

## **Collagen X Fracture Biomarker Suggests Surgical Intervention for Fracture Nonunion Drives Endochondral Bone Response**

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**INTRODUCTION:** Surgical intervention for fracture nonunion theoretically results in re-initiation of fracture healing, and therefore potentially future union. No validated method exists to monitor the results of surgical intervention in nonunion care. Growing evidence supports collagen X as a biomarker (CXM) for endochondral bone healing. We hypothesized that CXM increases following surgical intervention for nonunion against nonsurgical treatment and continued observation.

**METHODS:** Patients presenting with clinically defined delayed or frank nonunion of tibial or femur fractures were enrolled in a longitudinal observational study. Study participants were treated with either observation or surgical intervention following shared clinical decision making. Dried blood spots (DBS) were collected at all clinic visits. DBS was analyzed using previously published Elisa test to detect the trimeric breakdown product of collagen X, the base protein involved in cartilaginous intermediate tissue along the fracture healing pathway. Wilcoxon rank sum tests were used to test the difference in CXM between both treatment and nonunion types.

**RESULTS:** Twenty-two (13 nonunion, 6 delayed union, 3 septic nonunion) patients who originally sustained fractures of the femur or tibia (mean age: 37 years [SD, 12]; 82% male, 45% smoker) enrolled and 9 underwent surgical intervention (median days after index care: 321 [265 - 1,442]). Mean CXM in observational treatment was 649 pg/mL compared to 778 pg/mL following nonunion fracture surgery ( $p = 0.11$ ). In the nonunion intervention population, the average CXM increased from 536 pg/mL preoperatively to 700 pg/mL postoperatively ( $p = 0.04$ ). CXM plotted against time since injury demonstrated that patients who received a nonunion intervention developed an increased endochondral response. Mean CXM in delayed unions was 1027pg/mL, higher than that of nonunion (606 pg/mL,  $p = <0.001$ ).

**DISCUSSION AND CONCLUSION:** Early stage research demonstrates Collagen X levels may present higher in delayed unions than in nonunions, likely signifying that delayed unions may continue to have ongoing endochondral activity. Additionally, and perhaps most importantly, patients receiving surgical intervention for fracture non and delayed union demonstrated higher CXM levels after treatment than during pretreatment observation, implying that CXM may capture the effect of treatment on union progression. Our findings support the theory that nonunion care continues to utilize an endochondral pathway. CXM may be an advantageous marker of fracture nonunion and prospective interventional studies.