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## From enantiomeric exchange to antimicrobial activity: the enzymatic secrets of salivary peptides

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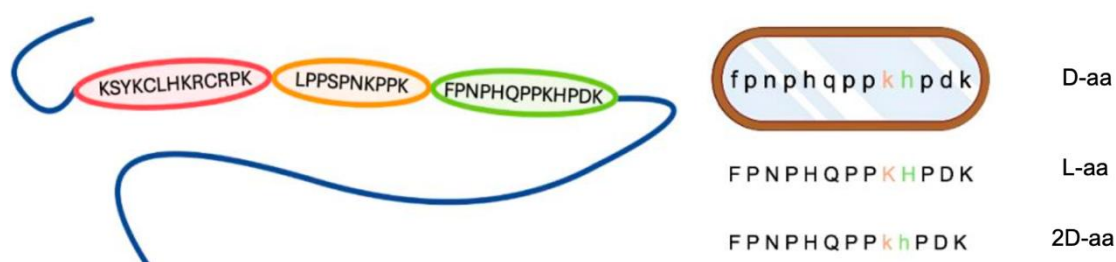
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The interaction between metal ions and antimicrobial peptides (AMPs) is a fundamental aspect of designing innovative bioinspired antimicrobial agents, offering new possibilities for overcoming antibiotic resistance [1]. Human salivary AMPs, particularly those derived from the MUC7 protein, are essential to the innate immune system and act as a primary defense against pathogens. Their antimicrobial activity is significantly enhanced when they form complexes with metal ions [2]. Due to their broad-spectrum action and low possibility of inducing resistance, AMPs are promising candidates for new therapeutic approaches. However, their clinical use is often restricted by limited enzymatic stability, which necessitates developing peptidomimetics to improve their stability and antimicrobial properties [3].

In this study, we investigate the enzymatic stability, thermodynamics, coordination behavior, structural properties, and antimicrobial activity of Cu(II) and Zn(II) complexes with salivary proline-rich peptides and their D-amino acid-substituted analogs (**Figure**).



**Figure:** Simplified structure of mucin (left) with highlighted relevant fragments. Analyzed peptidomimetics of the FPN peptide (right), with enzymatically vulnerable regions marked in color. Lowercase letters in the sequence indicate D-amino acids.

A comprehensive analytical approach, integrating potentiometric titration, spectroscopic techniques (UV-Vis, CD, EPR), mass spectrometry, and HPLC, was employed to investigate

the coordination behavior and stability of metal-peptide and peptidomimetic complexes. Biological assays further assessed their antimicrobial potential.

Our findings reveal that metal coordination preferences vary depending on the applied modifications. Enantiomeric substitution of amino acids significantly enhances the thermodynamic stability of Cu(II) and Zn(II) complexes, while the enzymatic stability of partially modified peptides remains unchanged. Notably, the fully D-amino acid analog exhibits exceptional resistance to proteolysis and, when complexed with metal ions, demonstrates the most favorable minimum inhibitory concentration (MIC) values, underscoring its potential for antimicrobial applications.

This study underscores the significance of enantiomeric modifications in regulating metal-peptide interactions and enhancing the stability of antimicrobial peptides. These modifications represent a promising strategy for the development of next-generation antimicrobial agents with improved therapeutic efficacy.

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