MX from an Industrial Perspective

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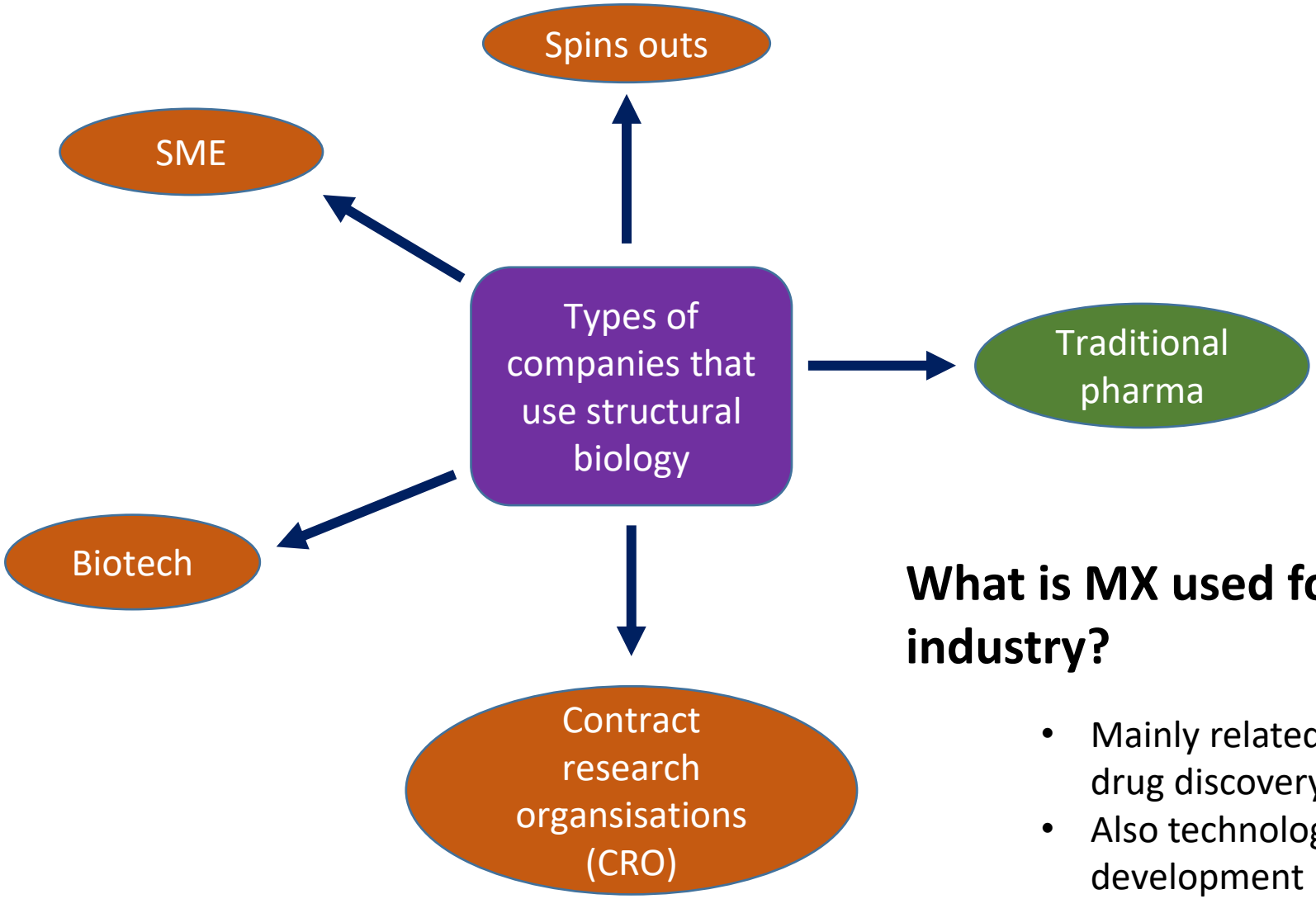
diamond

Types of industry and advantages of using MX

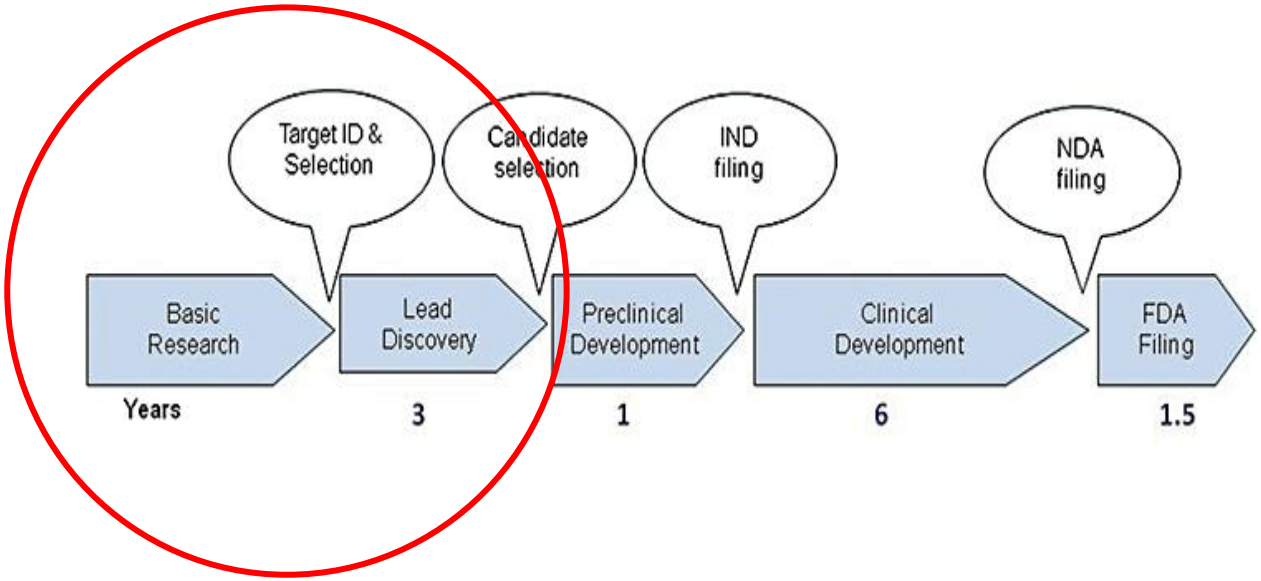


What is MX used for in industry?

