Rational design to bioactive peptides in the development of biotechnological tools for therapies against human pathogens

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Recently, several peptides have been studied regarding the defence process against pathogenic microorganisms, which are able to act against different targets, with the purpose of developing novel bioactive compounds. The present presentation focuses on the structural and functional evaluation of the palindromic antimicrobial peptide *Pa*-MAP2 and *Pa*-MAP1.9, designed based on the peptide *Pa*-MAP from *Pleuronectes americanus*. For a better structural understanding, molecular modelling analyses were carried out, together with molecular dynamics and circular dichroism, in different media. Antibacterial activities against Gram-negative and positive bacteria were evaluated, as well as cytotoxicity against human erythrocytes, RAW 264.7, Vero and L6 cells. *In silico* docking experiments, lipid vesicle studies, atomic force microscopy (AFM) imaging and bacterial biofilm assays were carried out to explore the activity of the peptide. *In vivo* studies on infected mice were also done. All peptide designed based in *Pa*-MAP primary sequence favoured an α-helix structure that was pH dependent, and only present on alkaline environment, with a dynamic N- and C-terminals that are stabilized in anionic media. *Pa*-MAP2 and *Pa*-MAP1.9 only showed activity against Gram-negative bacteria, with a MIC of 3.2 and 6.0 μM, and without any cytotoxic effect. *In silico*, lipid vesicles and AFM studies confirm the preference for anionic lipids (POPG, POPS and LPS) for both synthetic peptides, with the positively charged lysine residues being essential for the initial electrostatic interaction. *In vivo* studies showed that *Pa*-MAP2 and *Pa*-MAP1.9were efficient against bacteria and increases to 100% the survival rate of mice infected with *Escherichia coli*.