Instituto de Medicina Molecular João Lobo Antunes

Master Project Proposal

Title: Exploring the biomedical potential of bacterial pigments for nano-based antimicrobial solutions against human pathogens

Synopsis:

The urgent global need to develop groundbreaking antimicrobial solutions has prompted the biotechnological prospection of the microbial biomolecular repertoire as source of novel bioactive compounds against human pathogens and, in particular, against multidrug resistant strains.

Microbial pigments play important roles in the survival and competitiveness of microorganisms. In particular, redox-active pigments, such as phenazines, prodigiosin and violacein, have also been studied by its versatile biomedical potential, including antimicrobial activity against Gram-negative and Gram-positive bacteria, yeasts, fungi and virus.

The combination of microbial pigments with nanosystems, such as metal nanoparticles (MNPs), may result in a synergy of their properties. However, the development and characterization of such systems have not been systematically carried out. Furthermore, nano-based formulations can be modulated towards improved physico-chemical stability, increased antimicrobial efficiency, and low toxicity/immunogenicity.

Our group at iMM (NSantos Lab) has a solid record studying the molecular mechanism of action of antimicrobial compounds and in the development of bioinspired nano-based antimicrobial solutions for biomedical application.

This project aims at characterizing the antimicrobial potential and mode of action of nano-based formulations integrating microbial pigments, as pure compounds and/or extracts, towards the development of biomedical solutions against multidrug-resistant human pathogens. The work plan will be carried out at iMM, in the scope of projects of the group, and will be adjusted according to the ongoing research, student's interests and the results obtained.

The work plan will include activities in the scope of 4 ongoing work packages:

WP1: Producing microbial pigments in bacteria and establishing the pigment extraction/purification methodology to enhance the yield, by testing different extraction solvents and by resorting to rotary evaporation. The pigment extracts produced will be characterized by spectrophotometry and chromatography coupled to mass spectrometry.

WP2: Characterizing the antimicrobial properties of the studied pigments against major human pathogens. The antimicrobial screenings will be assayed with model strains susceptible and resistant to common antimicrobials (Biosafety Levels 1 and 2; *e.g., Pseudomonas aeruginosa, Staphylococcus* spp., *Streptococcus* spp., *Candida* spp.), by standardized methods to estimate minimum inhibitory concentrations (MIC), minimum biocidal concentrations (MBC) and minimal concentration inhibiting biofilm formation.

WP3: Synthesizing nano-based formulations integrating microbial pigments and metal nanoparticles. The synthesized nanosystems will be characterized by in-house biophysical techniques to determine their charge, size and stability (*e.g.,* UV-Vis spectrophotometry, dynamic light scattering, zeta potential measurements, transmission electron microscopy).

WP4: Evaluating the antimicrobial mode of action and cytotoxicity. The mode of action of the developed nano-based formulations will be characterized against a prioritized panel of pathogens and eukaryotic cell lines, by in-house flow-cytometry, confocal microscopy and

atomic force microscopy (AFM) approaches, well-established in the group, on cell integrity, changes in biochemical and biomechanical properties of cells, redox stress and DNA damage.

Thesis writing will be done throughout the 10 months.

Given the novelty and exploratory character of some aspects of this research, alternative paths will be implemented in case of sub-optimal performance or dead-ends, to ensure the success of the MSc thesis.

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Bibliography:

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