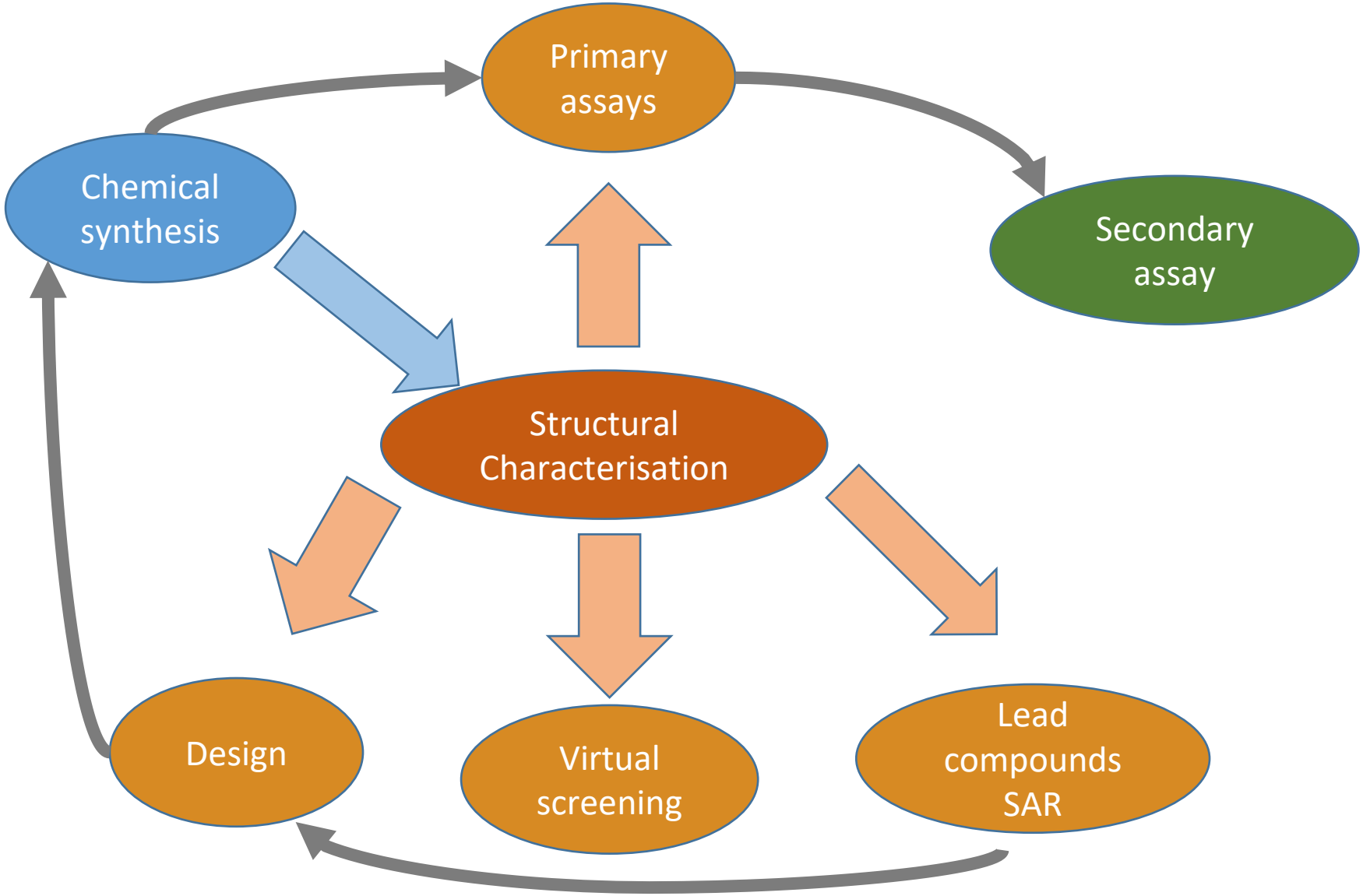
- Mainly related to drug discovery
- Also technology development

Why use structural biology

Invaluable insights at the drug discovery phase



- Protein structures can be used for the following:
  - Basic research – characterisation of the protein target (academic and industrial)
  - Lead discovery – identification of compounds (industrial and academic)



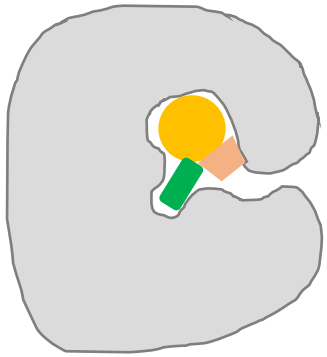
Structure is integral to many steps of the discovery process

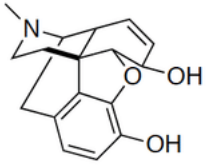
How structures contribute to screening

Structural aided drug design	• Crystal structures help design molecules	• Bound ligand in structure used to help predict where modifications could be added to provide increased potency or selectivity • Often used as an adjunct to other screening strategies within big pharma
Virtual screen	• Docking models: use of a virtual compound library with the X-ray structure of the protein • If have a known ligand, as a base to develop further compounds on	• Can provide the starting structures for a focused screen without the need to use expensive large library screens • Can also be used to look for novel patent space around existing compound structures
Fragment screen	• Soak small compounds into crystals with often low mM activity	• Identify new binding sites • Identify novel chemistry for known sites • Join selected fragments together to fit into the chemical space to increase potency

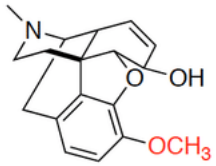
SAR – Structure activity relationships

- Link between the drug target and the lead compound.
- Structures of the target with and without the lead compound act as a guide that can allow computational chemists and molecular modellers to identify binding site interactions

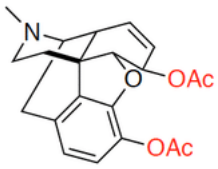




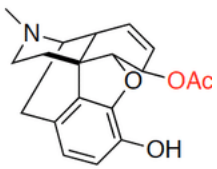
morphine
relative potency = 1
addictive



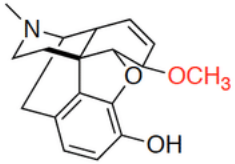
codeine
relative potency = 0.2
addictive



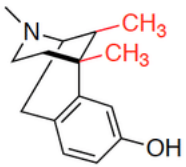
heroin
relative potency = 2
addictive



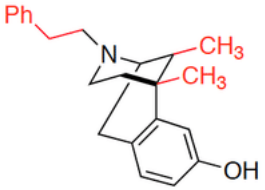
acetylmorphine
relative potency = 4
addictive



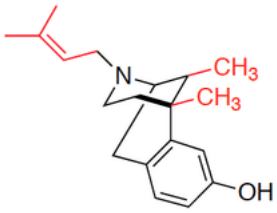
heterocodeine
relative potency = 5
addictive



metazocine
relative potency = 1
addictive



phenazocene
relative potency = 4
non-addictive



phenazocene
relative potency = 0.33
non-addictive

Crysalin Ltd

R&D based spin out

Venture capital funded spin out from University of Oxford

Founded by John Sinclair and Martin Noble

- Aim: to provide rapid determination of protein structures, irrespective of target class, at sub 3Å resolution

How to achieve this?

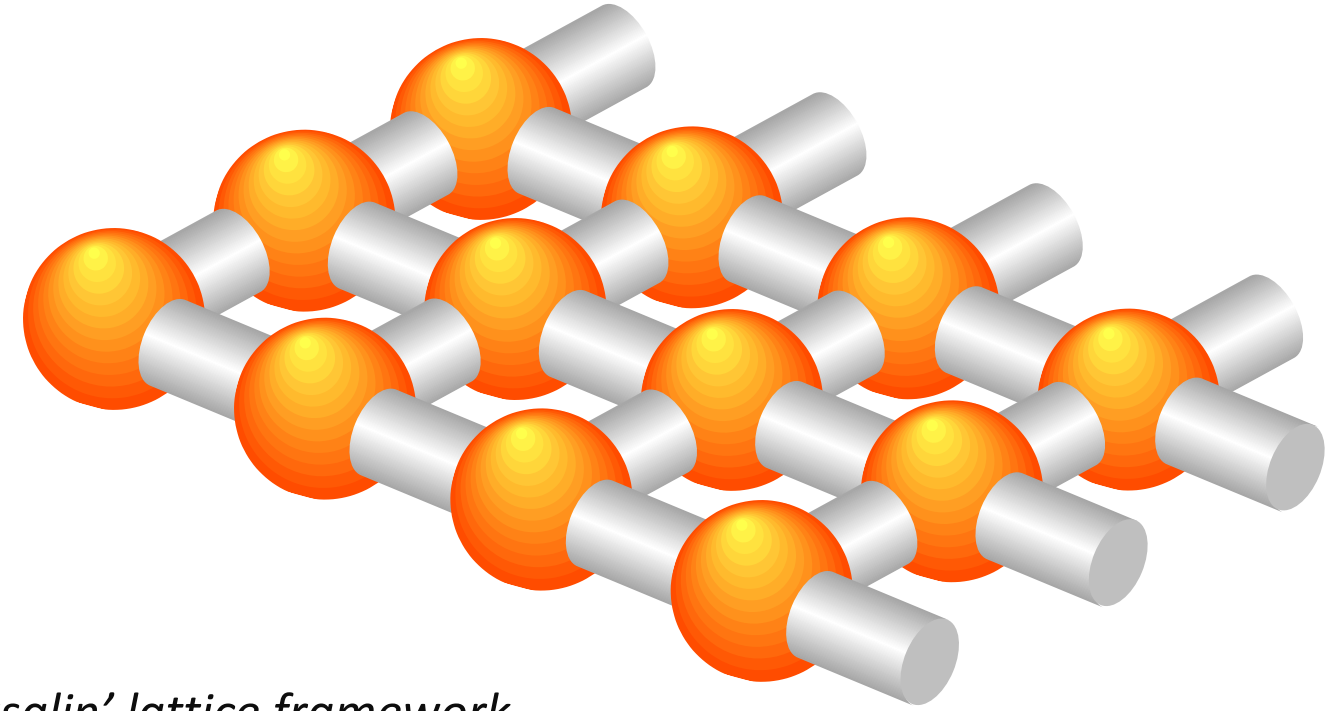
Crysalins are a biological nanomaterial – a self assembling protein lattice

