Sea urchin inspired adhesive proteins – expression and purification of Nectin structural domains

Place of work: **Protein Misfolding and Amyloids in Biomedicine laboratory (Lab 8.5.56), BioISI, FCUL**

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MSc Research Plan:

Bioadhesion is vital for sea urchins, since it is through the production of adhesive secretions that these marine invertebrates attach, move, feed and defend themselves. Besides the interest from a biological perspective, this process **has a biotechnological importance** since wet-effective, biocompatible and ecological adhesive materials are applied **for medical and biotechnological applications**, such as surgical adhesives or promoters of cellular adhesion for in-vitro cultures/3D organ printing.

In recent years knowledge in bioadhesion in sea urchins and other marine organisms has increased however molecular details remain largely unknown. Understanding these processes and what defines the uniqueness of bioadhesives from these organisms is extremally important.

The **Bioadhesion and Biomimicry Research Group** has been dedicated to the study of the Sea-urchin *Paracentrotus lividus.* Importantly, they have identified an adhesive protein, Nectin, which is highly over-expressed in the adult's tube foot discs and secreted into the adhesive footprint [1, 2]. This protein contains six tandemly repeated discoidin-like domains that bind molecules with galactose and N-acetylglucosamine carbohydrate moieties, a LDT motif predicted to interact with an α4/β7 integrin receptor and a putative sulfation site at Tyr77 which can be important for secretion [3-5]. Recently, in collaboration with the **Protein Misfolding and Amyloids in Biomedicine laboratory** we have **expressed** in *E. coli* **and purified the full length recombinant Nectin**, and partially characterized its structural and adhesive features.

To advance our understanding of the molecular basis of sea urchin reversible adhesion and to develop biomimetic adhesives inspired in the sea urchin Nectin model, **here we propose to depict the importance of the different discoidin-like domains for the adhesive properties** of this inspired sea urchin protein. To achieve our goal, we propose to:

1) Optimize recombinant protein expression, in *E. coli*, for 3 different constructs of the Nectin domains (
2) Establish purification protocols for the different constructs using a combination of chromatographic methodologies (his-tag affinity or ion exchange columns and gel filtration columns);
3) And ultimately perform a biochemical and structural analysis (using different spectroscopic techniques) of the best hits and evaluate their adhesive properties in relation to the full length recombinant Nectin.

MSc project will be developed at the **Protein Misfolding and Amyloids in Biomedicine** laboratory, BioISI, under the supervision of Dr. Bárbara Henriques. The proposed work is part of a collaborative project between the host laboratory and Dr. Romana Santos (co-supervisor).

Students selected for this project, after thesis registration, are eligible to apply to the BioISI Junior Programme (supporting 8 students with a 6-month Scholarship (BII), being the selection criterium the academic merit of the candidates).

References

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4. Zito, F., et al., Matrix Biol, 2010. **29**(5): p. 341-5.
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