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In vivo visualization of the dorsal penile nerve using 3.0-Tesla DTI and fiber tractography

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Introduction

Male sexual dysfunction—including erectile dysfunction (ED) and premature ejaculation (PE)—represents a major challenge in global reproductive health. Epidemiological projections estimate that the number of men affected by ED will exceed 322 million by 2025 (1). Organic ED arises from complex interactions among neural, vascular, and endocrine systems, with the dorsal penile nerve (DPN)—a terminal branch of the pudendal nerve—playing a critical role in maintaining normal erectile function through its structural integrity and functional status (2). Although international guidelines recommend nocturnal penile tumescence (NPT) monitoring and penile duplex Doppler ultrasound for the assessment of ED (3), these modalities rely on indirect parameters and cannot provide quantitative evaluation of neural microstructure.

Notably, even with nerve-sparing radical prostatectomy (NS-RP) or intensity-modulated radiotherapy (IMRT), postoperative ED rates remain as high as 25–75% (5), underscoring the inadequacy of current imaging techniques for detailed neural assessment. Similarly, selective dorsal penile neurectomy—used in some centers to treat PE (6)—would greatly benefit from more precise nerve imaging to guide surgical planning and postoperative evaluation.

Diffusion tensor imaging (DTI) tractography is a powerful tool for visualizing and analyzing structural connectivity within the human brain. DTI leverages the anisotropic diffusion of water molecules to noninvasively assess the integrity of nerve fibers, and has been successfully applied to peripheral neuropathies such as sciatic nerve injury (7–9). DTI information can be used to map the orientation of axon fibers using a technique termed DTI tractography. This technology has been used for neurosurgical planning to aid in the removal of brain tumors, as well as for neural pathway mapping of the brain, spinal cord, and brachial plexus.

The DPN, as the terminal branch of the pudendal nerve (originating from the S2-S4 nerve plexus), presents unique characteristics—tortuous course, intimate adjacency to the corpora cavernosa sinusoids, and marked inter-individual variability (10). Occasionally, small nerve may not be well visualized by conventional DTI, primarily because of limited spatial resolution. Partial-volume effects from pulsatile blood flow blur the nerve-vascular interface, while inherently low signal-to-noise ratio (SNR) further impairs fiber tracking accuracy. Consequently, standard DTI protocols fall short of the precision required for reliable DPN visualization. Studies have demonstrated that employing data acquisition with reduced voxel dimensions is beneficial for enhancing tensor calculation accuracy. This is attributed to the minimization of

partial volume averaging. However, due to constraints such as acquisition time, imaging coverage, signal-to-noise ratio (SNR), and reconstruction performance, DTI tractography generally utilizes sections thicker than 2 mm (11) .

In response to these challenges, this study assessed the feasibility of a 1.5-mm isotropic voxel DTI sequence in vivo visualization of the dorsal penile nerve by comparing the images with those obtained with a conventional DTI.

Materials and Methods

1. Study Design and Participants

This prospective, single-center technical feasibility study was conducted from December 2024 to February 2025 with approval from the institutional ethics committee (Guizhou International Hospital Ethics Committee) ; All methods were performed in accordance with the relevant guidelines and regulations; All participants provided written informed consent. Healthy male participants aged 18–40 years with no history of erectile dysfunction (International Index of Erectile Function-5 score ≥ 22) were recruited consecutively. Exclusion criteria included: (a) history of spinal or pelvic surgery or trauma; (b) contraindications to MRI (e.g., metallic implants or severe claustrophobia); (c) acute genitourinary infection, neurological, or metabolic disease; and (d) inability to tolerate the required scanning position.

2. MRI Equipment and Sequence Parameters

All examinations were performed on a United Imaging U-880 3.0 T superconducting MRI scanner equipped with a 24-channel flexible body phased-array coil. Each participants underwent two sequential DTI sequences with FSE-MX anatomical references: before (original) and after (modified) parameter modification—using the settings detailed in Table 1.

3. Image Post-Processing and Analysis

3.1 Post-Processing Platform

DTI data were transferred to the scanner's built-in uWS-MR post-processing workstation (version R005.7.0.1322062) and analyzed using the MR Neuro Analysis (DTI/DTT) module.