- Remove need for protein crystallisation
- No reproducibility issues (poor/variable resolution, etc)
- No target class restriction (ion channels/GPCRs)
- Remove need for large quantities of highly purified protein
- Increase speed of process
- Increase certainty of result

An industrially robust and consistent process with predictable results

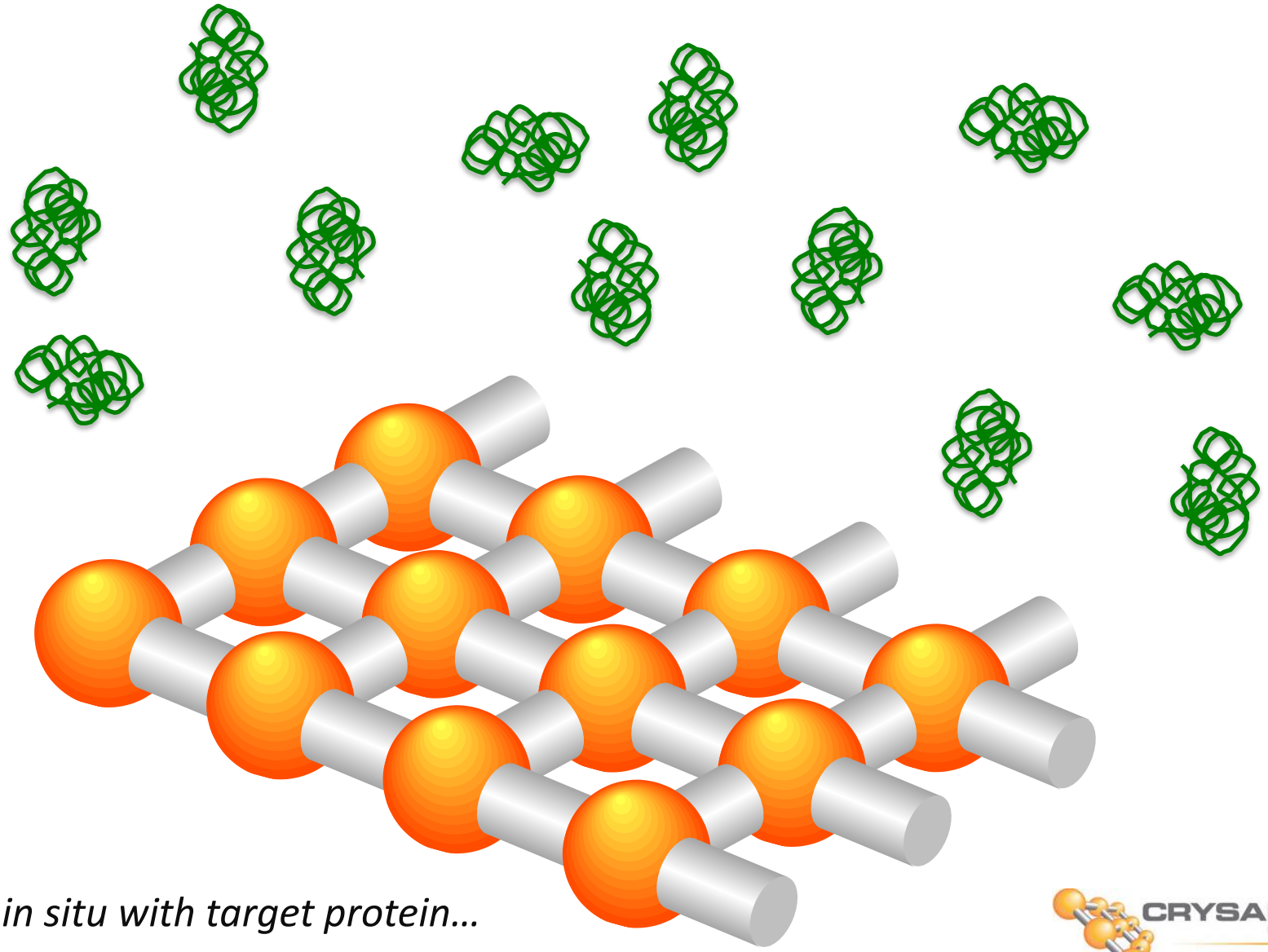
The freedom to choose which target to work on due to scientific validity of the target not the technical limitations of the methodology

