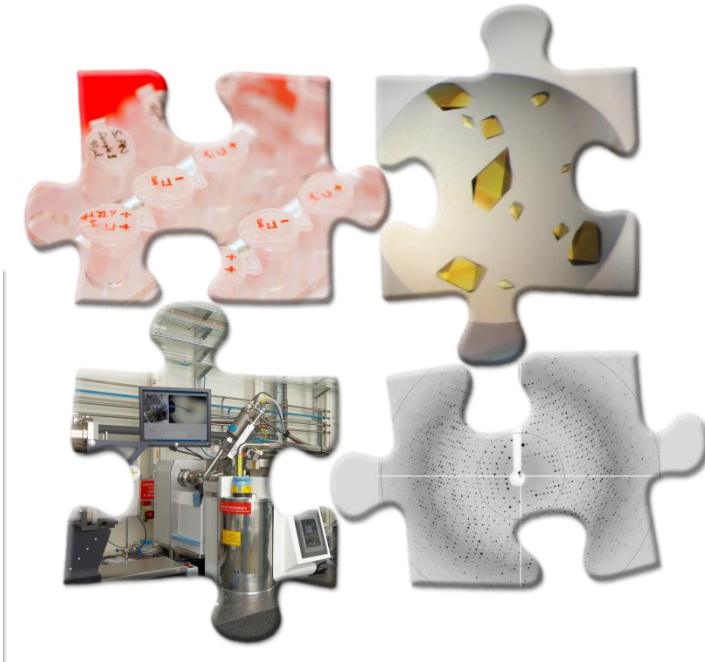


# Integrated Pipelines for Ligand Screening

*... and more*



**Jose A. Marquez**

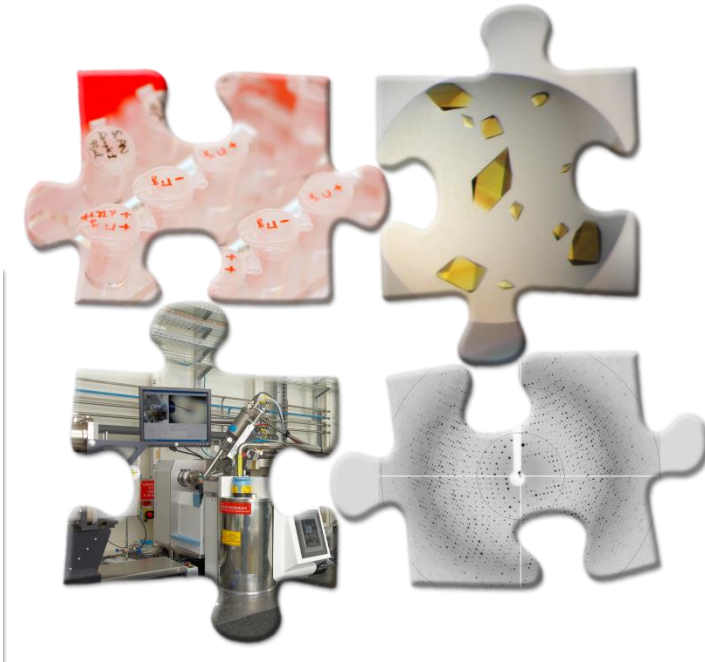
*Head of the Crystallization Facility*

*EMBL-Grenoble*

# Integrated Pipelines for Ligand Screening

*... and more*

- Crystallization and data collection occur at highly automated facilities ...