3.2 Processing Workflow

Two board-certified radiologists with 8 and 5 years of pelvic MRI experience, respectively, independently evaluated all images while blinded to the acquisition protocol.

-ROI Placement and Fiber Tracking:

On coronal FSE-MX images, the dorsal penile vein served as the anatomical landmark for targeting the bilateral DPN (Figure 1 A-C). At the level of the penile root, identical circular ROIs were manually drawn each DPN (Figure 1 E). Deterministic fiber tracking was then performed along the principal diffusion direction. The SuperFlex flexible coil geometry was incorporated to generate high-fidelity three-dimensional fiber bundle reconstructions, clearly depicting the nerve's course relative to the corpora cavernosa (Figure 1 D&F).

-Objective Image Quality Metrics:

Signal-to-noise ratio (SNR) and contrast-to-noise ratio (CNR) were calculated on both the raw b-value (b1000) images.

The SNR is defined as:

$$SNR = \frac{S_{cc}}{\sigma_{Noise}}$$

The CNR is defined as:

$$CNR = \frac{|S_{cc} - S_{Tunica}|}{\sigma_{Noise}}$$

S_{cc} denotes the mean signal intensity of the corpus cavernosum, and σ_{Noise} represents the standard deviation of background (air) noise; and S_{Tunica} denotes the mean signal intensity of the tunica albuginea.

Subsequently, three-dimensional fiber tract reconstructions were subjectively evaluated for

diagnostic image quality and nerve depiction using the following scoring system: (a) image quality, 1–5 (1 = non-diagnostic, unusable for nerve assessment; 5 = excellent, artifact-free); (b) nerve visualization, 0–3 (0 = not visible; 1 = proximal one-third visualized; 2 = proximal two-thirds visualized; 3 = excellent, including clear depiction of distal branches); and (c) nerve continuity, 0–2 (0 = no continuous tract despite multiple seed points; 1 = continuous tract only with multiple seeds; 2 = uninterrupted tract from a single seed point).

-DTI Quantitative Metric Extraction: Fractional anisotropy (FA), mean diffusivity (MD), relative anisotropy (RA), and volume ratio (VR) were recorded for each ROI.

3.3 Repeatability

Intra-observer Repeatability: Reader A performed two independent assessments of all images with a 4-week interval to minimize recall bias.

Inter-observer Agreement: Reader B independently evaluated the same parameters concurrently with Reader A's initial measurements.

4. Statistical Analysis

5. Normality was tested using the Shapiro–Wilk test. Subjective scores were compared using the Wilcoxon signed-rank test with Cliff's delta effect size. For objective metrics, normally distributed data were analyzed by paired t-tests (mean $\pm$ SD), and non-normal data by Wilcoxon tests (median, IQR). Weighted Cohen's kappa assessed consistency of subjective scores, and intraclass correlation coefficients (ICC) evaluated agreement for objective metrics. All statistical analyses were performed in SPSS 26.0, and all figures were generated in MedCalc. A two-tailed $P < 0.05$ was considered statistically significant.

Results

1. Participant Characteristics and Image Quality Assessment

A total of 21 healthy male participants were included (Table 2), with a median age of 30 years (IQR: 26–38). Following parameter optimization, both nerve visualization scores (median 3 vs. 1; $P < 0.001$) and nerve continuity scores (median 2 vs. 1; $P < 0.001$) increased significantly, with Cliff's delta effect sizes of 1.00 (very large) and 0.905 (large), respectively. Although the overall image quality score remained unchanged (median 4 vs. 4; $P = 0.014$), objective measures of image contrast improved markedly: the optimized sequence yielded substantially higher SNR (158.38 vs. 61.57) and CNR (101.57 vs. 34.96), both $P < 0.001$ (Table 2, Figure 2). Figure 3 provides a direct visual comparison of dorsal penile nerve imaging quality in the same Participant before and after sequence optimization.

