Separation of Sodium Signals Between Mono- and Bi-Exponential T_2 Decays via Multi-TE Single-Quantum Sodium (^{23}Na) MRI

Yongxian Qian,[1] Ying-Chia Lin,[1] Xingye Chen,[1,2] Tiejun Zhao,[3] Karthik Lakshmanan,[1] Yulin Ge,[1] Yvonne W. Lui,[1,4] Fernando E. Boada [1,5]

[1] Bernard and Irene Schwartz Center for Biomedical Imaging, Department of Radiology, New York University Grossman School of Medicine, New York, NY 10016.
[2] Vilcek Institute of Graduate Biomedical Sciences, NYU Grossman School of Medicine, New York, NY 10016.
[3] Siemens Medical Solutions USA, New York, NY 10016.
[4] Department of Radiology, NYU Langone Health, New York, NY 10016.
[5] Now at Department of Radiology, Stanford University, Stanford, CA 94305.

Running title: Mono- and bi-T2 sodium separation
Manuscript type: Research Article **Word Count (body):** 5451
Key Words: sodium MRI, single-quantum MRI, triple-quantum MRI, neuroimaging, neurodegeneration

Correspondence: Yongxian Qian, PhD
Center for Biomedical Imaging (CBI)
660 First Ave, 4th Floor
New York, NY 10016, USA
Phone: 212-263-1159
Email: Yongxian.Qian@nyulangone.org

This work was partially presented in the 25th Annual Meeting of ISMRM, Honolulu, Hawaii, USA, 2017.

NIH-Funding Required Statement: Research reported in this publication was supported in part by the National Institute On Aging of the National Institutes of Health under Award Number

1. INTRODUCTION

In human brains, sodium ions (Na^+), when exposed to an electric field gradient of negatively charged macromolecules and proteins, experience nuclear quadrupolar interaction that results in bi-exponential decay in transverse (T_2) relaxation of nuclear spins when the ions are not in fast motion, a situation in which correlation time between sodium ions and electric field gradient is much shorter than the inverse of Larmor frequency, $\tau_c << 1/\omega_0$.[1,2] Sodium ions in fast motion cancel out the effect of quadrupolar interactions, resulting in mono-exponential T_2 decay.[1-4] Sodium ions in bi-exponential T_2 decay were historically, but incorrectly, considered as invisible "bound" (chemically) sodium [5,6] because their short-T_2 components were not detectable by then-NMR (nuclear magnetic resonance) techniques.[1-4] The terminology however remains in use in today's sodium MRI, although concerns have been raised recently by some researchers, with no better substitutes yet.[7] For convenience, this article refers "bound" (not chemically) sodium to those showing bi-exponential T_2 decay and "free" sodium to those showing mono-exponential T_2 decay. The free and bound sodium ions can appear in both intra- and extra-cellular spaces,[8-10] depending on their relative correlation time with electric field gradient.[1,2]

Sodium (^{23}Na) MRI currently acquires signals from both free and bound sodium ions, and quantifies total sodium concentration (TSC) at voxels of an image. TSC is unique measure for non-invasive assessment of disruption in ionic homeostasis of cells in, or recovery from, pathological conditions such as stroke, tumor, multiple scleroses, epilepsy, bipolar disorder, and mild traumatic brain injury.[8-12] However, TSC is dominated by free sodium from cerebrospinal fluid (CSF) which has a high sodium concentration (~145 mM) and overshadows alteration in intracellular sodium which has a much lower concentration (~15 mM). Separation of mono- and bi-T_2 sodium signals can remove CSF impact and highlight intracellular alterations, especially at early stage of a disease happening at cellular level or in early (cellular) response to a treatment.

The difference in T_2 relaxation was extensively explored in sodium MRI as a means to separate the two populations of sodium in the brain. Triple quantum filtering (TQF) was considered a standard for human studies, in which MR signals were generated solely from triple-quantum (TQ) transitions.[13] TQF techniques, however, require multiple radiofrequency (RF) pulses for excitation and multi-step phase cycling to eliminate single-quantum (SQ) signals,[13-17] leading to long scan time (20–40 min) and high specific absorption rate (SAR, causing a safety concern).

More problematic is that TQF has much low signal-to-noise ratio (SNR) about 10 times lower than SQ.[15-17] These difficulties hamper TQF to be widely used on humans.

Alternative approaches were proposed. Inversion recovery (IR), adopted from proton (^{1}H) MRI, exploits a difference in T_1 relaxation between the mono- and bi-T_2 sodium ions, and suppresses signals from the mono-T_2 sodium of longer T_1 time.[18-20] The IR approach needs an extra RF pulse for the suppression and worsens SAR issue, not favorable to human studies. It also suffers from incomplete suppression of the mono-T_2 sodium signals which are ~10 times higher than the bi-T_2 sodium signals, due to spatial inhomogeneity of B_1^+ field although adiabatic pulses are usually used, and complicates quantification of the bi-T_2 sodium owing to unknown residual mono-T_2 sodium signals.[9,11,18] To overcome these drawbacks, another alternative approach, called short-T_2 imaging, was proposed in which SQ images were acquired at multiple echo times (TEs) and then subtracted from each other to produce an image of the short-T_2 component of bi-T_2 sodium.[21-24] In such a way, SAR was reduced to, and SNR was increased to, the level of SQ images, favorable to human studies in clinic. Unfortunately, the subtraction could not completely eliminate mono-T_2 sodium signal (~20% in residual), degrading accuracy of bi-T_2 sodium quantification.[24]

