

RESEARCH ARTICLE

Sericin improves alginate-gelatin hydrogels' mechanical properties, porosity, durability, and viability of fibroblasts in cardiac spheroids

Supplementary File

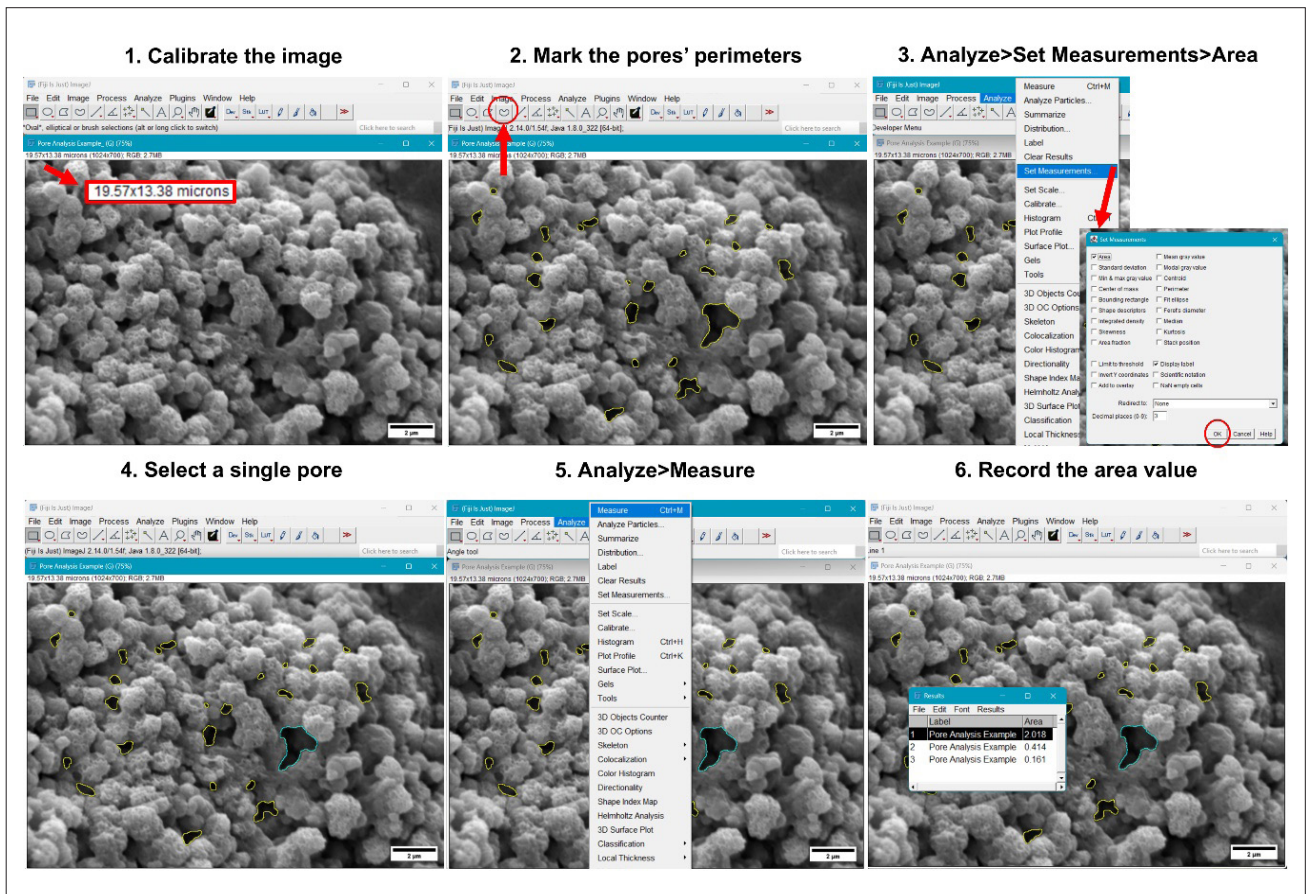
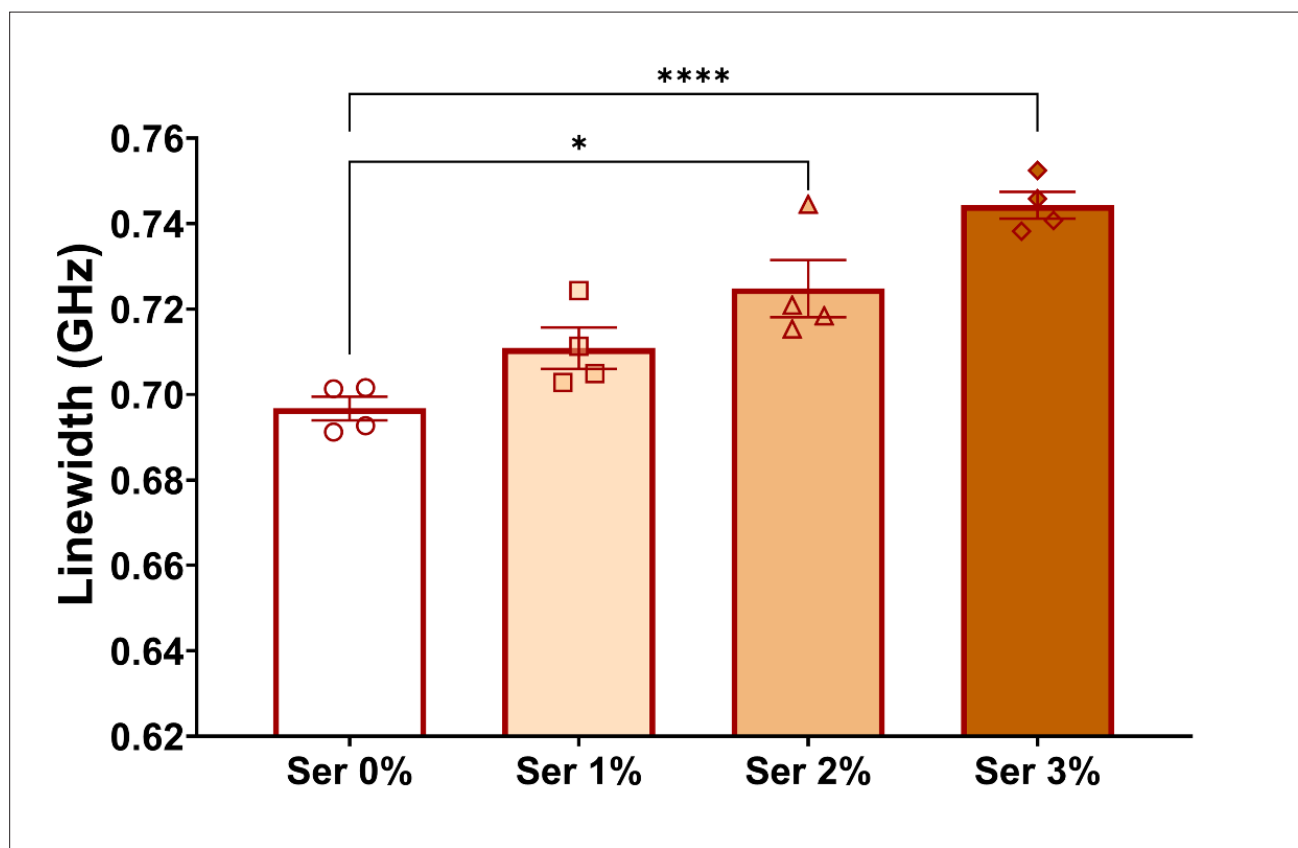
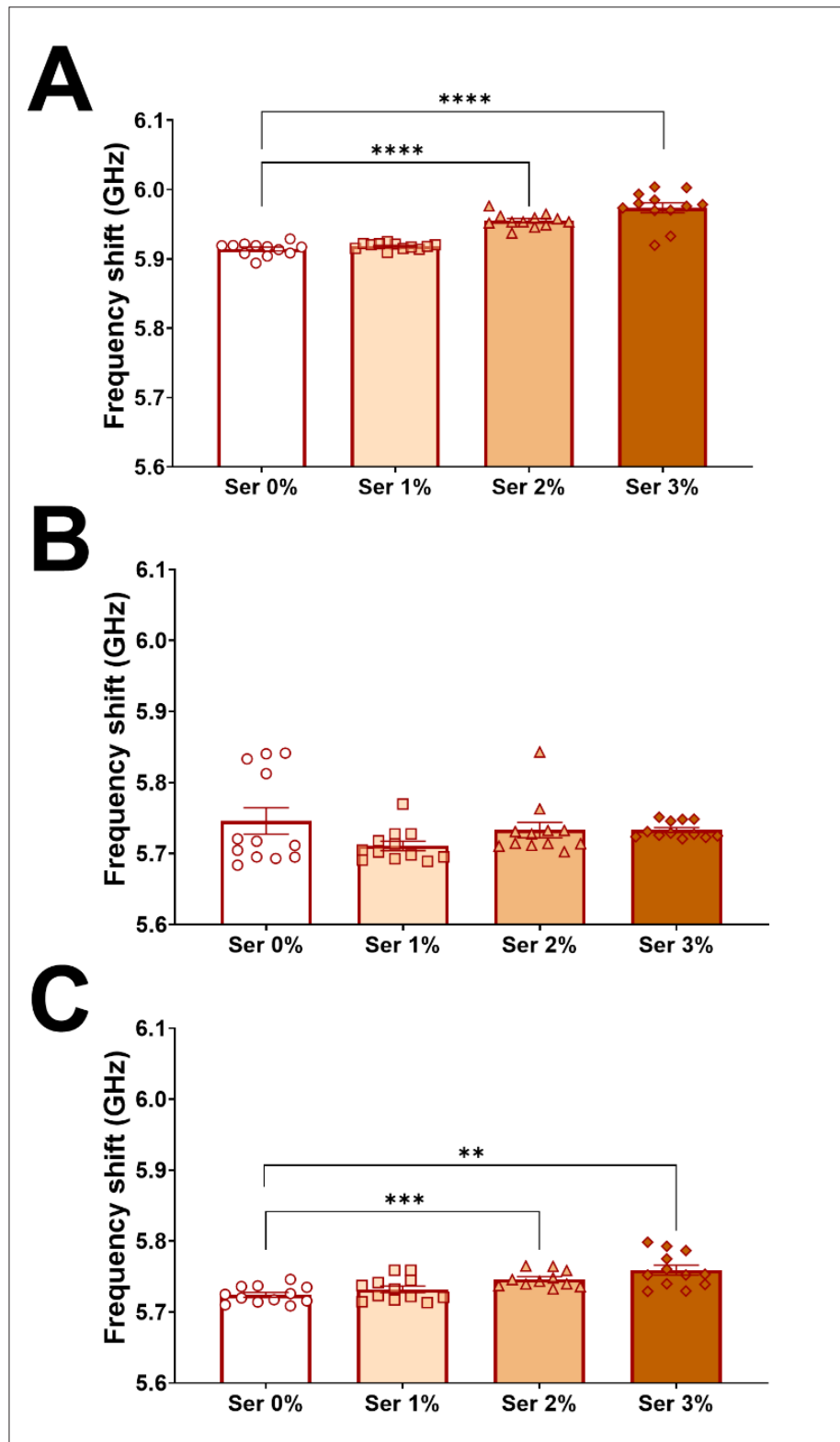