...but these operations are separated by manual steps

✓ **Crystal mounting**

- Manpower & training
- Loss of diffraction power
- Time

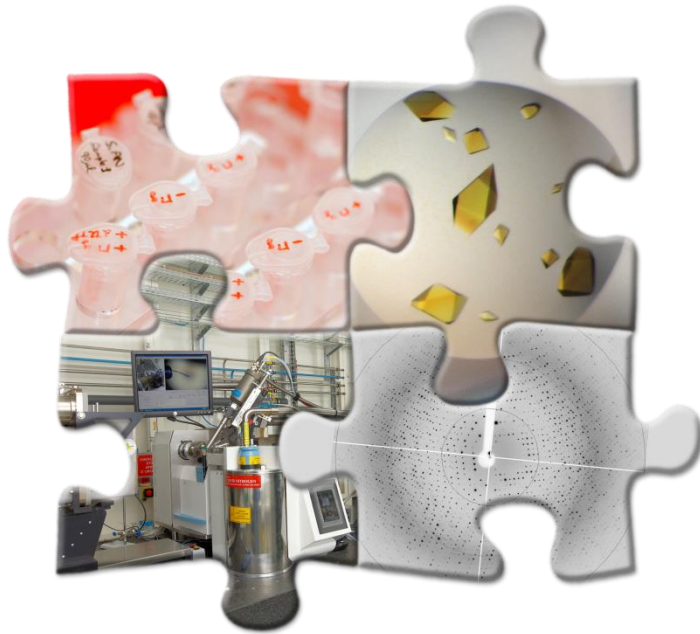
✓ **Data collection**

- Synchrotrons are sparsely distributed
- Access to beam time every 1-3 months

# Integrated Pipelines for Ligand Screening

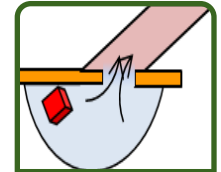
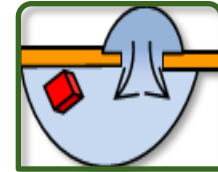
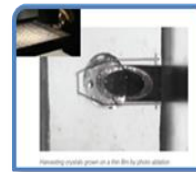
*... and more*

Integrated protein to X-ray  
data collection pipelines



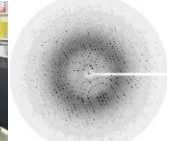
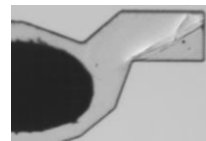
## CrystalDirect

Crystal Mounting & Processing



## MASSIF

Automated Data Collection



# Integrated Pipelines for Ligand Screening

## Four Key Components

### The HTX Lab

From Pure samples to Crystals



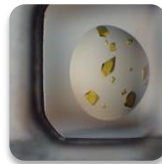
The HTX lab has supported 791 Scientists since 2003