2. Repeatability Analysis

Intra-observer repeatability for the optimized protocol was enhanced compared with the original sequence (Table 3). For example, SNR yielded a Cronbach's α of 0.887 and an ICC of 0.864 (95% CI: 0.629–0.947) post-optimization, versus $\alpha = 0.173$ and ICC = 0.152 pre-optimization. Similar improvements ($P < 0.001$) were observed for CNR, FA, MD, and other metrics. Inter-observer agreement for subjective scores (Table S1) was perfect (weighted $\kappa = 1.000$) for nerve visualization and continuity under the optimized protocol, while image quality agreement remained high ($\kappa = 0.618$ vs. 0.745). Bland–Altman analysis (Figure S1) confirmed low bias (<5% mean difference) and narrow 95% limits of agreement for all optimized metrics.

3. Diffusion Tensor Imaging Metrics

Optimized imaging parameters produced significantly higher fractional anisotropy (FA: 0.203

vs. 0.184; $P = 0.007$) and lower volume ratio (VR: 0.841 vs. 0.956; $P = 0.002$) compared with the original protocol. Mean diffusivity (MD) was also markedly reduced in the optimized sequence (0.724×10^{-3} vs. 1.332×10^{-3} mm²/s; $P < 0.001$) (Table 2, Figure 2).

Discussion

In this study, we systematically optimized the DTI sequence parameters and, for the first time, achieved high-resolution in vivo visualization of the DPN. The optimized protocol employed several key adjustments: smaller isotropic voxels ($1.50 \times 1.50 \times 1.50$ mm³ vs. $1.88 \times 1.88 \times 4.00$ mm³) to enhance spatial resolution; an extended repetition time (TR) to 6917 ms (vs. 5717 ms) to allow more complete T_1 relaxation and boost overall signal; a shortened echo time (TE) from 79.8 ms to 56.1 ms, reducing T_2 -decay-related signal loss and susceptibility artifacts while preserving tissue contrast; and a reduced echo train length (ETL) from 51 to 42 to minimize blurring from T_2 decay. Lowering the radiofrequency pulse density further mitigated magnetization-transfer-induced contrast shifts (12). Combined with iterative reconstruction algorithms, these modifications increased SNR and CNR by 157% (158.38 vs. 61.57) and 191% (101.57 vs. 34.96), while channel compression and in-plane merging further suppressed noise and accelerated reconstruction.

Conventional DTI sequences balance scan efficiency by using a single excitation, which is adequate for Cerebral white matter fiber bundle but insufficient for imaging fine structures such as the DPN. In this study, we employed multi-excitation (increasing excitations from 1 to 3), which is less sensitive to magnetic field inhomogeneities and thus reduces geometric distortion caused by air-tissue interfaces around the penis (13). However, multiple excitations can induce signal-averaging effects that lower the absolute values of diffusion metrics such as mean

diffusivity (MD) and volume ratio (VR) (12). Consistent with this, the modified protocol yielded significantly lower MD (0.724×10^{-3} vs. 1.332×10^{-3} mm²/s) and VR (0.841 vs. 0.956) compared with the original sequence (both $P < 0.05$). The optimized sequence also demonstrated excellent intra-observer consistency (e.g., SNR ICC increased from 0.152 to 0.864) and strong inter-observer agreement, confirming its reliability.

Several limitations warrant discussion. First, as the inaugural study of in vivo DPN DTI, no established normative ranges exist for quantitative metrics such as FA. Although FA and MD differed significantly pre- and post-optimization, their clinical relevance must be validated against histopathological findings. Second, our sample size was modest and limited to healthy volunteers; future studies should expand the cohort to include patients, age- and pathology-specific reference intervals, and correlate DTI metrics with sexual function scores. Finally, despite the clear improvement in dorsal penile nerve visualization, scan duration increased (9:51 vs. 5:31). Subsequent work should leverage parallel imaging techniques or deep-learning-based acceleration algorithms to enhance time efficiency.

Conclusion

Combined optimized DTI and fiber tractography enables the first high-resolution 3D visualization of in vivo DPN, providing a novel tool for microstructural assessment in male sexual dysfunction.