Crysalin concept 2D



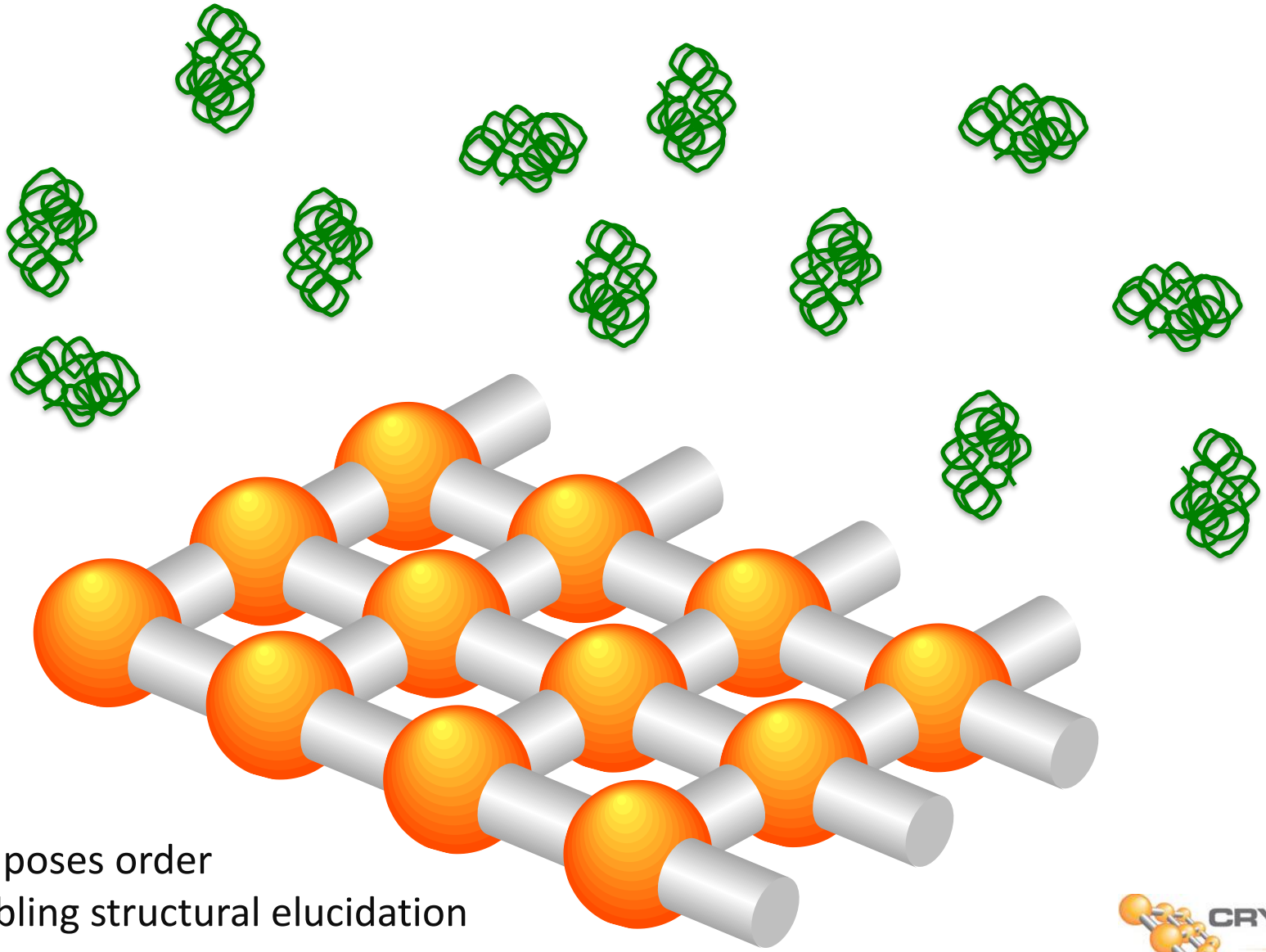
The 'Crysalin' lattice framework…

Technical concept 2D



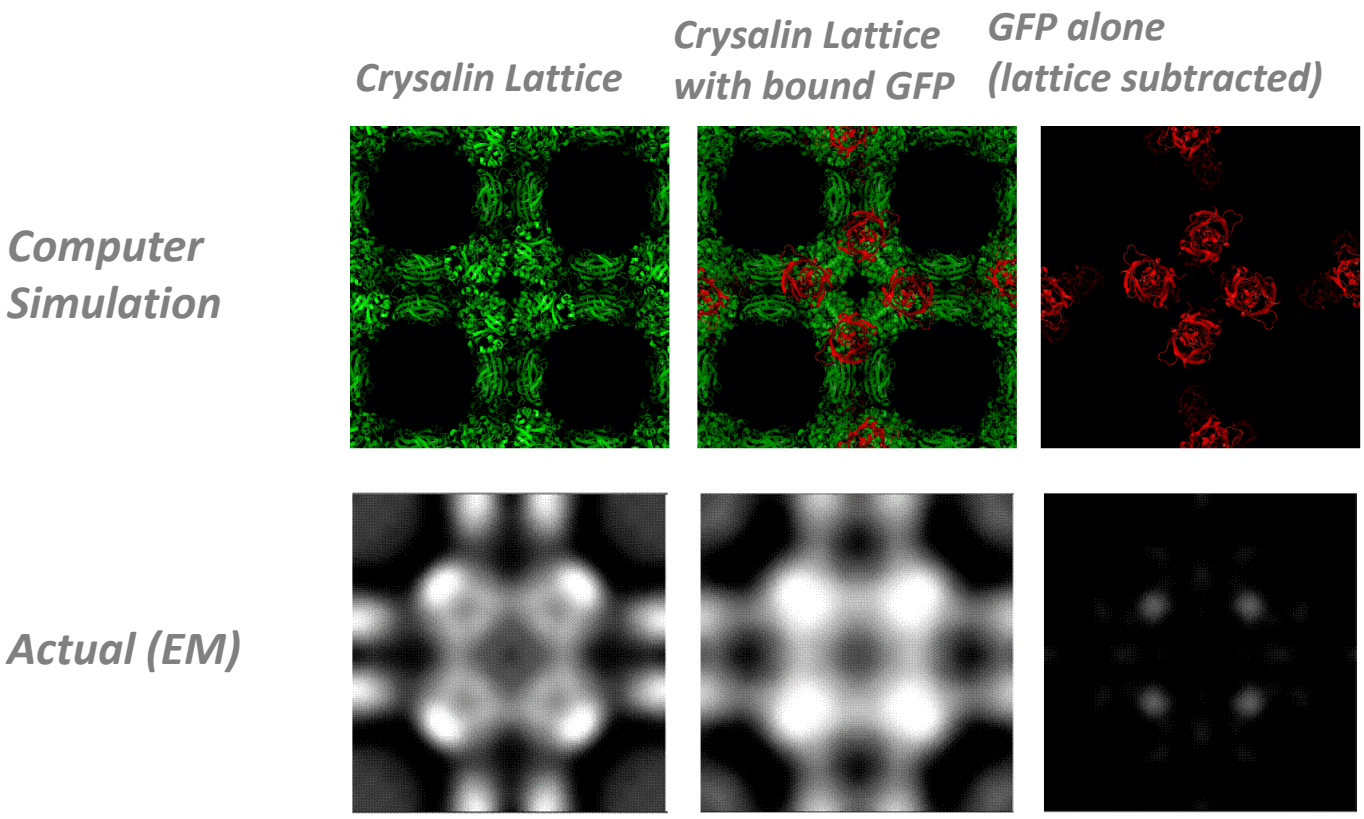
…placed in situ with target protein…

Technical concept 2D



…imposes order
enabling structural elucidation

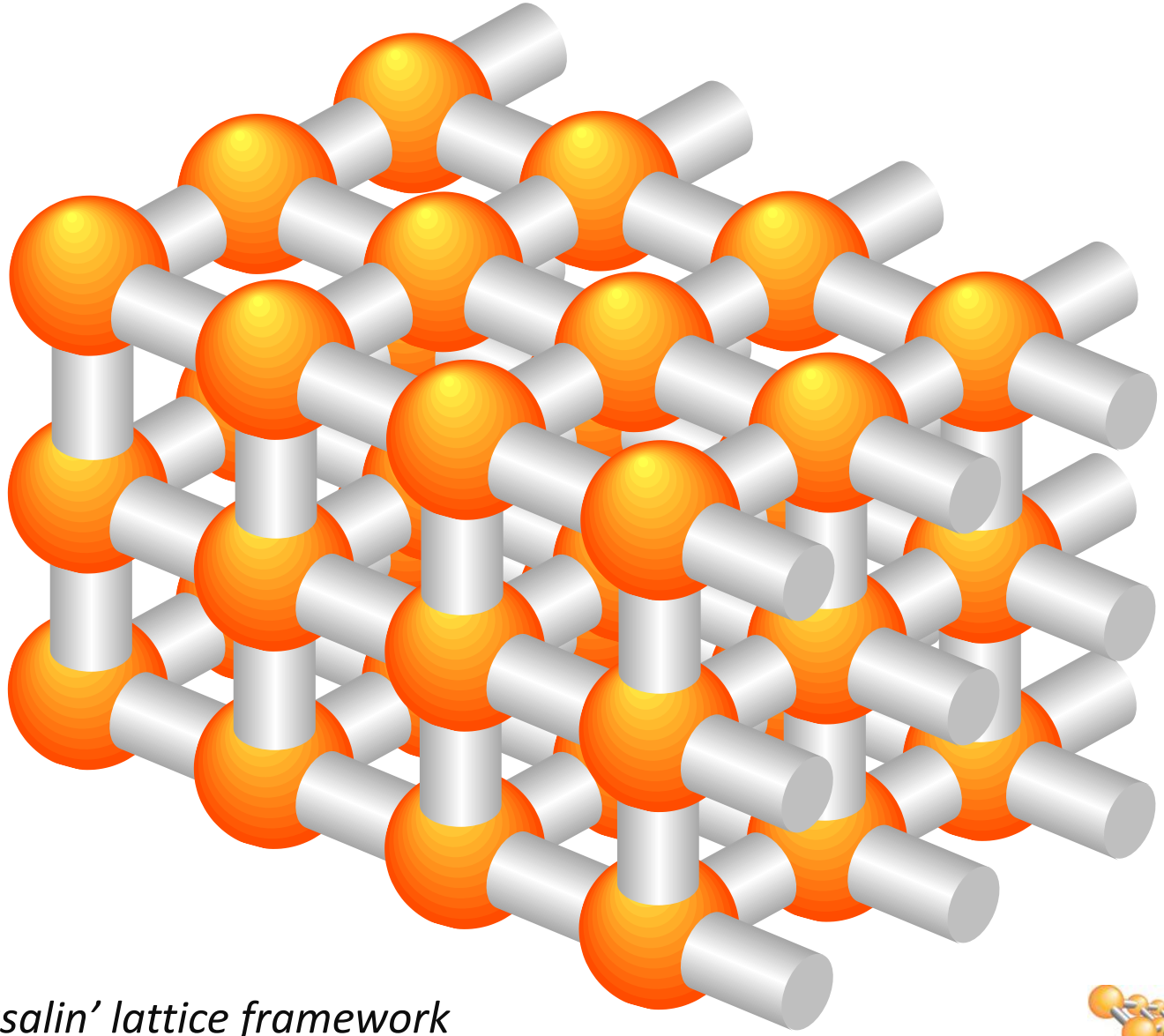
2D Crysalin lattice



Crysalin lattice manufacture sufficient to generate medium resolution structures (circa 12Å)*

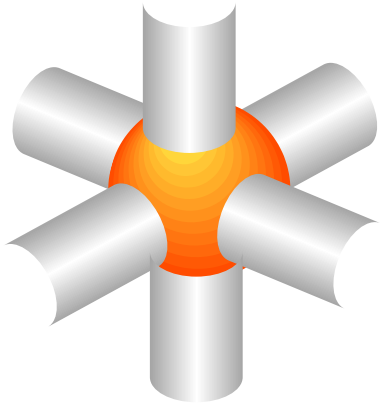
* *Generation of protein lattices by fusing proteins with matching rotational symmetry*, John C. Sinclair, Karen M. Davies, Catherine Vénien-Bryan & Martin E. M. Noble, Nature Nanotechnology 6, 558–562 (2011)

Technical concept 3D

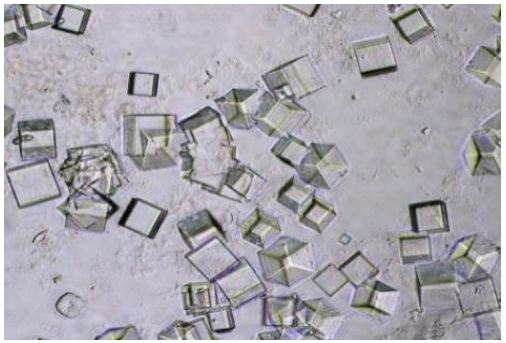