200 Regular Users  
Over 600 registered  
scientists



1.900 samples processed  
per year



990.000 Crystallisation  
Experiments per year



BioStructx



## Access to European Researchers Through



# Integrated Pipelines; *Four Key Components*

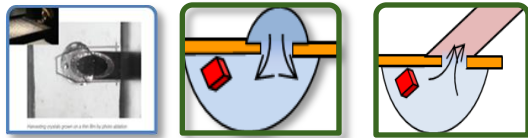
## The HTX Lab

From Pure samples to Crystals

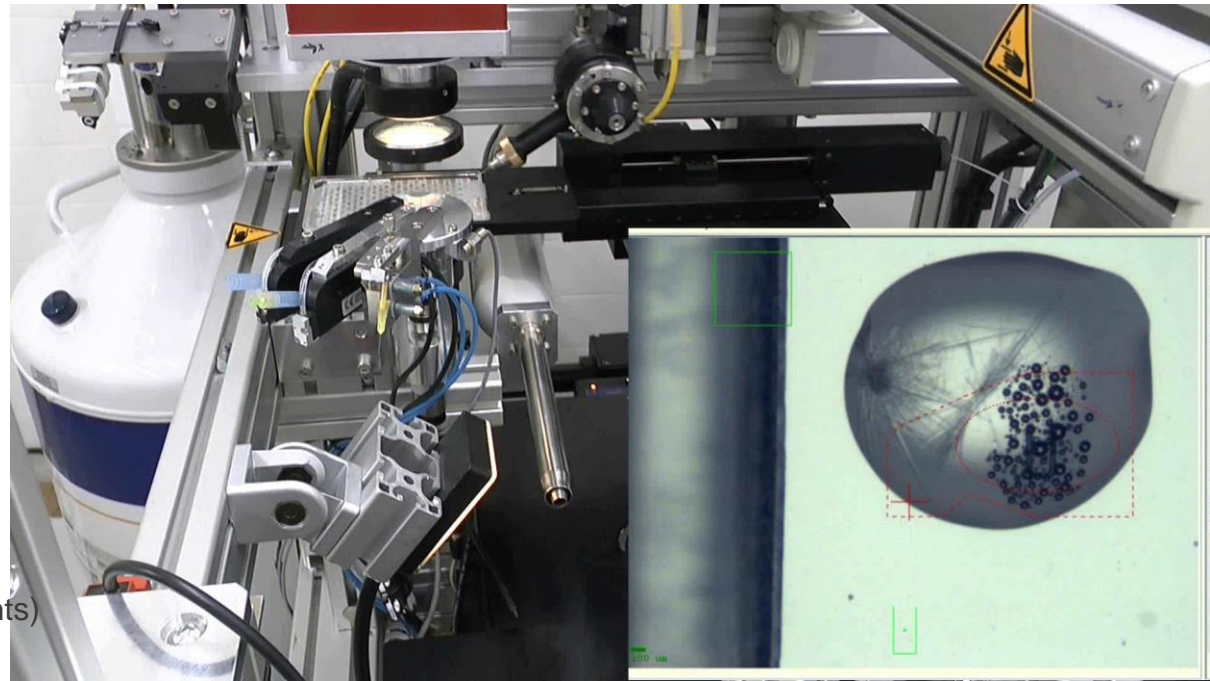


## CrystalDirect

Crystal Mounting & Processing



- Automated Crystal Harvesting
- Direct Cryocooling (no cryo protectants)
- Reduced mechanical stress
- Reduced background
- Multicrystal mounting
- Crystal Surgery



