Master Thesis Proposal

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Lab/Institution: Mass Spectrometry - ITQB
TITLE: Dynamics of circulating proteins during star-fish nerve regeneration

BACKGROUND

The potential for regeneration, including the central nervous system, has its maximum expression in echinoderms. Preliminary studies of regeneration in echinoderms were based on the determination of growth rates and on the morphological, histological and cellular basis of this phenomenon. More recently, some advances have been made in the characterization of the molecular mechanisms involved in the regeneration process of tissues and organs. Studies that have been developed in our laboratory focus on a star-fish species common in Portuguese coastal areas and with high regeneration capability, the Marthasterias glacialis. Coelomic fluid, the main intra-tissue communicating medium in echinoderms for which it is hypothesized a relevant role during the regeneration process. This biological fluid is reach in proteins, particularly glycoproteins, potentially fundamental for the regeneration process.

CF Franco et al Proteomics (2011) 11: 1359-1364; CF Franco et al Proteomics (2011) 11: 3587-92; CF Franco et al Electrophoresis (2012), 33, 3764–3778; C Franco et al J Proteomics (2014), 99: 1-25; C Franco et al Proteomics (2013), 13, 686-709 (Review article)

OBJECTIVES

Changes in the proteome of the cell-free coelomic fluid will be characterized during nerve regeneration. The obtained results will contribute for a deeper knowledge on factors and biological processes involved in the regeneration of the central nervous system of starfish, with potential applications in regenerative medicine.

The process of sea-stars handling, including the induction of regeneration and tissue collection, will be made at the Aquário Vasco da Gama (Dafundo, Oeiras).

Task 1: Extraction and quantification of proteins will be performed according with an already developed protocol.
Task 2: Tryptic digest mixtures obtained before and after cleavage of N-linked oligosaccharides by PNGase F will be analysed by high-resolution LC-MSMS. The detected proteins will be identified and a relative quantification among the several regeneration time-points will be performed using an adequate software.
Task 3: Peptide profiles obtained with and without PNGase F digestion will be compared allowing a characterization of the glycoproteins detected in the coelomic fluid.
Task 4: Quantification date will be evaluated by uni- and multivariate analysis to determine the proteins relevant for the several regeneration steps
Task 5: Pathway analysis will be performed using the proteins identified with expression differences between regeneration time-points
Task 6: Writing of Master Thesis

TIMELINE (use fill tool for the cells)

	Month 1	Month 2	Month 3	Month 4	Month 5	Month 6	Month 7	Month 8	Month 9	Month 10
Task 1										
Task 2										
Task 3										
Task 4										
Task 5										
Thesis										