Demineralized Bone Matrix Bioinks with Enhanced Odontogenic Differentiation: Synthesis and Characterization

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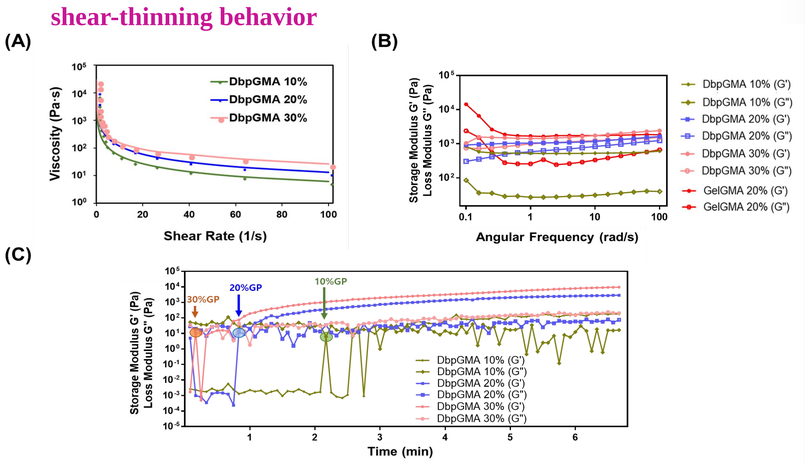
Abstract

Demineralized bone powder (DBP) have demonstrated substantial bioactivity for hard tissue regeneration, particularly in dental tissue engineering. However, the influence of DBP particle size and concentration on functional performance of bioinks remains underexplored.

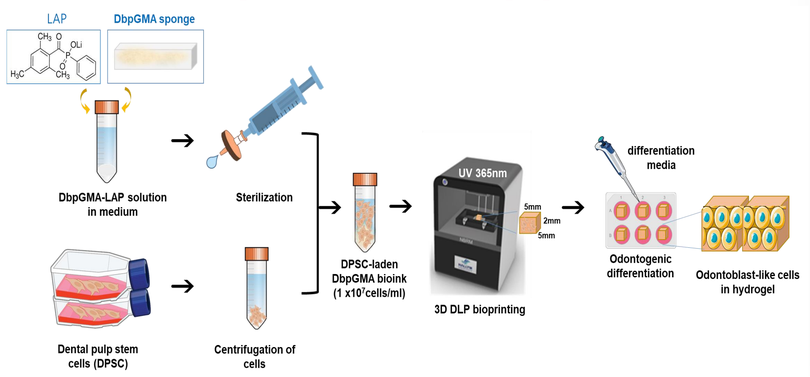
This study aims to elucidate how distinct DBP particle sizes ranges, with elemental composition analyzed before and after demineralization. DBP was chemically modified with GMA to yield DbpGMA, we fabricated hydrogels using DLP bio-printing. The resulting constructs were evaluated for their structural integrity, rheological behavior, and surface morphology. DPSCs encapsulated within the hydrogels were assessed for proliferation and odontogenic differentiation, using DMP-1 and DSPP as key markers. Results revealed that 20% DbpGMA hydrogels with optimized particle size achieved superior printability, balanced mechanical and rheological properties, and promoted cell proliferation and odontogenic marker expression.

The findings underscore the critical role of controlled DBP particle size and concentration in enhancing bio-printing fidelity and bio-functionality of dental tissue constructs. This approach demonstrates the potential of tailored DBP based bioinks for advanced applications in dental regenerative medicine.

F4 **Rheological, mechanical, and structural characterization of DbpGMA (pre-)hydrogels. Viscosity as a function of shear rate for 10 %, 20 %, and 30 % DBP-GMA pre-hydrogel, demonstrating shear-thinning behavior**

