3D-PRINTED SKIN DRESSINGS MANUFACTURED WITH FISH SKIN COLLAGEN ENRICHED WITH UNSATURATED FATTY ACIDS FOR ENHANCED WOUND HEALING

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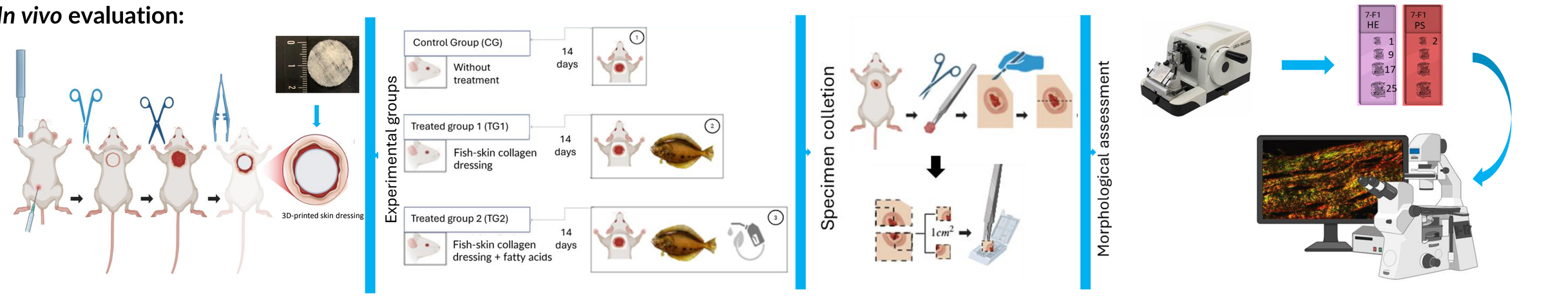
INTRODUCTION AND OBJECTIVE

Skin injuries compromise skin integrity and demand high public health costs. Tissue regeneration depends on a favourable microenvironment, with structural support and biological stimulus. Bioactive dressings, such as those produced by 3D printing with fish skin collagen [1,2] and unsaturated fatty acids [3], can promote efficient re-epithelialization. This study **aims** to develop and characterize 3D-printed collagen-based skin dressings combined with unsaturated fatty acids and to evaluate the *in vivo* effects of these components on wound healing morphology in rats.

MATERIAL AND METHODS

Marine collagen was extracted from *Syacium* spp. and mixed with sodium alginate and a blend of fatty acids, resulting in a bioink used to manufacture dressings through 3D printing. The material was characterized by **Fourier Transform Infrared Spectroscopy (FTIR)** and **Scanning Electron Microscopy (SEM)**. **Cytocompatibility** was assessed *in vitro* using the Alamar Blue assay with L929 murine fibroblast cells and HDFn human dermal fibroblasts. ANOVA and Tukey post hoc.

***In vivo* evaluation:**



RESULTS

Structural analysis was performed using Fourier Transform Infrared Spectroscopy (FTIR)

FTIR analysis confirmed the presence of collagen, alginate, and lipid components, each showing characteristic peaks.

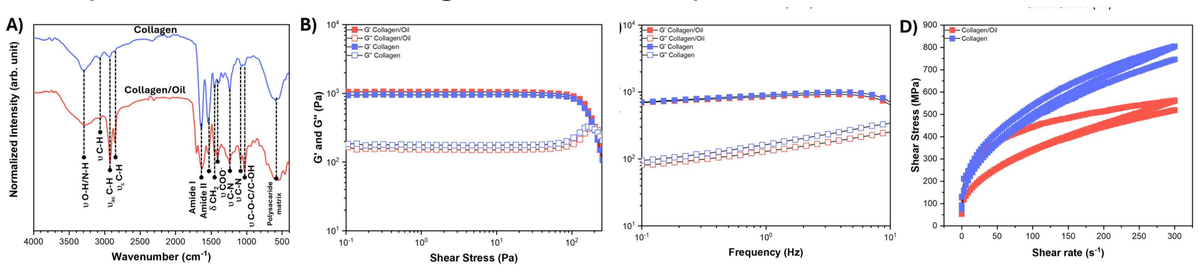
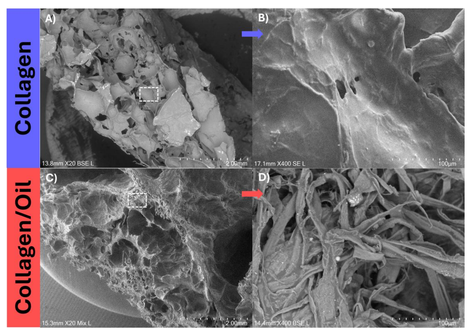


Figure 1. FTIR from 3D printed skin dressings. Infrared Spectroscopy (**A**); The moduli G'/'G' as a function of shear stress (**B**); The moduli G'/'G' as a function of frequency (**C**); Shear Stress em função do Shear Rate - Loop Tixotrópico (**D**).

Scanning Electron Microscopy (SEM)



SEM analysis revealed that the collagen–alginate matrix exhibited a porous structure, which became less porous with the addition of FA.

Figure 2. Scanning Electron Microscopy of collagen-based dressings.

