( β \) were generated. The fitting strategy for multiple diffusion time data is to solve:

\[
(D_β, β) = \arg\min_{i=1,2,...,n} \sum_{j=1,2,...,J_i} [S_{ij} - \text{SUB} \left( \bar{\Delta}_i, q_j; D_β, β \right)]^2
\]

where \( S_{ij} \) is the signal at the \( i \)th effective diffusion time \( \Delta_i \), and the \( j \)th q-space parameter \( q_j \), \( \text{SUB} \) is the sub-diffusion model (Equation 3) at \( \Delta_i \) and \( q_j \) for a given set of \( (D_β, β) \), \( n \) is the number of diffusion times, and \( J_i \) is the number of q-values corresponding to \( \Delta_i \) in data acquisition. This objective function allows incorporation of an arbitrary number of diffusion times, each having arbitrary number of q-values. Parameters were bounded to the ranges of \( 0 < \beta \leq 1 \) and \( D_β > 0 \) for all the parameter fittings. Model parameters were found to be insensitive to the choice of initial values. Parameters \( D^\star \) and \( K^\star \) were computed analytically using the estimated \( D_β \) and \( β \) according to (Equation 8) and (Equation 9).

**Goodness-of-fit and region-based statistical analysis**

For all of the simulation results, the coefficient of determination, referred to as \( R^2 \), was used to assess how well the mean kurtosis values were able to be fitted. This value was calculated using a standard \( R^2 \) formula

\[
R^2 = 1 - \frac{\sum_i (K_{Simulated,i} - K_{Fitted,i})^2}{\sum_i (K_{Simulated,i} - K_{Simulated})^2}
\]

where \( K_{Simulated} \) is the mean simulated \( K \) value.

For human data, we computed the region specific mean and standard deviation for each subject, and reported the weighted mean and pooled standard deviation along with the coefficient of variation (CV), defined as the ratio of the standard deviation to the mean. The weights were the number of voxels in the associated regions in each subject. Following Barrick et al., 2020, the tissue contrast (TC) is computed as \( TC = \left[ \mu_{WM} - \mu_{GM} \right] / \sqrt{\frac{1}{2} \sigma_{WM}^2 + \sigma_{GM}^2} \), where \( \mu_{WM} \) and \( \mu_{GM} \) are the mean parameter values in white and grey matters; and \( \sigma_{WM} \) and \( \sigma_{GM} \) are the standard deviations of parameter values. Higher TC values indicate greater tissue contrast.
Human data were analysed voxelwise, and also based on regions-of-interest. We considered three categories of brain regions, namely sub-cortical grey matter (scGM), cortical grey matter (cGM), and white matter (WM). The scGM region constituted the thalamus (FreeSurfer labels 10 and 49 for left and right hemisphere), caudate (11, 50), putamen (12, 51), and pallidum (13, 52). The cGM region was all regions (1000–2999) and separately analysed the fusiform (1007, 2007) and lingual (1013, 2013) brain regions, whilst the WM had white matter fibre regions from the cerebral (2, 41), cerebellum (7, 46), and corpus callosum (CC; 251–255) areas. The average number of voxels in each region were 3986 (scGM), 53326 (cGM), 52121 (WM), 1634 (thalamus), 831 (caudate), 1079 (putamen), 443 (pallidum), 2089 (fusiform), 1422 (lingual), 48770 (cerebral WM), 2904 (cerebellum WM), and 447 (CC). For each brain region, a t-test was performed to test for significant differences in mean kurtosis population means under the optimal and sub-optimal b-value sampling regimes presented in Table 5 compared to the benchmark parameter values presented in Table 2.

Scan–rescan analysis using intraclass correlation coefficient (ICC)

For each of the six subjects, both the first (scan) and second (rescan) scan images were registered to the first scan images of Subject 1 using inbuilt MATLAB (Mathworks, Version 2022a) functions (imregtform and imwarp). We used 3D affine registration to account for distortions and warps common in DW-MRI data. Cubic spline interpolation was applied to resample both the scan and rescan DW-MRI data for each subject onto Subject 1’s first scan data grid. The FreeSurfer labels for Subject 1’s first scan were used for brain region analysis. For each voxel, the ICC measure was applied to assess scan–rescan reproducibility of mean kurtosis, as described by Duval et al., 2018 and Fan et al., 2021,

$$\text{ICC} = \frac{s_{\text{inter}}^2}{s_{\text{intra}}^2 + s_{\text{inter}}^2}$$