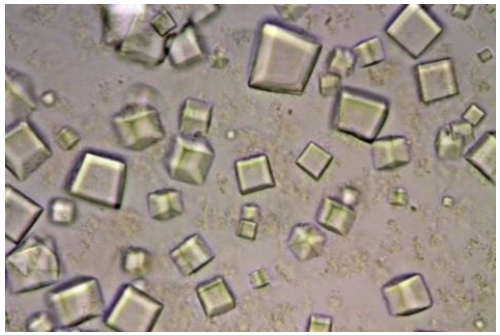
The 3D 'Crysalin' lattice framework

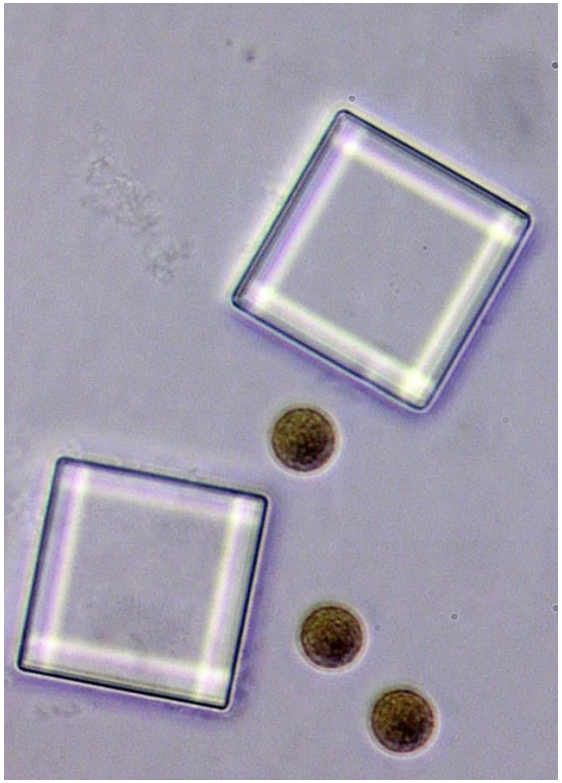
Crysalin formation



- Make the lattice components
- Crysalin lattice self assembles under the correct conditions
- Crysalins form as crystalline cubes varying in size from 20µm to 300µm
- Typical diffraction ranges of base lattice from 3.5-2.7Å (best diffraction to 2.5Å)

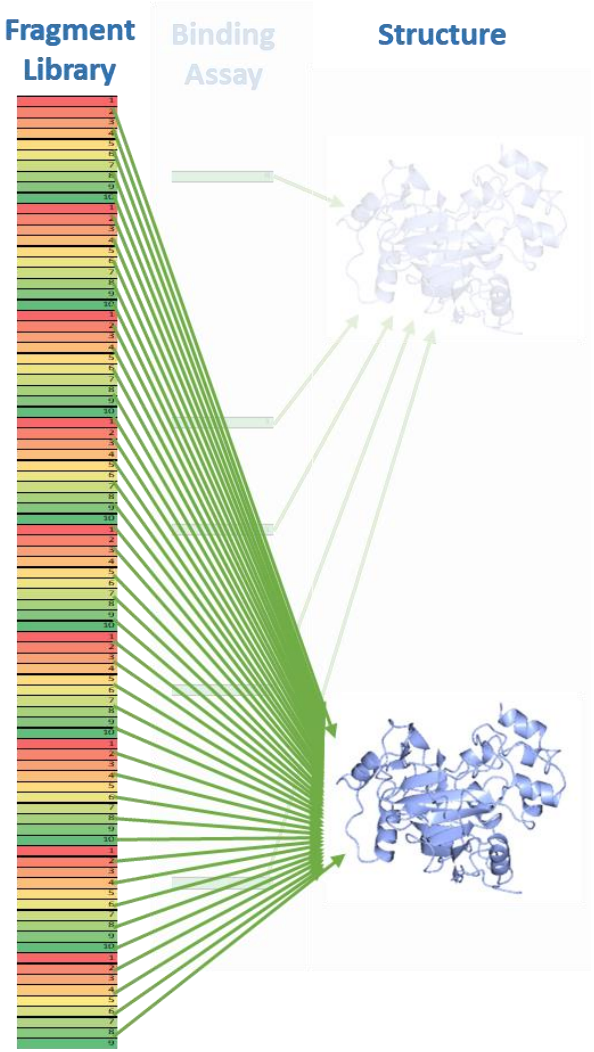






XChem Platform

Fragment Screening Platform - XChem



Standard practice

- Cascade of biophysical methods
  - SPR, NMR, MST…
- Require crystal structures

Via crystallography

- Very sensitive method
- High compound concentration
- Significant experimental overheads

