

Short Term report August 2020 – March 2021

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This proposal provides user access to new services based on the combination of EMBL's HTX Lab services (CrystalDirect) and automated data collection at ESRF's MX beamlines supported by the EMBL-ESRF Joint Structural Biology Group (JSBG). These access modalities were developed through **LTP MX-1819**.

15 projects involving 41 users have been positively reviewed through iNEXT Discovery all of which were granted access to the services above. This includes 11 Online Crystallography and 4 large scale Ligand/fragment screening projects. 10 of these projects have sent samples and benefited from beam time (8 Online Crystallography and 2 large-scale ligand/fragment screening). Full-text applications stored by the iNEXT-Discovery management system can be provided upon request.

Online Crystallography (protein-to-structure pipeline):

Anja Winter, Keele University, U.K. : Structural Analysis of novel Galactokinase from *L. Donovanii*: Multiple samples analysed. Crystal hits identified.

Livia Tepshi, Curie Institute, France: Structural analysis of Myosin domains. Multiple samples analysed

Chole Zubieta, INRA, France: Structural studies of MADS transcription factor-phylllogen effector protein interactions. Multiple samples analysed. One structure solved, one publication.

Ramesh Pillai, University of Geneva, Switzerland: Molecular insights into the RNA modifications and small RNA machinery. Crystals for three complexes obtained, one structure solved, publication in preparation.

Nicolas Reynoird, Institute for advanced biosciences (IAB), CNRS France: Characterization of a novel methyl binding domain. Crystal hits obtained.

Maria Dimarogona, University of Patras, Greece: Deciphering structure-function relationships of enzymes implicated in plastic degradation. Several ligand-complexes analysed multiple datasets obtained. One ligand-bound structure solved.

Virginie Ropars, I2BC CEA Saclay, France: Assembly of non-homologous End Joining pathway (NHEJ) at double-strand breaks and characterization of new inhibitors. Crystal hits for several NHEJ complexes, several datasets obtained, MS in preparation

Sophie Zinn-Justin, CEA Saclay, France: BRCA1/2 complexes in mitosis and meiosis. Multiple samples analysed, two structures solved, two publication, 3 PDB depositions

Large Scale Ligand/fragment screening

Andres Palencia, Institute for advanced biosciences (IAB), France: Validation of cryptosporidium hominis poly-adenosine polymerase as a novel therapeutic target to treat gastrointestinal diseases. A

collection of 500 nucleotide-related compounds selected through in silico studies has been screened. Data analysis is ongoing.

Walter Nickel, Heidelberg University, Germany: Fragment-based screening for FGF2 for the development of FGF2 secretion inhibitors. Screening of a 700-fragment library provided by the facility has been completed. Data analysis ongoing.

Samples for two additional fragment screening projects are expected shortly:

Ramon Hurtado-Guerrero, University of Zaragoza, Spain: FUT8 Inhibitors to tackle Cancer.

Sebastian Guettler, The Institute of Cancer Research, London U.K.: Crystallographic screening to identify binders to tankyrase substrate recruitment modules.

Additional results:

- A new method for the rapid analysis of membrane proteins in LCP based on the CrystalDirect technology and serial data collection has been developed (<https://doi.org/10.1101/2021.06.03.446146>).
- One of EMBL CrystalDirect harvesters has been integrated into the MASSIF1 beam line opening the prospect for new and unique experimental opportunities (<https://doi.org/10.1063/1.5084627>).

These access modalities have been advertised through invited presentations (American Crystallography Association Meeting 2020, IUCR meeting 2021, Instruc-Eric webinar series 2021, iNEXT Discovery webinar series 2020, ...) and dedicated publications (<https://doi.org/10.3791/62491>; <https://doi.org/10.1002/1873-3468.13062>)

Beam line performance.

The performance of the beam lines has been excellent. The benefits of the EBS upgrade are clearly visible and very impressive. We recommend to invest effort in enabling ISPyB to use automated sample PUC identification based on RFID tags (already available commercially). This will avoid errors in PUC assignments during beam line operation benefiting not only high throughput operations but also users of the remote beam line operation mode.

PDBs: 22

7ABM, 7NDY, 7BDX, 6STB, 6ST9, 6ST8, 6STA, 5QTK, 6SHV, 6SI5, 6H3, 6TBZ, 6TCE, 6FZS, 6ZMN, 6FZT, 6YXH, 6YXG, 6YXF, 6YXD, 6YX9, 7NB0

Publications= 8

The intervening domain is required for DNA-binding and functional identity of plant MADS transcription factors Xuelei Lai, Rosario Vega-Léon, Veronique Hugouvieux, Romain Blanc-Mathieu, Froukje van der Wal, Jérémy Lucas, Catarina S Silva, Agnès Jourdain, Jose M

Muino , Max H Nanao , Richard Immink , Kerstin Kaufmann , François Parcy , Cezary Smaczniak , Chloe Zubieta.

Nat Commun. 2021 Aug 6;12(1):4760. doi: 10.1038/s41467-021-24978-w.

BRCA2 binding through a cryptic repeated motif to HSF2BP oligomers does not impact meiotic recombination. Ghouil R, Miron S, Koornneef L, Veerman J, Paul MW, Le Du MH, Sleddens-Linkels E, van Rossum-Fikkert SE, van Loon Y, Felipe-Medina N, Pendas AM, Maas A, Essers J, Legrand P, Baarends WM, Kanaar R, Zinn-Justin S, Zelensky AN.

Nat Commun. 2021 Jul 29;12(1):4605. doi: 10.1038/s41467-021-24871-6.

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Structural Bases for the Allergenicity of Fra a 1.02 in Strawberry Fruits. Orozco-Navarrete, B.; Kaczmarska, Z.; Dupeux, F.; Garrido-Arandia, M.; Pott, D.; Perales, A. D.; Casañal, A.; Márquez, J. A.; Valpuesta, V.; Merchante, C. *J. Agric. Food Chem.* 2020, 68 (39), 10951–10961. <https://doi.org/10.1021/acs.jafc.9b05714>.

Fragment-Based Discovery of Non-bisphosphonate Binders of Trypanosoma Brucei Farnesyl Pyrophosphate Synthase. Münzker, L.; Petrick, J. K.; Schleberger, C.; Clavel, D.; Cornaciu, I.; Wilcken, R.; Márquez, J. A.; Klebe, G.; Marzinzik, A.; Jahnke, W. *ChemBioChem* 2020, 21 (21), 3096–3111. <https://doi.org/10.1002/cbic.202000246>.

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