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From chicken feathers to antimicrobial peptides for smart, self-disinfecting surfaces

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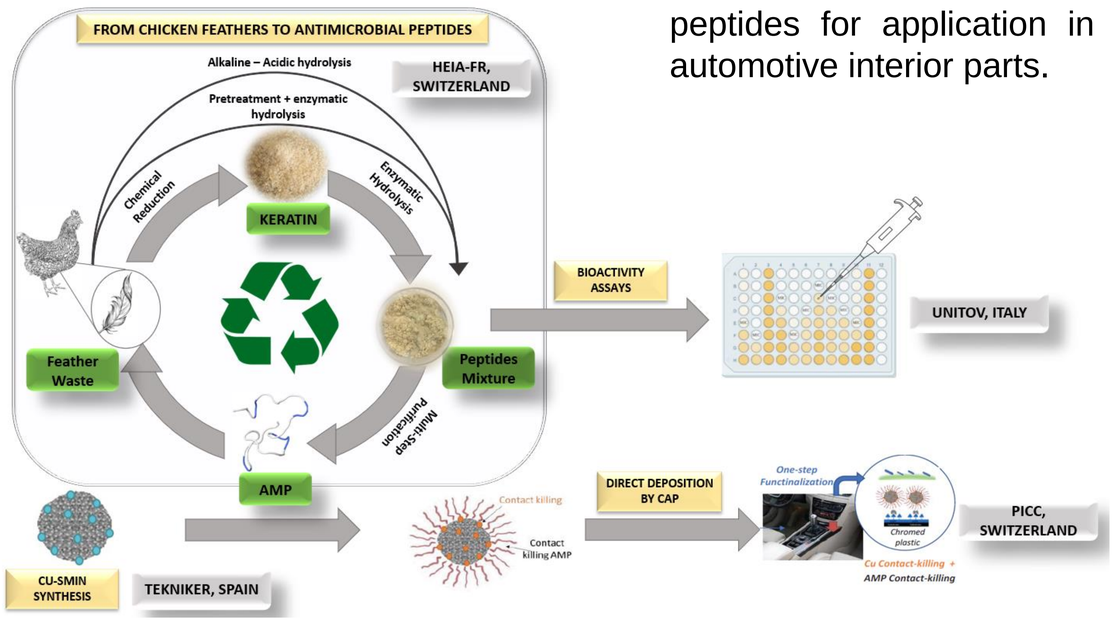
Introduction

Microbial colonization of frequently touched surfaces burdens our society by causing significant cost to human lives and economy. The EU project RELIANCE aims to design and develop an innovative smart response self-disinfectant antimicrobial coatings that act by contact killing and prevent therefore the spread of infections. The nanocoating consists of copper-functionalized mesoporous silica nanoparticles modified with **Antimicrobial Compounds** such as peptides extracted from keratin-rich waste streams.

At HEIA-FR, we are deeply involved in the extraction, isolation and purification of **Antimicrobial Peptides** (AMP) from chicken feathers. Multiple extraction approaches are investigated and a spectrum of analytical methods are used for the purification and characterization of the extracted peptides.

Overview

Chicken feathers, a keratinous waste source, very abundant in nature but underexploited due to the high stability of keratin. Therefore, various promising approaches are employed in the extraction and isolation of sustainable antimicrobial peptides for application in automotive interior parts.



Peptide's isolation and purification

In ultrafiltration process, a semipermeable membrane with defined pore sizes allowing smaller molecules, such as salts and contaminants, to pass through while retaining peptides.

This method not only ensures the purity of peptides by eliminating impurities but also allows to separate peptides into distinct ranges, such as 1-5 kDa, 5-10 kDa, and >10 kDa, enabling precise isolation according to size.

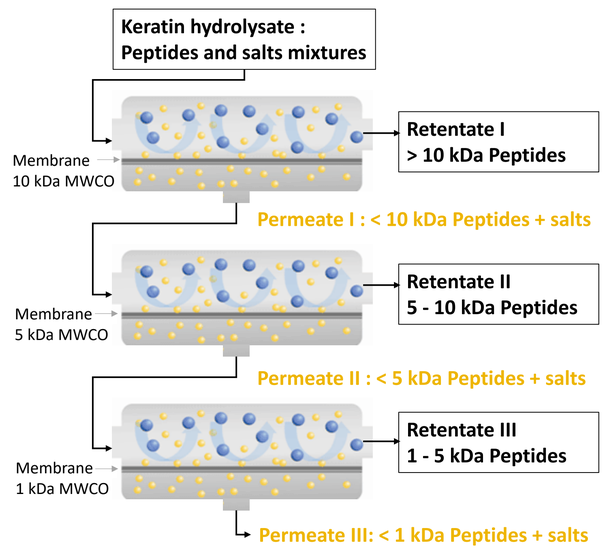


Figure 1. Overview of our global UF approach.

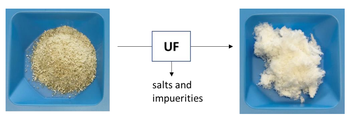


Figure 2. Keratin extract : crude (left), and after desalting by UF with a 2 kDa MWCO (right).