XChem platform

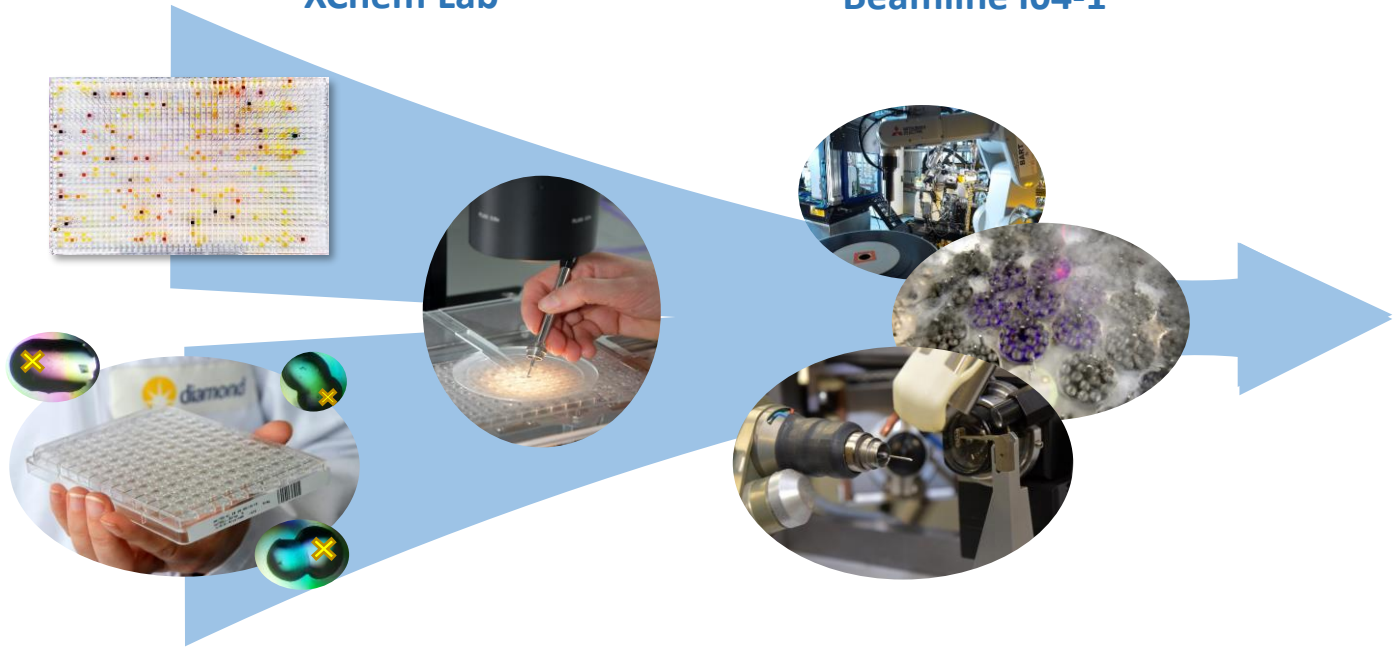
- >100 campaigns completed
  - 2-20% hit rate (project dependent)

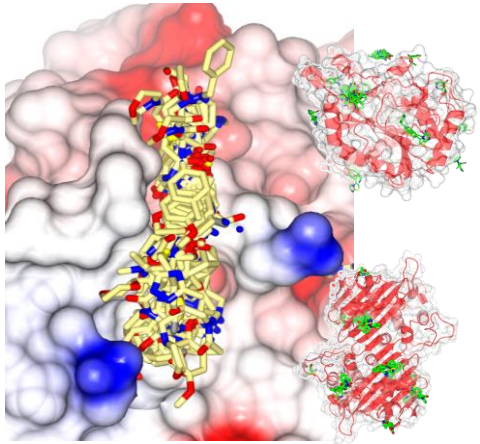
Overview of process

XChem Lab

Beamline I04-1

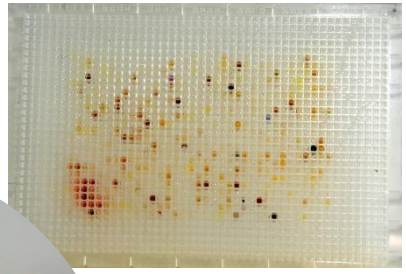
Example readout

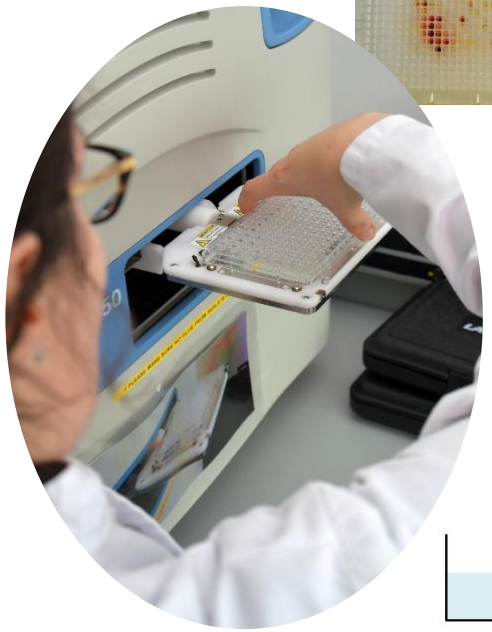


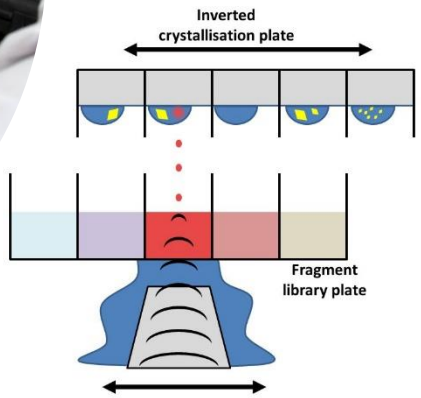


Libraries and Soaking

DSiP library in 1536 plate







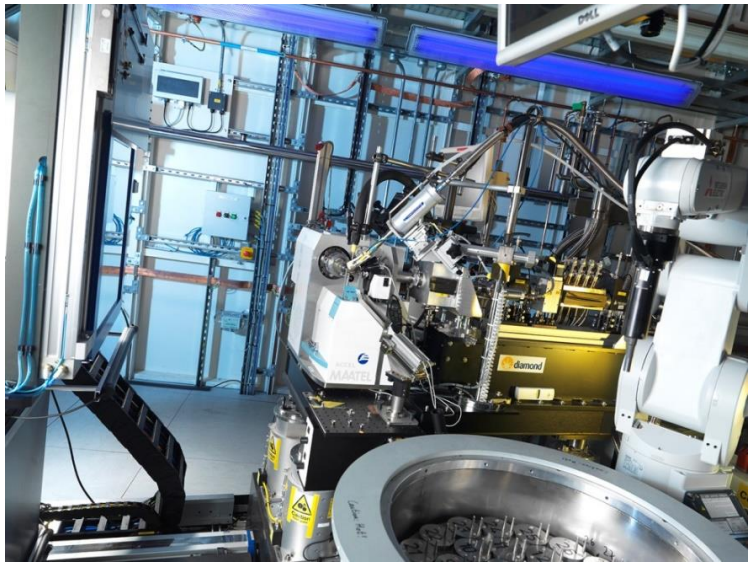
- Libraries
  - DSiP
    - ~950 compounds
    - 500mM in DMSO
  - External libraries
  - Cherry pick compounds
- Echo (Labcyte)
  - ~10min to transfer our DSIP library

