

# ***In vitro* renal cortex elasticity and viscosity measurements with Shearwave Dispersion Ultrasound Vibrometry (SDUV) on swine kidney**

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**Abstract**— Renal fibrosis threatens kidney viability and fibrosis has been associated with altered tissue structure affecting the biomechanical properties of the kidney, quantifiable as elasticity and viscosity. Importantly, early detection of renal fibrosis may guide therapy and eliminate invasive biopsy procedures. The ability to detect fibrosis early and monitor it regularly with sufficient sensitivity and specificity is an active area of research. A newly emerging method called Shearwave Dispersion Ultrasound Vibrometry (SDUV), that quantifies both elasticity and viscosity by evaluating dispersion of shear wave propagation speed versus its frequency, offers a potential tool to determine renal elasticity and viscosity *in vivo*.

The purpose of this study was to evaluate the feasibility of SDUV for *in vitro* measurements of renal cortex elasticity and viscosity in the kidney.

## I. INTRODUCTION

Fibrosis alters the tissue structure and thus the biomechanical properties of the organ. For instance, *in vivo* studies in humans have shown that normal liver tissue is elastic compared to fibrotic tissue [1]. Similarly, changes in kidney elasticity, with Magnetic Resonance Elastography (MRE) [2] and ultrasound-based [3] elasticity imaging, had indicated renal scarring in *in vivo* and *in vitro* animal models, respectively.

Renal fibrosis is a common pathway of Chronic Kidney Disease (CKD) leading to the destruction of kidney parenchyma and ultimately end-stage renal disease (renal failure), a condition that requires dialysis or kidney replacement [4]. Early detection of renal fibrosis, a consequence of an excessive accumulation of extracellular matrix components that alter the elastic properties of the tissue, could provide critical prognostic information required to guide therapy or alternatively to avoid invasive procedures. However, a major obstacle is the inability to detect fibrosis early and monitor it regularly with sufficient sensitivity and specificity.

Conventional functional measurements such as renal biopsy can determine the degree of fibrosis in a kidney, but frequent biopsies are not feasible for screening because of the associated risk, sampling error and cost. Indirect screening by serum creatinine concentration is used to

calculate an estimated Glomerular Filtration Rate (GFR) [9], which is the most accepted measure of renal function. However, neither creatinine levels nor GFR are specific measures of renal fibrosis. Furthermore, significant kidney dysfunction may be present despite a normal creatinine level, especially when renal disease is unilateral [10].

Radiological kidney examinations by ultrasound [11], computed tomography [12] or magnetic resonance imaging [13] aid the diagnosis and prognosis based on anatomical changes and morphology. In addition, functional examinations associated with these modalities may provide functional diagnostic measures. However, these methods do not offer assessment of the mechanical properties of the tissue. Thus far, the evaluation of renal fibrosis by conventional imaging modalities has been limited. New methods of imaging renal tissue have the potential to significantly improve the management of renal fibrosis and limit the need for invasive procedures.

A newly emerging method called Shearwave Dispersion Ultrasound Vibrometry (SDUV), that quantifies both shear modulus and shear viscosity by evaluating dispersion of shear wave propagation speed versus its frequency, could potentially improve evaluation of renal disease.

The purpose of this study is to evaluate the feasibility of SDUV for *in vitro* measurements of renal cortex shear modulus and shear viscosity on normal swine kidney.

## II. METHODS

### A. Principle of SDUV

Shearwave Dispersion Ultrasound Vibrometry (SDUV) [14]-[15] applies an external localized force to generate harmonic shear waves that propagates outward from the vibration center.

For a homogenous Voigt model the shear wave propagation speed  $c_s$  depends on the frequency of shear wave  $\omega_s$ :

$$c_s(\omega_s) = \sqrt{2(u_1^2 + \omega_s^2 \mu_2^2)} \rho \left( u_1 + \sqrt{\mu_1^2 + \omega_s^2 \mu_2^2} \right) \quad (1)$$

where  $\rho$ ,  $\mu_1$  and  $\mu_2$  are the density, shear modulus and shear viscosity of the medium, respectively. The external localized force is generated by a 'Push Transducer' (refer to Fig. 1) that transmits repeated tone bursts of ultrasound.

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Typically, a push sequence consists of 10 tonebursts that exert a force of constant amplitude every 10 ms.

