Introduction

Urinary bladder tissue has many components, and its neuromuscular nature indicates that both signals from the brain and muscle contractions play a role in its function. The wall tissue layers include the urothelium, lamina propria, detrusor, and serosa. Starting inside the bladder, the urothelium mucosal layer is that which is exposed directly to the bladder’s contents, protecting the tissue from any toxic effects. On top of this layer is the lamina propria, which is also sometimes referred to as the submucosal layer, which serves as a connective tissue full of the major blood vessels feeding the bladder. Next, the most contributing level of the tissue is located: the detrusor. This layer is the muscular component of the bladder wall, which contracts upon signal to void the bladder’s contents. The final layer, surrounding the outside of the urinary bladder, is known as the serosa, and this layer is dense with collagen fibers. These are the main levels involved with any species, but there can be many differences between species.1

Aside from storing the urine, the urinary bladder is also responsible for the appropriate emptying of its contents. The micturition reflex occurs when the pressure buildup in the bladder wall travels to the sacral region of the spinal cord, and when the wall becomes stretched enough, neurons from the spinal cord send a message to the muscle to contract. Also involved is the transmission of the message to the brain, which causes the innate desire to urinate. Although short term inhibition is possible, this eventually will allow for the voiding of the bladder’s contents.2

The extracellular matrix in the bladder includes both elastin and collagen, with both types I and III collagen fibers being present. These fibers allow for the bladder’s compliance and stretching ability. The type III collagen coils have been found to uncoil in a specific way as the bladder fills, allowing the fluids to accumulate properly with minimal pressure buildup. Strain is initially compensated by using these unfolding coils and a thinning mucosal layer, but the smooth muscle causes the strain to reach a maximal point at which voiding is necessary.3 The exact behavior of these fibers is still not completely understood, but some different types of testing have been used in an attempt to understand this and relate it to overall bladder biomechanics.

Uniaxial testing has been popular in the past, during which a thin piece of tissue is placed into a custom made device that pulls on the tissue with a set pressure in one direction, while the results are recorded with a computer. However, the stress-free nature of the other side has led current researchers to use biaxial testing instead, during which the same general process is applied in two directions. Due to the more accurate
replication of true bladder conditions, this means of testing has proven to be more effective. Mechanical properties in combination with structural properties are the basis for a mathematical model of the bladder wall tissue. The current study will focus on the structural aspect, i.e. on the orientation of the fibers in the wall tissue using a small angle light scattering (SALS) device after the whole bladder has been put under different filling, and hence stretching, states. This device, pictured below, uses a laser to direct light through a thin sample of tissue, and the resulting scattering pattern is captured via a digital camera as the laser scans the sample (Figure 1). The calculations involved with processing the data are based on the theory of single slit diffraction, and because of potential interference, the data will only be useful for tissue samples under approximately 0.5 millimeters thick. Custom-made software will be used to make several angle measurements and calculations to calculate the fiber orientation from the experimental bladder tissue.

Figure 1: Small angle light scattering (SALS) device’s composition and setup.
Methods/Goals

Whole rat bladders will be obtained, and these will be carefully injected with known volumes of colored silicone gel through connected catheters. Each bladder will undergo a decellularization protocol, which will dissolve the smooth muscle cells and leave only the components of the extracellular matrix, collagen and elastin. Markings will be attached to the outside of the bladder using glue to allow for later determination of the coordinates. A laser scan arm (FARO USA, Lake Mary, FL) will then scan the entire surface of each bladder, thereby determining the three dimensional surface coordinates. From this information the exact three dimensional coordinates of the markers will be identified. The bladders will then be carefully cut in sections in such a way as to minimize curvature and maximize the ability to ascertain where exactly each piece was located in the original three dimensional structure.

The pieces will be “cleared” using a glycerol solution to remove molecules that may impede the laser’s function, and then these pieces will be placed in the device itself to obtain the light measurements. These images will be analyzed and the fiber orientation at each position will be obtained. At this point a method will be employed to determine the three dimensional coordinates of the tissue samples on each section, and this data will need to be mapped back onto the original three dimensional surface map of the bladder.

If all goes well, this project will demonstrate the fiber orientation in the bladder as a whole, instead of only at the microscopic level. It is hoped that seeing the data fall together in this way will be able to shed more insight on the behavior of the organ’s tissue, especially on the regional level. In addition, this information will be useful for future finite element modeling of the urinary bladder during filling and voiding.

References