In this study, the short-T_2 imaging is generalized to multi-TE single-quantum (MSQ) imaging to improve accuracy of the separation between mono- and bi-T_2 sodium signals, by replacing the subtraction with a matrix inversion. To develop MSQ technique, we optimized the TEs for data acquisition, investigated impact of T_2 values on accuracy of the separation, and acquired the free induction decay (FID) signals to generate T_2* spectrum for the matrix equation. To test MSQ technique, we implemented numerical simulations, phantom studies, and human studies. The results were supportive of the proposed MSQ technique. We also itemized limitations of MSQ technique and potential pitfalls in interpretation of separated sodium signals.

2. THEORY

2.1. Model of sodium signals

A two-population model is used to describe single-quantum sodium signal $m(t)$ evolving with time t at an imaging voxel ΔV.

$$m(t) = m_{fr}\, Y_{fr}(t) + m_{bd}\, Y_{bd}(t),\ t \geq 0 \qquad \text{Eq. [1a]}$$

$$m_{fr} \geq 0,\ m_{bd} \geq 0, \text{ and } m_{fr} + m_{bd} = m(0)$$

$$Y_{fr}(t) \equiv \exp(-t/T_{2,fr}) \quad \text{Eq. [1b]}$$

$$Y_{bd}(t) \equiv a_{bs} \exp(-t/T_{2,bs}) + a_{bl} \exp(-t/T_{2,bl}) \quad \text{Eq. [1c]}$$

m_{fr} and m_{bd} are signal intensity proportional to volume fraction v_q and sodium concentration C_q, i.e., $m_q \propto \Delta V(v_q C_q)$, q=fr and bd, for the mono-T_2 (or free) and bi-T_2 (or bound) sodium populations in a voxel ΔV, respectively. $Y_{fr}(t)$ is relaxation decay of the free sodium of time constant $T_{2,fr}$, and $Y_{bd}(t)$ is relaxation decay of the bound sodium with $a_{bs} = 0.6$ for the short-T_2 component ($T_{2,bs}$) and $a_{bl} = 0.4$ for the long-T_2 component ($T_{2,bl}$). The split of 60% vs. 40% in intensity is from theoretical and experimental results for individual sodium nuclear spins.[1-4,49] The T_2 values are in an order of $T_{2,bs} << T_{2,bl} \leq T_{2,fr}$. Model Eq. 1 doesn't include free sodium of short T_2.

2.2. Separation of the mono- and bi-T_2 sodium signals

Given SQ sodium images at $\{TE_1, TE_2, \ldots, TE_N\}$, Eq. 1a becomes a matrix equation Eq. 2.

$$\mathbf{M} = \mathbf{Y}\,\mathbf{X} \quad \text{Eq. [2a]}$$

$$\mathbf{M} \equiv (m_1, m_2, \ldots, m_N)^T, \qquad m_i = m(TE_i), i = 1,2,\ldots,N \quad \text{Eq. [2b]}$$

$$\mathbf{X} \equiv (m_{fr}, m_{bd})^T \quad \text{Eq. [2c]}$$

$$\mathbf{Y} \equiv \begin{pmatrix} Y_{fr}(TE_1) & Y_{bd}(TE_1) \\ Y_{fr}(TE_2) & Y_{bd}(TE_2) \\ \vdots & \vdots \\ Y_{fr}(TE_N) & Y_{bd}(TE_N) \end{pmatrix} \quad \text{Eq. [2d]}$$

Superscript T is operator for matrix transpose. A solution to Eq. 2 is given by Eq. 3 via an established algorithm called non-negative least-squares (NNLS)[25] in which non-negative condition on $\mathbf{X}$ is incorporated into the solution.

$$\mathbf{X} = NNLS(\mathbf{YX} - \mathbf{M}) \quad \text{Eq. [3]}$$

2.3. Measurement of T_2 values for the mono- and bi-T_2 sodium populations

The T_2 values ($T_{2,fr}$, $T_{2,bs}$, $T_{2,bl}$) in Eqs. 1b and 1c are required to perform the separation. They can be measured by acquiring FID signal $s(t)$ on whole imaging volume and using effective T_2 (i.e., T_2*) decay model of multi-exponential components to fit the FID signal in magnitude, i.e.,

$$|s(t)| = \sum_j A_j \exp(-t/T_{2,j}^*) \quad \text{Eq. [4]}$$

Hereinafter, T_2* replaces T_2 as spin echo is not favorable to sodium MRI. The curve-fitting is accomplished also through the NNLS algorithm [25] when T_2* values are pre-distributed in a range

of interest [T_2*$_{min}$, T_2*$_{max}$] at uniform or non-uniform intervals $\{\Delta T_{2,j}^*, j = 1, 2, ...\}$. Amplitudes $\{A_j\}$, called T_2* spectrum, determine relative incidence of T_2* components in the imaging volume which counts all T_2* components from both mono- and bi-T_2 sodium populations. To pair up the short- and long-T_2* components of the bi-T_2 sodium, the 60-40 split in intensity is a helpful guidance.

