Magnesium-iron doped hydroxyapatite/collagen scaffolds for enhanced antimicrobial activity and bone regeneration

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Introduction

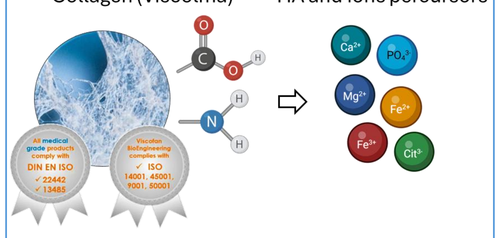
Bone injuries represent a challenge in healthcare, and researchers are increasingly focusing on biomimetic and smart materials as a promising solution [1]. Unlike conventional treatments, such as autografts or allografts, biomaterials could offer a more effective approach. Bone's complex structure and composition should inspire the development of scaffolds that support regeneration [2]. Collagen, being a natural component of the extracellular matrix, is widely used, but its fast degradation and weak mechanical properties limit its application.

<u>AIM</u>: This study explores collagen scaffolds with hydroxyapatite (HA) doped with magnesium (Mg) and/or iron (Fe), demonstrating that ion substitution can simultaneously fine tune scaffold physicochemical properties, enhance cytocompatibility, provide antibacterial activity, confer magnetic responsiveness, and promote bone regeneration.

Methodology

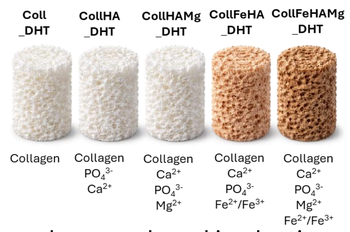
1. Hydrogel formulation

Collagen (Viscolma) — HA and ions percursors



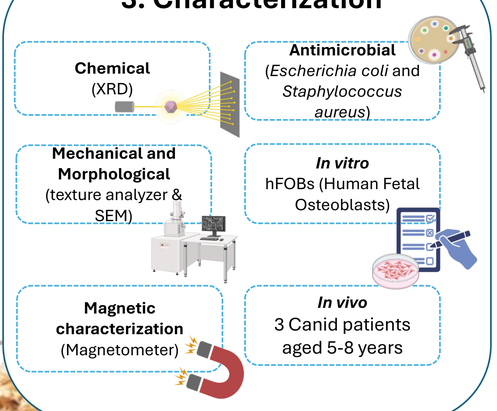
Heterogeneous nucleation of Mg and/or Fe-doped HA microparticles onto collagen type I.

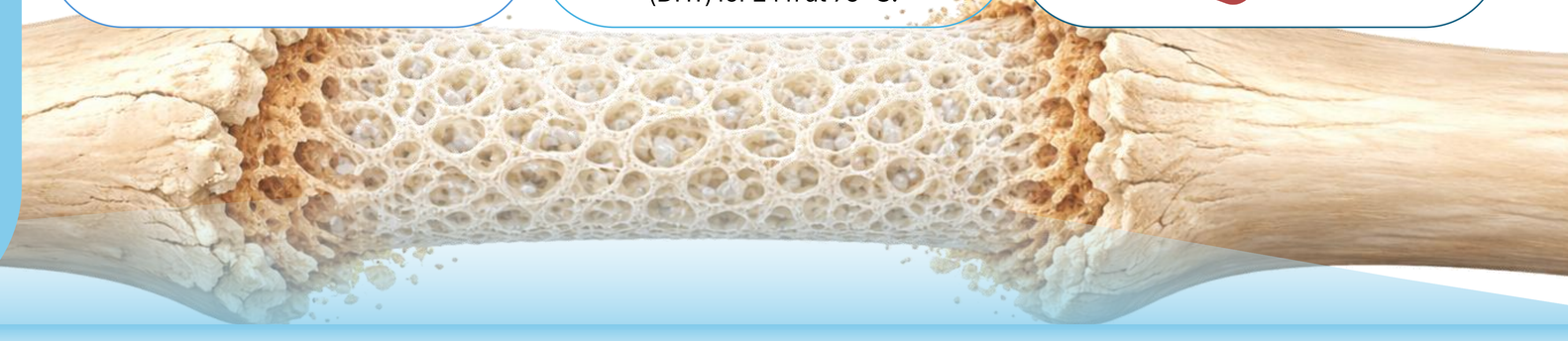
2. Scaffold fabrication & crosslinking



Hydrogels were placed in plastic molds, fabricated via freeze-drying, and crosslinked by dehydrothermal treatment (DHT) for 24 h at 75 °C.

3. Characterization





Results

Increased **mechanical properties**.

