UFR de Chimie et de Biologie
UNIVERSITÉ Grenoble Alpes

Master's degree in Biology – Chemistry-Biology Department

Master 2 internship project
Year 2020-2021

Laboratory/Institute: Institut de Biologie Structurale **Director:** Winfried Weissenhorn
Team: IRPAS **Head of the team:** Nicole Thielens
Name and status of the scientist in charge of the project: Véronique Rossi, Christine Gaboriaud.
HDR: yes ■ **no** ☐
Address: IBS - 71 Avenue des Martyrs CS 10090 38044 Grenoble cedex 9
Phone: **e-mail:** veronique.rossi@ibs.fr

Program of the Master's degree in Biology:

■ Immunology, Microbiology, Infectious Diseases ■ Integrative Structural Biology
☐ Physiology, Epigenetics, Differentiation, Cancer ☐ Neurosciences and Neurobiology
☐ Planta International

Title of the project:

Complement C1s in diseases

Objectives (up to 3 lines):

Explore molecular and functional consequences of patient mutations on complement protease C1s. Develop nanobodies, ie Immunoglobulin molecular tools, directed against the protease C1s in its active state, to be be used as tools for detection in fluids and tissues and to block its activity.

Abstract (up to 10 lines):

The highly controlled C1s serine protease is known as an innate immune trigger. It associates with a homologous protease (C1r) and a defense collagen (C1q) to form C1, the initiation complex of the complement cascade. Triggering of the cascade by C1, through activation of the proenzyme proteases C1r and C1s, leads to the elimination of pathogens or self-debris via different biological effects. Thanks to its mosaic structure, C1s is a highly specialized serine protease, with only two main substrates, the complement components C2 and C4. However, recent observations have shown new implications of C1s with uncontrolled proteolytic non-canonical functions independent of complement in pathological processes such as severe periodontal Ehlers-Danlos syndrome, or within renal tumors. For that reason, active C1s will be of particular interest.
Depending on the state of the project and on the student interest, the tasks will mainly be:
◊ to obtain the recombinant C1s variants using site-directed mutagenesis and to produce them in a mammalian expression system. To characterize the C1s variants for their enzymatic activity compared to the wild type C1s and to check for the functional impact of the mutations on the cleavage of other new potential targets that could be linked to the disease.
◊ to produce new minimal anti-C1s antibody fragments (called nanobodies, Nb), as molecular tools for improved detection of this enzyme in fluids and tissues, for analyses and diagnostic purposes. Once produced in bacteria and purified, the Nbs will be characterized for their interaction with the catalytic domain of C1s in different states of activation, also assessing the impact on its catalytic activity.

Methods (up to 3 lines):

Depending on the chosen main objective: Bacterial or mammalian expression (HEK293 cells), Biochemistry for purification and sample preparations (FPLC), SDS-PAGE analyses, Biophysics for interaction characterization (SPR/BLI), Enzymatic assays

Up to 3 relevant publications of the team:

[1] Bally I., Dalonneau F., Chouquet A., Gröbner R., Amberger A., Kapferer-Seebacher I., Stoiber H., Zschocke J., Thielens N., Rossi V., Gaboriaud C. (2019) Two different missense C1S mutations, associated to periodontal Ehlers-Danlos syndrome, lead to identical molecular outcomes. Front. Immunol 10, 2962.
[2] Moreau C., Bally I., Chouquet A., Bottazzi B., Ghebrehiwet B., Gaboriaud C., Thielens N. (2016) Structural and functional characterization of a single-chain form of the recognition domain of complement protein C1q. Front. Immunol 7, 79.
[3] Rossi V, Bally I, Lacroix M, Arlaud GJ, Thielens NM. (2014) Classical complement pathway components C1r and C1s: purification from human serum and in recombinant form and functional characterization. Methods Mol Biol. 1100:43-60.

Requested domains of expertise (up to 5 keywords):

Background in biochemistry and interest in immunology or biophysics