

Introduction

- Lichen sclerosis (LS) is an idiopathic chronic inflammatory disease of the genital epithelium
- Affected men develop severe urethral stricture disease which is refractory to conventional treatment
- The pathophysiology of inflammation and fibrosis in LS is unknown, precluding development of new treatments.
- Other fibrotic diseases display increased collagen content and decreased fiber width reflective of increased collagen cross linking.

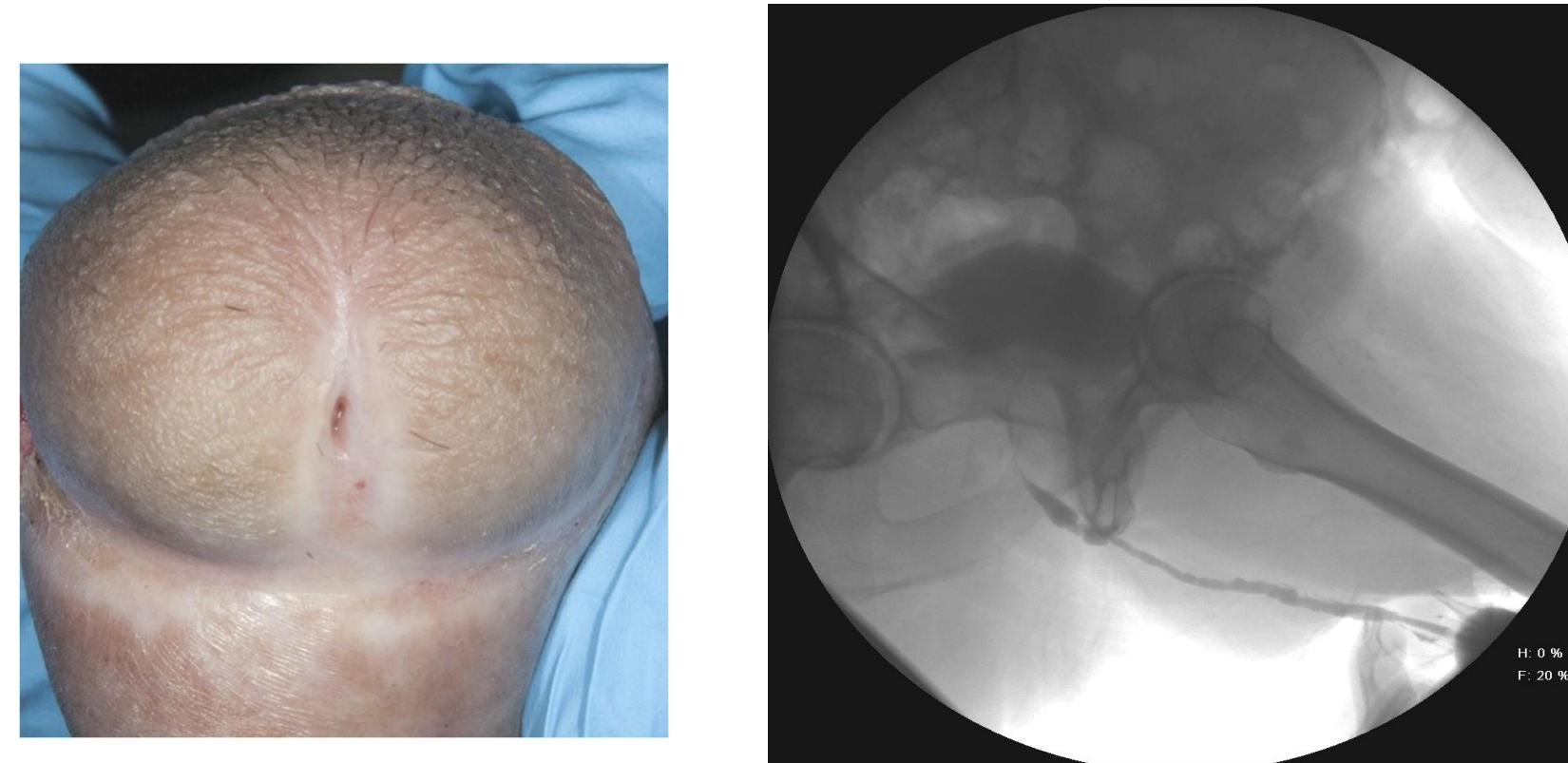


Figure 1: Representative clinical images of lichen sclerosis and related urethral stricture disease. (A) Clinical photograph of the characteristic whitish discoloration of the periurethral glans penis. (B) A retrograde urethrogram demonstrating the characteristic involvement of the entire male anterior urethra with sparing of the proximal bulbar urethra.

Hypothesis

We hypothesize that genital skin and male urethra affected by LS will have increased total collagen content and decreased fiber width compared to patients without LS, recapitulating the pattern seen in other fibrotic diseases

Methods

- Urethral, vulvar, and foreskin surgical pathology specimens were obtained following vulvar biopsy (women), and circumcision or urethroplasty (men).
- All specimens were re-reviewed with a Genitourinary Pathologist to determine the diagnosis of LS
- Tissue sections were stained with picosirius red & imaged with fluorescent microscopy
- An observer blinded to LS status and tissue type acquired five 20x stromal images from each specimen
- Collagen content was quantified using Image J as the mean proportion of image area stained.
- Mean collagen fiber width, length, and count were obtained using CT-FIRE image analysis software.

