UNIVERSITY OF LEICESTER

3 Year PhD Studentship available for September 2018

Department: Molecular and Cellular Biology

Supervisors: **Dr. Alfredo De Biasio** (adb43@leicester.ac.uk)

Eligibility: UK /EU applicants only

Project Title: Architecture and function of human DNA lesion bypass machines

Project Description:

Aims:

To reveal the molecular architecture and function of the human DNA lesion bypass machinery, so to define novel therapeutic targets for cancer treatment.

Background:

Cellular responses to DNA damage and/or replication stress can affect genome stability in cancers and influence the response of patients to therapy. Beside direct repair, DNA damage tolerance (DDT) is one of the genomic maintenance programs that can contribute to the etiology of cancer. Translesion DNA synthesis (TLS) is a critical component of DDT that allows the bypass of DNA lesions and other replication barriers, and thus it can directly influence the effectiveness of anti-cancer drugs inducing DNA damage[1,2]. TLS is carried out by polymerases specialized in replicating damaged templates, and which require the processivity factor PCNA and the oncogenic modulator $p15^{PAF}$ to function[3]. Because TLS polymerases can render genotoxic anti-cancer drugs less effective *via* direct bypass of the lesions caused by treatment, and they promote tumor heterogeneity by contributing to mutagenesis, these enzymes have emerged as an attractive target for the development of new anti-cancer therapies[4,5].

However, due to the lack of structural and mechanistic information on the protein complexes orchestrating TLS, the TLS process is still poorly understood at the molecular level, and this represents an important limitation for the development of novel and effective TLS inhibitors. To shed light on the molecular basis of TLS, we have already determined the structure of human PCNA bound to $p15^{PAF}$ (Ref. 6) and DNA[7], and this sets the ground for a more complete structural and functional investigation of the TLS machinery.

Approaches:

In this project, the student will:

1) Express recombinant DNA replication and TLS proteins (*e.g.*, polymerase δ, η and ι, PCNA, $p15^{PAF}$, RFC) in bacterial cells, and purify them to homogeneity

2) Determine the 3D structure of the produced recombinant proteins and their assemblies using X-ray crystallography and Cryo-EM, working with the new Leicester Cryo-EM facility established at the Leicester Institute of Structural and Chemical Biology (LISCB).

3) Elucidate the functional role of $p15^{PAF}$ in TLS, using *in vitro* DNA replication assays.

4) Participate in a collaboration with Prof. Joachim Jose at the Pharmaceutical and Medicinal Chemistry Institute of the University of Münster, Germany, to apply the "Autodisplay" technology platform[8] for the development of specific inhibitors of $p15^{PAF}$ function.

References:

1. Waters, L.S et al. (2009) Microbiol Mol Biol Rev 73:134–154

2. Ghosal, G. et al. (2013) Transl Cancer Res 2:107–129

3. Povlsen, L.K. et al. (2012) Nat Cell Biol; 14(10):1089-98

4. Korzhnev, D.M. & Hadden, K.M. (2016) J Med Chem 59:9321–9336; 13

5. Sail, V. et al. (2017) ACS Chem Biol 12(7):1903-191

6. De Biasio, A. et al. (2015) Nat Commun 6:6439; 5

7. De March, M. et al. (2017) Nat. Commun. 8, 1393

8. Jose, J. (2006) Appl Microbiol Biotechnol. 69(6):607-14

Funding details:

This studentship is fully funded for 3-years by the University of Leicester, starting in September 2018. This will provide a Tuition Fee waiver at UK/EU rates and stipend at RCUK rates (for 2018 this will be £14,777) for 3 years.

Entry requirements:

Applicants are required to hold/or expect to obtain a UK Bachelor Degree 2:1 or better in a relevant subject. The University of Leicester English language requirements apply where applicable.

How to apply:

You should submit your application using our online application system.

> **Apply for Molecular and Cell Biology Research / September 2018**
>
> In the funding section of the application please indicate you wish to be considered for A **College of Life Sciences Studentship**
>
> In the proposal section please provide the **name of the supervisor and project title.** Include a personal statement explaining why you want to be considered for this project.

Project / Funding Enquiries:

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