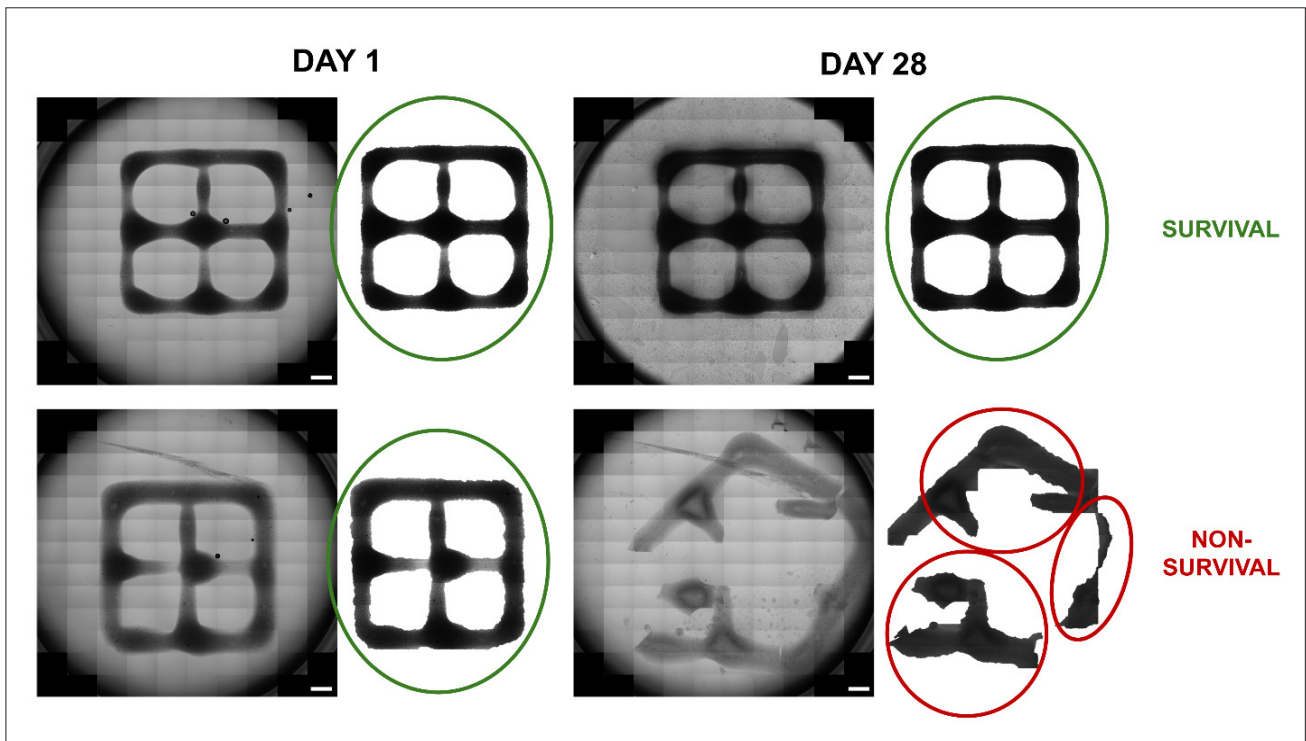
Figure S1. Schematic representation of the steps taken to analyze hydrogel pores by using ImageJ software.