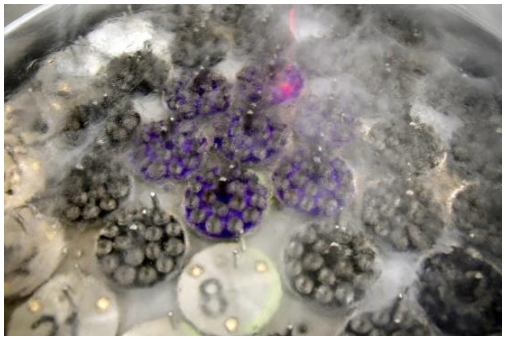
Crystal Harvesting

Crystal SHIFTER, commercialised by OLT

- Crystal SHIFTER
  - Motorised X&Y stage under a microscope
  - Allows for rapid crystal harvesting
  - Touch screen graphical interface
    - Record events (time stamps)
      - Crystal mounted
      - Crystal status
      - Compound status
  - Linked to SoakDB
    - XChem data management system

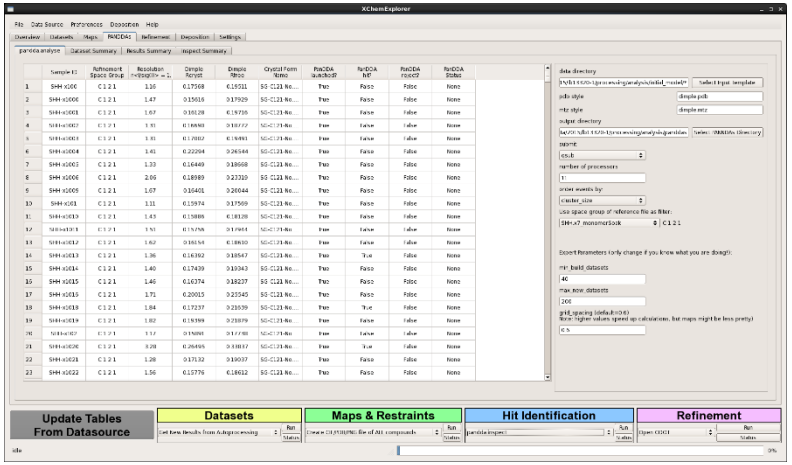
Fully Automated X-ray Data Collection



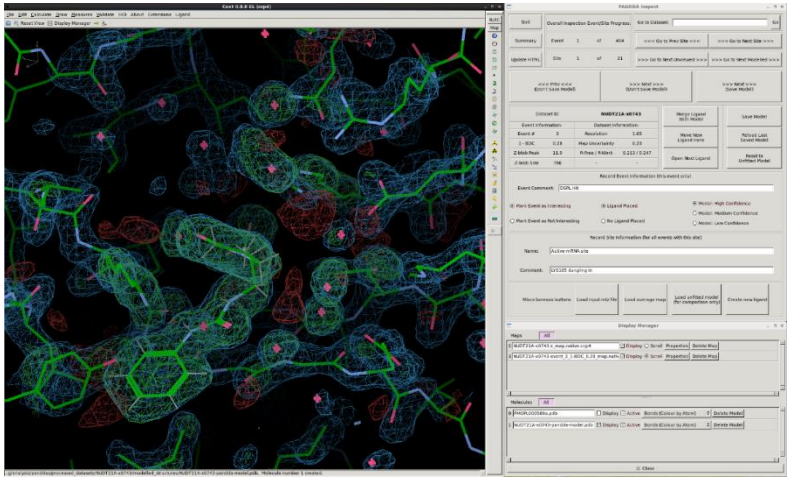


- I04-1 MX beamline
  - Automated loop centring
    - Optical or X-ray
  - Bart robot
    - Dewar capacity ~600 samples
  - Unattended data collection
  - Process up to 32 samples per hour
  - Eiger detector
    - 15s or 60s data collection
  - Auto-processing pipelines
    - Xia2, Dials and AutoProc

