Polycaprolactone/bioactive glass 45S5 microparticles for bone regeneration: from in vitro characterization to in vivo validation in a cranial defect model

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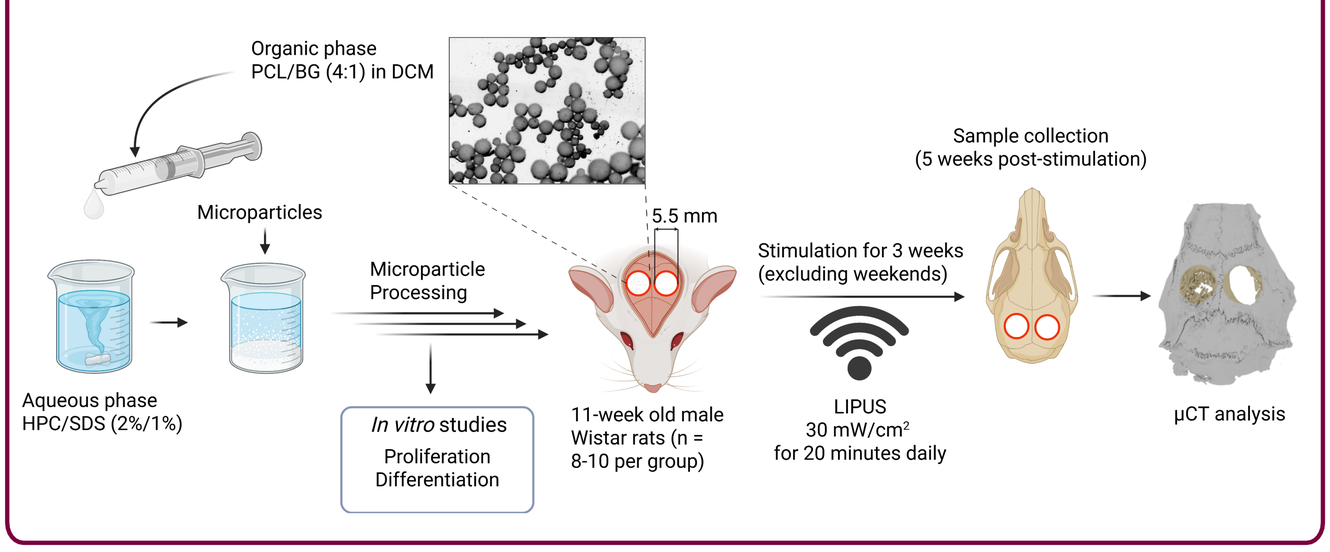
Introduction

Successful healing of critical-size bone defects remains a major challenge, as conventional grafts are limited by donor-site morbidity, immune responses, and availability. This underscores the need for synthetic, bioactive materials that support bone regeneration.

Polycaprolactone (PCL) combined with bioactive glass 45S5 (BG) represents a promising biodegradable and osteoinductive system. In addition, low-intensity pulsed ultrasound (LIPUS) is a non-invasive therapy known to enhance bone healing, though its synergy with such materials remains unclear.

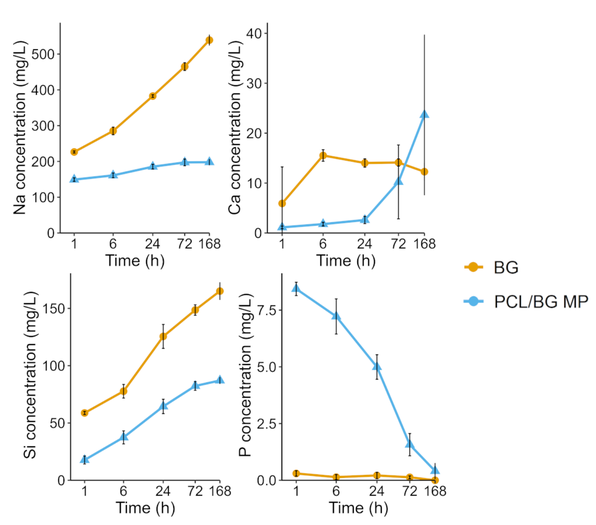
Here, we evaluated PCL/BG microparticles (MP) in vitro for their effect on mesenchymal stem cell response, followed by in vivo assessment in a rat critical-size calvarial defect model to determine whether MPs together with LIPUS stimulation improve bone regeneration.

