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# **Application of shear wave elastography to estimate the stiffness of the male striated urethral sphincter during voluntary contractions**

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Keywords: elastography, ultrasound imaging, male, pelvic floor, stiffness, striated urethral sphincter

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## **Abstract**

**Objectives:** To investigate whether increases in stiffness can be detected in the anatomical region associated with the striated urethral sphincter during voluntary activation using shear wave elastography; to identify the location and area of the stiffness increase relative to the point of greatest dorsal displacement of the mid urethra (i.e. striated urethral sphincter); and to determine the relationship between muscle stiffness and contraction intensity.

**Subjects and methods:** Ten healthy men participated. A linear ultrasound transducer was placed mid-sagittal on the perineum adjacent to a pair of electromyography electrodes that recorded non-specific pelvic floor muscle activity. Stiffness in the area expected to contain the striated urethral sphincter was estimated via ultrasound shear wave elastography at rest and during voluntary pelvic floor muscles contractions to 5%, 10% and 15% maximum. Still image frames were exported for each repetition and analysed with software that detected increases in stiffness above 150% of the resting stiffness.

**Results:** Pelvic floor muscle contraction elicited an increase in stiffness above threshold within the region expected to contain the striated sphincter for all participants and contraction intensities. The mean(SD) ventral-dorsal distance between the centre of the stiffness area and region of maximal motion of the mid-urethra (caused by striated urethral sphincter contraction) was 5.6(1.8), 6.2(0.8), and 5.8(0.7) mm for 5%, 10% and 15% MVC respectively. Greater pelvic floor muscle contraction intensity resulted in a concomitant increase in stiffness, which differed between contraction intensities(5% vs. 10%;  $P<0.000$ , 5% vs. 15%;  $P<0.000$ ; 10% vs. 15%;  $P=0.003$ ).

Conclusion: Voluntary contraction of the pelvic floor muscles in men is associated with an area of stiffness increase measured with shear wave elastography, which concurs with the expected location of the striated urethral sphincter. The increase in stiffness occurred in association with an increase in perineal surface EMG activity, providing evidence that stiffness amplitude relates to general pelvic floor muscle contraction intensity. Future applications of shear wave elastography may include investigations of patient populations in which dysfunction of the striated urethral sphincter is thought to play an important role, or investigation of the effect of rehabilitation programmes that target this muscle.

# Introduction

The striated urethral sphincter (SUS) contributes to maintenance of urinary continence in men by compression of the mid-urethra [1-3]. This muscle loops around the ventral aspect of the urethra providing dorsally directed pressure and motion [4], which contrasts the ventrally directed force of the puborectalis/puboperinalis which course from the pubic symphysis to a midline attachment posterior to the rectum and prostate [5]. The importance of the SUS for continence is thought to increase after removal of the prostate (which removes prostatic urethral smooth muscle) [1, 2], and is a common target for conservative [6] and surgical treatment [7] for stress urinary incontinence in men after radical prostatectomy. Detailed understanding of function of the SUS in continent and incontinent men is needed. However, most methods to assess the function or dysfunction of the SUS are highly invasive (e.g. needle EMG [8], intra-urethral surface EMG [3], urethral pressure) and impractical for investigation of SUS function in clinical populations, or in large-scale studies of healthy populations. Although changes in muscle morphology and tissue displacement observed with non-invasive B-mode ultrasound imaging (US) can be used to infer activation of the SUS and other pelvic floor muscles [4], it does not provide information on the tension/stress developed within the individual peri-urethral muscles during contraction. This is because the relationship between muscle displacements and muscle stress is not straightforward. Quantification of muscle stress is crucial to understand SUS function because it is likely to inform how muscle activation is transformed to urethral pressure.