(14)

with $s_{\text{intra}}^2 = \text{mean}_i \left( \frac{1}{2} \cdot ((m_{i,\text{scan}} - \bar{m}_i)^2 + (m_{i,\text{rescan}} - \bar{m}_i)^2) \right)$ and $s_{\text{inter}}^2 = \text{var}_i(\bar{m}_i)$. Here, $m_{i,\text{scan}}$ is the kurtosis value measured in the voxel for subject $i$, and $\bar{m}_i$ is the average value between scan and rescan. An ICC histogram and the mean and standard deviation descriptive statistics were generated for all brain regions analysed.

Conclusion

The utility of DKI for inferring information on tissue microstructure was described decades ago. Continued investigations in the DW-MRI field have led to studies clearly describing the importance of mean kurtosis mapping to clinical diagnosis, treatment planning and monitoring across a vast range of diseases and disorders. Our research on robust, fast, and accurate mapping of mean kurtosis using the sub-diffusion mathematical framework promises new opportunities for this field by providing a clinically useful, and routinely applicable mechanism for mapping mean kurtosis in the brain. Future studies may derive value from our suggestions and apply methods outside the brain for broader clinical utilisation.

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Author contributions

Megan E Farquhar, Software, Formal analysis, Validation, Investigation, Visualization, Methodology, Writing – original draft, Writing – review and editing; Qianqian Yang, Viktor Vegh, Conceptualization, Software, Formal analysis, Supervision, Funding acquisition, Validation, Investigation, Methodology, Writing – original draft, Project administration, Writing – review and editing

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Ethics

This study uses a publicly available MGH Connectome Diffusion Microstructure Dataset. The institutional review board of Mass General Brigham approved the study protocol and ensured that all relevant ethical regulations were complied with. Written informed consent was obtained from all participants. More details about the MGH Connectome Diffusion Microstructure Dataset are available at https://doi.org/10.1038/s41597-021-01092-6.

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Additional files

Supplementary files
- MDAR checklist

Data availability

The Connectome 1.0 human brain DW-MRI data used in this study is part of the MGH Connectome Diffusion Microstructure Dataset (CDMD) (Tian et al., 2022), which is publicly available on the figshare repository: https://doi.org/10.6084/m9.figshare.c.5315474. MATLAB codes generated for simulation study, parameter fitting, and optimisation of b-value sampling are openly available at https://github.com/m-farquhar/SubdiffusionDKI (copy archived at Farquhar, 2023).
The following previously published dataset was used:

| Author(s) | Year | Dataset title | Dataset URL | Database and Identifier |
|-----------|------|---------------|-------------|-------------------------|
| Tian Q, Fan Q, Witzel T, Polackal MN, Ohringer NA, Ngamsombat C, Russo AW, Machado N, Brewer K, Wang F, Setsompop K, Polimeni JR, Keil B, Wald LL, Rosen BR, Klawiter E, Nummenmaa A, Huang SY | 2022 | Comprehensive diffusion MRI dataset for in vivo human brain microstructure mapping using 300 mT/m gradients | https://doi.org/10.6084/m9.figshare.c.5315474 | figshare, 10.6084/m9.figshare.c.5315474 |

**References**

Aggarwal M, Smith MD, Calabresi PA. 2020. Diffusion-time dependence of diffusional kurtosis in the mouse brain. *Magnetic Resonance in Medicine* **84**:1564–1578. DOI: https://doi.org/10.1002/mrm.28189, PMID: 32022313

Barrick TR, Spilling CA, Ingo C, Madigan J, Issacs JD, Rich P, Jones TL, Magin RL, Hall MG, Howe FA. 2020. Quasi-diffusion magnetic resonance imaging (QDI): A fast, high b-value diffusion imaging technique. *NeuroImage* **211**:116606. DOI: https://doi.org/10.1016/j.neuroimage.2020.116606, PMID: 32032739

Cheng J, Shen D, Yap P-T, Bassier PJ. 2018. Single- and multiple-shell uniform sampling schemes for diffusion MRI using spherical codes. *IEEE Transactions on Medical Imaging* **185**:199–209. DOI: https://doi.org/10.1109/TMI.2017.2756072

Chuhutin A, Hansen B, Jespersen SN. 2017. Precision and accuracy of diffusion kurtosis estimation and the influence of b-value selection. *NMR in Biomedicine* **30**:377. DOI: https://doi.org/10.1002/nbm.3777, PMID: 28841774