Characterization by FTIR and SDS-PAGE

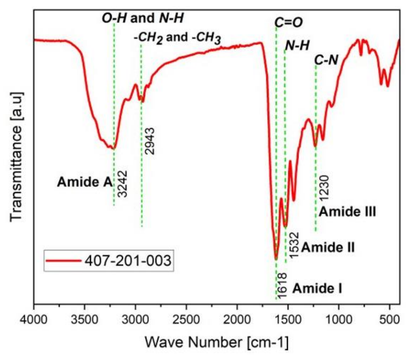


Figure 3. FTIR spectra of keratin.

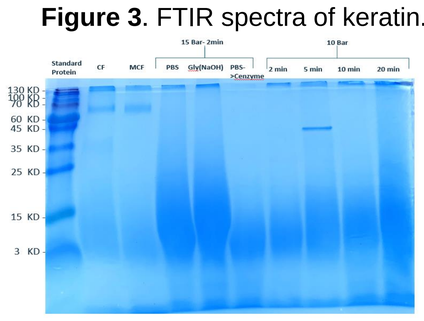


Figure 4. SDS-PAGE profiles of various peptides mixtures

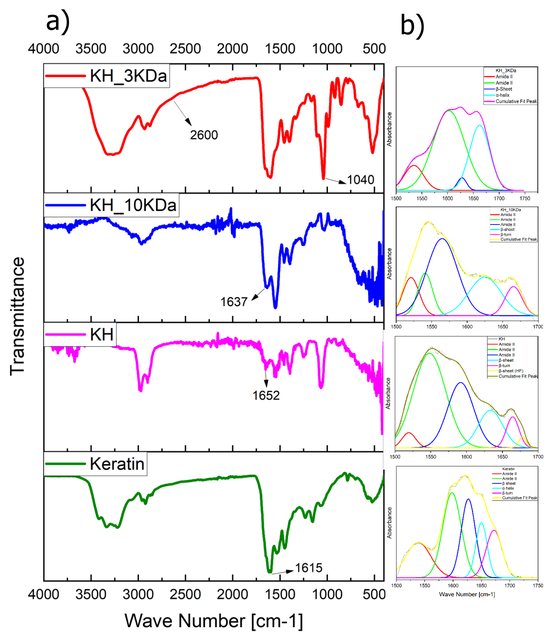


Figure 5. a) FT-IR spectra of Keratin and Keratin hydrolysates, b) Deconvolution of the amide I region for four samples

Scaling-up separation by prep. HPLC

This preparative HPLC chromatogram shows the successful scaling-up of the purification process. By separating samples based on polarity, the method efficiently handles larger quantities. Each separated fraction will undergo bioactive testing to further explore their properties.

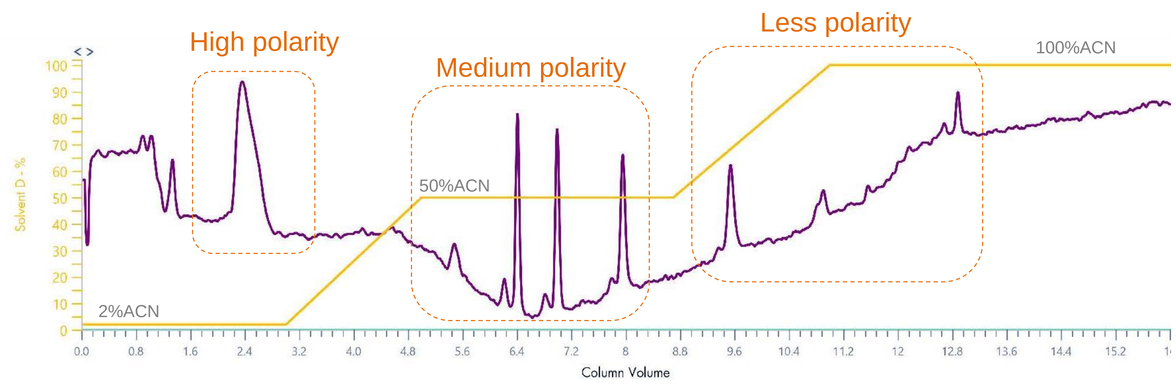


Figure 6. Chromatograms from preparative HPLC using a C18 column (US5C183-150/300). The flow rate was set at 20 mL/min with a mobile phase gradient of water and acetonitrile (yellow line). A 40 mg sample was injected, and the wavelength detector at 275 nm was used. The peptide hydrolysate was fractionated based on their polarities.

Summary & Outlook

This work evaluates various hydrolysis methods to identify the best extraction approach for producing peptides with optimal antimicrobial activity from poultry waste. Several extracted AMP mixtures demonstrate antimicrobial activity and are subsequently subjected to further purification to isolate the most active peptide fraction. This multi-step purification process advances the understanding of the activity-structure relationship and allows the selection of a sustainable scalable process for the production of keratin-based AMPs.

Acknowledgements

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