Tissue stiffness in skeletal muscle (animal models) has traditionally been studied through surgical implantation of sensors within muscles [9, 10]. A new non-invasive US imaging method has been developed that can be used to study human skeletal muscle, but has never been applied

to the context of the urethral control in men. Shear wave elastography (SWE) quantifies the shear modulus (stiffness) of a localised area of tissue. Like traditional US, it visualizes internal structures, but concurrently quantifies their stress/stiffness. An advantage of SWE over other elastography techniques is that it can quantify changes in tissue stiffness in real-time, including during slow dynamic tasks [11]. Recent studies of other skeletal muscles have shown that muscle stiffness measured by SWE is strongly linearly related to muscle stress/force [12, 13] and muscle activity [14]. SWE could provide a novel, non-invasive solution to quantify the spatial distribution of stress (and applied ureteral pressure) in peri-urethral striated muscles with concurrent estimation of muscle activity. Together, stress/muscle activity data would aid resolution of understanding of the effect of SUS contraction on the urethra, and test interpretations of how much, and where, the SUS muscle increases tension to apply pressure to the urethra. The technique must first be validated in healthy men.

The purpose of this study was to assess the feasibility of ultrasound SWE to quantify stiffness of the male SUS during voluntary pelvic floor muscle contractions. The first aim was to investigate whether increased stiffness could be detected in the anatomical region associated with SUS shortening and activation. If an increase in stiffness was observed, the second aim was to calculate the location and area of the stiffness increase relative to the location previously used to interpret SUS activity from motion of the urethra. The third aim was to determine the relationship between muscle stiffness and contraction intensity.

## **Subjects and Methods**

### **Subjects**

Ten healthy men with mean(SD) age 34(5) years, height 176(8) cm and weight 78(16) kg and no history of neurological or urological disease/symptoms participated. All participants provided written consent. This study was conducted in conjunction with a similar investigation in females (Aljuraifani et al, manuscript submitted), and the protocol was approved by the Institutional Medical Research Ethics Committee (#2010000545), and conducted in accordance with the principles of the Declaration of Helsinki.

## Measurement

Participants sat on a plinth with knees extended and a back rest at approximately 60° from horizontal. A linear transducer (SL10-2 MHz, Supersonic imagine, Aix-en-Provence, France) was set at 5MHz and placed on the perineum between the anus and scrotum in the mid-sagittal plane. SWE and B-mode US data were collected in video format (frame rate: 1 Hz) and stored on the device (Aixplorer V9, Supersonic imagine, Aix-en-Provence, France). The region of interest over which stiffness could be calculated within the B-mode image was set to maximum (3 x 3 cm) and positioned to include the dorsal pole of the pubic symphysis (PS) and the ventral and dorsal borders of the mid-urethra (Fig. 1A). A pair of EMG surface electrodes (Ambu Blu sensor N, N-00-s125, Ballerup, Denmark) was placed on the perineum to provide a general estimate the overall intensity of the pelvic floor muscle activity and was not intended to reflect SUS EMG. An electrode was placed on either side, 1 cm anterior to the anus with an inter-electrode distance of ~2 cm. Although this electrode configuration does not lie along the orientation of muscle fibres it was found to provide a more stable recording of general muscle activation than electrodes placed in a dorsal-ventral direction, which were confounded by ultrasound gel. A reference electrode was placed over the iliac crest. EMG data were bandpass



filtered (20-1000 Hz), amplified 2000 times (Neurolog, Digitimer, Welwyn Garden City, UK), and sampled at 4 kHz using Spike2 software and a Power1401 data acquisition system (Cambridge Electronic Design, Cambridge, UK).