Duval T, Smith V, Stikov N, Klawiter EC, Cohen-Adad J. 2018. Scan-rescan of axcaliber, macromolecular tissue volume, and g-ratio in the spinal cord. *Magnetic Resonance in Medicine* **79**:2759–2765. DOI: https://doi.org/10.1002/mrm.26945, PMID: 28994487

Fan Q, Polackal MN, Tian Q, Ngamsombat C, Nummenmaa A, Witzel T, Klawiter EC, Huang SY. 2021. Scan-rescan repeatability of axonal imaging metrics using high-gradient diffusion MRI and statistical implications for study design. *NeuroImage* **240**:118323. DOI: https://doi.org/10.1016/j.neuroimage.2021.118323, PMID: 34216774

Farquhar M. 2023. SubdiffusionDKI. swi:1:rev:d1b3d199a21f9861bd103bb2bf413e91ca38c8fc. Software Heritage. https://archive.softwareheritage.org/swi:1:dir:f7594648d8752f82098dbb999d22868df6c3066;origin=https://github.com/m-farquhar/SubdiffusionDKI;visit=swi:1:snp:bd43d430f2ddc02bdc674803f857dbf9104b968;anchor=swi:1:rev:d1b3d199a21f9861bd103bb2bf413e91ca38c8fc

Goryawala M, Mellon EA, Shim H, Maudsley AA. 2023. Mapping early tumor response to radiotherapy using diffusion kurtosis imaging*. *The Neuroradiology Journal* **36**:198–205. DOI: https://doi.org/10.1177/19714092192112204, PMID: 3600488

Gudbjartsson H, Patz S. 1995. The Rician distribution of noisy MRI data. *Magnetic Resonance in Medicine* **34**:910–914. DOI: https://doi.org/10.1002/mrm.1910340618, PMID: 8598820

Guo M, Shen B, Li J, Huang X, Hu J, Wei X, Wang S, Yuan R, He C, Li Y. 2022a. Diffusion abnormality in temporal lobe epilepsy patients with sleep disorders: a diffusion kurtosis imaging study. *Frontiers in Psychiatry* **13**:885477. DOI: https://doi.org/10.3389/fpsyt.2022.885477, PMID: 35693954

Guo J, Sun W, Dong C, Wu Z, Li X, Zhou R, Xu W. 2022b. Intravoxel incoherent motion imaging combined with diffusion kurtosis imaging to assess the response to radiotherapy in a rabbit VX2 malignant bone tumor model. *Cancer Imaging* **22**:47. DOI: https://doi.org/10.1186/s40644-022-00488-w, PMID: 36064445

Hansen B, Lund TE, Sangill R, Jespersen SN. 2013. Experimentally and computationally fast method for estimation of a mean kurtosis. *Magnetic Resonance in Medicine* **69**:1754–1760. DOI: https://doi.org/10.1002/mrm.24743, PMID: 23589312

Hansen B, Shemesh N, Jespersen SN. 2016. Fast imaging of mean, axial and radial diffusion kurtosis. *NeuroImage* **142**:381–393. DOI: https://doi.org/10.1016/j.neuroimage.2016.08.022, PMID: 27539807

He Y, Chen H, Zhang H, Grimm R, Zhao C, Guo X, Liu Y, Yuan Z. 2022. Optimization of scan parameters to reduce acquisition time for RESOLVE-based diffusion kurtosis imaging (DKI) in nasopharyngeal carcinoma (NPC). *The British Journal of Radiology* **95**:20210641. DOI: https://doi.org/10.1259/bjr.20210641, PMID: 35704453

Heidemann RM, Porter DA, Anwander A, Feiweier T, Heberlein K, Knösche TR, Turner R. 2010. Diffusion imaging in humans at 7T using readout-segmented EPI and GRAPPA. *Magnetic Resonance in Medicine* **64**:9–14. DOI: https://doi.org/10.1002/mrm.22480, PMID: 20577977
Henriques RN, Jespersen SN, Jones DK, Veraart J. 2021. Toward more robust and reproducible diffusion kurtosis imaging. Magnetic Resonance in Medicine 86:1600–1613. DOI: https://doi.org/10.1002/mrm.29515

