**Supplementary Material:**

Smooth Muscle Regional Contribution to Vaginal Wall Function

Gabrielle L. Clark1, Anastassia P. Pokutta-Paskaleva2, Dylan J. Lawrence1, Sarah H. Lindsey4, Laurephile Desrosiers5, Leise R. Knoepp5, Carolyn L. Bayer1, Rudolph L. Gleason Jr. 2,3 and Kristin S. Miller1

1 Department of Biomedical Engineering, Tulane University

2 The Wallace H. Coulter Department of Biomedical Engineering, Georgia Institute of Technology

3 The George W. Woodruff School of Mechanical Engineering, Georgia Institute of Technology

4 Department of Pharmacology, Tulane University School of Medicine

5 Department of Female Pelvic Medicine & Reconstructive Surgery, University of Queensland Ochsner Clinical School

Supplemental material presenting the methodologies and data for balloon catheterization to measure intravaginal pressures *in vivo*. Further, contractility results following elastase digestion.

**Balloon Catheterization for Intravaginal Pressure**

Female mice (n=6; 4-6 months) were anesthetized with 4% isoflurane in 100% oxygen () at estrus to measure intravaginal pressure with a balloon catheter (1-4). A custom 3 mm-diameter balloon was fabricated with polyvinyl chloride and secured with 6-0 suture to a 1.25 mm-diameter aluminum tube (Fig 1A). The balloon catheter was attached to a 3-way stop cock, disposable pressure transducer, and 3mL-syringe (Fig 1A). The disposable pressure transducer was integrated via system components and laptop (PowerLab and LabChart8, ADInstruments, Colorado Springs, CO,USA) (5). The column was filled with water and the balloon was distended to 3 mm obtaining a baseline pressure reading then inserted into the vaginal canal passing the introitus to measure the change in pressure (3). This was performed 3 times on each animal. The intravaginal pressure readings were averaged over the 3 trials to eliminate user variability (Fig 1B).

**Figure 1**. Schematic for the set-up of measuring intravaginal pressure with a balloon catheter (A). Measured *in vivo* intravaginal pressure (n=6) reported as mean +/- standard deviation (B).

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**Figure 2.** Change in outer diameter with contraction under the physiologic length and mean pressure, pre- (close) and post- (open) elastase digestion (A). Change in axial force with contraction under the physiologic length and mean pressure, pre- (grey) and post- (white) elastase digestion. Axial force was significantly reduced following elastase digestion (B). Data is reported as mean +/- SEM. Statistical significance is denoted by \* (p<0.05).



**B**

**A**

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