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## Introduction

- Tissue engineering is a promising strategy for penile tunica albuginea (TA) regeneration as a treatment for Peyronie's disease
- Design criteria for engineered tissue must be established, but there is a dearth of information regarding the biochemical makeup of the TA
- By analyzing TA biochemistry, tissue engineers can use measurements as a gold standard for engineering tissues in animal models and the clinic

## Peyronie's Disease

Peyronie's Disease (PD) is caused by scar tissue in the tunica albuginea, and causes painful, curved erections. One potential solution is to replace the tunica albuginea with a tissue engineered prosthesis.



Healthy erect penis



Bending caused by PD

## Objective

**To perform an initial quantitative characterization of the matrix components and crosslinks of porcine TA**

- Collagen is the protein that give structure to many different collagenous tissues
- Quantification of collagen content and its crosslinks are particularly relevant to tissue engineering
- Elastin, which allows for large strains of tissues, is also necessary to quantify, because its deposition correlates to the hyperelastic nature of the tissue

## Materials & Methods

- 1 cm x 0.5 cm pieces of porcine tunica albuginea were isolated from the penises of six skeletally mature male domestic farm pigs
- Wet weights (WW) were followed by lyophilization and measurement of dry weights (DW)
- Collagen (COL) was measured with hydroxyproline assay
- Pyridinoline (PYR) and desmosine (DES) were quantified with a liquid chromatography-mass spectrometry assay
- Elastin (ELN) was estimated with a molar ratio of DES
- Three biological replicates were tested from each of the six pigs and averaged for each data point
- Hematoxylin and eosin (H&E) staining was performed following the manufacturer's instructions, and a representative image is shown under 4x magnification.

## Results (Histology)

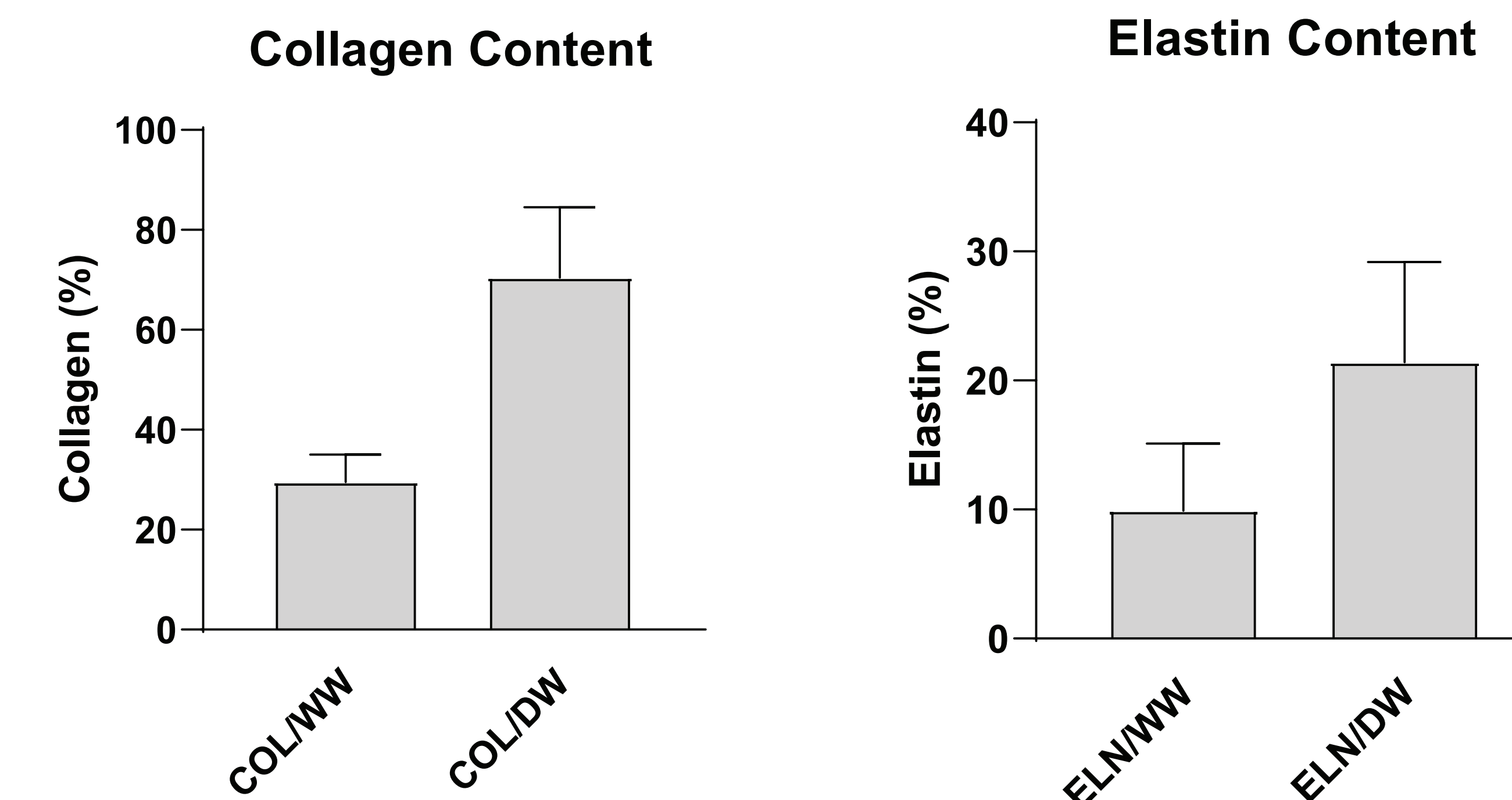
- Tunica albuginea (TA) and corpus cavernosum (CC) are visualized under H&E staining
- Both sections stain positive for eosin. Cell nuclei are present in the CC, collagen fibrillation in the TA is visible