*Zander et al., Acta Cryst D, 2016, in Press*



# Integrated Pipelines; *Four Key Components*

*Available to all users of the THX lab*

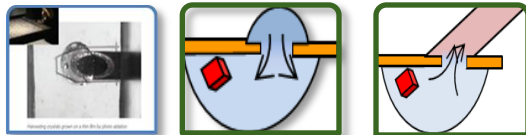
## The HTX Lab

From Pure samples to Crystals

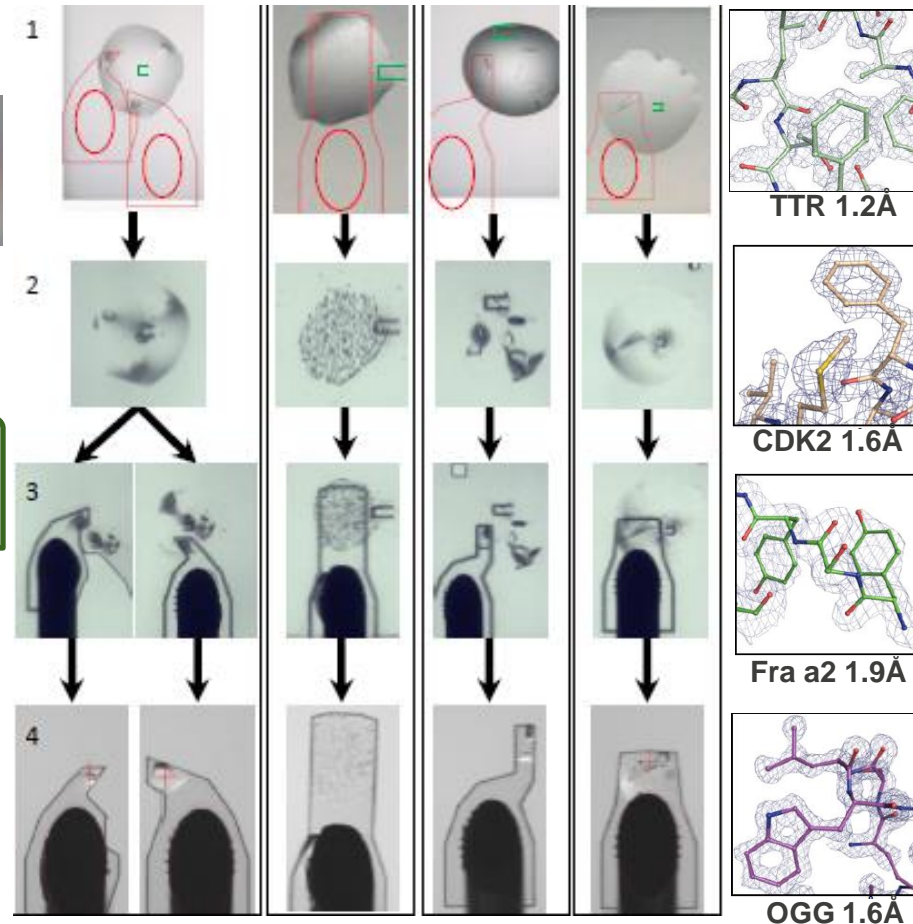


## CrystalDirect

Crystal Mounting & Processing



Zander et al., Acta Cryst D, 2016, in Press



### Transthyretin

- human transport protein
- Alycia Yee, Trevor Forsyth

### Cyclin-dependent kinase 2

- involved in cell cycle progression
- J-P Marquette, Magali Mathieu

### Fra a2

- strawberry allergen protein
- Ana Casañal, Victoriano Valpuesta

### 8-Oxoguanine glycosylase

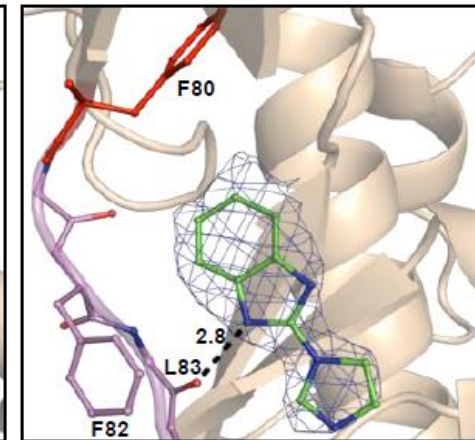
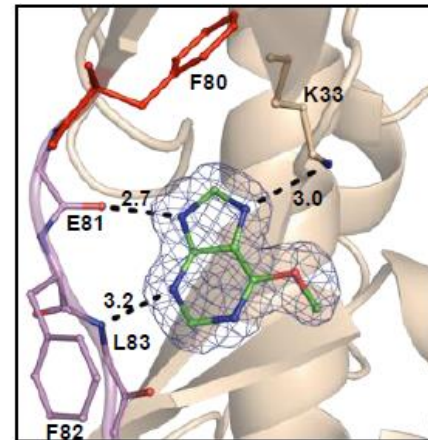
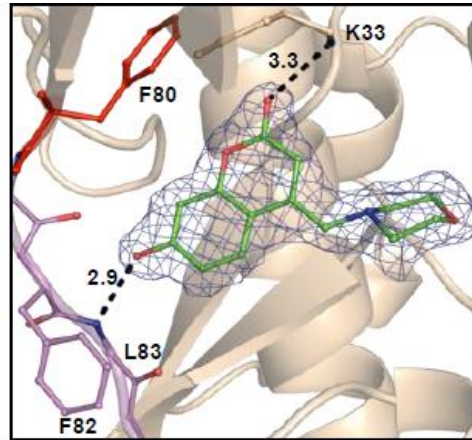
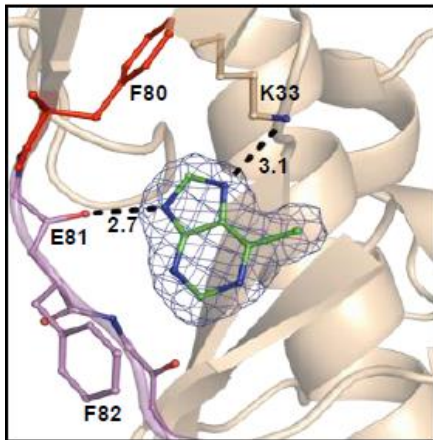
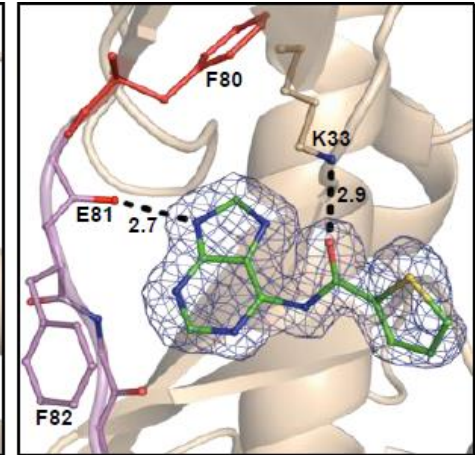
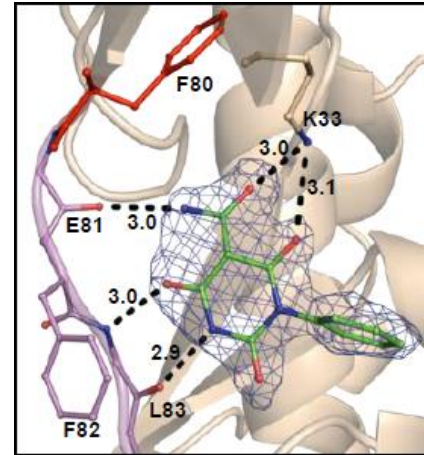
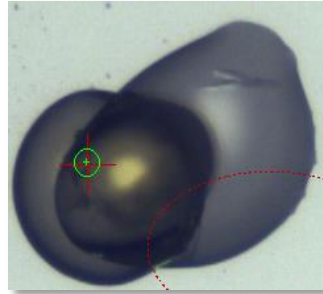
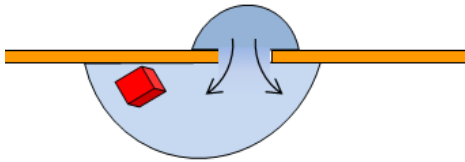
- DNA repair protein
- Mari Ytre-Arne, Bjørn Dalhus