Hu R, Kim H, Kim J, Allen JW, Sun PZ. 2022. Fast diffusion kurtosis imaging in acute ischemic stroke shows mean kurtosis-diffusivity mismatch. Journal of Neuroimaging 32:941–946. DOI: https://doi.org/10.1111/jon.13000

Huang SY, Witzel T, Keil B, Scholz A, Davids M, Dietz P, Rummert E, Ramb R, Kirsch JE, Yendiki A, Fan Q, Tian Q, Huang SY, Kim H, Kim J, Allen JW, Sun PZ. 2022. Diffusion kurtosis imaging and dynamic contrast-enhanced MRI for the differentiation of parotid gland tumors. European Radiology 32:2748–2759. DOI: https://doi.org/10.1007/s00330-021-08312-y

Huang N, Helpern JA, Ramani A, Lu H, Kaczynski K. 2005. Diffusional kurtosis imaging: the quantification of non-Gaussian water diffusion by means of magnetic resonance imaging. Magnetic Resonance in Medicine 53:1432–1440. DOI: https://doi.org/10.1002/mrm.20508, PMID: 15963600

Ingo C, Magin RL, Parrish TB. 2014. New insights into the fractional order diffusion equation using entropy and kurtosis. Entropy 16:5838–5852. DOI: https://doi.org/10.3390/e16115838, PMID: 25334436

Ingo C, Sui Y, Chen Y, Parrish TB, Webb AG, Ronen I. 2015. Parsimonious continuous time random walk models and kurtosis for diffusion in magnetic resonance of biological tissue. Frontiers in Physics 3:11. DOI: https://doi.org/10.3389/fphys.2015.00011, PMID: 25334472

Istratov AA, Vyvenko O. 1999. Exponential analysis in physical phenomena. Review of Scientific Instruments 70:1233–1257. DOI: https://doi.org/10.1063/1.1149581

Jeliascu IO, de Skowronski A, Geffroy F, Palombo M, Novikov DS. 2022. Neurite Exchange Imaging (NEXI): A minimal model of diffusion in gray matter with inter-compartment water exchange. NeuroImage 256:119277. DOI: https://doi.org/10.1016/j.neuroimage.2022.119277, PMID: 35523369

Jensen JH, Helsper JA, Ramani A, Lu H, Kaczynski K. 2005. Diffusional kurtosis imaging: the quantification of non-gaussian water diffusion by means of magnetic resonance imaging. Magnetic Resonance in Medicine 53:1432–1440. DOI: https://doi.org/10.1002/mrm.20508, PMID: 15963600

Jensen JH, Helsper JA. 2010. MRI quantification of non-Gaussian water diffusion by kurtosis analysis. NMR in Biomedicine 23:698–710. DOI: https://doi.org/10.1002/nbm.1518, PMID: 20632416

Kiselev VG. 2010. The cumulant expansion: an overarching mathematical framework for understanding diffusion NMR. Diffusion MRI Oxford University Press. DOI: https://doi.org/10.1093/med/9780195369779.003.0010

Kraff O. 2017. 7T: Physics, safety, and potential clinical applications. Journal of Magnetic Resonance Imaging 46:1573–1589. DOI: https://doi.org/10.1002/jmri.25723, PMID: 28370675

Kuder TA, Quick HH. 2017. 7T: Physics, safety, and potential clinical applications. Journal of Magnetic Resonance Imaging 46:1573–1589. DOI: https://doi.org/10.1002/jmri.25723, PMID: 28370675

Lebel C, Treit S, Beaulieu C. 2019. A review of diffusion MRI of typical white matter development from early childhood to young adulthood. NMR in Biomedicine 32:e3778. DOI: https://doi.org/10.1002/nbm.3778, PMID: 28886240

Le Bihan D, Mangin JF, Poupon C, Clark CA, Pappata S, Molko N, Chabriat H. 2001. Diffusion tensor imaging: concepts and applications. Journal of Magnetic Resonance Imaging 13:534–546. DOI: https://doi.org/10.1002/jmri.1076, PMID: 11276097

Le Bihan D, Johansen-Berg H. 2012. Diffusion MRI at 25: exploring brain tissue structure and function. NeuroImage 61:324–341. DOI: https://doi.org/10.1016/j.neuroimage.2011.11.006

Lee HH, Papaoianou A, Novikov DS, Fieremans E. 2020. In vivo observation and biophysical interpretation of time-dependent diffusion in human cortical gray matter. NeuroImage 222:117054. DOI: https://doi.org/10.1016/j.neuroimage.2020.117054, PMID: 32585341