Alternatively, empirical estimates of the brain tissues may be applicable to T_2* values, because solutions to Eq. 2 are not so sensitive to T_2* values due to exponential decay (see Sections 2.4 and 4.2 for details).

FID signals, when acquired with an array coil, may have unique initial phases $\{\varphi_{0,l}, l=1, 2, \ldots, N_c\}$ at individual elements, and need to be aligned to produce a resultant FID signal. Alignment (via phase correction) can be towards a reference phase such as zero phase, one of the initial phases, or mean phase across elements. In addition, signal intensity at individual elements needs to be scaled via "FFT factor" which is stored in the header of a raw FID data file.

FID signals at the first few samples are distorted by hardware filtering during analog-to-digital conversion (ADC). The number of affected samples are in a range of 3–10 points, depending on sampling bandwidth, with the first sample having the largest distortion. This distortion alters measurement of T_2* components, especially the short T_2* components which are critically important to the bi-T_2 sodium. Correction for the distortion can be performed using an established exponential extrapolation (see Appendix A for details).

2.4. Impact of T_2 values on the mono- and bi-T_2 sodium separation*

Ideally, T_2* values are measured at individual voxels. Practically, the measurement is time consuming and not favorable in clinical studies. A fast estimate is necessary. Intuitively, solutions $\{m_{fr}, m_{bd}\}$ to Eq. 1 are not very sensitive to T_2* values due to exponential decays in Eqs. 1b and 1c. This observation can be verified theoretically and numerically. Theoretically, small changes in T_2* values, $\{\delta T_{2,q}^*, q=fr, bs, bl\}$, lead to small changes $\{\delta m_p, p=fr, bd\}$ in m_{fr} and m_{bd} under the same $m(t)$, that is,

$$0 = \delta(m_{fr}Y_{fr}) + \delta(m_{bd}Y_{bd}) \quad \text{Eq. [5a]}$$

$$-m_\delta = Y_{fr}\delta m_{fr} + Y_{bd}\delta m_{bd} \quad \text{Eq. [5b]}$$

$$m_\delta \equiv m_{fr}\delta Y_{fr} + m_{bd}\delta Y_{bd} \quad \text{Eq. [5c]}$$

$$\delta Y_q = \left(\frac{tY_q}{T^*_{2,q}}\right)\left(\frac{\delta T^*_{2,q}}{T^*_{2,q}}\right) \leq e^{-1}\left(\frac{\delta T^*_{2,q}}{T^*_{2,q}}\right), \quad q=fr,\ bs,\ bl \qquad \text{Eq. [5d]}$$

$$\delta Y_{bd} = a_{bs}\,\delta Y_{bs} + a_{bl}\,\delta Y_{bl} \leq e^{-1}\left[a_{bs}\left(\frac{\delta T^*_{2,bs}}{T^*_{2,bs}}\right) + a_{bl}\left(\frac{\delta T^*_{2,bl}}{T^*_{2,bl}}\right)\right] \qquad \text{Eq. [5e]}$$

Where δ is difference operator.

Numerically, errors in m_{fr} and m_{bd} can be calculated, given a series of $\{T^*_{2,q}, \delta T^*_{2,q}; q=fr, bs, bl\}$ at a specific pair (m_{fr}, m_{bd}). This creates a plot showing how the computed (m_{fr}, m_{bd}) change with $\{\delta T^*_{2,q}; q=fr, bs, bl\}$.

2.5. Optimalization of the number of TEs

In principle, the more TEs the better differentiation between the T_2* relaxations of the mono- and bi-T_2 sodium populations, and the better solutions to Eq. 2. In practice, the number of TEs is restricted by total scan time (TA), SNR, signal decay, and risk of motion artifacts across TEs. Therefore, a trade-off must be made for the number of TEs. To determine an optimal number of TEs, it is necessary to understand noise propagation in Eq. 2. Let singular value decomposition (SVD) [26] of the matrix **Y** in Eq. 2a be

$$\mathbf{Y} = \mathbf{U}\,\mathbf{\Sigma}\,\boldsymbol{V}^T \qquad \text{Eq. [6a]}$$

$$\mathbf{\Sigma} = \mathrm{diag}(\sigma_1, \sigma_2) \qquad \text{Eq. [6b]}$$

$$\mathbf{X} = \mathbf{V}\,\mathbf{\Sigma^{-1}}\,\mathbf{U}^{\mathrm{T}}\,\mathbf{M} \qquad \text{Eq. [6c]}$$

Singular values (σ_1, σ_2) determine noise transfer (amplification or suppression) in Eq. 6c from the measured TE-images **M** to the separated mono- and bi-T_2 sodium images **X**.

However, Eq. 6c allows negative values in **X** when random noise contaminates **M**. This violates the "non-negative" condition on **X**. Therefore, SVD analysis is applicable only to **X** elements with SNR ≥ 2 where the elements, with Gaussian noise, have 95.4% of chance in the territory of non-negative value.[27]

3. METHODS

The proposed MSQ technique is graphically illustrated in Fig. 1. The inputs are multiple TE images and an FID signal. The outputs are the mono-T_2 (free), bi-T_2 (bound), and total sodium images, as well as the map of field inhomogeneity ΔB_0 and the map of single-T_2*. In between are the data

processing functionalities for the T_2* spectrum, mono- and bi-T_2 sodium separation, and mapping of ΔB_0 and single-T_2*. Motion correction (MoCo) across multi-TE images is optional. Also optional is the low-pass (LP) filtering, which is a 3D averaging over a size of 3×3×3 voxels for instance, to reduce random noise on the bi-T_2 sodium. The ΔB_0 and single-T_2* maps present spatial distributions of the B_0 field inhomogeneity and the short- and long-T_2* components, and provide indications for uncertain short-T_2* decays possibly caused by the B_0 inhomogeneity. These maps are critical and complimentary to quantification and explanation of the separated mono- and bi-T_2 sodium signals.