**Over 5000 crystals processed !**

# CrystalDirect Crystal Soaking Through Controlled Diffusion

Zander et al., *Acta Cryst D*, 2016, in Press

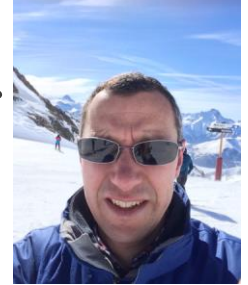
- Phasing agents
- Ligands



# Integrated Ligand Screening

## Four Key Components

Matthew Bowler  
Staff Scientist



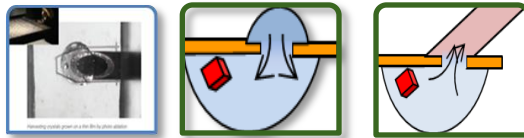
### The HTX Lab

From Pure samples to Crystals



### CrystalDirect

Crystal Mounting & Processing

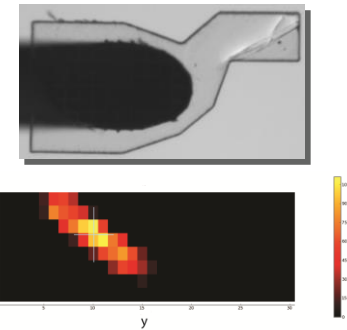
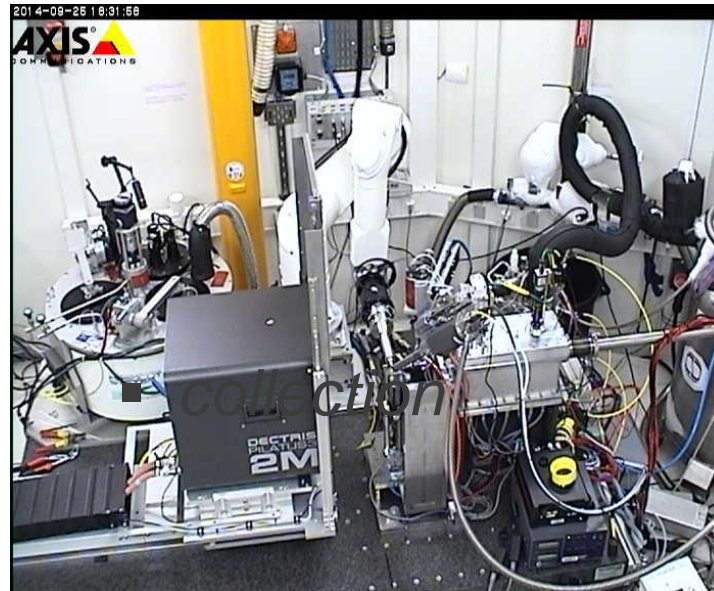