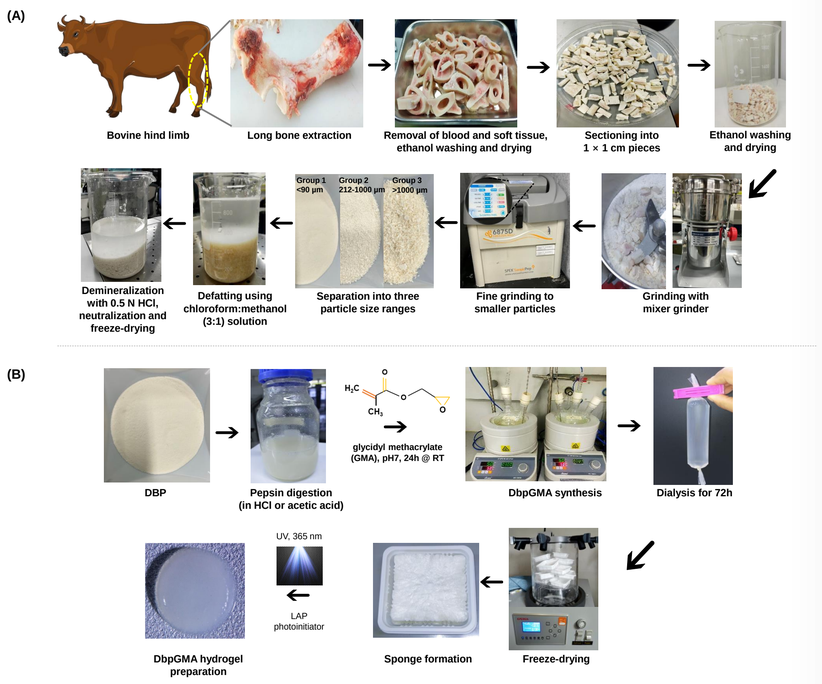


F5 **Schematic illustration of the fabrication process and odontogenic differentiation of DPSC-laden DbpGMA hydrogels. The DbpGMA precursor solution was mixed with LAP and sterilized using a syringe filter. Dental pulp stem cells (DPSCs) were then added to the sterilized DbpGMA-LAP solution to prepare the bioink.**