## **Procedure**

Men were trained to contract pelvic floor muscles without activation of the abdominal muscles using B-mode image to provide biofeedback of dorsal displacement of the mid-urethra. Participants performed a 3-s maximal voluntary contraction (MVC) of the pelvic floor muscles for 2 repetitions using the instruction to “retract the penis into the body as strongly as possible” which we have shown previously leads to optimal activation of the SUS [15]. The peak MVC EMG amplitude was used to calculate contraction intensity targets set at 5, 10 and 15% MVC. This relatively low level of activation was used for several reasons. First, it has been reported that at high levels of activation some of the pelvic floor muscles fatigue rapidly as a result of central fatigue [16]. A low level target was selected to circumvent this issue and enable participants to maintain a stable contraction for 5s at all target levels. Second, estimations of muscle stiffness with SWE have been reported to be less accurate at high contraction amplitudes (40-70% MVC, depending on the muscle studied) because the relationship between muscle stiffness and force is no longer linear [17]. Low level contractions were therefore necessary to enable detection of stiffness changes between intensities. Third, performance of MVC would likely elicit activation of surrounding muscles and the associated increase in stiffness might affect the capacity to estimate the location of the area of stiffness increase related to SUS. Targets were visually displayed on a computer monitor with a real-time trace of the pelvic floor EMG (root-mean-square amplitude [time constant - 0.1 s]). Following initiation of the SWE

video recording, participants performed 2 repetitions at each contraction intensity, with each contraction maintained for 5 s to provide sufficient time to generate three separate SWE frames during the steady-state hold for analysis (see below).

## Data analysis

Four image frames were exported from the video data for each repetition of pelvic floor muscle contraction; one image at rest and three during contraction. Images were imported into a custom-written image analysis program (Matlab r2012b, The Mathworks, Natick, USA) to calculate the change in tissue stiffness associated with voluntary contraction at each intensity. This analysis was based on comparison of the stiffness map at rest and during contraction. This process involved selection of a *region of interest* (ROI) from the stiffness map that was based on anatomical landmarks that define boundaries for the region expected to contain the SUS muscle, based on anatomical dissections and MRI [1, 18] and motion detected with B-mode imaging [19, 20] (Fig. 1). In brief, the anatomy of the SUS is described as an omega-shaped loop of striated fibres that insert dorsally into the perineal body and encircles the anterior and lateral aspects of the urethra [1]. Measures made with transurethral ultrasound imaging indicates muscle thickness in healthy men is approximately 6 mm [21]. The anatomy implies change in muscle stiffness with voluntary contraction should occur ventral to the urethra. It has been suggested that the location of SUS along the mid-urethra can be identified by the point that undergoes the greatest dorsal motion during voluntary contraction (i.e. referred to as the “SUS point of interest” [POI]) [19]. Based on the muscle anatomy, the maximum ventral boundary of the ROI was set at 10 mm ventral to the SUS POI, and the cranial boundary of the ROI was 5 mm from the SUS POI. The dorsal boundary for the ROI was the middle of the urethra (Fig. 1). For analysis the ROI was first

defined within the resting B-mode image and an ellipse was approximated to the location of the pubic symphysis to mask the stiffness associated with this structure. This region was transposed to the SWE map for calculation of the average stiffness for the rest image (Fig. 2A-B) and used to define the threshold stiffness applied to detect an increase in stiffness within the images recorded during contraction. On the basis of pilot data this threshold was set at 150% of the average resting stiffness.

For analysis of the images recorded during contraction (Fig. 2C), the anatomical boundaries for the ROI were visually identified on the B-mode image and transposed onto the stiffness map as described for the rest image. Increases in stiffness above the threshold were detected automatically (Fig. 2D) for each contraction repetition, and averaged across the three images to produce a single image of increased stiffness. Each stiffness map identified several regions of increased stiffness within the ROI. The location and amplitude of each regions of increased stiffness within the averaged image was quantified with the following outcome variables: (i) the X (ventral-dorsal) and Y (cranial-caudal) distance between the centre of gravity (COG) of the region of increased stiffness and the region of maximum mid-urethral dorsal displacement (SUS POI), (ii) the average stiffness value within each region of increased stiffness, and (iii) the cross-sectional area of each region of increased stiffness. For each area of stiffness, the “region stiffness” was calculated by multiplying its area by its average stiffness to provide a single measure representative of both properties. Summation of the region stiffness for each region of increased stiffness within the ROI provided “cumulative stiffness”. Cumulative stiffness was calculated for each contraction intensity (5, 10 and 15% MVC). Data from separate

repetitions were used to evaluate the repeatability of the measure and were averaged for the final analysis to reduce any variation in the data.