List of abbreviations

The dorsal penile nerve (DPN), diffusion tensor imaging (DTI), signal-to-noise ratio (SNR), contrast-to-noise ratio (CNR), fractional anisotropy (FA), mean diffusivity (MD), relative anisotropy (RA), volume ratio (VR), erectile dysfunction (ED)

Reference:

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Table 1. DTI Parameter Optimization Details

Imaging Parameter	Original	Modified
Voxel Size	1.88×1.88×4.00 mm	1.50×1.50×1.50 mm
Scan Time	5:31	9:51
Number of Slices	34	90
Slice Thickness (mm)	4	1.5
TR (Repetition Time, ms)	5717	6917
TE (Echo Time, ms)	79.8	56.1
Multislice Factor	1	2
FAST	1D	-
Phase Acceleration Factor	2	-
Multislice Mode	Sequential	-
Number of Excitations	-	3
Echo Train Length	51	42
Echo Spacing (ms)	0.72	0.83
Zoom Imaging	PP	-
RF Mode	Standard	-
Fat Suppression Mode	Dual-Pulse	-
Iterative Mode	-	Optimized
Reconstruction Mode	Advanced	-
Channel Compression	-	√
Max Reconstruction Channels	-	10
In-plane Merging	-	√
Raw Images	√	-
B-value	1000	1000

Notes.—"√" indicates enabled/available, "-" indicates not applicable/unavailable.

Abbreviations retained as per original (e.g., PP, FAST).

Characteristic	Original	Modified	Cliff's Delta (δ)	Z-Value (T-Value)	P Value
Age (y)	30 (26-38)	30 (26-38)	-	-	-
No. of male participants	21	21	-	-	-
SNR	61.573(25.686-137.596)	158.376(42.958-495.102)	-	-4.015	<0.001*
CNR	34.963(11.733-97.872)	101.570 (27.620-352.857)	-	-3.841	<0.001*
Score of	4 (3-5)	4 (3-5)	0.286	-2.449	0.014*

Table 2. Participant Characteristics and Image Quality Assessment

image quality Score of neural visualization	1 (1-2)	3 (2-3)	1.000	-4.158	<0.001*
Score of nerve continuity	1(0-2)	2(1-3)	0.905	-4.264	<0.001*
FA	0.184±0.049 [†]	0.203 (0.147-0.381)	-	-2.711	0.007*
MD				21.141	
(10 ⁻³ mm ² / s)	1.332±0.124 [†]	0.724±0.080 [†]	-	(T-Value)	<0.001*
RA	0.155±0.038 [†]	0.216 (0.103-0.782)	-	-2.520	0.012*
VR	0.956±0.019 [†]	0.841 (0.356-0.984)	-	-3.136	0.002*

Note.—Data are medians with IQRs in parentheses, unless otherwise noted.

[†]As the data are normally - distributed, values are shown as means ± SDs.

Table 3. Repeatability Analysis of Objective DTI Parameters for Dorsal Penile Nerve Evaluation: Intra-Observer Consistency Based on Intraclass Correlation Coefficient (ICC)

Parameter	Cronbach's Alpha	Average ICC	95% CI	F Value	P Value
SNR (Pre)	0.173	0.152	(-0.755, 0.627)	1.209	0.337
SNR (Post)	0.887	0.864	(0.629, 0.947)	8.837	<0.001*
CNR (Pre)	0.996	0.996	(0.990, 0.998)	240.022	<0.001*
CNR (Post)	0.998	0.997	(0.993, 0.999)	400.061	<0.001*

Parameter	Cronbach's Alpha	Average ICC	95% CI	F Value	P Value
FA (Pre)	0.983	0.980	(0.942, 0.992)	59.168	<0.001*
FA (Post)	0.969	0.931	(0.343, 0.982)	32.552	<0.001*
MD (Pre)	0.960	0.962	(0.906, 0.985)	25.154	<0.001*
MD(Post)	0.899	0.900	(0.757, 0.959)	9.872	<0.001*
RA (Pre)	0.976	0.973	(0.929, 0.989)	40.872	<0.001*
RA (Post)	0.997	0.997	(0.993, 0.999)	364.592	<0.001*
VR (Pre)	0.932	0.934	(0.838, 0.973)	14.626	<0.001*
VR (Post)	0.999	0.999	(0.998, 1.000)	1136.450	<0.001*

