Creation of antimicrobial biopolymers by the use of recombinant DNA technology

André da Costa[1], Raul Machado[1], José Carlos Rodríguez-Cabello[2,3], Andreia C. Gomes[1], and Margarida Casal[1]

[1]CBMA (Centre of Molecular and Environmental Biology), Department of Biology, University of Minho, Campus de Gualtar, 4710-057 Braga, Portugal

[2]Bioforge (Group for Advanced Materials and Nanobiotechnology), Edicio LUCIA, Universidad de Valladolid, Valladolid, Spain

[3]Networking Research Centre on Bioengineering, Biomaterials and Nanomedicine (CIBER-BBN), E-47011 Valladolid, Spain

*andrecosta@bio.uminho.pt

The spread of antimicrobials resistant microorganisms has triggered the search for new ways to treat infections. One of these ways is the creation of antimicrobial devices and surfaces that kill or prevent the spread of microorganisms. In the present work we explored the properties of different antimicrobial peptides (AMPs) for the creation of biopolymers with broad antimicrobial activity. Antimicrobial recombinant protein-based polymers (rPBPs) were designed by cloning the DNA sequence coding for the different AMPs in frame with the N-terminus of the elastin-like recombinamer consisting of 200 repetitions of the pentamer VPAVG, here named A200. The new rPBPs, CM4-A200, BMAP-18A200 and Hep-A200, were purified via a simplified nonchromatographic method, making use of the thermoresponsive behavior of the A200 polymer. While the Hep-A200 polymer displayed antibacterial activity in the soluble state, CM4-A200 and BMAP-18A200 were poorly effective in solution. However, when CM4-A200 and BMAP-18A200 were processed into free-standing films high antimicrobial activity against Gram-positive and Gram-negative bacteria, yeasts and filamentous fungi was observed. The antimicrobial activity of these films was dependent on the physical contact of cells with the film surface. Furthermore, the antimicrobial films did not reveal a cytotoxic effect against both normal human skin fibroblasts and human keratinocytes. Finally, we have developed an optimized ex vivo assay with pig skin demonstrating the antimicrobial properties of the CM4-A200 cast films for skin applications.

Acknowledgments

This work was supported by the strategic programme UID/BIA/04050/2013 (POCI-01-0145-FEDER-007569) funded by national funds through the FCT I.P. and by the ERDF through the COMPETE2020 - Programa Operacional Competitividade e Internacionalização (POCI). By the Spanish Minister of Economy and Competitiveness (MAT2012-38043-C02-01) and Junta de Castilla y León-JCyL (VA152A12-2 and VA155A12-2), Spain. AC and RM, acknowledge FCT for SFRH/BD/75882/2011 and SFRH-BPD/86470/2012 grants, respectively.