## **Statistical analysis**

To determine whether an increase in stiffness could be detected in the anatomical region expected to contain the SUS during voluntary pelvic floor muscle contractions using SWE (Aim one), descriptive statistics were used to present the features of the regions of increased stiffness. To quantify the location of the area of increased stiffness relative to the SUS POI for each individual (Aim two) measures of the X and Y distance (mean and standard deviation) between the COG of the region of increased stiffness and the SUS POI were calculated for each individual. The location was also compared between the 5%, 10% and 15% MVC contraction intensities with a repeated measures analysis of variance (ANOVA) to determine whether location changed with contraction intensity. The repeatability of the cumulative stiffness measures between the two repetitions of contraction at each intensity was assessed by calculation of the intra-class correlation coefficients (ICC (2,1)). To investigate whether an increase in contraction intensity resulted in a concomitant increase in stiffness (Aim three), a repeated measures ANOVA was used to compare cumulative stiffness between the contraction intensities. To enable investigation of the relationship between the increase in stiffness and contraction intensity, the cumulative stiffness was expressed as a proportion of the sum of cumulative stiffness across all intensities (e.g.  $\text{proportion 5\% MVC} = \frac{\text{cumulative stiffness}(5\% \text{ MVC})}{\sum \text{cumulative stiffness}(5\%, 10\%, 15\% \text{ MVC})}$ ) for each participant, and then averaged across the group for each intensity. The relationship between cumulative stiffness and contraction intensity was then tested using linear regression.

## Results

Voluntary contraction of the pelvic floor muscles elicited an increase in stiffness (above 150% resting stiffness threshold) within the ROI for all participants and all contraction intensities. As this region was based on the location of greatest dorsal motion of the urethra, and is the region expected to contain the SUS this supports that notion that this stiffness increase is caused by SUS contraction (Aim one). For most participants, multiple regions of increased stiffness were identified within the ROI. The location and region stiffness of each of these regions of increased stiffness is shown in Fig. 3 for each participant. A single region of greatest region stiffness could be identified for each individual; referred to as the “dominant” area. Other areas of increased stiffness were observed outside the ROI (and therefore not analysed in detail). For instance there was commonly an area of increased stiffness between the urethra and the perineum (Fig. 2C). This is most likely explained by stiffening of the corpus cavernosum by contraction of the bulbocavernosus muscle. Another area that was commonly observed to show increased stiffness was between the pubic symphysis and urethra more ventral than our ROI. Again, this may be explained by compression of penile tissue by ischiocavernosus or bulbocavernosus contraction.

When the location of the dominant region was considered relative to the SUS POI at each contraction intensity (Aim two), the mean(SD) ventral-dorsal (X) distance between the COG and the SUS POI was 5.6(1.8), 6.2(0.8), and 5.8(0.7) mm for 5%, 10% and 15% MVC respectively. The mean(SD) cranial-caudal (Y) distance between the COG and the SUS POI was 0.7(1.2), 0.4(2.1), and -0.1(0.9) mm for 5%, 10% and 15% MVC respectively (distances cranial to the

SUS POI are shown as positive numbers). Thus, the location of the COG of the region of greatest stiffness did not differ between contraction intensities (Main effect [X distance]:  $P=0.52$ , [Y distance]:  $P=0.30$ ) and when averaged over the contraction intensities 5.9 mm ventral and 0.3 mm cranial to the SUS POI (Fig. 3).

