

Safety And Immunogenicity Of Human Decellularized Bone Graft Using Supercritical Carbon Dioxide In An *In Vivo* Study

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INTRODUCTION:

Supercritical carbon dioxide (SCCO₂) technology offers a non-toxic, efficient method for decellularization, preserving graft integrity while enhancing osteogenic potential. Unlike traditional methods, SCCO₂ minimizes residual chemicals and immunogenicity risks. This study evaluates the safety and immunogenicity of SCCO₂-DBG (Decellularized Bone Graft) *in vivo*, aiming to establish its clinical potential as a superior alternative to fresh frozen bone graft (FFBG).

MATERIALS & METHODS:

Twelve male New Zealand white rabbits were randomly divided into two groups, undergoing femoral trochlea defect surgery with graft implantation. Blood samples were collected to analyse cytokine levels (IL-6, TNF- α , IFN- γ) using ELISA. Histological analysis of kidney and spleen tissues and systemic toxicity assessments, including spleen index and body weight, were conducted.

RESULTS:

Both groups showed comparable weight gain and no significant pyrogenic responses or spleen index differences. Histological analysis revealed minimal inflammatory changes in the kidney and spleen. Cytokine profiles showed significantly lower IL-6 and TNF- α levels in the SCCO₂-DBG group at Day 14, indicating reduced systemic inflammation.

DISCUSSION:

Histological analysis revealed no significant abnormalities in the kidneys or spleen for both graft types, supporting their safety. SCCO₂-DBG showed significantly reduced IL-6 and

TNF- α reactivity, indicating a milder inflammatory response compared to FFBG. Hematological stability and fewer spikes in eosinophil and basophil counts further highlight SCCO₂-DBG's reduced systemic immune activation. Additionally, stable systemic parameters, including weight gain, spleen index, and pyrogen testing presenting it as a safer alternative for clinical grafting applications.



CONCLUSION:

This study highlights that histological, cytokine, and hematological analyses showed minimal systemic impact and lower inflammatory responses for SCCO₂-DBG¹. These findings support its potential in bone grafting, emphasizing the importance of decellularization in enhancing graft safety and clinical outcomes.

References:

1) Visser, N., Rezaie et al (2019). Effects of surgical angiogenesis on segmental bone reconstruction with cryopreserved massive-structural allografts in a porcine tibia model.