### MASSIF

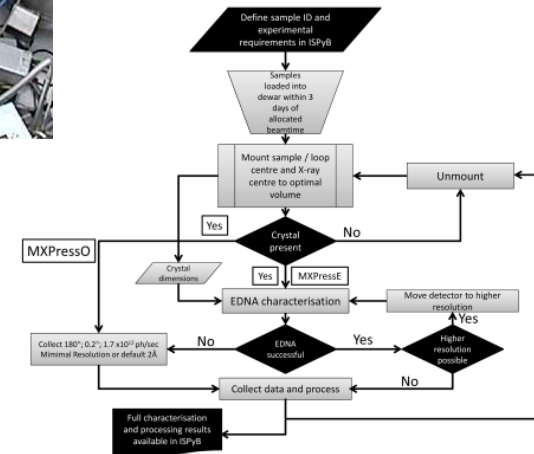
Automated Data Collection



### ESRF MASSIF 1



### Automated workflows



- Hands-off data collection
- 150 Samples per session



# Integrated Pipelines for Ligand Screening

## Four Key Components

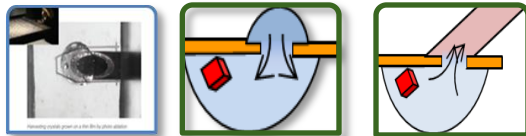
### The HTX Lab

From Pure samples to Crystals



### CrystalDirect

Crystal Mounting & Processing



### MASSIF

Automated Data Collection

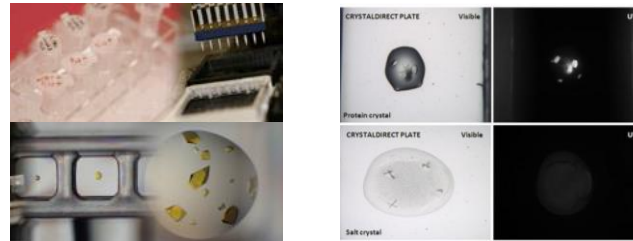


### Software

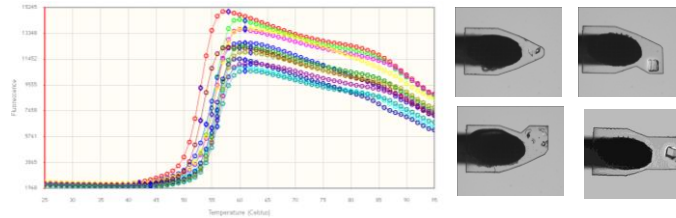
CRIMS & ISPyB



## The Crystallization Information Management System



- Sample evaluation
- Screening
- Optimization
- Crystal Mounting



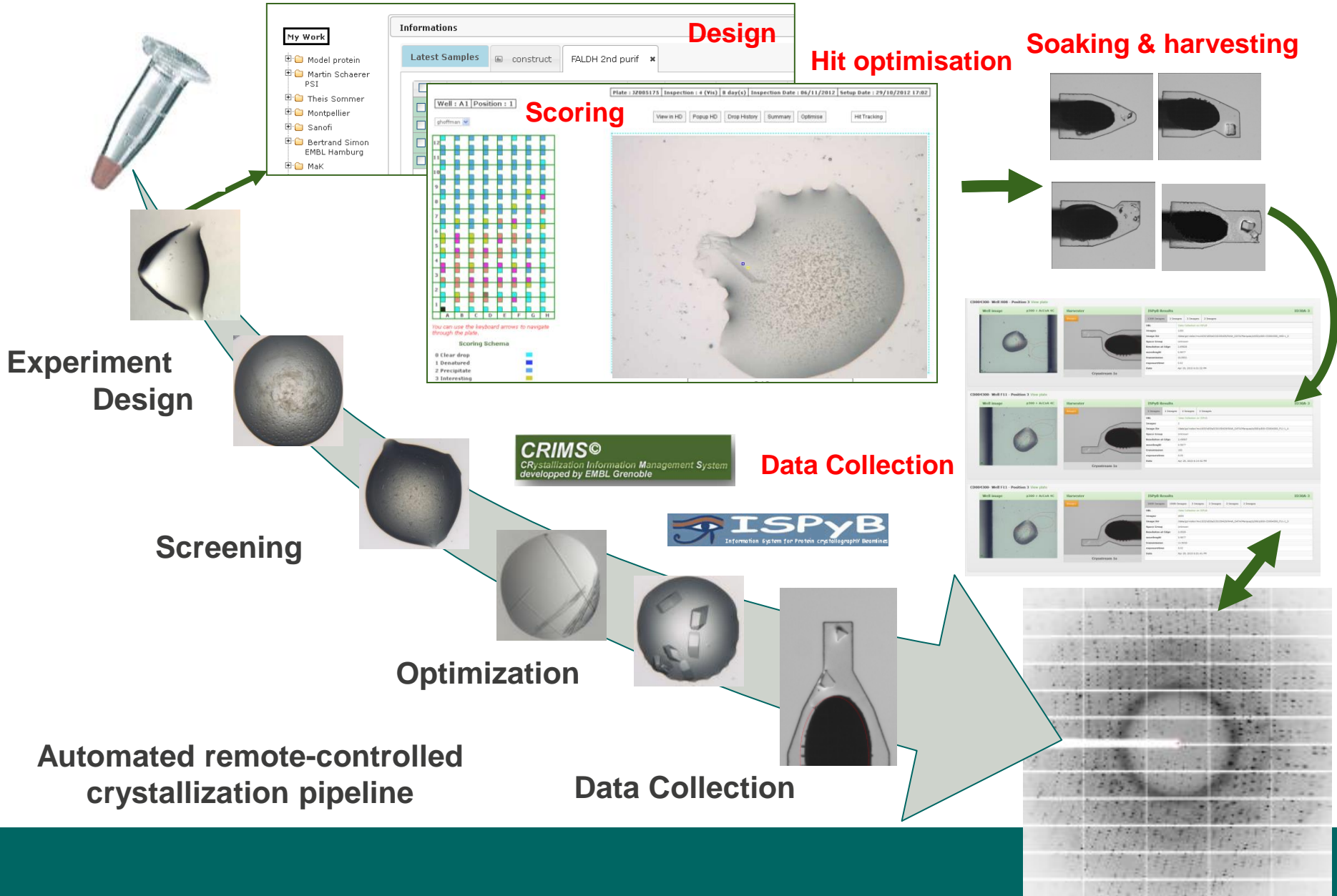
- From pure sample to mounted crystals
- Operating at EMBL, HD, HH & GR
- Licensed to 10 other laboratories

## ISPyB Beamline Data management system



- Data Collection
- Data Processing
- Reporting

# Remote Crystallography Pipeline



# Integrated Ligand Screening

HTS screening

Provided by User

Fragment Library

Provided by HTX lab

High Throughput  
Crystallisation