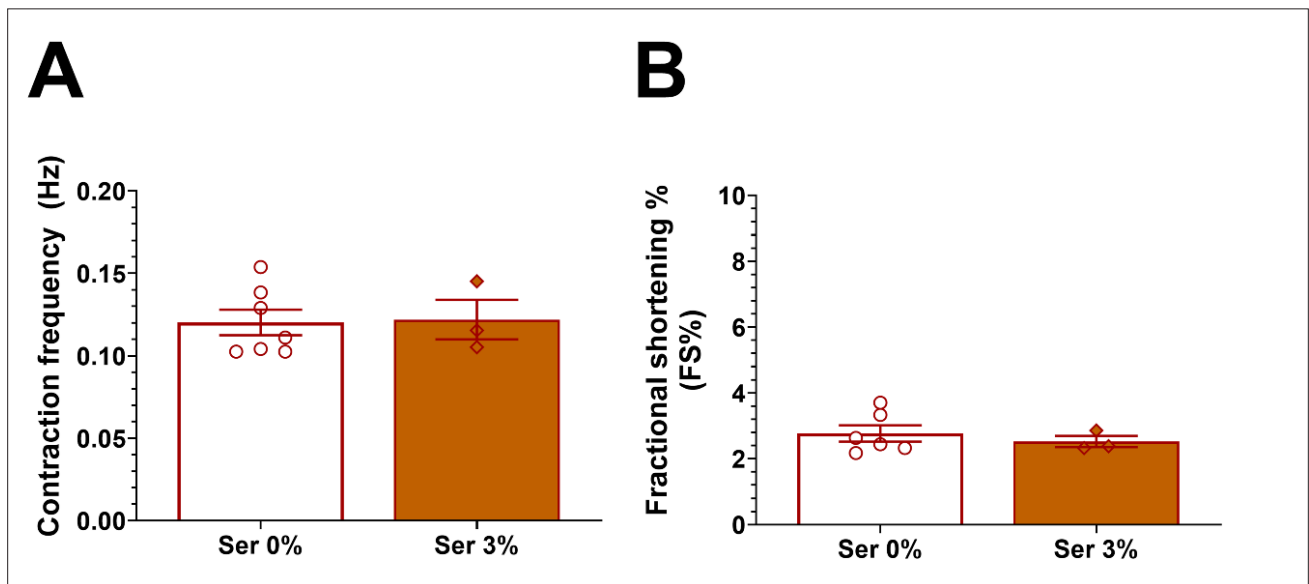
**Figure S2.** Brillouin microspectroscopy linewidth analyses on sericin-containing alginate-gelatin hydrogels before crosslinking. Brown-Forsythe and Welch ANOVA test was used to compare the groups ( $*p < 0.05$ ,  $****p < 0.0001$ ). Error bars represent the mean  $\pm$  SE and are within the symbols if not visible ( $n = 4$ ).