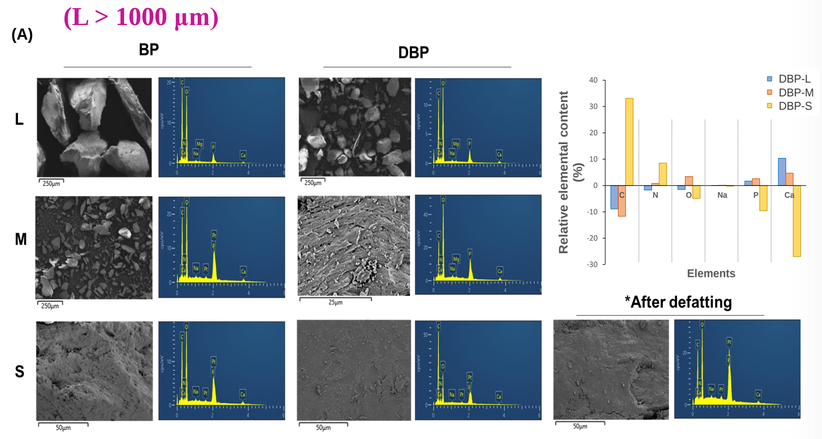


Experiments and Results

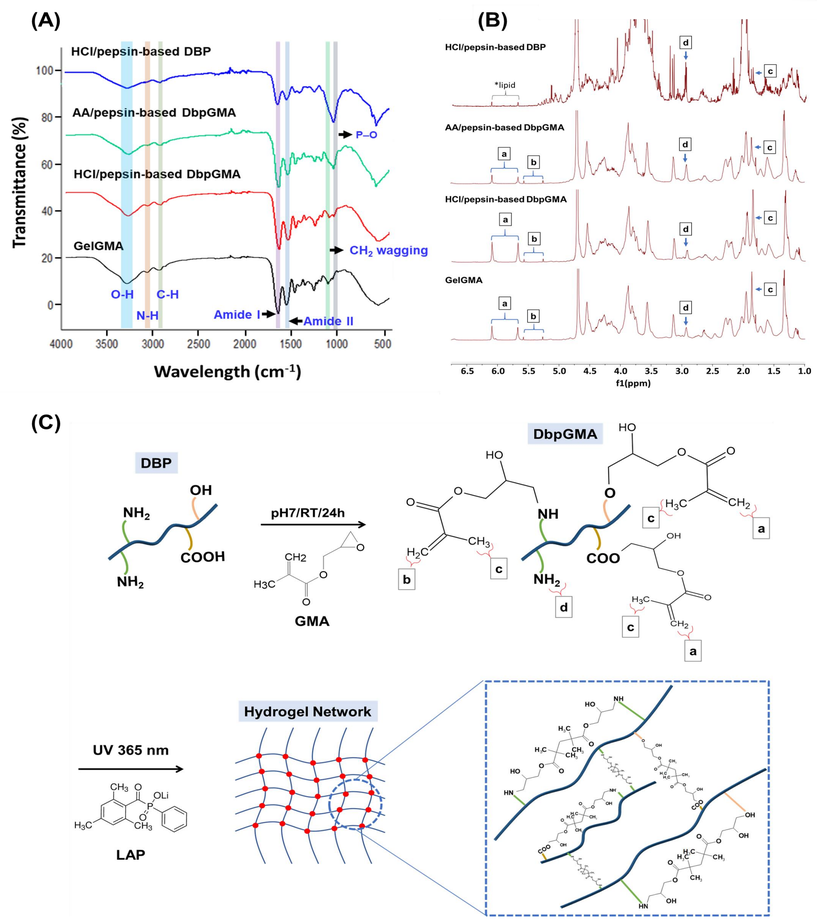
F1 **Preparation process of DBP and DbpGMA hydrogels. Fresh bovine long bones (tibiae and femurs) were cleaned to remove blood and soft tissues, followed by ethanol washing and drying**



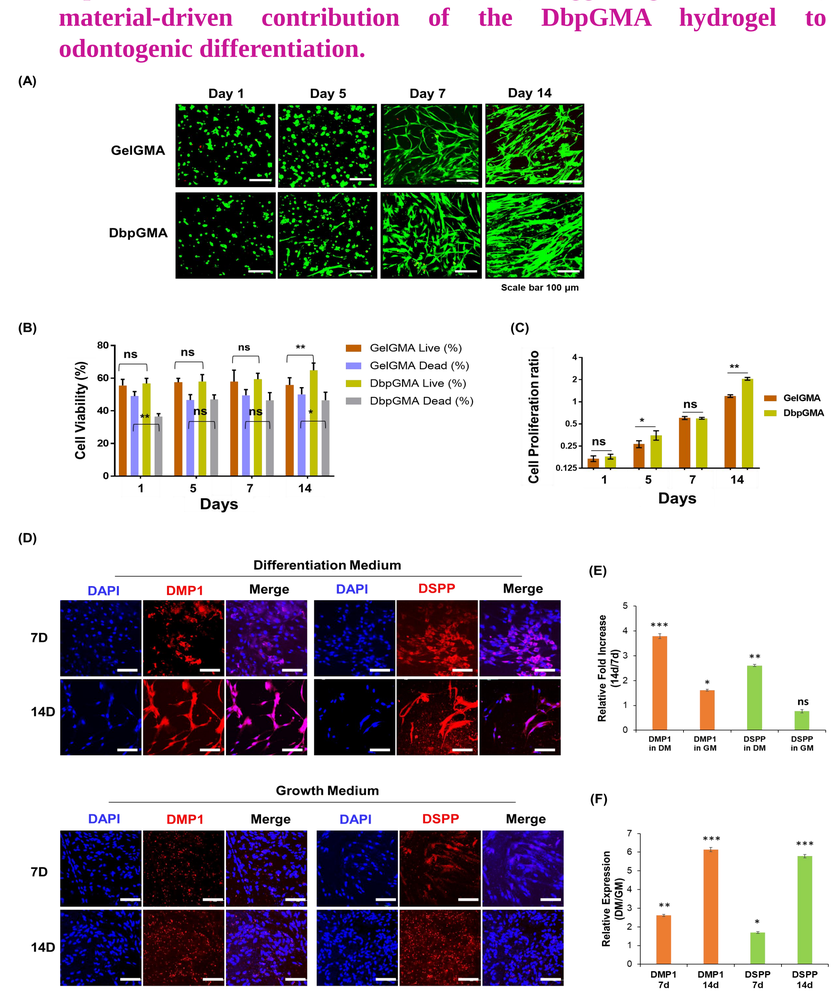
F2 **EDX elemental analysis of DBP. Small particles (S < 90 μm), medium particles (210 < M < 1000 μm), and larger particles (L > 1000 μm)**



