Development of a New Biomaterial for Bone Tissue Engineering: Enzyme Cross-linked Collagen from Salmon *(Salmo salar)*

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CONTEXT

- **Bone is a tissue with various roles** (support, protection, movement, mineral storage, blood cell synthesis…) and **in constant remodeling** helping to heal [1,2]

> 2 million bone grafts are realized per year worldwide. Current implants are inorganic (e.g. ceramics or titan) or organic (e.g. mammalian collagen matrix) [1,3,4]

- **Why collagen in bone tissue engineering?**
  - Biocompatible, biodegradable and low immunogenicity [4]
  - Most abundant protein in connective tissues

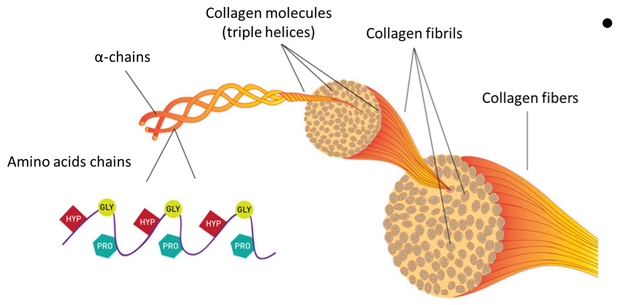


Fig.1. Fibrous structure of collagen [A]

- **Why use marine collagens?** [5,6]
  - Close compositions to mammalian collagens
  - Limit zoonosis
  - Value marine by-products
  - No cultural or religious issues

Salmo salar [B]

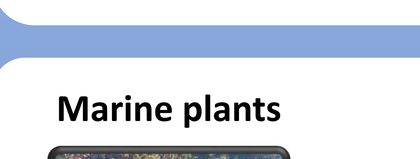
Lower mechanical properties and **stability** than mammalian collagens

How to obtain a salmon collagen matrix with similar properties to mammalian collagens?

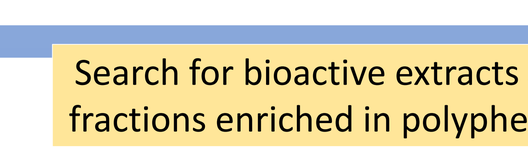
Enzymatic crosslinking with **microbial transglutaminase** (mTG): ↗ bond network ✚ ↗ mechanical properties ✚ No cytotoxic residues ⫽ Impact on pore size? [5,7]

Does mTG crosslink salmon collagen?

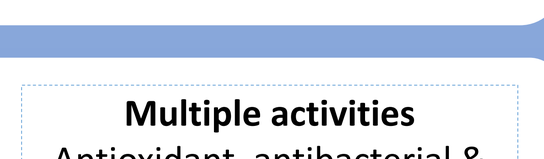
Dry native collagen → (Crosslinking reaction) → Hydrogels → (Freeze-drying) → Dry scaffolds → (Rehydration) → Rehydrated scaffolds



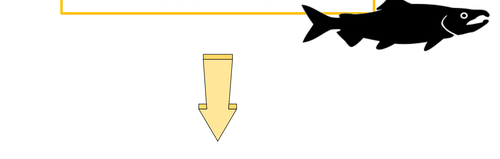
Composed of high molecular weights



Similar macroscopically BUT cross-linked collagen keeps its shape at RT unlike native



Dry cross-linked collagen is more retracted than native



Cross-linked scaffold stay under solid form while native solubilizes

→ **Improved bond network**?

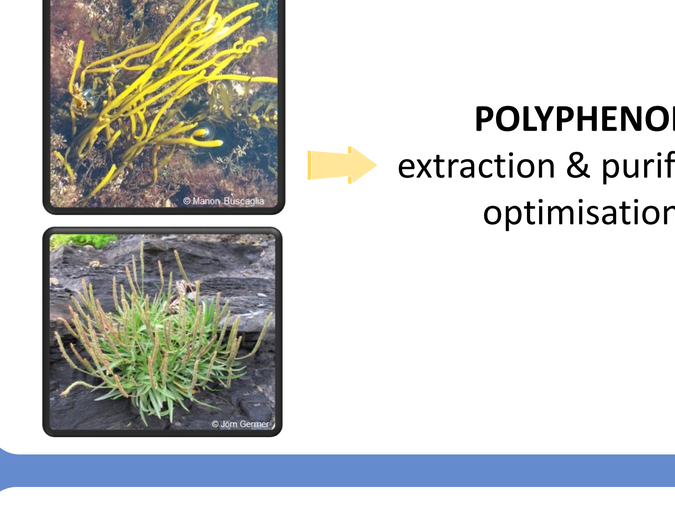
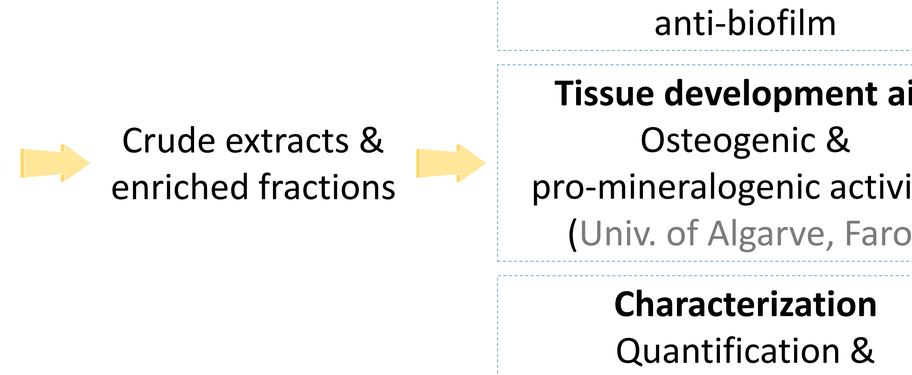


Fig.2. SEC-HPLC profile of native salmon collagen in distilled water filtered on 0.22µm



Macroscopic analysis
Appearance & texture

Microscopic analysis
Structure & pore size

Rheological analysis
Viscoelastic properties

Our role in the BlueHuman project

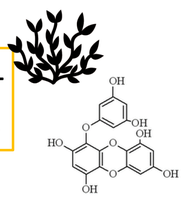
"**BLUE** biotechnology as a road for innovation on **HUMAN**'s health aiming smart growth in Atlantic Area"

http://bluehuman.cetmar.org/

Marine collagen scaffold

BIOMATERIAL FOR BONE TISSUE ENGINEERING

Active polyphenol-enriched fractions



Search for bioactive extracts and fractions enriched in polyphenols

Marine plants

POLYPHENOLS
extraction & purification optimisations

Crude extracts & enriched fractions

Multiple activities
Antioxidant, antibacterial & anti-biofilm

Tissue development aid
Osteogenic & pro-mineralogenic activities (Univ. of Algarve, Faro)

Characterization
Quantification & characterization

CONCLUSION & PERSPECTIVES

- Salmon collagen crosslinking is effective with mTG at several temperatures and the created network supports rehydration.
- Adjustments will be done to improve collagen properties (hydrogels and scaffolds) in comparison to mammalian collagen (porcine).
- Microscopic and rheological analyses will follow to determine structure and mechanical properties (IRDL & PIMM at UBO).
- Finally, this new biomaterial will be combined with active polyphenols from marine plants with antioxidant, antibacterial or pro-osteogenic activities to improve bone regeneration conditions and limit the supply of antibiotics that can have multiple negative effects during long-term treatments [2,8].

REFERENCES

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