The between-repetition repeatability of the cumulative stiffness measures had ICCs (2,1) of 0.80, 0.79 and 0.61 for 5%, 10% and 15% MVC respectively, which indicates good to excellent reliability [22]. When participants contracted the pelvic floor muscles with increasing intensity, there was a concomitant increase in cumulative stiffness (Main Effect:  $P<0.001$ ) (Aim three). When cumulative stiffness (at each intensity) was normalised to the sum of cumulative stiffness across all intensities, mean(SD) values were 21(7)%, 34(3)% and 45(7)% for 5%, 10% and 15% MVC respectively and differed significantly between each intensity (Post hoc: 5% vs. 10%;  $P<0.000$ , 5% vs. 15%;  $P<0.000$ ; 10% vs. 15%;  $P=0.003$ ). A strong linear correlation was observed between normalised cumulative stiffness and contraction intensity ( $R^2=0.76$ ,  $P=0.001$ ). The mean(SD) cumulative stiffness of the un-normalised data was 788(501), 1231(617), and 1597(681)  $\text{kPa}\cdot\text{mm}^2$  for 5%, 10% and 15% MVC respectively. Normalised individual and group data are shown in Fig 4.

## Discussion

These data show that voluntary contraction of the pelvic floor muscles using the instruction to “retract the penis into the pelvis” in men is associated with an area of stiffness increase measured with non-invasive SWE. This area concurs with the expected location of the SUS muscle, as defined by previous anatomical studies and the location where dorsal

displacement of the urethra is observed on transperineal B-mode ultrasound imaging. The increase in stiffness in conjunction with increased contraction intensity provides further evidence that this stiffness increase relates to SUS muscle activation. Together these support the notion that SWE provides a method to assess SUS function and may play a role in evaluation of pelvic floor exercise programs that aim to train this muscle for treatment of disorders such as urinary incontinence after prostatectomy.

Previous investigations have quantified motion of the mid-urethra with transperineal ultrasound imaging and related the motion to function of the SUS [4, 19, 20]. Motion at this region was quantified relative to a coordinate system based around the pubic symphysis. This study used the same point (defined here as the SUS POI) [19] to serve as a reference for quantification of the location of the regions of increased stiffness. The average location of the COG of the area of stiffness increase across contraction intensities was 5.9 mm ventral and 0.3 mm cranial to the SUS POI. This location is comparable to measures reported for the location of the muscle using other imaging methods. Strasser et al [2] reported decreases in SUS thickness with voluntary contraction using transurethral ultrasound imaging (in older men post-prostatectomy). Although no group data were presented the authors reported a distance between the urethral wall and innermost aspect of the SUS muscle of 6.2 mm at rest in one participant. Wang et al [21] quantified anatomical properties of the SUS in 86 young Chinese men using MRI and reported SUS thickness ranged from 4.3-6.9 mm when measured between the ventral urethral border and the outer border of the muscle. Assuming a small distance between the ventral urethral border and the innermost border of SUS (2 mm), the SUS boundaries in the axial plane should be located in the range of 2.0-8.9 mm. These limits are similar to the SUS COG

location reported here. This supports the assertion that SWE provides an accurate estimation of the location of SUS.

When a muscle contracts there is a concomitant increase in tissue stiffness. This increase can be estimated by SWE. Studies of skeletal muscle have shown the increase in stiffness is linearly related to both active muscle contraction and passive muscle lengthening [12, 13]. As the SUS exerts force on the urethra, we argue that the increased stiffness of this muscle will provide an indirect estimate of increased urethral pressure. Although likely in most contexts this may be compromised in certain conditions such as urethral wall fibrosis or urethral stricture. Further, generation of urethral pressure will depend not only on the tension generated by SUS but also the radius of the urethra (LaPlace's law), thus, any situation that modifies these variables would affect the net urethral pressure.

