Protein Crystallization Setup and Workflows in the BI Structural Research Group

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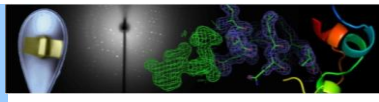


The Boehringer Ingelheim Biberach Structural Research Group has integrated protein expression and purification, biophysics and protein crystallography units. Focus of the group is to elucidate, characterize and quantify the interaction between ligands and their target proteins. We apply our technologies in preclinical stages of drug discovery projects and support projects in the target assessment, lead identification and lead optimization phases. We have a state-of-the-art crystallization laboratory equipped with high-throughput instrumentation that allows for the conduction of efficient crystallization campaigns. In our poster, we show details of the setup in our lab and give some insight into the strategies and workflows that are implemented.

Protein Expression & Purification

- baculovirus/ insect cell and E. coli expression systems
- wavebag cell culture
- Techfors 10L fermentors
- Multifors 1L fermentors
- range of Äkta systems

Biophysics

- **S**tatic **L**ight **S**cattering
- **M**asspectrometry
- **I**sothermal **T**itration **C**alorimetry
- Thermofluor
- Microscale Thermophoresis
- NMR
- **C**ircular **D**ichroism

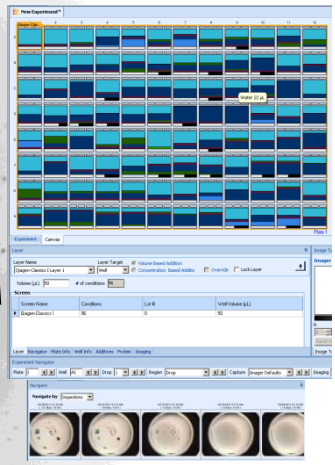
Crystallization

X-ray Hardware

Rigaku Micro7 HFM
- Varimax-HF, AFC11, Saturn944+, Actor, Cobra Cryosystem
- Varimax-HF, Mar345DTB, Cobra Cryosystem

Incoatec Microfocus Source IμS
- Incoatec Optics, Mar345DTB, Cobra Cryosystem

Crystallization Software: Rockmaker



PCT

Pre-**C**rystallisation-**T**est to optimize protein-concentration

Primary Screens

- hardware: Phoenix
- 12 primary screens on hand
- incubation at 2 temperatures
- +/- ligands
- various drop-mixtures
- 96 well MTP

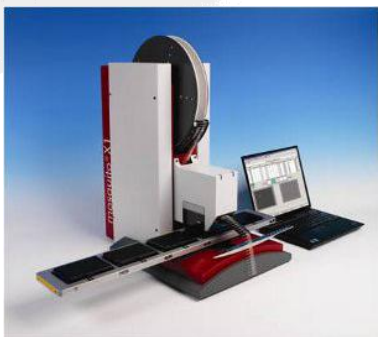
Finescreens

- Hardware: Formulator and Mosquito
- 96 and 24 well plates

Crystal Production

- hanging drop
- sitting drop
- seeding (various methods)
- 24 well VDX plates

RockImager



Our Aims

- provide a flexible, multifaceted protein crystallization service
- provide (high) crystallization throughput adapted to project needs
- crystallize on demand freshly prepared protein without delay
- give advice on crystallization options and optimization protocols

Our Group Mission

- identify and characterize protein structures and ligand binding sites
- validate and prioritize **H**igh **T**hroughput **S**creening hits
- support **L**ead **O**ptimization process by structure-based molecular design
- supply novel chemical matter by **F**ragment **B**ased **S**creening