**Figure S3.** Brillouin microspectroscopy analyses sericin-containing alginate-gelatin hydrogels generated by using PureSilk® Sericin (Fibrothelium). Frequency shift measurements of hydrogels' samples (A) before crosslinking, (B) after crosslinking, and (C) after incubation with the medium. Brown-Forsythe and Welch ANOVA test was used to compare the groups (\*\* $p < 0.01$ , \*\*\* $p < 0.001$ , \*\*\*\* $p < 0.0001$ ). Error bars represent the mean  $\pm$  SE and are within the symbols if not visible ( $n = 11$ ).



**Figure 4.** Schematic representation of the bioprinted patches' durability evaluation through a visual inspection of the pictures obtained by using EVOS M7000 Imaging System. At each time point, the shape and structure of each patch were compared to the initial shape obtained immediately after crosslinking to detect any deviation. When the patch appeared intact without any fragmentation, we considered it “survival.” On the contrary, when we detected a fragmentation, the patch was recorded as “non-survival.” Scale bars: 2 mm. .



**Figure 5.** Contractile activity of CSs embedded in Ser0% and Ser3%. Statistical analyses of (A) contraction frequency and (B) fractional shortening % of CSs were performed using unpaired *t*-test with Welch's correction to compare the groups. Error bars represent the mean  $\pm$  SE and are within the symbols if not visible ( $n \geq 3$ ). Abbreviation: CSs, cardiac spheroids.

## Supplementary videos

**Video S1.** Full-length 3D-rendering videos of a representative CS embedded in Ser0% hydrogel.

**Video S2.** Full-length 3D-rendering videos of a representative CS embedded in Ser1% hydrogel.

**Video S3.** Full-length 3D-rendering videos of a representative CS embedded in Ser2% hydrogel.

**Video S4.** Full-length 3D-rendering videos of a representative CS embedded in Ser3% hydrogel.