Data Analysis: XChemExplorer & PanDDA



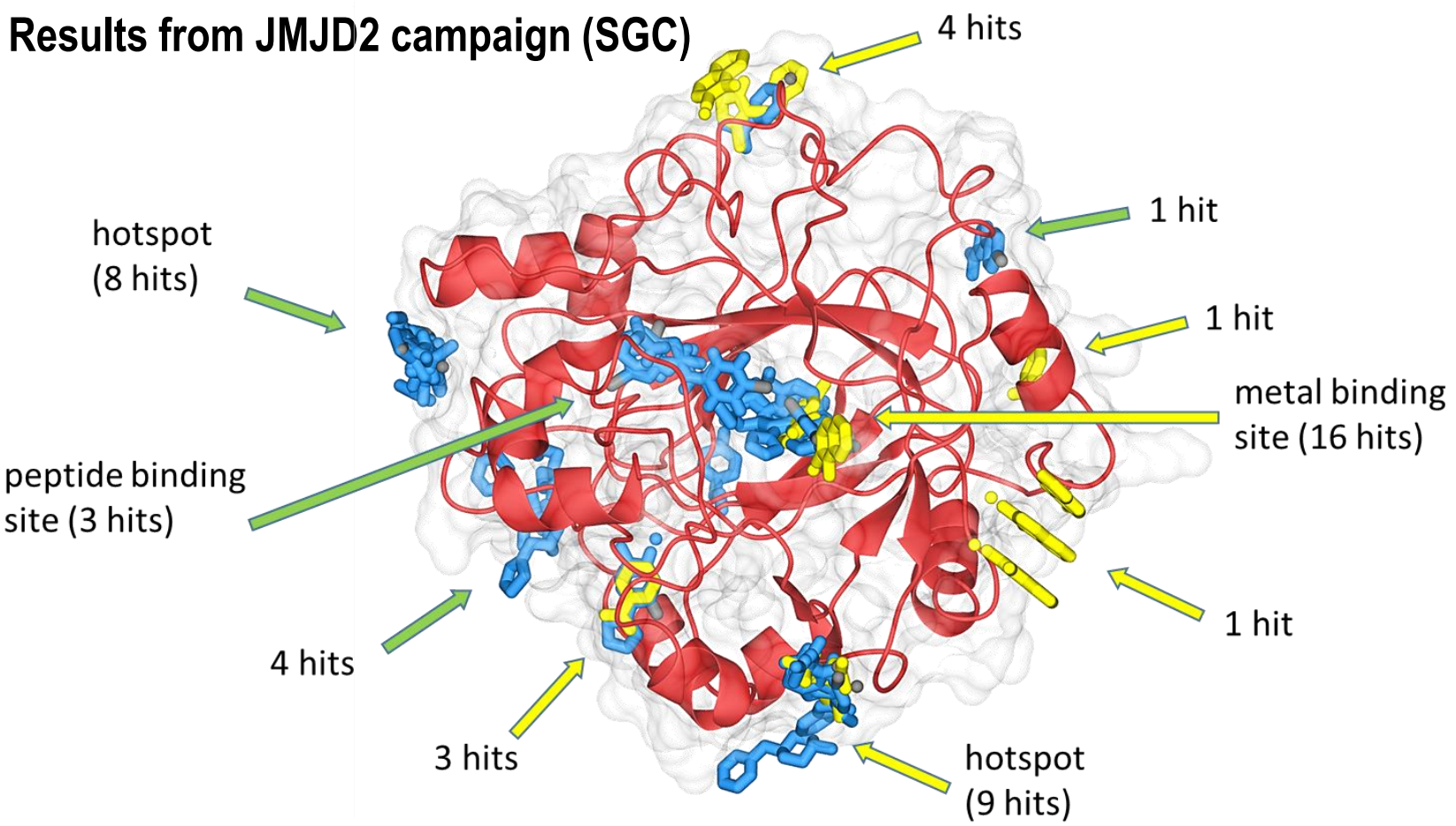
Krojer *et al*, 2017, ActaD



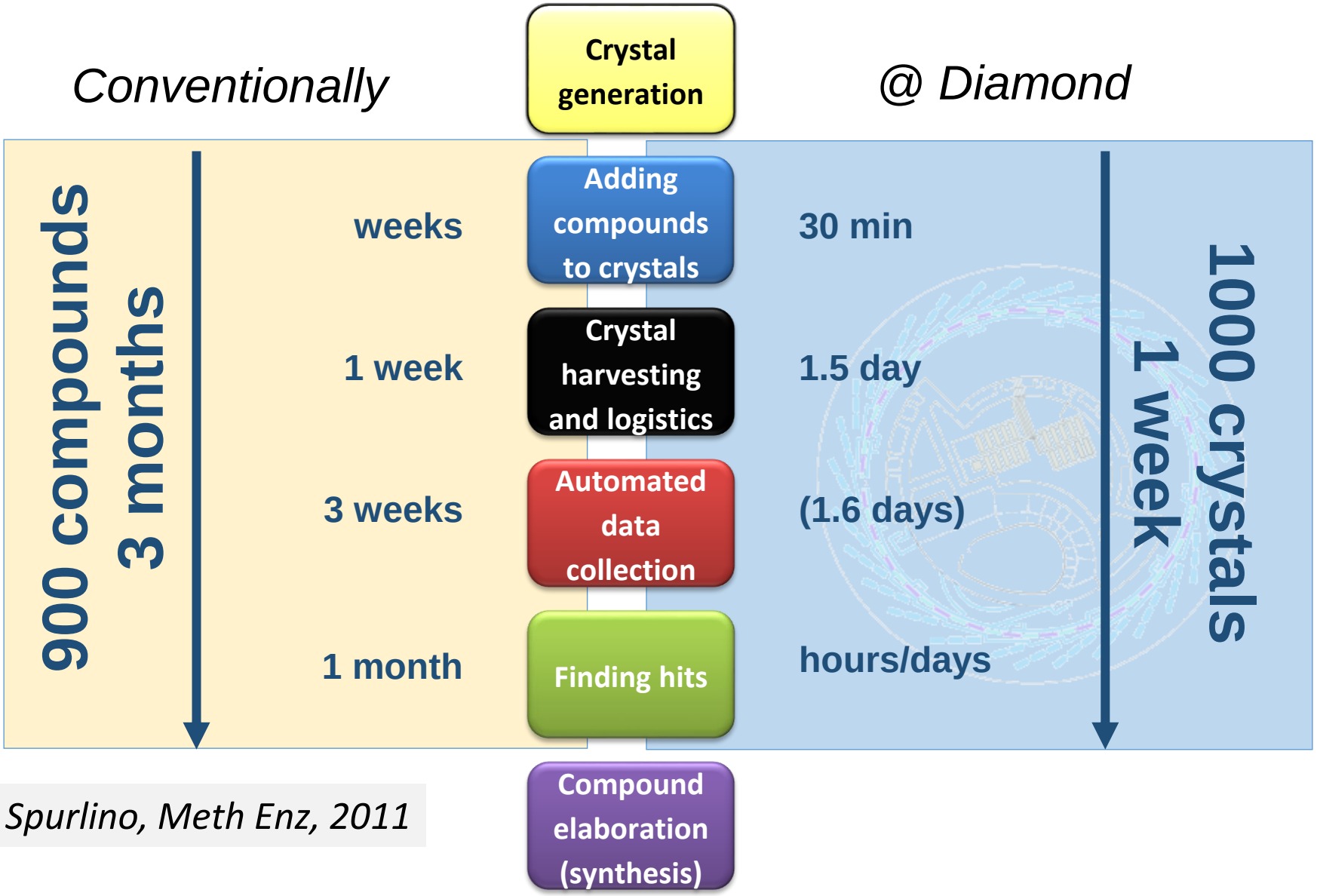
PanDDA. Pearce et al, 2017, Nat. Comm.

- XChemExplorer
  - Record all information
    - Same SQLite database as SoakDB
  - Rapid review of data collection
    - Selection of processed files
  - Generation of maps
    - Run Dimple on user's PDB model
  - PanDDA user interface
    - Pre-PanDDA
      - Generate accurate PDB model of "background state"
    - Coot Plugin
      - Facilitate ligand fitting
      - Quickly browse through events and record comments

Fragment Screening Platform - XChem

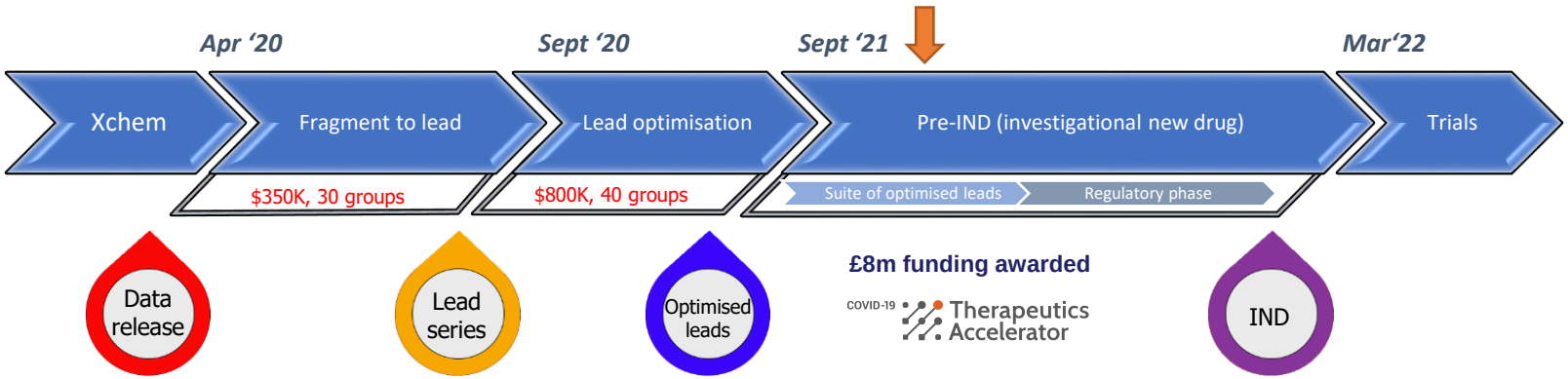


Order of magnitude speedup



Spurlino, Meth Enz, 2011

Fragment Screening for COVID-19



<18 months:

- 4 lead series
- ~500 crystal structures on fragalysis
- Progressed from >200 μM hits to <50 nM inhibitors
- Philanthropic funding or in-kind contributions

Current status:

- Potent despite simplicity (<20nM)
- Strong antiviral inhibition (<100nM)
- Inhibits new strains
- Highly encouraging safety prediction

Optimising:

- Oral exposure (blood levels)
- Metabolic stability

Imminent:

- Efficacy in mice

Future:

Establishing:

- Strategy to pharmacy
- Pre-IND delivery
- Keystone partner
- Delivery partners
- Healthcare system
- Funding sources

XCHEM Diamond Fragment Screening

COVID Moonshot

How is the platform used by industry?

- Trained on the platform and use it with support in place
- Send projects for “full service” access
  - Project run by members of Diamond’s ILO team
  - This often includes data analysis

- Companies run multiple campaigns per year
- Incorporating into their general screening protocols
- Frequently have follow up compounds sent for testing

- Over subscribed – often running 12-14 campaigns at different stages at any one time

- Increasing enquires from virtual and AI companies

Working in MX for industry

- Rapid turnover of data
- Different approach to data collection
- Quick analysis of data to report to client/manager/board
- Steer the scientific direction
- Can be working on a well defined system, but also developing that system
- Expectation of success
- If a project isn't working, will be stopped
- Interactions across teams
- Rapid problem solving
- Working on multiple projects
- Development of transferable skills, eg.
  - Management
  - Business development

Questions?