To implement the MSQ technique, a computer software was custom-developed in MATLAB (R2021a, MathWorks, Natick, MA) on a laptop computer (MacBook Pro, 16GB memory, Apple M1 chip, Apple Inc., Cupertino, CA).

The numerical simulations in this study were performed in MATLAB on the MacBook Pro laptop or Windows desktop, otherwise specified. Random noise of Gaussian distribution was generated using MATLAB function *randn(n)*, while the NNLS algorithm was implemented using [*x*, *resnorm*, *residual*] = *lsqnonneg(C, d)*.

3.1. Measurement of T_2 values*

FID signals were acquired on whole brain of the study subjects right before sodium imaging. A product pulse sequence, either *AdjXFre* embedded in the manual shimming task or independent *fid_23Na*, was employed with parameters: TE=0.35–1.0ms, TR=100–300ms, and averages =1–128, and TA=0.2–39s. When a dual-tuned (^{1}H-^{23}Na) 8-channel Tx/Rx head array coil [28] was used, there was a difference in initial phase and in FFT scale factors across channels. The initial phases were measured on channel image at central slice, and removed by aligning to zero phase. Channel signals were weighted with FFT scale factors and complex-value combined into a resultant FID. An additional step was performed to correct for distortion at the first few points of the resultant FID signal. Finally, a spectrum of T_2* values was calculated according to Eq. 4, at a resolution of 0.5ms for T_2* in 0.5–100ms. Assignment of T_2* peaks to $\{T_2^*_{fr}, T_2^*_{bs}, T_2^*_{bl}\}$ was based on their relative positions and intensities, i.e., $T_2^*_{fr} \geq T_2^*_{bl} >> T_2^*_{bs}$ and intensity ratio 6:4 for the bi-T_2 sodium. Repeatability of the T_2* measurement is addressed in the Supporting Information.

3.2. Sensitivity to T_2 values*

As a testing, a set of T_2* values, $\{T^*_{2,fr}, T^*_{2,bs}, T^*_{2,bl}\}$ = {50.0, 3.5, 15.0}ms, most commonly encountered in our human brain studies, was employed. Then, an error of $\delta T^*_{2,q}$, *q=fr*, *bs*, *bl*, in a range of ± 20% was added to the testing values. Finally, $\{m_{fr}, m_{bd}\}$ and errors $\{\delta m_{fr}, \delta m_{bd}\}$ relative to the true values in a range of $0.1 \leq m_{fr} \leq 0.9$, were calculated according to Eq. 3. The relationships between $\{\delta m_{fr}, \delta m_{bd}\}$ and $\{\delta T^*_{2,q}; q=fr, bs, bl\}$ were plotted. To focus on the relation of "$\delta T_2^* - \delta m$", TEs were sampled in an ideal case, i.e., TE_0= 0, ΔTE=1ms, and 80 TEs covering entire T_2* decays.

3.3. Optimization of TEs

The simulations were implemented via Eq. 6 for three cases: an ideal case serving as reference, practical case 1 having a large number of TEs, and practical case 2 having a small number of TEs. The ideal case had 80 TEs, i.e., TE = (0, 1, 2, …, 79) ms, to cover entire range of T_2* decays. The two practical cases, suggested by existing human studies,[24,29-31] had total scan time (TA) limited to 22min for 8 TE-images, and the TEs were chosen to be most sensitive to T_2* decays.[29] Thus, the case 1 had 8 TEs = (0.5, 1, 2, 3, 4, 5, 7, 10) ms, and the case 2 only had two TEs = (0.5, 5.0) ms but 4 averages at each TE. The SVD singular values ($\sigma_1 \geq \sigma_2 \geq 0$) were calculated for each set of TEs via Eq. 6. The optimal set of TEs would be the one that had σ_2 producing the minimum amplification of random noise in the separated mono- and bi-T_2 sodium images.

3.4. Computer simulations

The mono- and bi-T_2 sodium separation was carried out via Eq. 3 at a typical set of T_2* values, $(T^*_{2,fr}, T^*_{2,bs}, T^*_{2,bl})$ = (50.0, 3.5, 15.0) ms, and an optimal two-TE scheme, TE = (0.5, 5.0) ms. Sodium signals were calculated via Eq. 1, with an additive Gaussian noise, $N(0, \sigma^2)$, at each of noise trials (independent from each other), $m(t) + n(t)$. The mono- and bi-T_2 signal amplitudes $\{m_{fr}, m_{bd}\}$ were simulated to vary in a normalized range of 0.0–1.0 at a step size = 0.1. The separation was implemented using the function *lsqnonneg*() in MATLAB, and repeated N_{noise} times at each of the specific amplitudes. Mean and standard deviation (SD) were reported as separated sodium signal. N_{noise}=1054 was chosen to detect a 10% of SD, or 0.1 effect size d=Δμ/SD, in difference between the mean and true value at 90% power and 5% significant level under the two-sided Student's *t*-test.[27]

