

**P15. Structural Integrity Of Cryopreserved Human Heart Valve Collagen And Elastin**

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**OBJECTIVES:** Cryopreservation is commonly used to prepare human heart valves for long-term storage. Despite the excellent hemodynamic performance and durability of cryopreserved allografts, reports have questioned the structural integrity of their constituent proteins.

**METHODS:** This study evaluates the effect of cryopreservation on collagen and elastin integrity within the leaflet and conduit of 6 aortic and 6 pulmonary human heart valves. Two-photon laser scanning confocal microscopy (LSCM) was used to assess structural changes through visualization of elastin autofluorescence [760 nm excitation; 468 nm emission] and collagen second harmonic generation (SHG) [800 nm excitation; 404 nm emission]. To permit pairwise comparisons, each valve was bisected longitudinally; half was imaged fresh and half was cryopreserved and stored in liquid nitrogen for 1-6 weeks before thawing, rinsing for >1.5 hr in buffered saline, and subsequent evaluation.

**RESULTS:** Qualitative analysis of all resultant images (3-5 sites per surface) indicated the maintenance of collagen and elastin structure within leaflet and conduit post-cryopreservation. Semi-quantitative assessment of collagen SHG and elastin autofluorescence signal intensity showed that average signal intensity was highly variable among both fresh and cryopreserved samples. Changes in average signal intensity between fresh and cryopreserved donor-paired samples exhibited no directional trend. The collective results shows no statistically significant change in collagen or elastin signal intensity due to cryopreservation, although there was a reduction in high intensity outliers.

**CONCLUSIONS:** These two-photon LSCM results do not support a detrimental effect of cryopreservation on collagen and elastin structural integrity in human heart valves.