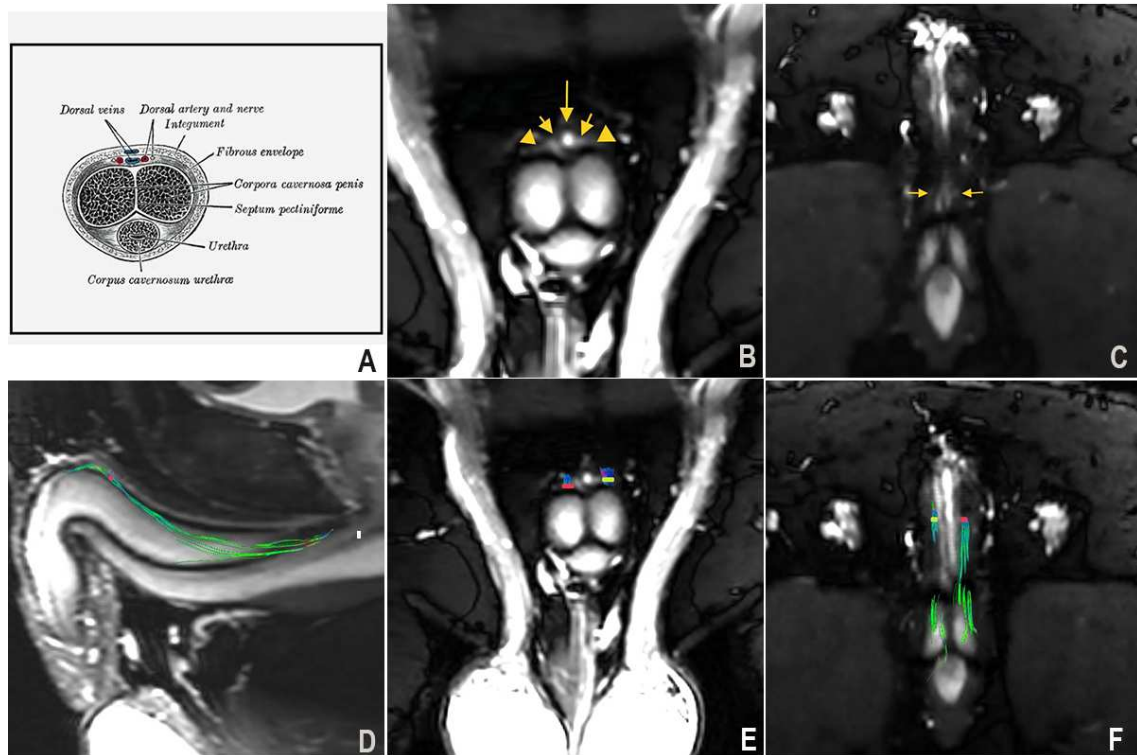


Fig 1. Anatomical Localization and High-Fidelity 3D Tractography Reconstruction

(A) DPN anatomical relationships Source: Adapted from Gray, H. (1918). *Anatomy of the Human Body* (20th ed.), Fig 1212. (B) Neurovascular identification on coronal FSE-MX image Long arrow: Dorsal penile vein, Short arrow: Dorsal penile artery, Triangle: DPN.(C) Bilateral DPN visualization on axial FSE-MX. (E) Manual placement of isometric circular ROIs at penile root level. (D&F) Three-dimensional tractography reconstruction demonstrating faithful rendering of the dorsal penile nerve trajectory along the corpora cavernosa and its parallel course to the dorsal penile arteries. *Note: All images acquired using a modified DTI protocol.*