Methods

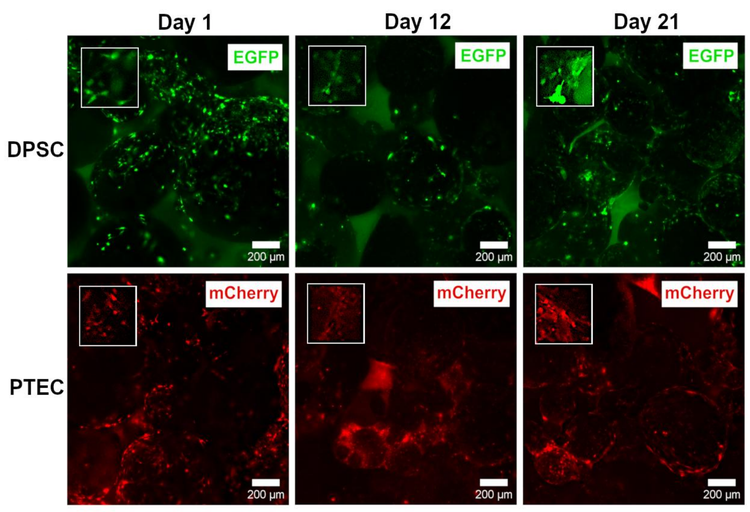


Results

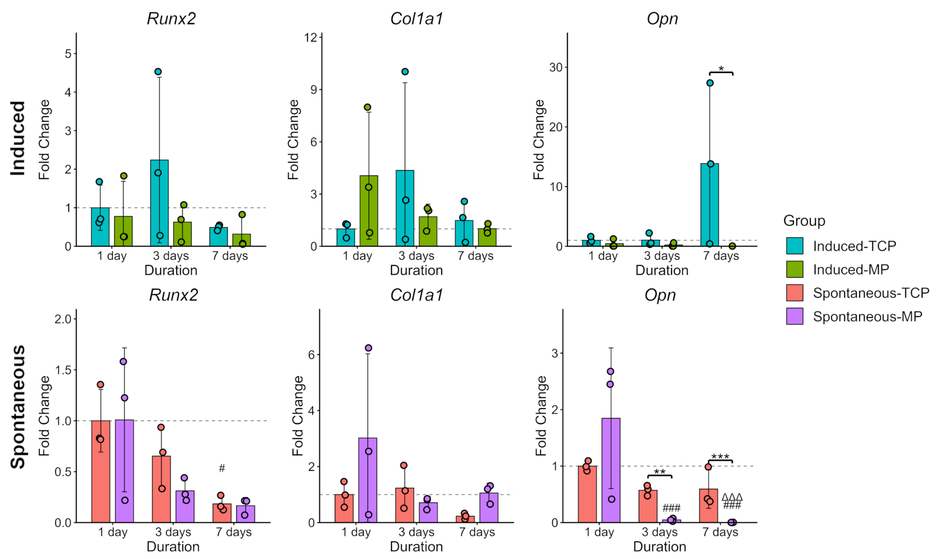
In vitro



Ion release from BG and PCL/BG MP over time. Elemental analysis shows increasing Na, Ca, and Si concentrations over 168 h, with sustained release from PCL/BG microparticles.

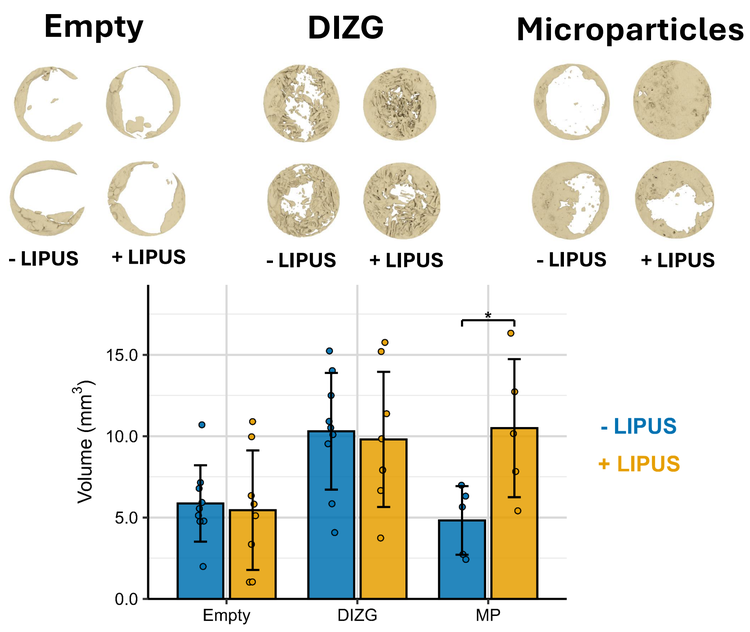


Spatiotemporal distribution of DPSC and PTEC on MP. Fluorescence imaging shows EGFP-labeled DPSC and mCherry-labeled PTEC from day 1 to day 21.

