# From mono-functional enzymatic coatings to bi-functional coatings to impair Staphylococci adhesion

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Despite the remarkable advances in modern healthcare, there are some drawbacks associated to the extended use of medical devices and biomaterial implants when microorganisms are able to reach their surface, forming biofilms and becoming the focus of biomaterial-associated infections (BAI) which are hardly to treat. The growing number of BAI has led to the need of developing novel antibacterial coatings for medical devices. The use of enzymes able to degrade biofilm matrix components, such as proteins and extracellular DNA, represents a promising approach to fight these infections.

The first aim of this study was to apply a biologically inspired strategy for covalent immobilization of different enzymes (lysozyme, proteinase K and DNAse I) on clinically relevant substrata (silicone) to obtain mono-functional coatings able to prevent staphylococci adhesion or to kill the adhered bacteria, depending on the enzyme used. The coating developed with the best anti-adhesive properties was afterwards combined with an antimicrobial peptide (colistin), generating a bi-functional coating.

Compounds immobilization was mediated by a polydopamine (pDA) coating and the anti-adhesive and antimicrobial performances of the generated surfaces were investigated for a clinical isolate of *Staphylococcus aureus* using fluorescence microscopy.

Results showed that unmodified silicone allowed the adhesion of bacteria without compromising their viability. Silicone modified with polydopamine coating had no significant effect on bacterial attachment and viability. Lysozyme immobilization was not able to reduce bacterial attachment or compromise their viability. On the other hand, proteinase K was able to reduce the percentage of bacterial attachment and a significant fraction of these adhered bacteria was found dead. Regarding the functionalization with DNAse I, these coatings presented the best anti-adhesive properties and since it is know that this enzyme is not cytotoxic, it was further combined with colistin and the bi-functional coating obtained proved to be more effective on reducing the fraction of bacterial attachment.

The overall results suggest that the use of coatings functionalized with enzymes is able to degrade biofilm matrix components and their conjugation with antimicrobial peptides presents a promising strategy for creating antibacterial surfaces to be applied in biomaterials for medical devices and implants.

**Acknowledgements:** The authors thank the project FCT PTDC/SAU-SAP/113196/2009/FCOMP-01-0124-FEDER-016012, the Strategic Project PEst-OE/EQB/LA0023/2013, the Project “BioHealth - Biotechnology and Bioengineering approaches to improve health quality", Ref. NORTE-07-0124-FEDER-000027, co-funded by the Programa Operacional Regional do Norte (ON.2 – O Novo Norte), QREN, FEDER, the project “RECI/BBB-EBI/0179/2012 - Consolidating Research Expertise and Resources on Cellular and Molecular Biotechnology at CEB/IBB”, Ref. FCOMP-01-0124-FEDER-027462, FEDER. The authors also acknowledge Diana Alves PhD Grant SFRH/BD/78063/2011.

**Keywords** biofilm-dispersive enzymes; antibacterial coatings; biomaterial-associated infections; polydopamine