STUDY OF ACTION MECHANISM OF HSP1 ANTIMICROBIAL PEPTIDE BY CIRCULAR DICROISM AND NUCLEAR MAGNETIC RESONANCE

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Several strategies have been investigated to combat microorganisms resistant to conventional antibiotics, among which stand out the use of antimicrobial peptides. Intensive structural studies show that these peptides exert their biological activity by interaction with lipid membranes acquiring a secondary structure. In order to understand the action mechanisms of peptides, circular dichroism (CD) and Nuclear Magnetic Resonance (NMR) are widely employed to obtain information about preferential conformation and interaction way with biomimetic environment. Recently, has been reported the isolation of antimicrobial peptide HSP1 from Hyla punctata species, which was studied in this work. CD spectra were obtained in different solutions containing phosphate buffer at pH 5.8. The results show that HSP1 adopts random conformation in aqueous medium. Furthermore, was observed that the addition of TFE to 38.15 mmol.L⁻¹ HSP1 results in a maximum wavelength at 190 nm and minimum intensity at 205 and 225 nm, indicating that the peptide adopts α-helix structure in this environment. The same behavior was observed in SDS and DPC micelles, revealing a maximum helicity of 60% and 58%, respectively. NMR experiments were carried out in DPC and SDS micelles medium, in which the peptide presented higher α-helical conformation. With the joint analysis of the TOCSY, NOESY and HSQC-CH contour maps was possible to realize the resonances assignment and identify inter-residues interactions features of α-helix structure. In DPC micelles it was possible to verify small folding in N-terminal region, while in SDS micelles presented a more extended conformation. Whereas in zwitterionic membranes HSP1 tends to insert in hydrophobic region, in negative membranes electrostatic interactions are predominant and promote a parallel orientation of HSP1 to the bilayer surface.

Key words: antimicrobial peptide; action mechanism; conformational analysis.