3.5. Phantom studies

Four phantoms, custom-built and described in previous work,[24] were studied. They were 50-mL centrifuge tubes filled with a mixture of distilled water, 10% w/w agar powder, and sodium chloride (NaCl) at three concentrations (90, 120, and 150mM) and at 150mM without agar, mimicking bi- and mono-T_2 sodium signals in the brain tissues. Sodium MRI was performed on a clinical scanner at 3T (MAGNETOM Trio Tim, Siemens Medical Solutions, Erlangen, Germany) with a dual-tuned (^{1}H-^{23}Na) volume head coil (Advanced Imaging Research, Cleveland, OH). The data acquisition was implemented using an SNR-efficient, three-dimensional (3D) pulse sequence called the twisted projection imaging (TPI),[32] with parameters: rectangular RF pulse duration = 0.8ms, flip angle=80º (limited by SAR and TR), field of view (FOV)=220mm, matrix size=64, nominal resolution=3.44mm (3D isotropic), TPI readout time=36.32ms, total TPI projections =1596, TPI *p*-factor=0.4, TR=100ms, TE_1/TE_2=0.5/5ms, averages=4, and TA=10.64min per TE-image. The image reconstruction was offline implemented on a desktop computer (OptiPlex 7050, 8GB memory, Windows 10, DELL, Round Rock, TX) using a custom-developed programs in C++ (MS Visual Studio 2012, Microsoft, Redmond, WA). Separation of the mono- and bi-T_2 sodium signals was implemented using a custom-developed program as described above.

3.6. Human studies

The human studies were conducted with the approval of local Institutional Review Board (IRB) at NYU Grossman School of Medicine, New York, NY, USA. The study subjects included nine healthy adults (age 39.6±21.4 years between 21–74 years; 3 males and 6 females) and six patients with diverse neurological disorders (1 bipolar disorder, 3 epilepsy, 1 multiple sclerosis, and 1 mild traumatic brain injury; age 30.5±15.1 years between 18–59 years; 3 males and 3 females), after the exclusion of one subject and one patient due to motion between the two TE-images. The study was performed on a clinical 3T MRI scanner (Prisma, Siemens Healthineers, Erlangen, Germany) with a custom-built 8-channel dual-tuned (^{1}H-^{23}Na) head array coil.[28] The same TPI pulse sequence as in the phantom studies was used for data acquisition. Images were reconstructed using the gridding algorithm,[33,34] off-line and channel-by-channel, and combined into a resultant image via the sum-of-squares (SOS) algorithm.[35] To decouple random noise across channels, an orthogonal linear transform (detailed in Ref. 30) was performed in which physical channel data

were transformed into virtual channels with random noise independent from channel to channel. This decoupling and denoising process also normalized signal amplitudes across channels by dividing noise standard deviation. Separation of the mono- and bi-T_2 sodium signals was implemented in the same way as in the phantom studies.

*3.7. Mapping of ΔB_0 and single-T_2**

To map ΔB_0 (or $\Delta f_0 = \gamma \Delta B_0/2\pi$), Hermitian product method [36] was performed via Eq. 7 at individual imaging voxels to calculate phase differences $\{\Delta\varphi_i, i = 1, 2, \ldots, N-1\}$ between TEs $\{TE_i, i = 1, 2, \ldots, N\}$. Image amplitude at individual channels were corrected with the FFT factors $\{w_l, l = 1, 2, \ldots, N_c\}$. Phase unwrapping was not performed due to small intervals in the TEs and, in general, small inhomogeneity in the B_0 field in sodium MRI. Computation for ΔB_0 map is fast (0.078s) on a Mac laptop computer for images of size 64×64×64 at two TEs.

$$\Delta f_0 = \frac{1}{2\pi(N-1)} \sum_{i=1}^{N-1} \Delta\varphi_i / \Delta TE_i \qquad \text{Eq. [7a]}$$

$$\Delta\varphi_i = phase\{\sum_{l=1}^{Nc} w_l^2 \cdot m_l^*(TE_i) \cdot m_l(TE_{i+1})\} \qquad \text{Eq. [7b]}$$

$$\Delta TE_i = TE_{i+1} - TE_i \qquad \text{Eq. [7c]}$$

To map single-T_2*, a MATLAB curve-fitting function *fit*(*x*, *y*, '*exp1*') was used to calculate single-T_2* values at each voxel via Eq. 8. A restriction ($T_2^*{}_{max}$<100ms) was enforced to exclude unreasonable values caused by noise. The computation time is acceptable (10min17s).

$$|m(TE_i)| = A_0 \exp(-TE_i/T_2^*), \ 0 \le T_2^* \le T_{2,max}^* \qquad \text{Eq. [8]}$$

3.8. Signal-to-noise ratio

In a region of interest (ROI), SNR was calculated via Eq. 9 in a simplified way for both volume and array coils by taking the ratio of mean intensity *S* to noise standard deviation (SD) in noise-only background regions. A factor of 0.655 was applied to noise SD to account for Rician distribution in magnitude images.[37] For SNR mapping, pixel signal is used in the calculation.