This study offers evidence that SWE provides an estimate of stiffness of tissues in the anatomical area expected to contain the SUS, and that the increase in stiffness differs between contraction intensities. Applications of this method may include investigations of patient populations in which dysfunction of SUS is thought to play an important role (e.g. stress urinary incontinence and post-prostatectomy incontinence). Given that rehabilitation programs focus on improving activation and strength of the SUS (to improve urethral pressure generation), SWE may be an effective tool to monitor progress and test the efficacy of clinical interventions.

Several methodological factors require consideration. First, as high amplitude contractions were not tested in this study, we cannot comment on the feasibility of SWE to estimate stiffness in the location of the SUS with contraction intensities above 15% MVC.



Second, current technology means that SWE images can be collected with a maximum frequency of 0.5 to 1 sample per second (1 sample per second was used in this study). This precludes estimation of tissue stiffness during dynamic tasks. Third, previous work has highlighted that muscle stiffness is most accurately estimated when the transducer is placed in parallel with muscle fibres [17]. As this was not possible for the SUS this may have reduced the accuracy of our measures. However, the observed increase in stiffness with small steps in contraction intensity of 5% MVC suggests that our SWE measures have high sensitivity to changes in SUS stiffness. Fourth, given the close proximity of the SUS fibres to adjacent tissues (e.g. vascular and connective) which may be compressed by contraction of SUS and other muscles of the pelvic floor, we cannot be certain the extent which the observed areas of stiffness relate to the SUS muscle alone. Fifth, technical limitations of the SWE device prevented consistent stiffness estimates of deeper muscles of the pelvic floor such as puborectalis. Ongoing improvements in SWE technology may enable this in future.

## **Conclusion**

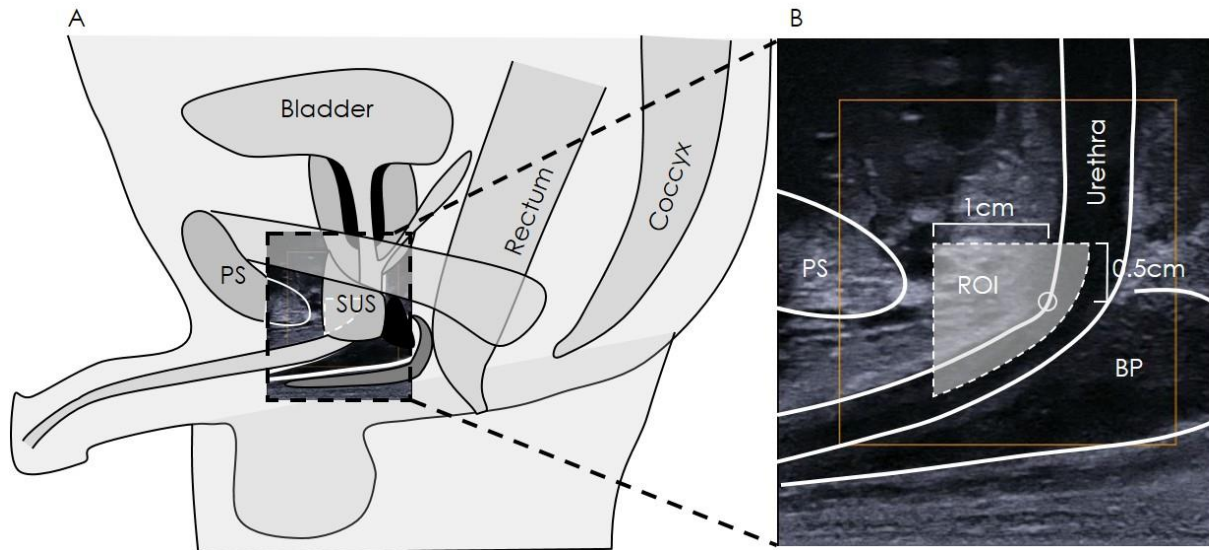
These data show that voluntary contraction of the pelvic floor muscles in men is associated with an area of stiffness increase measured with non-invasive SWE. Further, the location of this increase concurs with the expected location of the SUS muscle. Systematic increase in stiffness with contraction intensity provides further evidence of a relationship to SUS muscle contraction. Future applications of SWE may include investigations of patient populations in which dysfunction of SUS is thought to play an important role, and investigation of the effect of rehabilitation programmes that target this muscle.