Li A, Yuan G, Hu Y, Shen Y, Hu X, Hu D, Li Z. 2022a. Renal functional and interstitial fibrotic assessment with non-Gaussian diffusion kurtosis imaging. Insights into Imaging 13:01215-6. DOI: https://doi.org/10.1186/s13244-022-01215-6, PMID: 35394225

Li HW, Yan GW, Yang J, Zhuo LH, Bhetuwal A, Long YJ, Feng X, Yao HC, Zou XX, Feng RH, Yang HF, Du Y. 2022b. Quantitative analysis for detection and grading of hepatocellular carcinoma: Comparison of diffusion kurtosis imaging, intravoxel incoherent motion and conventional diffusion-weighted imaging. Oncology Letters 24:403. DOI: https://doi.org/10.3892/ol.2022.13523, PMID: 36276491

Li Q, Cao B, Liu K, Sun H, Ding Y, Yan C, Wu P, Dai C, Rao S, Zeng M, Jiang S, Zhou J. 2022d. Detecting the muscle invasiveness of bladder cancer: An application of diffusion kurtosis imaging and tumor contact length. European Journal of Radiology 151:110329. DOI: https://doi.org/10.1016/j.ejrad.2022.110329, PMID: 35487092

Li Q, Zhang GMY, Peng X, Li X, Sun H, Chen L. 2022. Diffusion kurtosis imaging as an imaging biomarker for predicting prognosis in chronic kidney disease patients. Nephrology Dialysis Transplantation 37:1451–1460. DOI: https://doi.org/10.1093/ndt/gfab229
Magin RL, Hall MG, Karaman MM, Vegh V. 2020. Fractional calculus models of magnetic resonance phenomena: relaxation and diffusion. Critical Reviews in Biomedical Engineering 48:285–326. DOI: https://doi.org/10.1615/CritRevBiomedEng.202003925, PMID: 33639049

Maiter A, Riemer F, Allinson K, Zaccagna F, Crispin-Ortuza M, Gehring M, McLean MA, Priest AN, Grist J, Matys T, Graves MJ, Gallagher FA. 2021. Investigating the relationship between diffusion kurtosis tensor imaging (DKTI) and histology within the normal human brain. Scientific Reports 11:8857. DOI: https://doi.org/10.1038/s41598-021-87857-w, PMID: 33893238

Maralakunte M, Gupta V, Grover S, Ahuja CK, Sahoo S, Kishore K, Vyas S, Garg G, Singh P, Govind V. 2023. Cross-sectional analysis of whole-brain microstructural changes in adult patients with bipolar and unipolar depression by diffusion kurtosis imaging. The Neuroradiology Journal 36:176–181. DOI: https://doi.org/10.1177/1971409221114446, PMID: 35817080

Metzler R, Klafter J. 2000. The random walk’s guide to anomalous diffusion: a fractional dynamics approach. Physics Reports 339:1–77. DOI: https://doi.org/10.1016/S0370-1573(00)00070-3

Olesen JL, Østergaard L, Shemesh N, Jespersen SN. 2022. Diffusion time dependence, power-law scaling, and exchange in gray matter. NeuroImage 251:118976. DOI: https://doi.org/10.1016/j.neuroimage.2022.118976, PMID: 35168088

Pohmann R, Speck O, Scheffler K. 2016. Signal-to-noise ratio and MR tissue parameters in human brain imaging at 3, 7, and 9.4 tesla using current receive coil arrays. Magnetic Resonance in Medicine 75:801–809. DOI: https://doi.org/10.1002/mrm.25677, PMID: 25820458

Poot DHJ, den Dekker AJ, Achten E, Verhoey M, Sijbers J. 2010. Optimal experimental design for diffusion kurtosis imaging. IEEE Transactions on Medical Imaging 29:819–829. DOI: https://doi.org/10.1109/TMI.2009.2037915

Shafto MA, Tyler LK, Dixon M, Taylor JR, Rowe JB, Cusack R, Calder AJ, Marslen-Wilson WD, Duncan J, Dalglish T, Henson RN, Brayne C, Matthews FE, Cam-CAN. 2014. The Cambridge centre for ageing and neuroscience (Cam-CAN) study protocol: a cross-sectional, lifespan, multidisciplinary examination of healthy cognitive ageing. BMC Neurology 14:204. DOI: https://doi.org/10.1186/s12883-014-0204-1, PMID: 25412575