$$SNR = 0.655\, S/SD \qquad \text{Eq. [9]}$$

3.9. Estimation of extra- and intracellular volume fractions

The estimates give the up-band of volume fractions when all the mono-T_2 sodium are assigned to extracellular space while the bi-T_2 sodium to intracellular space, in the case that mono- and bi-T_2

sodium may co-exist in both extra- and intracellular spaces. The estimates were made in ROIs of the gray and white matters via Eq. 10 at the concentrations C_{ex}=145mM for extracellular and C_{in} =15mM for intracellular spaces.

$$V_{ex} = 1/(1 + a) \quad \text{Eq. [10a]}$$

$$V_{in} = a/(1 + a) \quad \text{Eq. [10b]}$$

$$a \equiv m_{bd}C_{ex}/m_{fr}C_{in} \quad \text{Eq. [10c]}$$

3.10. Statistical Significance

A regular statistical significance (P=0.05) was applied to the comparisons, via Student's *t*-test, between the two sets of data in this work. Minimum sample size for the *t*-test is 16, with 80% power, 5% significance level, two-sided test, and 1.0 effect size.[27]

4. RESULTS

4.1. Measurement of T_2 values*

Fig. 2 presents a representative of T_2* values on a healthy subject (52 years old, male), with and without correction for the distortion at the first five ADC samples (Figs. 2a1,b1). The correction removed distortion and reduced overall residual fitting error from 2.33% to 1.49% (Figs. 2a2,b2). The correction also improved resolution of short-T_2* components: from singlet at 2.5ms to doublet at 0.5ms and 2.5ms (Figs. 2a3,b3). A high resolution in T_2* was achieved at 0.5ms, with residual fitting error less than 1.5%. Notably, a few of sparse peaks appear in the T_2* spectrum, indicating that T_2* values are well clustered in the human brain and that a single set of T_2* values is applicable to the separation of mono- and bi-T_2 sodium.

4.2. Sensitivity to T_2 values*

Fig. 3 shows the computed impact of T_2* values on the accuracy of separation of the mono- and bi-T_2 sodium signals (m_{fr}, m_{bd}). The calculation was performed at a typical set of (T_2*$_{fr}$, T_2*$_{bs}$, T_2*$_{bl}$) = (50.0, 3.5, 15.0) ms in two extreme cases: the mono-T_2 sodium dominating, m_{fr} = 0.9, and the bi-T_2 sodium dominating, m_{bd} = 0.9. The impact of individual T_2* components are shown in columns. In column A, an error in T_2*$_{fr}$ caused an error in m_{fr} or m_{bd} much smaller for the dominant one (e.g., $\Delta m_{bd} < 2.2\%$ when ΔT_2*$_{fr} < 20\%$, and $\Delta m_{fr} < 2.9\%$ when ΔT_2*$_{fr} < 5.0\%$). In column B,

an error in T_2*_{bs} had a small impact on both m_{fr} and m_{bd} (e.g., when dominating, $\Delta m_{bd} < 4.8\%$ and $\Delta m_{fr} < 0.04\%$ when $\Delta T_2*_{bs} < 20\%$). In column C, an error in T_2*_{bl} led to an error in m_{fr} or m_{bd} much smaller for the dominant one (e.g., $\Delta m_{bd} < 5.2\%$ and $\Delta m_{fr} < 0.6\%$ when $\Delta T_2*_{bl} < 20\%$). The best case is in column B where the T_2*_{bs} had small impact (<4.9%) on both mono- and bi-T_2 sodium signals. The worst case is in Fig. 3a1 where the T_2*_{fr} had a large impact on the bi-T_2 sodium signal, $\Delta m_{bd} = 35.6\%$ when $\Delta T_2*_{fr} = -5\%$. In other words, when the mono-T_2 sodium is very dominating, T_2*_{fr} value should be as accurate as possible (usually achievable in practice) to attain the best separation for the bi-T_2 sodium.

4.3. Optimization of TEs

The ideal scheme of 80 TEs is presented in Fig. 4a on $Y_{fr}(TE)$ of the free sodium at a typical T_2*_{fr}=50ms and on $Y_{bd}(TE)$ of the bound sodium at a typical $\{T_2*_{bs}, T_2*_{bl}\} = \{3.5, 15.0\}$ms. The intuitively-favorable scheme of 8 TEs was presented in Fig. 4b, while the optimal candidate of two TEs was presented in Fig. 4c. Singular values (σ_1, σ_2) of the three TE schemes were compared against each other in Fig. 4d. In Fig. 4e is singular values of the 2-TE scheme, slowly changing with the TE_2 increasing. The σ_2 is less than 1.0 for the 8-TE and 2-TE schemes, leading to an amplification of noise. Therefore, a better choice for less noise amplification is the 2-TE scheme, in which TE_2 at 5ms produced a value near maximum of σ_2 while preserving higher signal than the larger TE_2. Thus, the 2-TE scheme is selected for the human studies.

4.4. Computer simulations

Fig. 5 demonstrates the simulated separation of mono- and bi-T_2 sodium signals (m_{fr}, m_{bd}) at a typical set of (T_2*_{fr}, T_2*_{bs}, T_2*_{bl}) = (50.0, 3.5, 15.0) ms and two-TE scheme TEs=(0.5, 5.0), at three SNRs: extra-high (100), high (50), and regular (25). The mean and standard deviation (SD) of the separated m_{fr} and m_{bd} were presented. The SD (error bar) consistently decreased with SNR increasing. There was an underestimate for m_{fr} or m_{bd} near the maximum value 1.0, but an overestimate near the minimum value 0.0, with an amount decreasing with SNR increasing.