Cytocompatibility

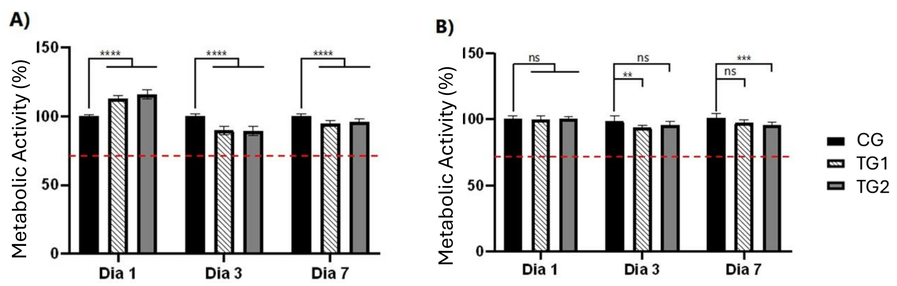


Figure 3. Cell viability graph. Cell viability of L929 murine fibroblast cell line (A) and HDFn human cells (B) though AlamarBlue assay for collagen-based dressings incubated 1, 3 and 7 days.. *p < 0,05, **p < 0,01, ***p < 0,001 e ****p < 0,0001.

In vivo analysis

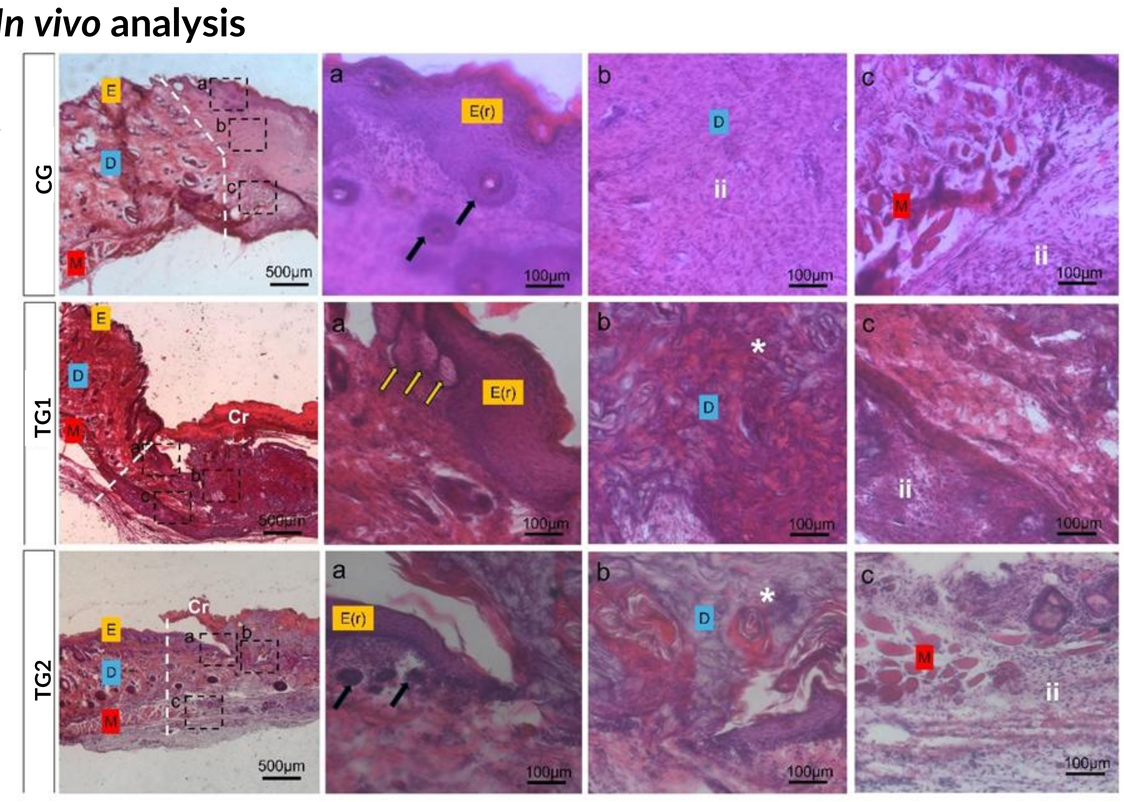


Figure 4. Histopathological aspects of rats skin lesions of CG, TG1 and TG2. Skin layers: Epidermis (**E**), dermis (**D**) and muscle layer (**M**). "**E(r)**"=Re-epithelialization process and (**Cr**) wound scab. Black arrows point hair follicles and yellow arrows point sebaceous glands."*"= collagen fibers and "ii"= inflammatory infiltrate.



Figure 5. Collagen fibers of the reticular dermis of rat skin from the all groups. In three photomicrographs, the collagen fibers are observed in a mesh-like pattern; those appearing red and yellow are type I collagen fibers, and those in green are type III collagen fibers. (Picrosirius under polarized light)

CONCLUSION

The 3D-printed collagen-based dressings incorporating fatty acids exhibited a favorable porous structure, biocompatibility, and stimulatory effects on cellular activity. The *in vivo* morphological findings demonstrated enhanced dermal collagen deposition and accelerated skin regeneration. These findings support the potential of marine collagen and fatty acid-based bioinks for use in advanced skin regeneration therapies.

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