Spilling CA, Howe FA, Barrick TR. 2022. Optimization of quasi-diffusion magnetic resonance imaging for quantitative accuracy and time-efficient acquisition. Magnetic Resonance in Medicine 88:2532–2547. DOI: https://doi.org/10.1002/mrm.29420, PMID: 36054778

Tabesh A, Jensen JH, Ardekani BA, Helpren JA. 2011. Estimation of tensors and tensor-derived measures in diffusional kurtosis imaging. Magnetic Resonance in Medicine 65:823–836. DOI: https://doi.org/10.1002/mrm.22655, PMID: 21337412

Tian Q, Fan Q, Witzel T, Polackal MN, Ohringer NA, Ngamsombat C, Russo AW, Machado N, Brewer K, Wang F, Setsompop K, Polimeni JR, Keil B, Wald LL, Rosen BR, Kwiter EC, Nummenmaa A, Huang SY. 2022. Comprehensive diffusion MRI dataset for in vivo human brain microstructure mapping using 300 mT/m gradients. Scientific Data 9:7. DOI: https://doi.org/10.1038/s41597-021-0192-6, PMID: 35042861

Tournier J-D. 2019. Diffusion MRI in the brain – theory and concepts. Progress in Nuclear Magnetic Resonance Spectroscopy 112–113:1–16. DOI: https://doi.org/10.1016/j.pnms.2019.03.001

Van Essen DC, Smith SM, Barch DM, Behrens TEJ, Yacoub E, Ugurbil K, WU-Minn HCP Consortium. 2013. The WU-Minn human connectome project: an overview. NeuroImage 80:62–79. DOI: https://doi.org/10.1016/j.neuroimage.2013.05.041, PMID: 23684880

Veraart J, Van Hecke W, Sijbers J. 2011a. Constrained maximum likelihood estimation of the diffusion kurtosis tensor using a Rician noise model. Magnetic Resonance in Medicine 66:678–686. DOI: https://doi.org/10.1002/mrm.22835, PMID: 21416503

Veraart J, Poot DHJ, Van Hecke W, Blockx I, Van der Linden A, Verhoey M, Sijbers J. 2011b. More accurate estimation of diffusion tensor parameters using diffusion Kurtosis imaging. Magnetic Resonance in Medicine 65:138–145. DOI: https://doi.org/10.1002/mrm.22603, PMID: 20878760

Wang M-L, Wei X-E, Yu M-M, Li W-B. 2022. Cognitive impairment in mild traumatic brain injury: a diffusion kurtosis imaging and volumetric study. Acta Radiologica 63:504–512. DOI: https://doi.org/10.1177/0284185121998317

Wu EX, Cheung MM. 2010. MR diffusion kurtosis imaging for neural tissue characterization. NMR in Biomedicine 23:836–848. DOI: https://doi.org/10.1002/nbim.1506, PMID: 20623793

Yablonskiy DA, Sukstanskii AL. 2010. Theoretical models of the diffusion weighted MR signal. NMR in Biomedicine 23:661–681. DOI: https://doi.org/10.1002/nbim.1520, PMID: 20886562

Yang Q, Reutens DC, Vegh V. 2022. Generalisation of continuous time random walk to anomalous diffusion MRI models with an age-related evaluation of human corpus callosum. NeuroImage 250:118903. DOI: https://doi.org/10.1016/j.neuroimage.2022.118903

Zelinski AC, Angelone LM, Goyal VK, Bonmassar G, Adalsteinsson E, Wald LL. 2008. Specific absorption rate studies of the parallel transmission of inner-volume excitations at 7T. Journal of Magnetic Resonance Imaging 28:1005–1018. DOI: https://doi.org/10.1002/jmri.21548, PMID: 18821601

Zhou Z, Chen Y, Zhao F; Sun Z, Zhu L, Yu H, Wang W. 2023. Predictive value of intravoxel incoherent motion combined with diffusion kurtosis imaging for breast cancer axillary lymph node metastasis: a retrospective study. Acta Radiologica 64:951–961. DOI: https://doi.org/10.1177/02841851221107626

Zong F, Du J, Deng X, Chai X, Zhuo Y, Vegh AV, Xue R. 2021. Fast diffusion kurtosis mapping of human brain at 7 Tesla with hybrid principal component analyses. IEEE Access 9:107965–107975. DOI: https://doi.org/10.1109/ACCESS.2021.3100546