4.5. Phantom studies

Fig. 6 summarizes the outcomes of phantom studies. Fig. 6a shows phantom arrangement of four tubes and sodium images at TE_1/TE_2=0.5/5ms. Fig. 6b shows FID signal from the four tubes at

averages=1, the residual fitting error, and the T_2* spectrum. Fig. 6c are the sodium images separated at (T_2*$_{fr}$, T_2*$_{bs}$, T_2*$_{bl}$) = (50, 5, 25) ms according to the T_2* spectrum in Fig. 6b3; the maps of ΔB_0, single-T_2*, and SNR; the signal intensities of the separated sodium signals (mean±SD) in the tubes; and the quantification of sodium concentration.

The separation in Fig. 6c3 recovered 95.8% of mono-T_2 sodium signal in the saline water tube, while leaving 4.2% to bi-T_2 sodium signal (much better than 20% left by the subtraction approach [24]). The separation recovered 72.5, 80.4, and 75.9% of bi-T_2 sodium signal in the agar tubes at sodium concentrations of 150, 120, and 90mM, respectively. The quantification of sodium concentration in Fig. 6c4, when calibrated at the saline water, showed a systematic bias in total and bi-T_2 sodium concentrations, leading to an underestimate of sodium concentrations.

4.6. Human studies

4.6.1. T_2 values in whole brain across subjects*

Fig. 7. presents a scattering plot of individual T_2* components from the T_2* spectra across all the subjects studied. Typical T_2* spectra and FID signals are shown in Fig. 7a for a healthy young subject (21 years old, male) and in Fig. 7c for an epilepsy patient (31 years old, male). The peaks in the T_2* spectra were sparse and just 2–4 peaks, suggesting that a global set of T_2* values (T_2*$_{fr}$, T_2*$_{bs}$, T_2*$_{bl}$) be a plausible estimate for the whole brain (Figs. 7a, c). However, these T_2* values are slightly different from subject to subject (Fig. 7b). The short-T_2* component is clearly crowded in a range of 1-5ms, while the long-T_2* is widely scattered in three bands centered at 10, 20, and 30ms, respectively. Interestingly, the long-T_2* component is shifted to lower values in the patient group, compared with the healthy group. There seems no difference between males and females in the healthy group. Therefore, the T_2* values are heterogeneous across the subjects.

4.6.2. Mono- and bi-T_2 sodium separation

Figs. 8 and 9 present two typical cases of the human studies in full implementation of the mono- and bi-T_2 sodium separation. Case 1 (Fig. 8) is from a 26-year-old healthy female, and includes 3D sodium images of the brain at TE_1/TE_2=0.3/5ms (Fig. 8a), FID signal of whole brain and associated fitting error and T_2* spectrum (Fig. 8b), the separated sodium images from the two-TE images using (T_2*$_{fr}$, T_2*$_{bs}$, T_2*$_{bl}$) = (50.0, 6.0, 19.0) ms (Figs. 8c1-c3), and inverse-contrast displays (Figs. 8c4-c5). In Fig. 8d are SNR, ΔB_0, and single-T_2* maps calculated from the two-TE

images in Fig. 8a. Case 2 (Fig. 9) is from a 59-year-old male patient with bipolar disorder. The separated sodium images were attained at (T_2*$_{fr}$, T_2*$_{bs}$, T_2*$_{bl}$) = (50.0, 2.5, 7.0) ms according to the peaks in Fig. 9b3.

Fig. 8 indicates that signals from CSF in the brain were effectively separated into the mono-T_2 sodium image (Fig. 8c2 or c4), while signals from brain tissues such as gray and white matters were separated into the bi-T_2 sodium image (Fig. 8c3 or c5). Notably and surprisingly, signal intensity across brain tissues looks more uniform in the bi-T_2 sodium images than in the mono-T_2 sodium images (Fig. 8c2), total sodium images (Fig. 8c1), and TE_1-images (Fig. 8a1). SNR in Fig. 8d1 are 25+ in most regions of the brain, assuring a robust separation as suggested in the simulations in Fig. 5. The field inhomogeneity ΔB_0 in Fig. 8d2 varied between ±20Hz across the brain, with the largest off-resonance in the prefrontal and occipital lobes, leading to visible blurring of the tissues in the bi-T_2 sodium images (Fig. 8c3 or c5, sagittal). The single-T_2* map in Fig. 8d3 provides a spatial distribution of short and long T_2* components across the brain, complementary to T_2* spectrum in Fig. 8b3. It also indicates that majority of long T_2* components are located in the prefrontal lobe in this particular case (Fig. 8d3, sagittal).