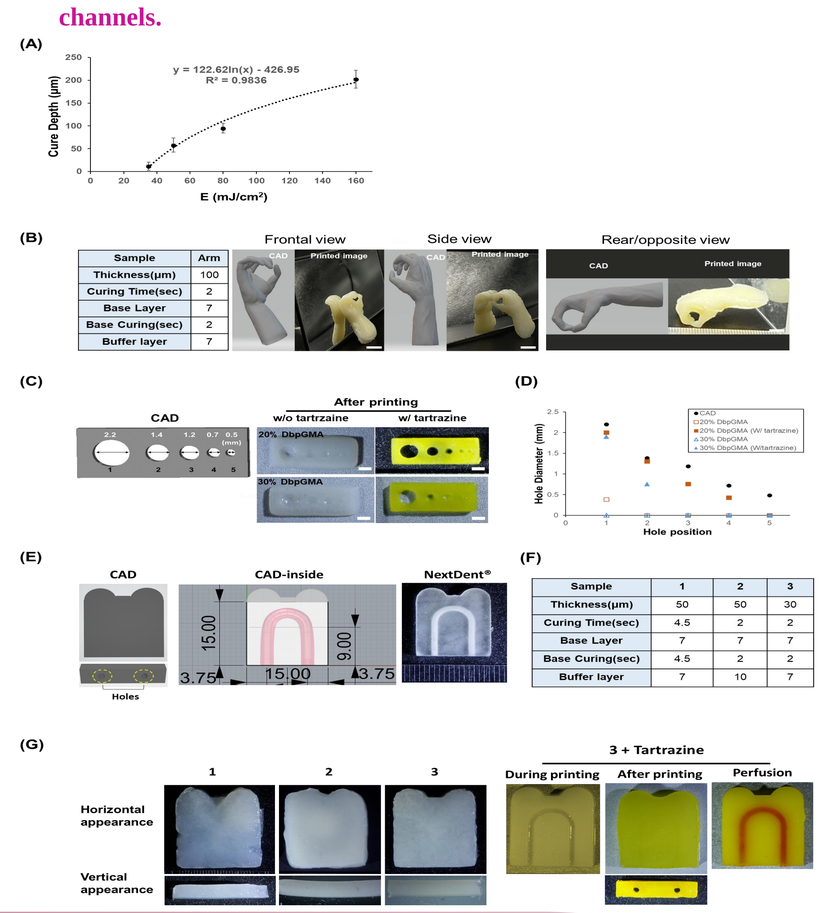
F3 **Structural characterization and proposed reaction mechanism of DbpGMA. FT-IR spectra of unmodified DBP, pepsin-digested DBP, and DbpGMA. Distinct peaks of amide I (1645 cm^{-1}) and amide II (1513 cm^{-1}) indicate preservation of the collagen backbone after modification**



F6 **Cytocompatibility and Odontogenic differentiation of DPSCs in DbpGMA hydrogels. While robust and time-dependent upregulation was observed under DM conditions, a moderate increase in DMP1 expression was also detected in GM, suggesting an intrinsic, material-driven contribution of the DbpGMA hydrogel to odontogenic differentiation.**



F7 **Printability optimization of DbpGMA hydrogels using DLP 3D printing. Parameter optimization improved geometric stability; however, overall shape fidelity remained limited. Incorporation of tartrazine enabled successful fabrication of continuous vascular channels.**



Conclusions

In this study, a novel photocrosslinkable hydrogel (DbpGMA) was developed by chemically modifying demineralized bone particles (Dbp) with glycidyl methacrylate (GMA). Among various particle sizes, DBP prepared using particles smaller than 90 μm effectively retained the collagenous organic matrix, yielding a high-quality DBP sol with improved handling properties. Collectively, DbpGMA represents a promising bone-derived biomaterial for regenerative medicine, particularly for dentin and hard tissue engineering using DLP 3D bioprinting.

Full paper & more info (scan QR)

Published in Volume 154, January 2026, Polymer Testing