Grenoble Alliance for Integrated Structural & Cell Biology

INTERNSHIP PROPOSAL

Institute and Group: Blackledge Group (Protein Dynamics and Flexibility by NMR) Institut de Biologie Structurale

Supervisor: Martin Blackledge (collaboration with Rob Ruigrok) **Phone:** 04 57 42 85 54 **Email**:martin.blackledge@ibs.fr

Research project title: Investigation of the structural and dynamic origin of membraneless viral replication factories

5 Keywords to describe the project: Intrinsically disordered proteins, protein dynamics, nuclear magnetic resonance spectroscopy, fluorescence spectroscopy, self-assembly

Description of the project (aims, experimental techniques, recommended background): 10 to 15 lines:

Paramyxoviruses constitute dangerous human pathogens that express their own machinery for transcription and replication, with their RNA genomes packaged into helical nucleocapsids, comprising a multitude of nucleoproteins (N). N interacts with the phosphoprotein (P) via distinct mechanisms during the viral replication cycle and these interactions represent important targets for drug development. Crucially, both N and P exhibit *extensive* intrinsic conformational disorder in their functional state, with unfolded regions from 100 to 500 amino acids in length. Intrinsically disordered proteins play crucial roles in biology, but their interaction mechanisms remain very poorly understood. Elaborating atomic resolution descriptions of highly disordered large-scale assemblies such as N:P complexes is extremely challenging, requiring the association of complementary solution state methods, one of the key aims of this project. In particular it has recently been shown that N and P proteins associate in infected cells to create highly dynamic *membraneless organelles*, liquid phases that constitute efficient viral replication factories and provide protection against the host immune system. The candidate will use NMR and fluorescence spectroscopy, combined with complementary biophysical approaches and simulation, to describe the function of these liquid droplets at atomic resolution, thereby providing new insight into the role of extensive conformational dynamics of N and P in the viral replication cycle. An interest in biophysics or biophysical chemistry is appropriate.

Justification that the internship's subject fits with the general theme of GRAL (3 lines):

The project aims to use NMR and fluorescence spectroscopies, in combination with electron microscopy and small angle scattering, to discover the molecular mechanisms underlying the formation of liquid-like membraneless organelles in cells infected by paramyxoviruses.

Relevant publications of the team (3 max):

Self-Assembly of Measles Virus Nucleocapsid-like Particles: Kinetics and RNA Sequence Dependence. Milles S, Jensen M, Communie G, Maurin D, Schoehn G, Ruigrok R, **Blackledge M** ***Angew Chemie Intl Edition***, 128, 9502–9506 (2016)

Visualizing the molecular recognition trajectory of an intrinsically disordered protein using multinuclear relaxation dispersion NMR. Schneider R, Maurin D, Communie G, Kragelj J, Hansen F, Ruigrok R, Jensen M, **Blackledge M** ***J Am Chem Soc*** 137, 1220 (2015)

Intrinsic disorder in measles virus nucleocapsids. Jensen M, Communie G, Ribeiro E, Martinez N Desfosses A, Salmon L, Mollica L, Gabel F, Jamin M, Longhi S, Ruigrok R, **Blackledge M** ***Proc Natl Acad Sci U S A*** 108, 9839 (2011)