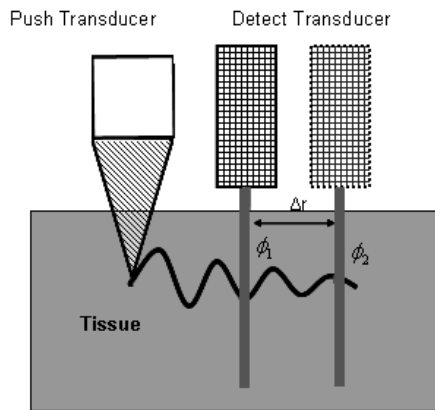


Fig. 1. SDUV applies a localized force generated by a ‘Push Transducer’ coupled to the tissue, transmitting repeated ultrasound tone bursts of ultrasound. A separated transducer acts as the detector (‘Detect Transducer’).

The shear wave speed is estimated from its phase measured at least at 2 locations separated by along its traveling path:

$$c_s(\omega_s) = \omega_s \Delta r / \Delta \phi_s \quad (2)$$

where  $\Delta \phi_s = \phi_1 - \phi_2$  is the phase change over the traveled distance  $\Delta r$ . The shear wave speed is then estimated with (2). The variation of  $c_s$  versus frequency is then fit by (1) to inversely solve for shear modulus and shear viscosity.

### B. Experiment

The swine kidney was obtained from a kidney Magnetic Resonance Elastography (MRE) study [16] in which the renal blood flow was obstructed by a vascular occluder reducing renal blood flow from baseline by 20% until total occlusion. MRE exams were conducted at each obstruction level. This study was approved by the Institutional Animal Care and Use Committee at Mayo Clinic.

The kidney was then removed immediately after sacrifice of the animal. Fig. 2 illustrates the experimental setup. The ‘Push Transducer’ is a focused single-element transducer with a diameter of 44 mm, a center frequency of 3 MHz, and a focal length of 70 mm. Shear waves generated at the transducer focal point propagate through renal tissue and vibration was detected by a single element transducer with a diameter of 12.7 mm, a center frequency of 5 MHz and a 50 mm focus length (‘Detect Transducer’). The ‘Push Transducer’ and ‘Detect Transducer’ were aligned confocally with a pulse echo technique using a small sphere as a point of target. The force was localized 5 mm deep into the kidney surface.

The renal cortex was excited at three different locations (white arrows on Fig. 3.). The pulse repetition frequency was 50 Hz. The propagation of the shear wave was tracked by the single element transducer in pulse-echo mode over a

range of 5 mm along the y-axis. The ultrasound echoes were digitized at 100 MHz and processed by the previously described method [17] to estimate the shear wave phase. The shear wave propagation speed was calculate by (2) and dispersion measurements at fundamental frequency of 50 Hz and its harmonics of 100 Hz, 150 Hz, 200 Hz, etc. were fit by (1) to solve for elasticity and viscosity.

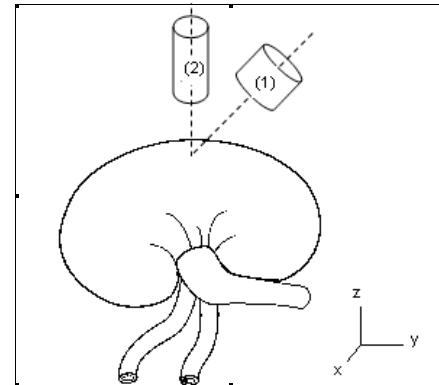


Fig. 2. Illustration of the experimental setup, (1) is the ‘Push Transducer’, (2) is the ‘Detect Transducer’.

During the experiment, the single element transducer was moved by 0.25 mm intervals 21 times along y-axis. This acquisition sequence was repeated 5 times in each of the Regions of Interest (ROI). Additionally, motion was recorded at 5 different ROI’s as depicted by the white squares in Fig. 3.. Three variability tests were performed in this study. In ‘Test A’, we evaluated the repeatability of SDUV at one location in a 5 mm by 5 mm patch (ROI1 in Fig. 3). Additionally, in ‘Test B’, we evaluated the variability of renal cortex shear modulus and shear viscosity measurements in a 5mm by 5mm patch, analyzing the shear wave propagation in the y-direction in 1 mm steps on z-axis deep into the kidney (ROI1 in Fig. 3). Finally, in ‘Test C’, we evaluated the variability of renal cortex shear modulus and shear viscosity measurements at 5 different locations (Fig. 3. ROI1, ROI2, ROI3, ROI4 and ROI5) in different 5 mm by 5 mm patches.