Automated Crystal  
Harvesting &  
Processing

Automated Data  
Collection  
MASSIF

Automated Data  
Processing &  
Ranking

In Progress

Hits

## Requirements

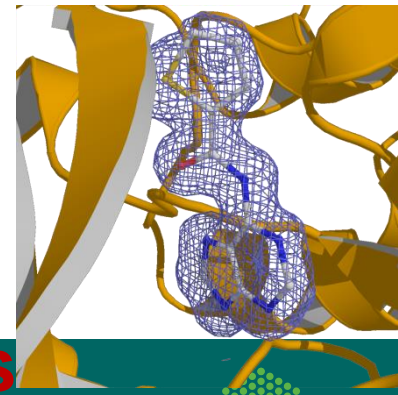
- ✓ Robust Crystal System stabilshed
  - Good cristallisation rate
  - Consistant diffraction  $<2.7\text{\AA}$
- ✓ High Biotechnological potential



iNEXT

European Commission  
H2020 program

Ligand soaking  
With Crystal Direct



# Thank you!

- **The HTX Lab**

- Irina Cornaciu
- Peter Murphy
- Guillaume Hoffmann
- Vincent Mariaule
- Sonia Rodriguez Puente
- Gael Seroul\*
- Aine Barry
- Zuzanna Kaczmarek



Diffraction instrumentation team

- **Florent Cipriani**
- Jérémy Sinoir
- Christophe Landret\*
- Gergely Papp
- Franck Felisaz
- Marcos Lopez Marrero
- Clement Sorez
- Christopher Rossi
- Robert Janocha

\* Former lab members

Synchrotron crystallography team

- **Andrew Mc Carthy**
- **Matthew Bowler**
- Alejandro De Maria Antolinos
- Max Nanao
- Adam Round

ESRF

- **Gordon Leonard**
- Christoph Mueller-Dieckmann
- Didier Nurizzo
- Olof Svensson
- Ulrich Zander
- Stephanie Malbet-Monaco