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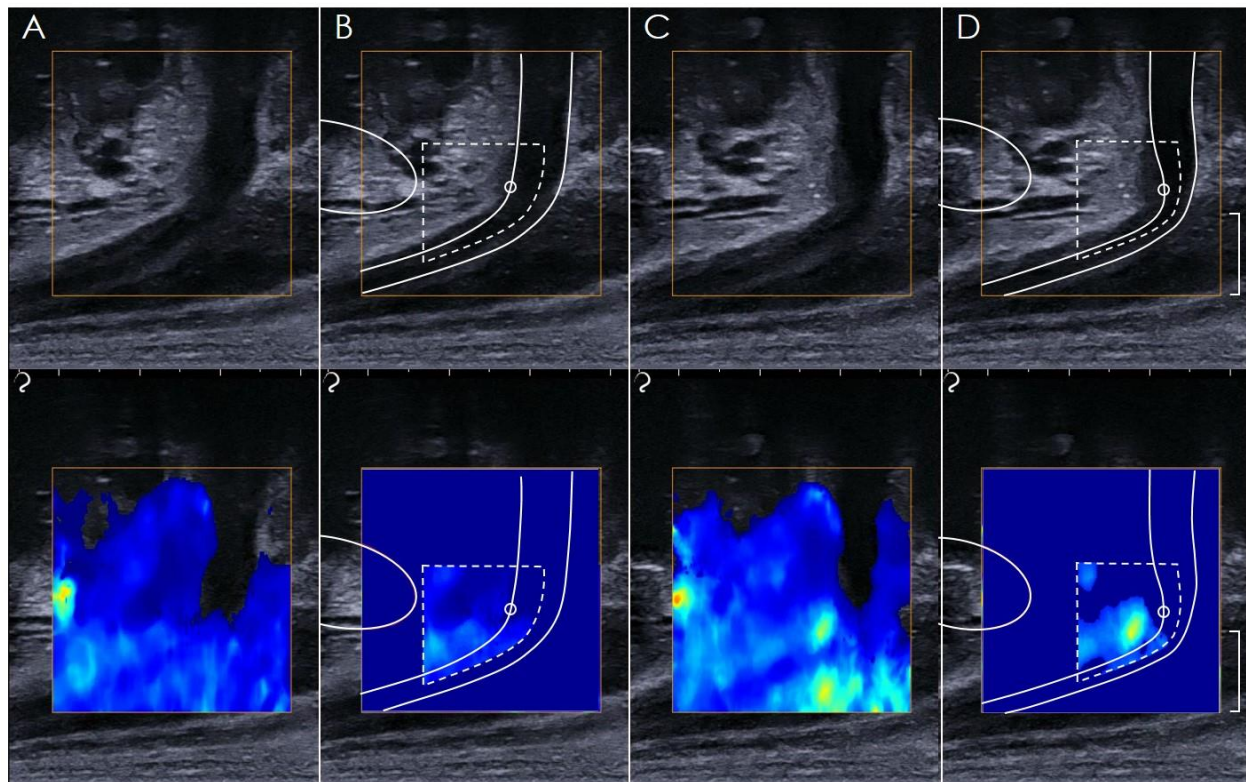
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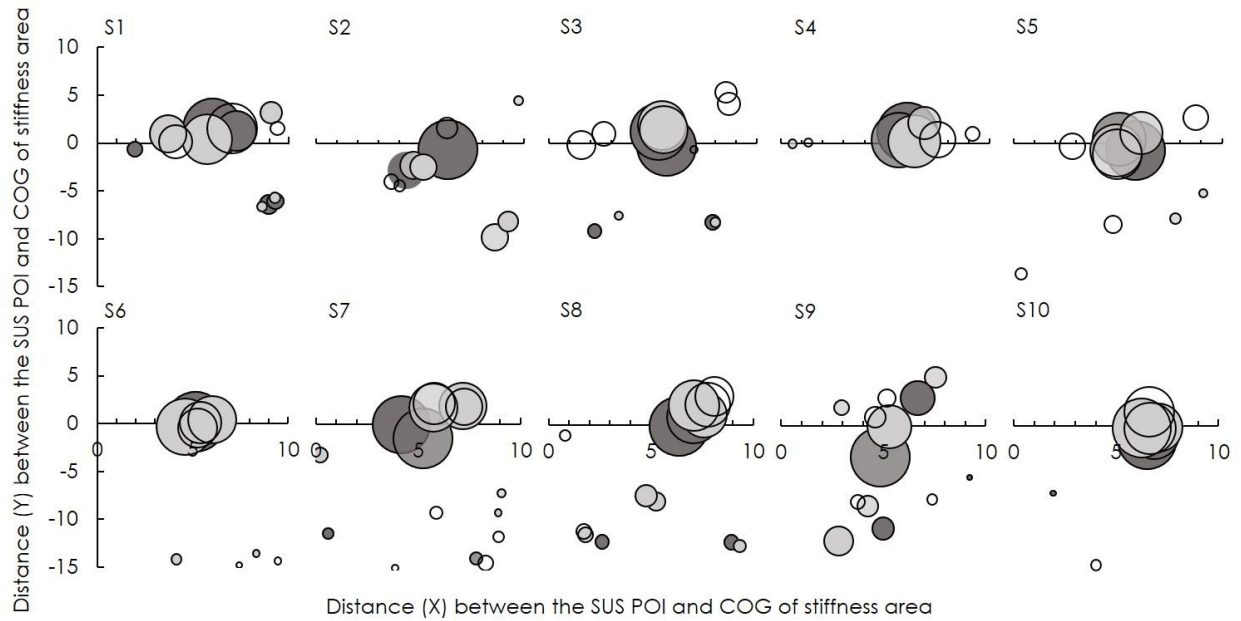
**Fig. 1** (A) Anatomical representation of the male pelvic floor in the sagittal plane. The inset depicts the anatomical area visualised with B-mode ultrasound imaging. (B) B-mode ultrasound image with borders of pelvic structures and region of interest (ROI) for stiffness measurement highlighted by solid white lines. SUS – striated urethral sphincter, PS – pubic symphysis, BP – bulb of penis.



**Fig. 2** Method for analysis of SWE images. B-mode ultrasound images are shown at the top of each panel with the corresponding stiffness map below. Images are shown at rest with (A) and without (B) identification of pelvic structure borders, and during contraction with (C) and without (D) identification of structure borders. The stiffness map corresponding to (D) is shown after the threshold for stiffness increase (150% average resting stiffness) was applied. The region of interest (ROI) used for stiffness calculations is indicated with a dashed line. Image scale – 1 cm.



**Fig. 3** Location and region stiffness of regions of increased stiffness with voluntary contraction. Distances shown are measured between the centre of gravity (COG) of the stiffness area relative to the location of the greatest dorsal motion of the urethra suggested to be caused by the striated urethral sphincter (SUS point of interest (POI)) in the X and Y planes. Data are shown separately for each participant at each contraction intensity (white – 5%; light grey – 10%; dark grey –15%)



**Fig. 4** The change in stiffness as a proportion of the sum of cumulative stiffness over the three contraction intensities. Mean(SD) group (solid bars) and individual (lines) data are shown. \* -  $p < 0.01$ . Dashed line shown for the relationship between stiffness and contraction intensity for individual data ( $R^2=0.75$ )