1 mm

## Results (Biochemistry)

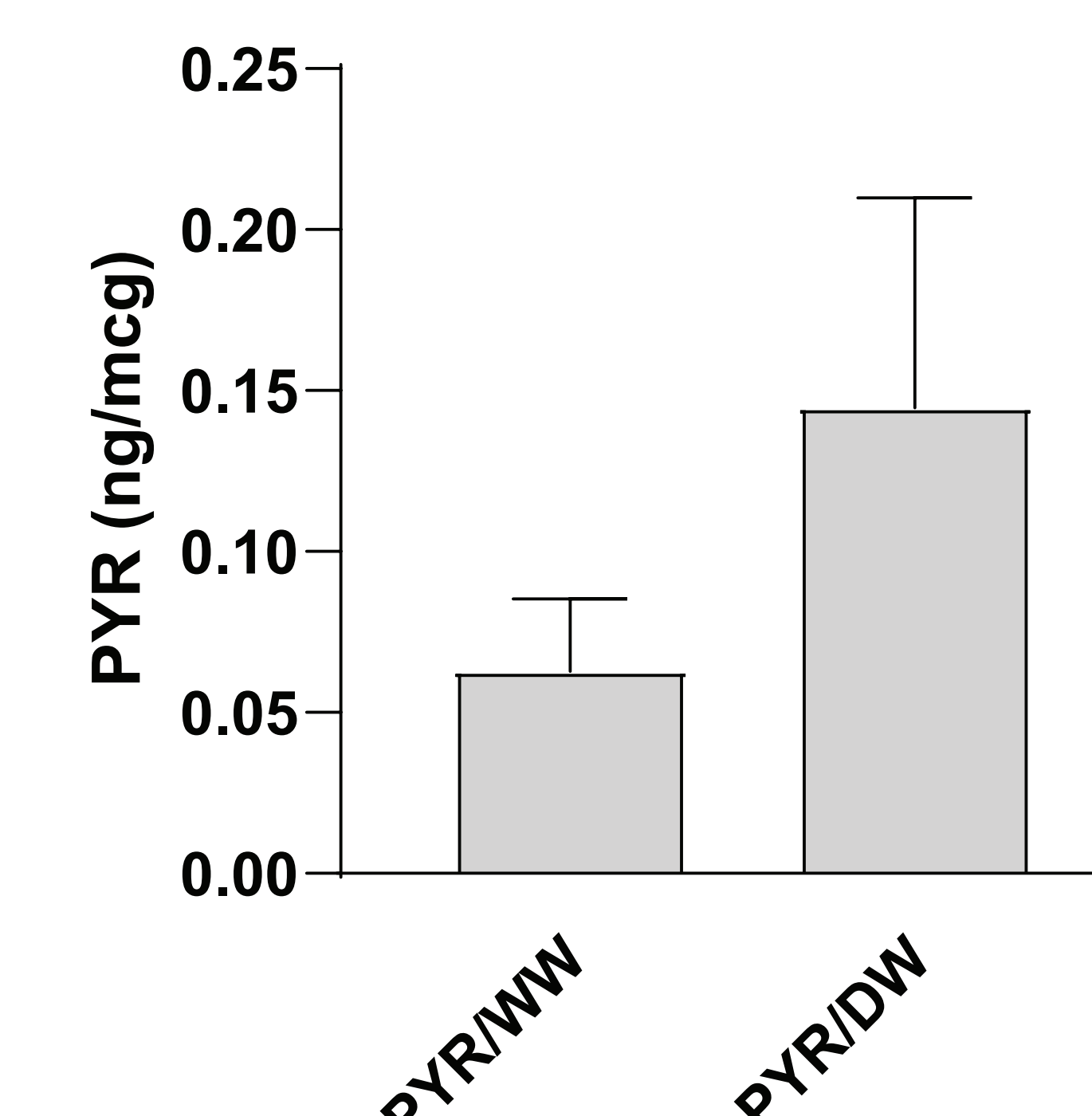
- Hydration =  $58.1 \pm 9.3\%$



- COL/WW =  $29.6 \pm 5.5\%$  and COL/DW =  $70.5 \pm 14.1\%$
- One pig had significantly less collagen content than the other five pigs (ANOVA with *post hoc* Tukey test)
- Collagen was the most abundant matrix component

- ELN/WW =  $9.9 \pm 5.2\%$ , ELN/DW =  $21.4 \pm 7.8\%$
- No differences in elastin content among all pigs
- The combination of collagen and elastin account for over 90% of the total dry weight

### Pyridinoline Content



- PYR/WW =  $0.06 \pm 0.02$  ng/μg
- PYR/DW =  $0.14 \pm 0.07$  ng/μg

## Discussion

- This study represents the first initial biochemical characterization of the porcine penile TA and establishes the abundance of collagen and elastin in healthy porcine tissue**
- Collagen and elastin together account for over 90% of the total dry weight of the TA, providing a strong and extensible extracellular matrix
- The high presence of elastin is similar to other tissues that contract and reshape, such as arteries and skin
- Mechanical characterization will also provide gold standards to ensure that engineered implants will be durable and effective in the *in vivo* environment

## Conclusions & Future Work

- The data from this work will assist with identification of a suitable animal model for penile tissue engineering and will determine standards for engineered TA
- An interspecies comparison is currently in progress that incorporates human cadaveric tunica albuginea
- Future characterization work also includes mechanical analysis of the tissue, and bottom-up proteomic analysis to fully understand all components that make up the TA

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