Pearl Programme for EArly-stage Researchers in Lille

Title of the thesis	Click chemistry and unnatural amino acids to illuminate oncogenic activities in living cells
Acronym	ONCOGIMAGING
Reference number	002

Hosting institution	**Employer**
Université de Lille Website: https://www.univ-lille.fr/home/	CNRS Website: http://www.cnrs.fr/en
Hosting research unit 1	**Hosting research unit 2**
Name: Unité de glycobiologie structurale et fonctionnelle Acronym: UGSF Identification number: UMR 8576 Address: Cité Scientifique Campus - Bât. C9 59655 Villeneuve d'Ascq Website: http://ugsf-umr-glycobiologie.univ-lille1.fr/	Name: Unité de glycobiologie structurale et fonctionnelle Acronym: UGSF Identification number: UMR 8576 Address: Cité Scientifique Campus - Bât. C9 59655 Villeneuve d'Ascq Website: http://ugsf-umr-glycobiologie.univ-lille1.fr/
Principal supervisor	**Co-supervisor**
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Thesis information

Keywords	oncoproteins, glycosylation, bioimaging, click-chemistry, unnatural amino-acids
Abstract	Visualizing a specific modification on a specific protein in living cells is a growing area of interest to address cellular functions played by individual proteins and to decipher the complex biological system as a whole. The activities of the teams - from the University of Lille and the Medical College of Wisconsin (MCW) - involved in this project are focused in a large part on the study of post-translational modifications (PTM) of key-proteins involved in tumor emergence. Cancer remains a major cause of morbidity and mortality in Western societies. Sedentary and poor nutrition have accentuated the incidence of this group of pathologies in recent decades. Indeed, many epidemiological studies have indicated that nutrition and lifestyle influenced their emergence. It is estimated that a non-negligible part of cancers still have no genomic or epigenomic origins. Beta-catenin and OTX2 are two oncoproteins actively involved respectively in the emergence of colorectal cancer (CRC) and medulloblastoma, an aggressive cancer of the central nervous system in children. While we are starting to understand the interference of nutrition on the oncogenic character of β-catenin, nothing is known currently for OTX2. Precision human health today appears as a priority both from a public health point of view and for the diagnosis and monitoring of patients. This goal in mind, this unique and innovative project aims to characterize the nutrient-dependent PTM O-GlcNAcylation of both oncogenes by developing a technique to visualize modified β-catenin and OTX2 in living cells. We wish to follow the *O*-GlcNAcylation status of each of these oncoproteins in different nutritional contexts. Our work and others have described a very strong connection between *O*-GlcNAcylation and nutrient levels. Generally, O-GlcNAcylation reduces proteins sensitivity to proteasome, as we showed for β-catenin. We hypothesize that aberrant glycosylation of OTX2 in interaction with

	excess nutrients is deleterious for patients suffering medulloblastoma. Much like our lab did with β-catenin, Dr. Olivier-Van Stichelen's lab has investigated the response of OTX2 to *O*-GlcNAcylation variation and the effect is similar to those observed on β-catenin, e.g. stabilization by this PTM. The biotech company e-Zyvec will partner on the conception and access to OTX2 plasmids necessary for those experiments. To follow in living cells β-catenin and OTX2, we will use a double labelling strategy. The first strategy is based on FRET phenomenon combining transfection of GFP-fused proteins and click chemistry. The second strategy will tag the protein by incorporation of unnatural amino acids. Together, these approaches will implement an innovative and original methodology to detect the protein *O*-GlcNAcylation in live cells. This project at the interface of oncology, protein biochemistry and chemical biology will provide valuable knowledge on nutritional induced oncogenic behaviors without genomic alteration. It should also open the field to a wide range of investigations in various scientific domains. The student in charge of this project will have at the end of the training a strong background in molecular and cell biology, biochemistry and organic chemistry. He/she will make exchanges between the University of Lille and the MCW. He/she will attend international conferences during which he/she will have the opportunity to present the progress of his thesis work.
Expected profile of the candidate	The candidate must have a solid training in cell biology and molecular biology. Expertise in cell imaging (confocal microscopy and FRET technology) is strongly desired. The candidate will be integrated into the teams "*O*-GlcNAcylation, signalling and cell cycle" (head: Tony Lefebvre) and "Chemical GlycoBiology" (head: Christophe Biot), and will have very strong connections with the team of Stéphanie Olivier-van Stichelen (MCW, USA). Strong interactions with the platform TISBio and the company e-Zyvec are to be considered. The candidate must be rigorous, have excellent organizational and teamwork skills, and be able to express himself / herself in English, written and spoken (the practice of French is not compulsory).
Application procedure	The application procedure is detailed on the European programme PEARL website www.pearl-phd-lille.eu. The funding is managed by the I-SITE ULNE foundation which is a partnership foundation between the University of Lille, Engineering schools, research organisms, the Institut Pasteur de Lille and the University hospital. The application file will have to be submitted before April 15, 2020 (10h Paris Time) and emailed to the following address : international@isite-ulne.fr.
Net salary and Lump Sum	A net salary of about €1,600 + €530 per month to cover mobility, travel and family costs.