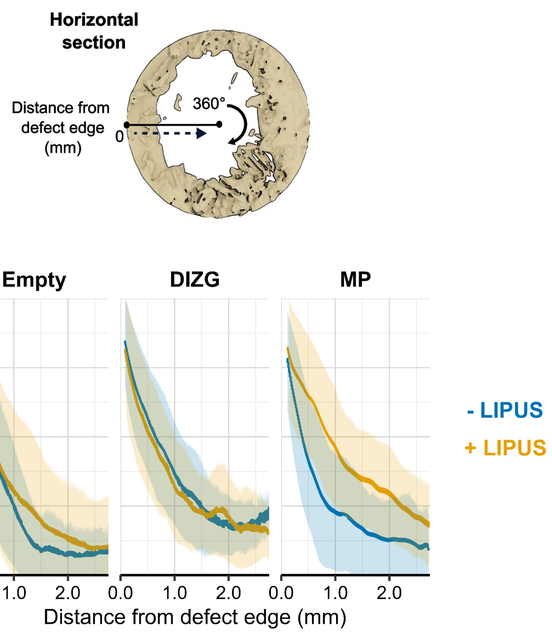


Osteogenic gene expression of cells on TCP and MP. qPCR analysis of Runx2, Col1a1, and Opn shows time-dependent changes under induced and spontaneous conditions.

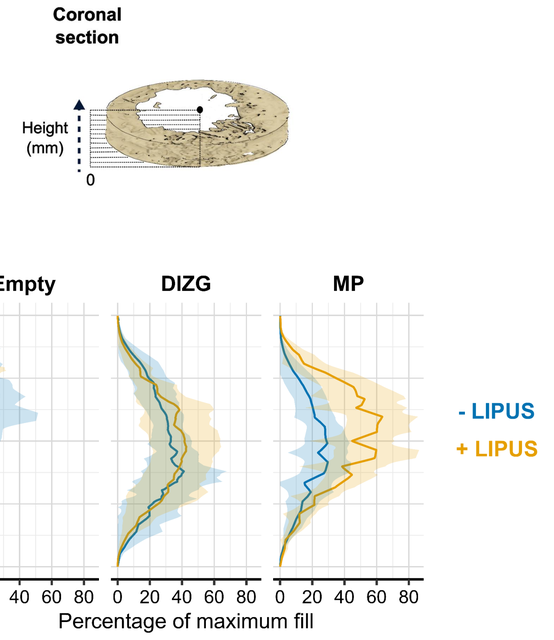
In vivo



Bone volume quantification and representative defect images at 8 weeks post-surgery. Representative 3D reconstructions (top) show defect healing in Empty, DIZG, and MP groups. Quantification (bottom) indicates increased bone volume in DIZG and MP, with MP + LIPUS showing the highest bone formation.



Bone ingrowth profiles along the defect width. Bone fill decreases with distance from the defect edge in all groups. MP + LIPUS shows the highest and most sustained defect fill across the width, followed by DIZG, while Empty groups exhibit limited bone formation largely confined to the periphery.



Bone ingrowth profiles from the defect base. Bone fill varies along the defect height, with MP + LIPUS showing the highest and most uniform fill across the defect. DIZG groups display moderate and relatively consistent bone formation, while Empty groups exhibit lower and more variable fill, indicating less organized regeneration.

Conclusion & Outlook

PCL/BG microparticles fabricated via emulsion solvent evaporation demonstrated good biocompatibility and supported mesenchymal stem cell adhesion, viability, and differentiation in vitro, with reduced proliferation indicating a shift toward osteogenic commitment and suppression of early adipogenic signaling. In vivo, the microparticles enabled bone tissue ingrowth and integration within critical-size defects. Notably, LIPUS stimulation combined with microparticles markedly increased overall bone volume, an effect not observed in other groups, while also promoting more mature tissue formation. These findings suggest a strong synergistic effect between PCL/BG microparticles and LIPUS, although further studies, including histological evaluation, are needed to better characterize the regenerated tissue.

Key findings

- PCL/BG MPs support MSC adhesion and differentiation *in vitro*
- MPs + LIPUS significantly increases bone volume
- PCL/BG MPs support bone ingrowth and defect integration *in vivo*

Acknowledgements

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