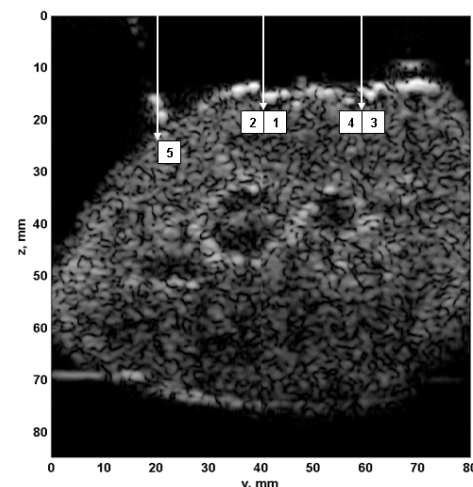


Fig. 3. B-mode image along zy-plane. The beam was focused at 3 locations (white arrows). The motion was tracked along 5 regions along y-axis (white squares) of 5 mm by 5 mm

### III. RESULTS

Fig. 4 shows the results from ‘Test A’, the shear wave propagation speed as a function of frequency measured along y-axis on 5 mm by 5 mm ROI 5 times. The wave speed values (circles) are an average of 5 acquisitions. The solid line is the fit from (1) that gives a shear modulus 2.3 kPa and a shear viscosity 2.2 Pa·s. The shear wave fit had a Mean Square Error (MSE) of 0.0027.

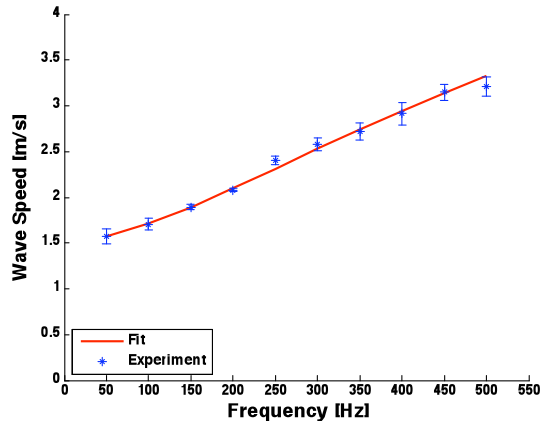


Fig. 4. Results from ‘Test A’, the shear wave propagation speed as a function of frequency measured along y-axis on 5 mm by 5 mm ROI 5 times

Results from ‘Test B’ are shown in Fig. 5, the shear wave propagation speed as a function of frequency measured along the y-axis on 5 mm by 5 mm patch on ROI1 by 1 mm steps on z-axis deep into the kidney. The solid line is the fit from (1) that gives a shear modulus 2.3 kPa and a shear viscosity 1.8 Pa·s. The shear wave fit had a Mean Square Error (MSE) of 0.0067.

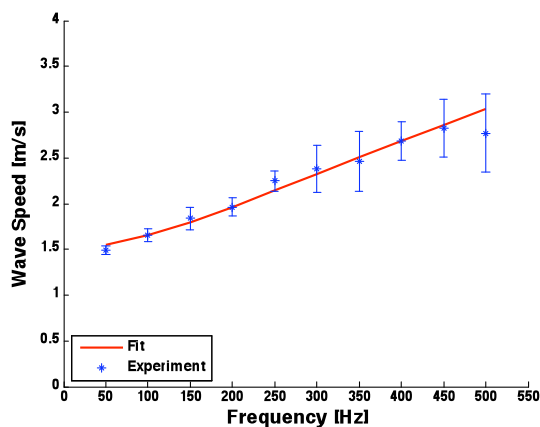


Fig. 5. Results from ‘Test B’, the shear wave propagation speed as a function of frequency measured along y-axis on 5 mm by 5 mm patch on ROI1 by 1 mm steps on z-axis deep into the kidney.

Fig. 6 shows results from ‘Test C’, the average shear wave propagation speed of 5 ROI’s as a function of frequency measured along y-axis on 5 mm by 5 mm. The solid line is the fit from (1) that gives a shear modulus 1.7

kPa and a shear viscosity 1.8 Pa·s. The shear wave fit had a Mean Square Error (MSE) of 0.0043.

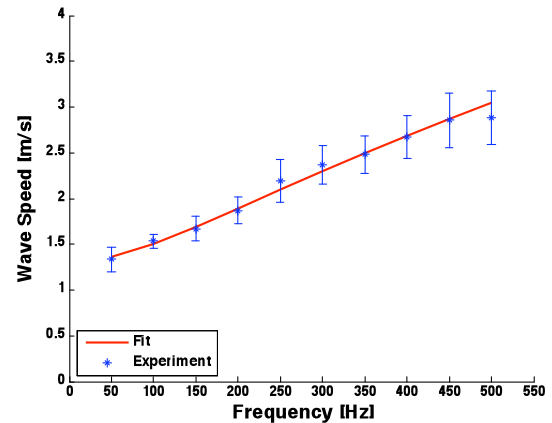


Fig. 6. Results from ‘Test C’, the average shear wave propagation speed of 5 ROI’s as a function of frequency measured along y-axis on 5 mm by 5 mm.