Results

	Lichen Sclerosis (n=29)	Non-Lichen Sclerosis (n=23)
Gender		
Male	20	14
Female	9	9
Tissue Type		
Vulva	9	9
Urethra	7	7
Foreskin	13	7

Table 1: Gender and tissue type of included lichen sclerosis and control patients

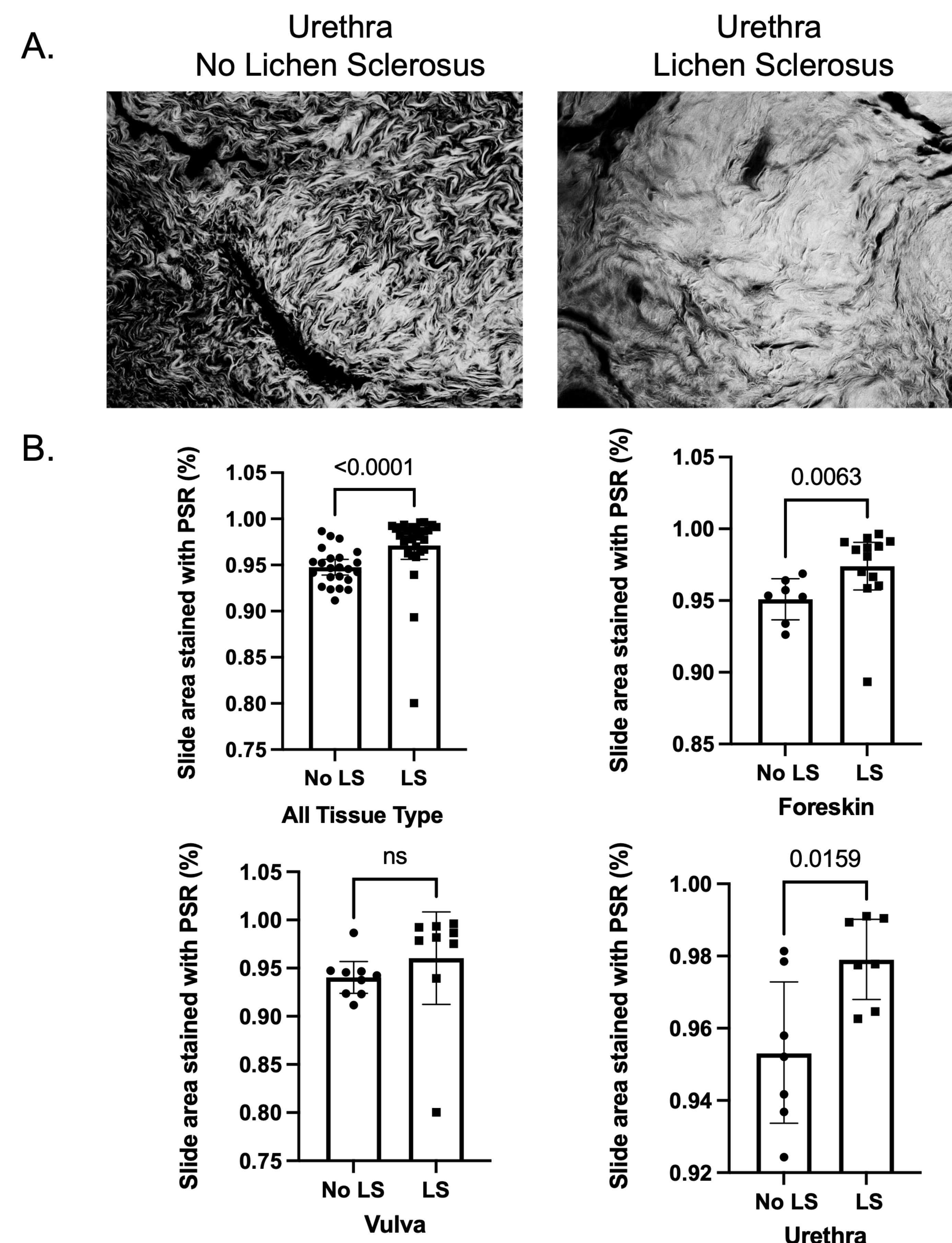


Figure 2: Comparison of collagen content in human vulva, urethra, and foreskin with and without LS. (A) Fluorescent microscopy images of PSR stained urethral collagen demonstrating increased collagen content and density in LS compared to non-LS tissues. (B) Comparison of total collagen content defined as proportion of slide area stained with PSR. Results are mean $\pm$ CI. Brackets indicate pairwise comparisons with p values listed above. ns = not significant, LS = lichen sclerosis, PSR = picosirius red.

Results (cont.)

