**Introduction:** Allograft bone from human cadavers is implanted in over 700,000 surgical procedures annually. These allografts have traditionally been stored in either a freeze-dried (FD) or frozen (FZ) state. Traditional frozen allografts require shipment on dry ice and storage in validated and monitored freezers, while freeze-dried allografts can be brittle and require lengthy rehydration times prior to use. This study investigates the effects of a recently introduced, glycerol-based preservation method, Preservon \(^6\) (PRVN), on the biomechanics and shelf-life of cortical bone. The benefits of this new preservation method include convenient graft shipment, storage at ambient temperature without expensive freezers, and minimal preparation prior to use. This process represents a significant change in the delivery of allograft tissues to healthcare professionals and their patients.

**Methods:** Biomechanics. Cortical bone was processed from the femoral shafts of seven donors into rectangular bars 4 mm x 4 mm x 80 mm and all groups were treated per the AllowashXG \(^6\) process (a patented cleaning, disinfection, and sterilization technology for allograft tissue \(^3\)). The control groups were either FD or FZ using current manufacturing processes. The experimental group was PRVN-treated (a patented preservation method for bone tissue \(^3\)). Each group contained 20 specimens and donors were evenly distributed among groups to account for donor-to-donor variation in mechanical properties. The FD specimens were rinsed for 30 seconds, 5, 15, and 60 minutes. The FZ specimens were rinsed for 2 minutes, and the PRVN-treated specimens were rinsed for 30 seconds. All specimens were rinsed in 0.9% isotonic saline. The testing procedure followed ASTM D790 – 07, a standardized 3-point bend test. Flexural strain, flexural strength, and flexural modulus were calculated.

**Shelf Life.** Allografts representative of the cortical, cancellous, and cortico-cancellous tissue types were chosen to be tested in axial compression. Nine millimeter iliac crest wedges (ICW), 20 mm fibular shaft segments (FS), and 14 mm Cloward dowels (CD) were processed, and at least 11 donors were represented in each group to account for donor-to-donor variation in mechanical properties. The control groups were treated per the AllowashXG process and FD. The experimental groups were treated per the AllowashXG process and PRVN-treated. Allografts were tested at time zero (T\(_0\)) and after one year of real-time aging (T\(_{1yr}\)) at ambient temperature. A minimum of 12 specimens were tested for each treatment per time period, and average compressive strength was calculated by dividing the load to failure by the cross sectional area.

**Results:** Biomechanics. Statistical analyses were conducted using a one-way analysis of variance (ANOVA) to compare biomechanical properties among groups, where p < 0.05 was considered statistically significant. In figures 1, 2, and 3, pairs of means grouped by a horizontal line are not significantly different from each other. For flexural strain, there was no statistical difference found between FZ and PRVN-treated bone; however, both these groups were found to be significantly and statistically less brittle than FD bone. For flexural strength, all three groups were statistically different from each other and PRVN-treated cortical bone was statistically stronger than both FD and FZ bone. For flexural modulus of elasticity, there was no significant difference between the FZ and PRVN-treated groups. The FD group was statistically higher than the FZ and PRVN-treated groups and therefore more brittle. Also, as evidenced in the following figures, FD bone was not able to recover its biomechanical properties even after rehydration for 60 minutes.

**Discussion:** The data presented here indicate that Preservon treatment offers advantages in handling and storage of allograft bone compared to freeze-dried and frozen preservation. Preservon treated bone was proven to be less brittle than freeze-dried bone and shown to exhibit similar brittle properties to frozen bone. In addition, Preservon treated allografts were equivalent in compressive strength to representative allografts currently produced, and Preservon treated allografts maintained their biomechanical integrity over time. Preservon treatment allows for ambient storage of fully hydrated allograft bone while maintaining biomechanical properties comparable to or higher than freeze-dried and frozen bone. Preservon treatment is a viable alternative to the time and temperature constraints associated with freeze-dried and frozen preservation methods for bone allografts.

**References:** 1. US Patents 5,556,379; 5,820,581; 5,977,034; 6,024,735. 2. US Patents 6,293,970; 6,544,289; 6,569,200; 7,063,726.