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Bone Tissue Engineering via Collagen Hydrogel-Coated PHBV-BG Scaffolds: Mimicking Both Rigid and Soft Tissues

Beatriz Aráoz[1], Santiago Crespo[2], Mercedes Pérez-Recalde[1] and Élida B Hermida[1]

[1]ITECA (CONICET-ECyT-UNSAM), Argentina. [2]ECyT-UNSAM, Argentina.

INTRODUCTION & AIM

Extensive bone damage often exceeds the body's natural regenerative capacity, making the use of implants essential. Bone itself is a combination of rigid tissues—formed by a collagen–hydroxyapatite matrix—and soft tissues such as bone marrow, whose extracellular matrix also contains collagen. The rigid phase can be mimicked using biodegradable polymers combined with bioactive materials, and 3D printing enables reproduction of its hierarchical porous structure. However, the stiffness contrast between these tissues prevents a single material or technique from mimicking both regions. We therefore propose post-processing strategies that anchor collagen gels onto 3D-printed scaffolds, better reproducing bone's complex architecture and supporting cell adhesion and differentiation.

METHOD

Scaffolds composed of polyhydroxybutyrate-co-valerate and bioactive glass (45S5), PHBV–BG, were 3D-printed (20% or/and 55% infill) by fused deposition modelling (FDM). Two surface treatments for collagen (0.4 %wt) adhesion were tested: PHBV (1.7 %w/w in dichloromethane), NaOH 1M for 1 min (PHBV/NaOH), followed by collagen, or BG suspension (BG) followed by collagen. Collagen deposition, morphology and wettability were assessed by SEM images, aniline blue staining and contact-angle measurements. Mechanical properties after treatments were evaluated in a DMA Q800 (tensile test, 1 mm/min).

RESULTS & DISCUSSION

1) Scaffold's morphology: biomimetic scaffold design

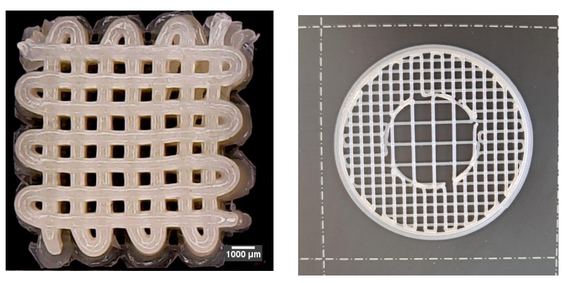

Figure 1: optical images of 3D-printed PHBV–BG scaffolds with dual-infill (a) and single-infill (b) architectures.

- **PHBV–BG filament shows excellent printability and dual-infill design achievable, enabling structural gradients within a single scaffold.**
- **Transversal bone architecture emulated (tibia-like internal morphology).**
- **Bone-like stiffness (600 ± 110 MPa, 55% infill).**

2) Wettability via contact angle

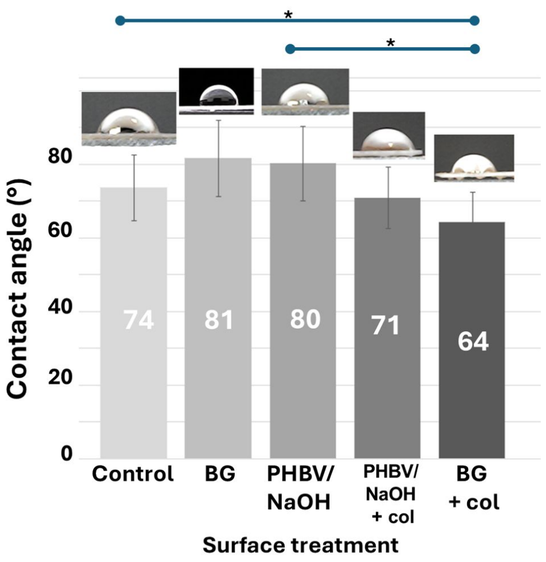

Figure 2: Contact-angle modulation induced by BG and PHBV/NaOH treatments; with collagen further enhancing surface wettability.