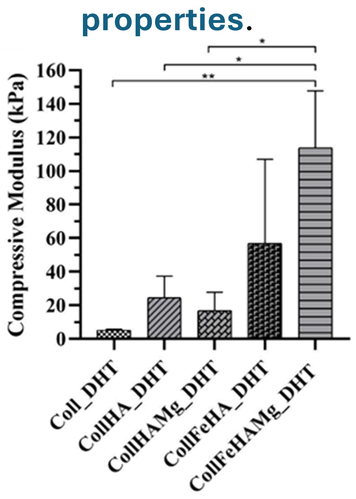


Fig. 1. Compressive modulus in wet state.

Low crystalline HA (~26° and 32° 2θ); iron-doped scaffolds form a magnetite/maghemite secondary phase.

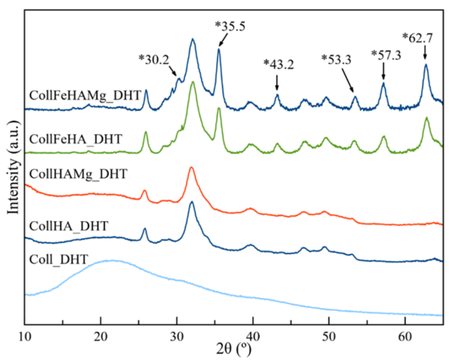


Fig. 2. X-ray powder diffraction profiles.

Fe-doped scaffolds exhibit **superparamagnetic behavior** due to Fe^{2+}/Fe^{3+} lattice doping and surface iron oxide particles.

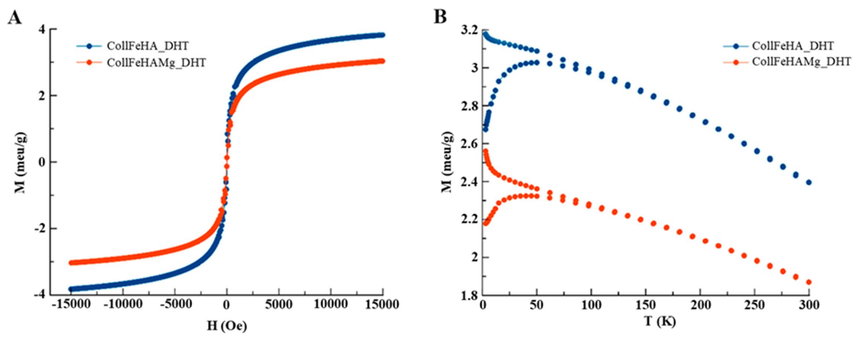


Fig. 3. (A) Magnetic curves as a function of the applied magnetic field at 300 K. **(B)** Zero-field-cooled and field-cooled magnetization curves as function of temperature measured under magnetic field of 1000 Oe.

Co-doping tunes ion release: Mg stabilizes the lattice and slows release, while Fe increases solubility and accelerates Ca and Fe delivery.

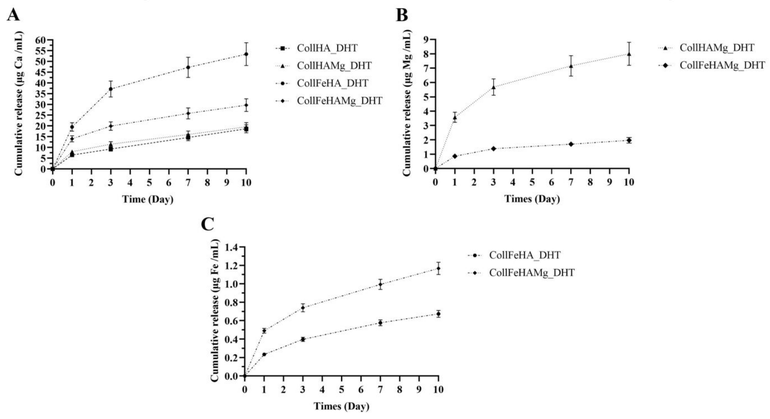


Fig.4. Cumulative release of Ca **(A)**, Mg **(B)**, and Fe **(C)** in the scaffolds.

Fe-doping confers strong **activity against *S. aureus***, but limited effect on *E. coli*.

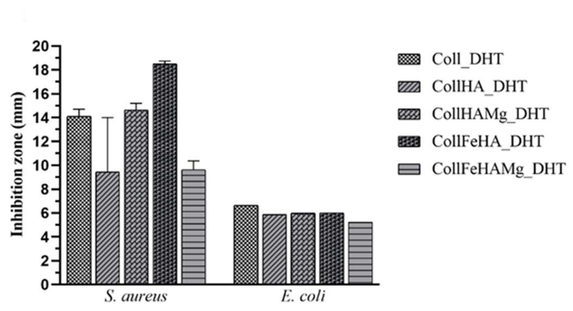


Fig. 5. Quantification of inhibition zone diameters (mm) for each scaffold.

hFOB cells **adhere and proliferates** on all scaffolds.