The shear modulus and shear viscosity values given by Fig. 4 (Test A), Fig. 5 (Test B) and Fig. 6 (Test C) are summarized in Table I.

TABLE I

SHEAR WAVE SPEED MEASURED ALONG Y-AXIS ON 5 mm BY 5 mm PATCH

Test	$\mu_1$ (kPa)	$\mu_2$ (Pa·s)	MSE
A	2.3	2.2	0.0027
B	2.3	1.8	0.0067
C	1.7	1.8	0.0043

Shear modulus and shear viscosity values from the shear wave propagation speed as a function of frequency measured along y-axis on 5 mm by 5 mm ROI 5 times (Test A), the shear wave propagation speed as a function of frequency measured along y-axis on 5 mm by 5 mm patch on ROI1 by 1 mm step on z-axis deep into the kidney (Test B), the average shear wave propagation speed of 5 ROI’s as a function of frequency measured along y-axis on 5 mm by 5 mm (Test C).

### IV. DISCUSSION

In this study the shear modulus of the renal cortex varied from 1.7 - 2.3 kPa and the renal cortex shear viscosity varied from 1.8 - 2.2 Pa·s. The shear wave speed versus frequency results in Fig. 4 fits well with the use of the Voigt model. Additionally, there is a small variability of our method within 5 mm by 5 mm patch, which suggest that SDUV could be capable of making *in vivo* shear modulus measurement of the renal cortex with sufficient precision.

On the other hand, Fig. 5 shows significant variation of shear wave speed for high frequencies (250-500 Hz), which could be attributed to anatomic variability of the renal cortex or a limitation of the model to fit the data at high frequencies. Furthermore, Fig. 6 suggests viscoelastic properties variation within the kidney, which again could be

attributed to anatomic variations since the kidney is a highly vascularized organ.

This study demonstrated that SDUV is capable of obtaining quantitative measurements of renal tissue shear modulus and shear viscosity. Although, there are limited studies regarding the *ex vivo* elastic properties of the kidney, an ongoing *in vivo* renal MRE study at 90 Hz frequency suggests shear elasticity changes with perfusion pressure (Fig. 7).

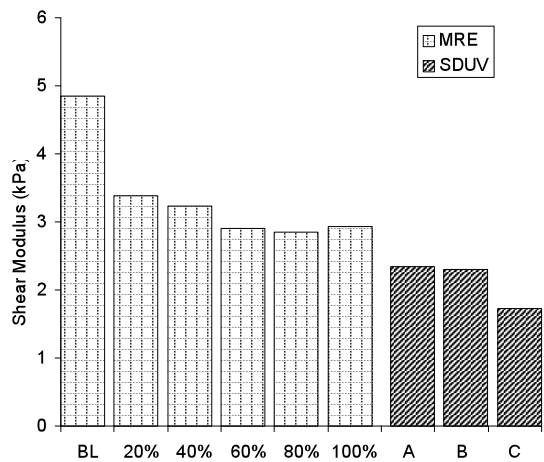


Fig. 7. Shear modulus measurements with *in vitro* SDUV (A = 5 mm by 5 mm patch at ROI1, B= 5 mm by 5 mm patch at ROI1 with 1 mm step deep into kidney, C= 5 mm by 5 mm patch at five ROI's) and *in vivo* MRE at Base Line (BL), 20% Renal Artery occlusion, 40% Renal Artery occlusion, 60% Renal Artery occlusion, 80% Renal Artery occlusion and 100% Renal Artery occlusion.

Importantly, the SDUV *in vitro* measurements fell below the range of MRE elasticity at total occlusion (100%), supporting the notion that SDUV is capable of quantifying renal tissue elasticity. Indeed, we would expect the *in vitro* shear modulus to be less than that at total occlusion since the renal tissue is comprised of residual perfusion at 100% occlusion, while the *in vitro* kidney is devoid of residual perfusion.

Although these physiological changes *in vivo* do not alter tissue composition, the results suggest that perfusion pressure may have effects on the tissue viscoelastic properties. The high temporal resolution capability of SDUV, 0.1 ms for each measurement, potentially offers an *in vivo* detection tool to assess the physiological changes on the kidney such as those related to perfusion pressure and feasibility to monitor development of renal fibrosis.

## V. CONCLUSION

This study showed the feasibility of SDUV to detect both shear modulus and shear viscosity of renal cortex *in vitro*. Future work includes *in vivo* measurements of renal viscoelastic properties with changes in perfusion pressure.

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