Fig. 9 demonstrates potential benefits from the bi-T_2 sodium images of patients with neurological disorders such as bipolar disorder which is known to cause abnormally-high intracellular sodium concentration in the brain but locations are unknown.[38,39] The bi-T_2 sodium images (Fig. 9c3 or c5) clearly highlighted brain regions of an elevated bi-T_2 sodium signal against surrounding tissues, with a ratio of 1.78 vs. 1.40 (or 27.1% increase) before the separation (Fig. 9c1). These regions have no visible contrast in the total or TE_1-images (Fig. 9a1 or c1). SNR in these regions is 40+ (Fig. 9d1), supporting a robust separation. The field inhomogeneity ΔB_0 in these regions is low (<5Hz, Fig. 9d2), excluding field-induced artifacts. The single-T_2* map in Fig. 9d3 shows abnormally low T_2* values in these regions, confirming an increase in short-T_2* components.

4.6.3. Estimates of extra- and intracellular volume fractions

In the healthy group (Fig. 10), the difference in volume fraction between the gray and white matters is significant (P=0.023): 89.6±4.5 % vs. 94.0±2.6 % for the intracellular space (in line with the literature, 75% vs. 92% [8]), and 10.4±4.5 % vs. 6.0±2.6 % for the extracellular space. No significant

difference (P=0.953) was found between the healthy and patient groups due to small samples (n=9 and 6).

5. DISCUSSION

The proposed MSQ technique has been demonstrated, using the computer simulations, physical phantoms, and human subjects, to be able to separate mono- and bi-T_2 sodium signals voxel-wise. The physics behind the technique is the intrinsic difference in T_2 relaxation between sodium nuclear spins: mono- vs. bi-exponential decay. In the restriction of total scan time, the two-TE scheme, instead of the eight-TEs, was selected for smaller transfer of random noise during the separation (Fig. 4). The measurement of T_2* spectrum from FID signals of entire brain and the application of a global set of T_2* values (T_2*_{fr}, T_2*_{bs}, T_2*_{bl}) were tested feasible to humans. In case of not plausible for a global set of T_2* values (i.e., T_2* spatially varying substantially [40-47]), multi-regional sets, or a linear combination of them, may be used.

The quantification of sodium concentration after the separation is not addressed in this study because it is involved in a complicated procedure of 1) calibration that transforms sodium signal into concentration and 2) correction for inhomogeneous sensitivity of coil elements. These processes deserve a separate study to deal with.

The limitations of MSQ technique are obvious, and understanding them is crucial to practice of the technique. The first limitation is the two-component model (mono- and bi-T_2 populations) which is of risk to produce a false-positive error for bi-T_2 sodium. If there are mono- and bi-T_2 sodium populations in a voxel, then they are separatable. If not, such as two mono-T_2 sodium decays at different T_2* values in a voxel, they would be falsely separated into bi-T_2 sodium because they can be combined mathematically (not physically) into a bi-exponential decay mimicking a true bi-T_2 sodium decay. This kind of false positive error stems from the fact that the separation is based on *mathematical* model, instead of *physical* model as does the TQF separation. Understanding this kind of false positive errors is critical to proper interpretation of the separated bi-T_2 sodium signals.

The second limitation is the misguided separation of single mono-T_2 sodium component of short T_2* value in a voxel, such as regions in nose and sinuses (Figs. 8c3 and 9c3). In this

situation, the MSQ separates it into bi-T_2 sodium of underestimated intensity. These mis-separated regions can be identified by means of the maps of ΔB_0 and single-T_2* (Figs. 8d and 9d).

The third limitation is the underestimate of bi-T_2 sodium signal caused by the TE_1 image, as illustrated in the phantom study (Fig. 6c3). The separation (Eq. 2) assumes TE_1-image intensity exactly at TE_1 (i.e., a very short readout time). Actual TE_1-image intensity is an average over readout time during which short-T_2* components decay significantly when readout is relatively long, such as readout T_s=36.32ms about ten times long of a short-T_2* at 3ms in this study. Therefore, the underestimation alters with readout time or pulse sequence. To mitigate the problem, two strategies may apply. One is to replace TE_1 value in Eq. 2 with an effective (larger) value that accounts for short-T_2* decay during the readout. The other is to shift T_2*_{bs} to a larger value, as did in previous work on the phantoms.[48] Alternatively, correction for the underestimates is integrated into calibration of image intensity for sodium concentration (Fig. 6c4).

6. CONCLUSION

The data presented in this study have demonstrated the feasibility of proposed multi-TE single-quantum sodium MRI technique to separate between mono- and bi-T_2 sodium signals in a fashion of voxel by voxel. The MSQ technique is based on a solid physics related to the intrinsic difference in T_2 relaxation between the two populations of sodium nuclear spins. The two-TE sampling scheme stands out for smaller noise transfer during the separation. A global set of T_2* values (T_2*_{fr}, T_2*_{bs}, T_2*_{bl}) measured on T_2* spectrum of whole brain was tested applicable to humans. However, the MSQ technique has limitations and requires cautions in practice.

7. ACKNOWLEDGMENTS

This work was partially presented in the 25th Annual Meeting of ISMRM 2017, Honolulu, Hawaii, USA. The authors would like to thank colleagues Dr. Ryan Brown for his support in the development of the 8-channel array sodium coil and Dr. Timothy Shepherd for his support in the recruitment of epilepsy patients. This work was financially supported in part by the National Institutes of Health (NIH) RF1/R01 AG067502, RF1 NS110041, R01 NS113517, R01 NS108491, and R01 CA111996; and by the General Research Fund from the Department of Radiology, NYU Grossman School of Medicine. This work was also performed under the rubric of the Center for