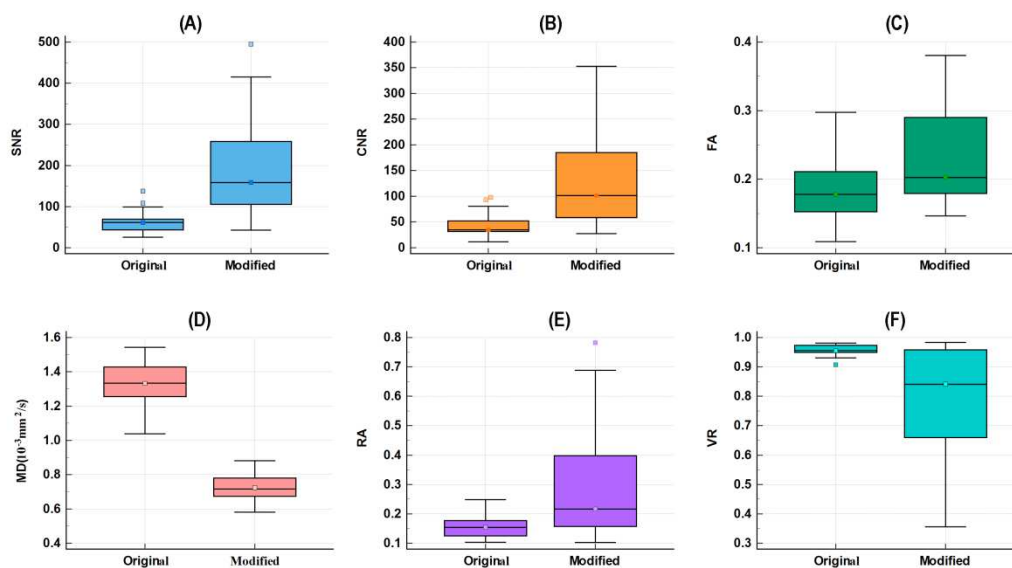


Fig 2. Comparative Boxplots of Objective and Quantitative Neural Metrics Before and After DTI Sequence Optimization

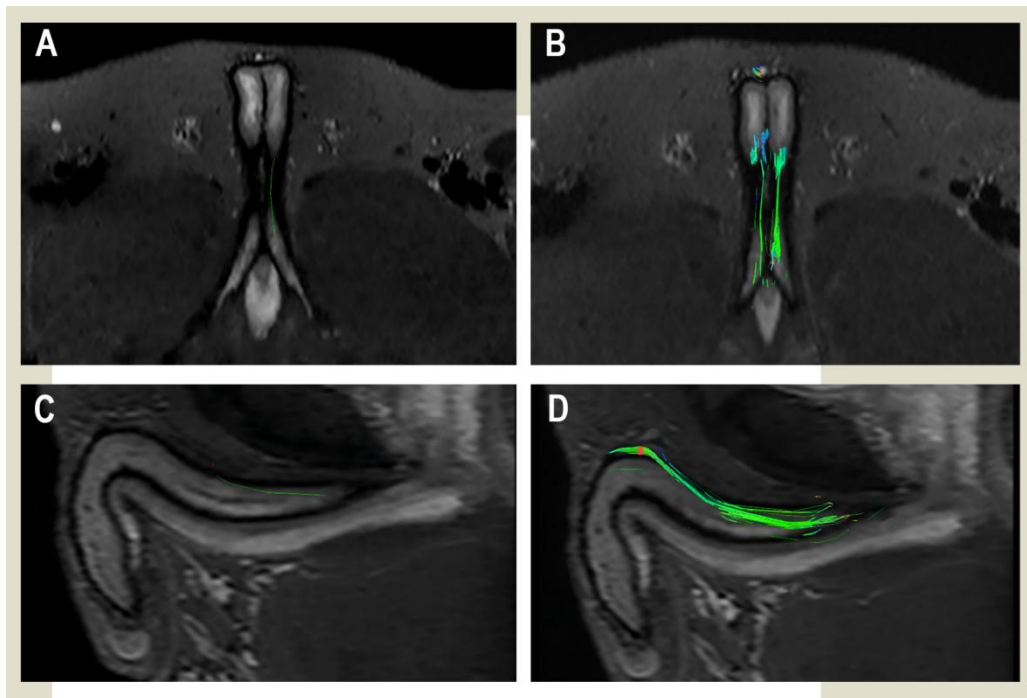


FIG 3. Representative DTI Fiber Tract Reconstructions of the Dorsal Penile Nerve Before and After Sequence Optimization

A (axial) and C (sagittal): The original DTI protocol with FSE-MX acquisitions in a 30-year-old male shows poor visualisation of DPN fiber bundles (voxel size: $1.88 \times 1.88 \times 4.00 \text{ mm}^3$). B (axial) and D (sagittal): A modified DTI protocol with FSE-MX (modified voxel size: $1.50 \times 1.50 \times 1.50 \text{ mm}^3$, increased excitations = 3, etc.) clearly delineates the bilateral DPN fiber bundles along the corpus cavernosum with enhanced spatial resolution.

Supplementary Files

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