- **Collagen coating consistently increased wettability.**
- **BG–collagen showed the highest wettability, significantly different from control.**
- **Wettability proved tunable across protocols, enabling control of cell-adhesive surface properties.**

3) Collagen adhesion

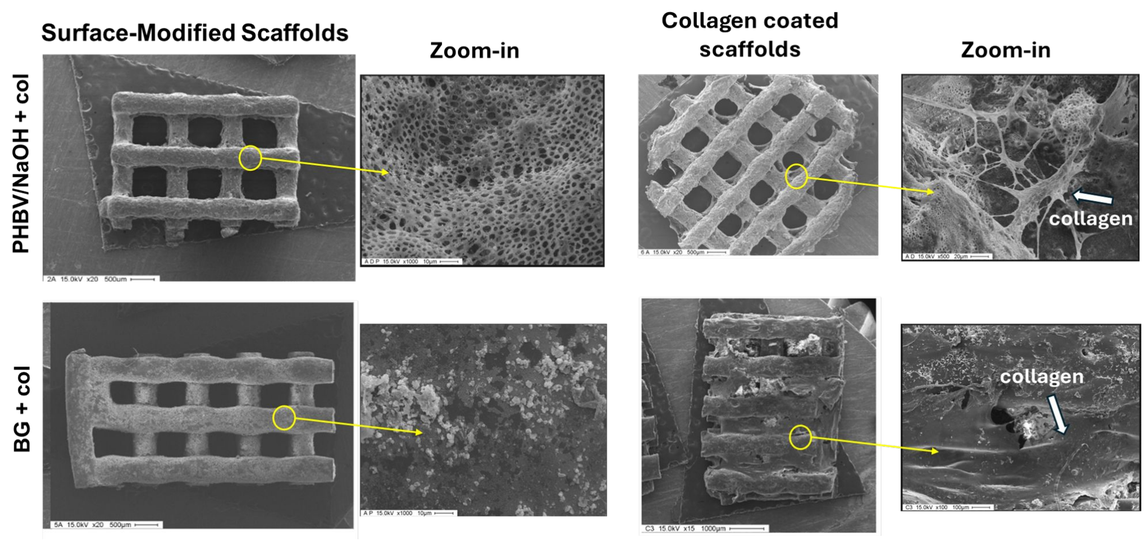

Figure 3: SEM images showing collagen attachment on BG- and PHBV/NaOH-treated scaffolds.

- **PHBV/NaOH treatment yields a porous surface, and BG treatment a rough one, with BG treatment producing greater collagen deposition.**
- **Adhesion tests indicated a good-to-moderate interfacial bonding, confirming that hydrogel detachment from the scaffold is mechanically difficult.**

4) Collagen stability

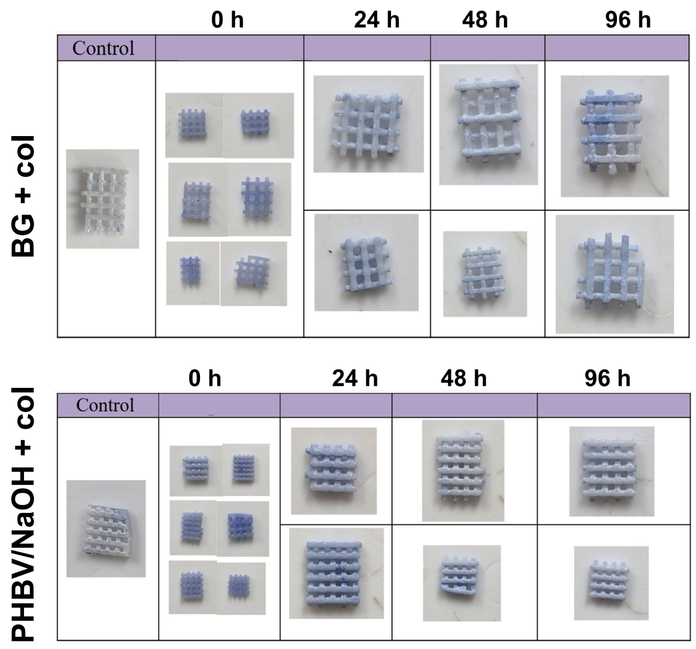

Figure 4: Optical images showing collagen stability on BG- and PHBV/NaOH-treated scaffolds after SBF exposure (0–96 h); the stained collagen-free control shows no blue signal.

- **Collagen-coated scaffolds incubated in SBF (37 °C) for 0–96 h.**
- **Protocols displayed different collagen retention over time.**
- **Aniline blue staining enabled visual tracking of collagen persistence.**
- **Fading of blue signal indicated gradual collagen loss under SBF exposure.**
- **Control scaffold, stained but collagen-free, showed no detectable coloration.**

CONCLUSION

The combined use of surface treatments and collagen deposition enabled precise modulation of scaffold wettability, with BG–collagen achieving the greatest improvement. These results show that post-processing can refine 3D-printed scaffold microenvironments, enhancing both cell adhesion and biomimicry

FUTURE WORK / REFERENCES

Future work will focus on evaluating cell adhesion and differentiation on treated scaffolds, optimising collagen crosslinking for improved stability, and integrating mechanical testing to assess the impact of surface modifications on structural performance.

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