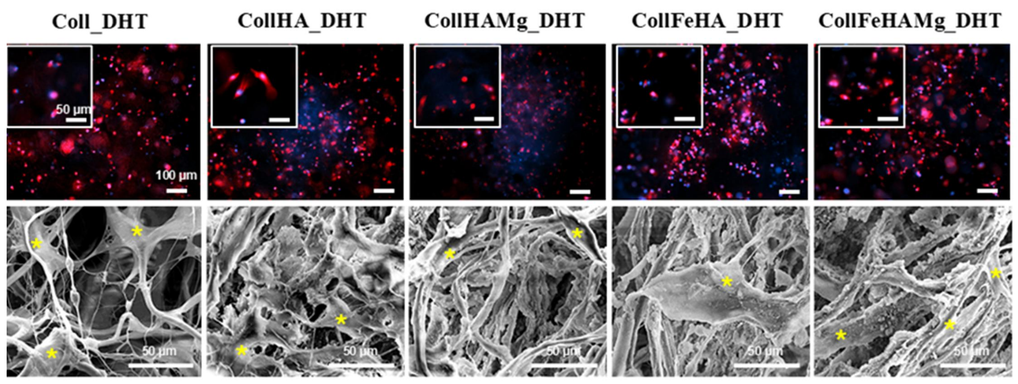


Fig. 6. Morphology of cells on the scaffolds. Top: cells stained for F-actin (red) and nuclei stained with DAPI (blue), at day 7 of culture; Bottom: SEM images at day 7 of culture. Yellow asterisks indicate cells embedded in the scaffold matrix.

Conclusions

Mg/Fe-doped HA/collagen scaffolds offer tunable mechanical properties, magnetic responsiveness, cellular compatibility, and antibacterial activity against *S. aureus*. In three canine clinical cases, scaffolds showed good integration and supported bone regeneration.

In three **canine clinical cases**, Fe-containing scaffolds accelerated bone radiodensity increase at 3 months with **good integration and no rejection**.

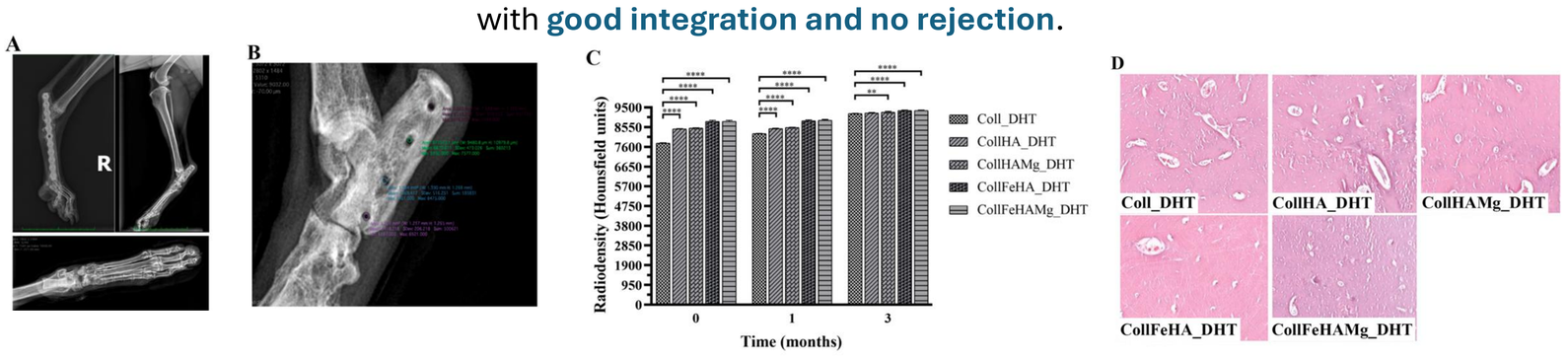


Fig.7. (A) Radiographic image of scaffold application sites. **(B)** Example of computer-assisted radiodensity measurement using Horos®. **(C)** Bone radiodensity in the defects at 3 months. **(D)** Histological sections of the samples obtained by biopsy/ trephine 1 mm 3 months.

References:
[1] J. Ye, et al, 3D printed porous magnesium metal scaffolds with bioactive coating for bone defect repair: enhancing angiogenesis and osteogenesis, Journal of Nanobiotechnology 23 (2025) 160. https://doi.org/10.1186/s12951-025-03222-3.
[2] J.W. Łuczak, M. Palusińska, D. Matak, D. Pietrzak, P. Nakielski, S. Lewicki, M. Grodzik, Ł. Szymański, The Future of Bone Repair: Emerging Technologies and Biomaterials in Bone Regeneration, International Journal of Molecular Sciences 25 (2024) 12766. https://doi.org/10.3390/ijms252312766.

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