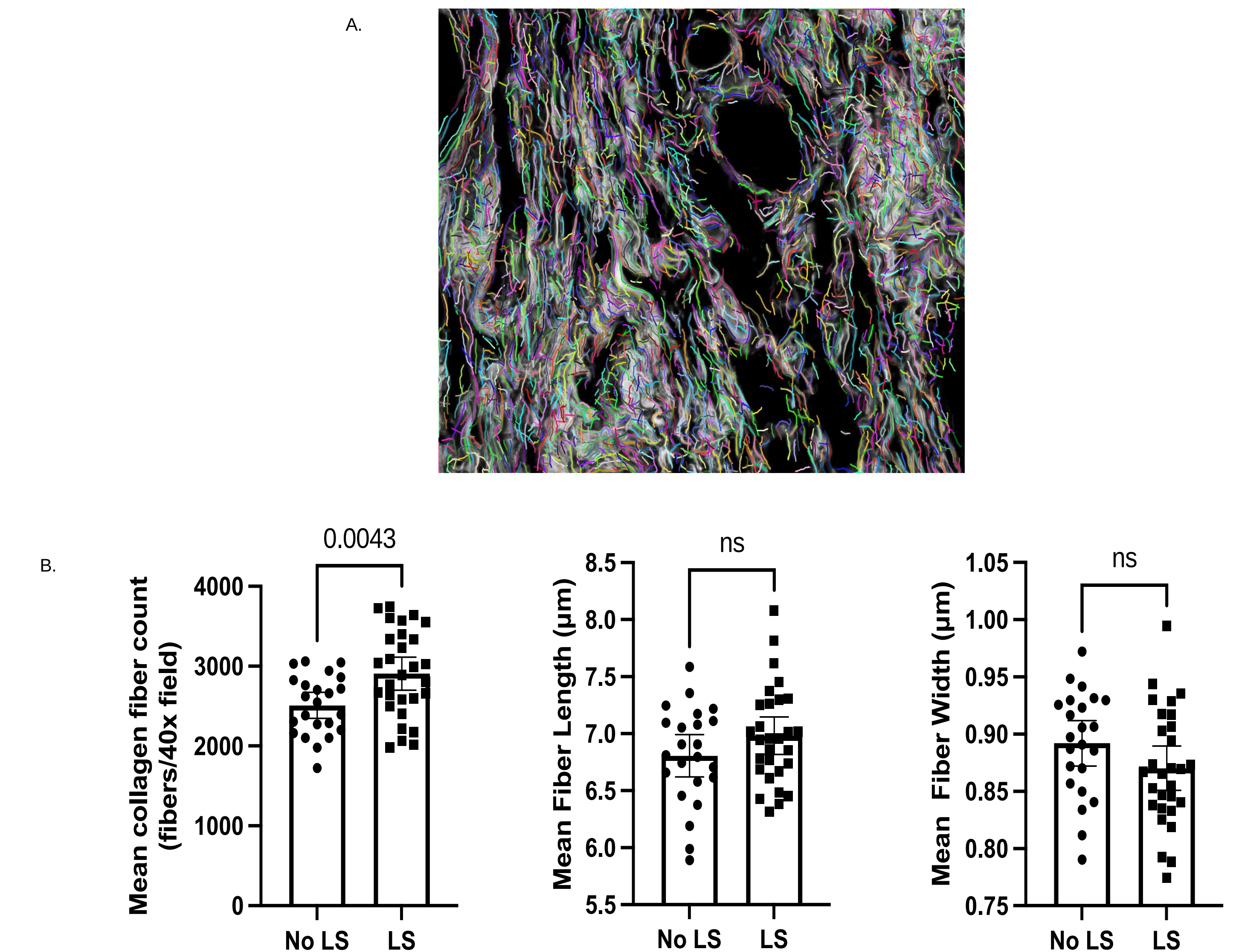


Figure 3: Comparison of collagen fiber count, length, and width in human vulva, urethra, and foreskin tissue with and without LS. (A) Representative 40x image demonstrating an area of subepithelial collagen deposition with superimposed curvelets from the CT-FIRE software package. Each colored curvelet is fit to an individual collagen fiber and analysis of these curvelets yield the data presented in the remainder of the figure. (B) Mean collagen fiber count is significantly increased in all LS tissues (mean = 2906; SE = 100.8) compared to all non-LS tissue (2509 $\pm$ 78.03; $p = 0.0043$). Mean fiber length and fiber width was not significantly different between all LS versus all non-LS tissue. Brackets indicate pairwise comparisons with p values listed above. ns = not significant, LS = lichen sclerosis.

Conclusion

- Total collagen content and fiber count is increased in LS tissues compared to non-LS tissues.
- We found that collagen fiber width and length were similar with respect to tissue type and LS status.
- However, the high density of collagen fibers may have precluded proper curvelet fit and prevented accurate assessment of fiber characteristics.
- To address poor curvelet fit, we plan to use texture analysis to quantify differences in collagen organization in LS tissues.

Acknowledgements

This research was supported by: K12DK100022NIH/NIDDK