MX from an Industrial Perspective

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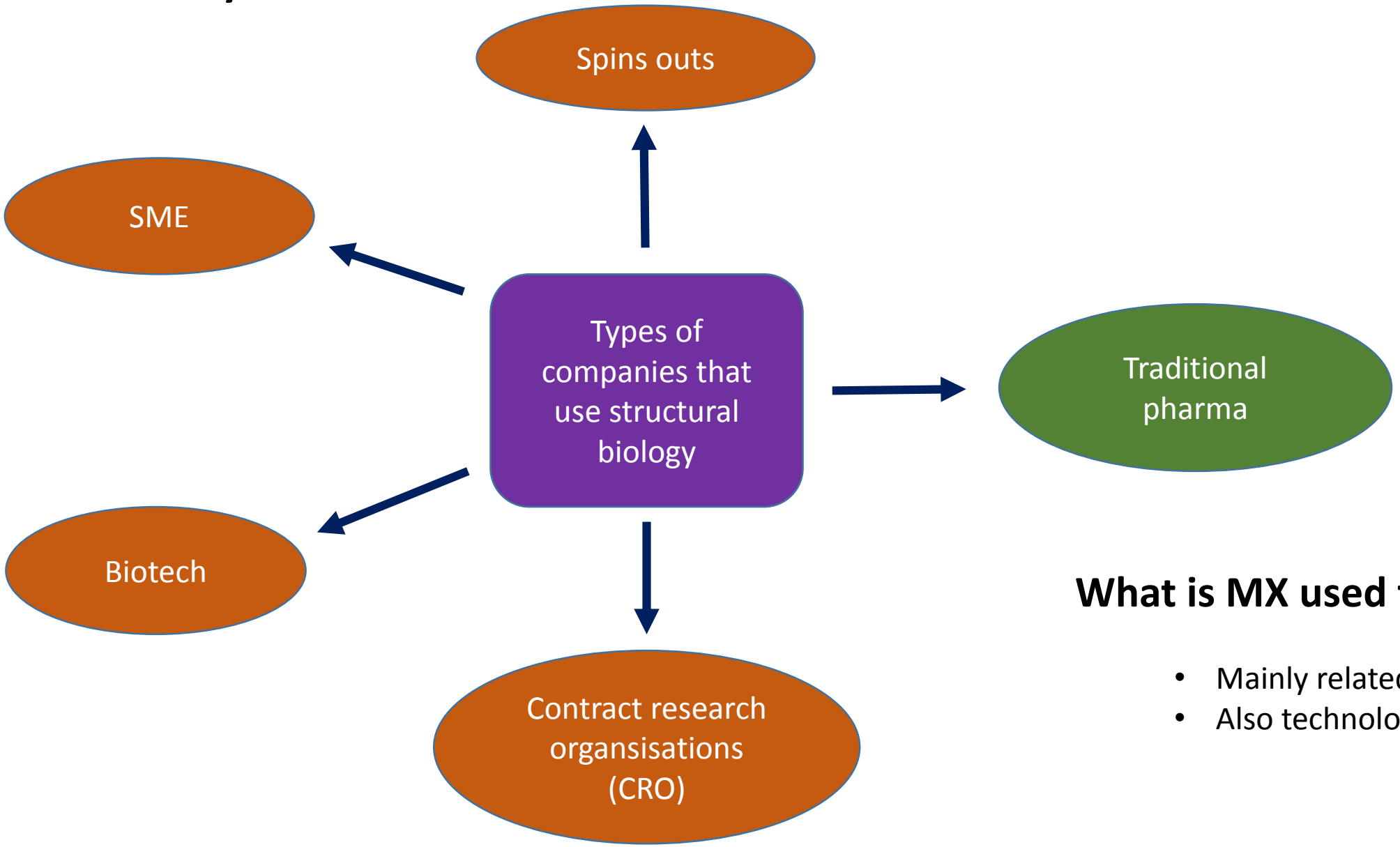
Is all industry the same?

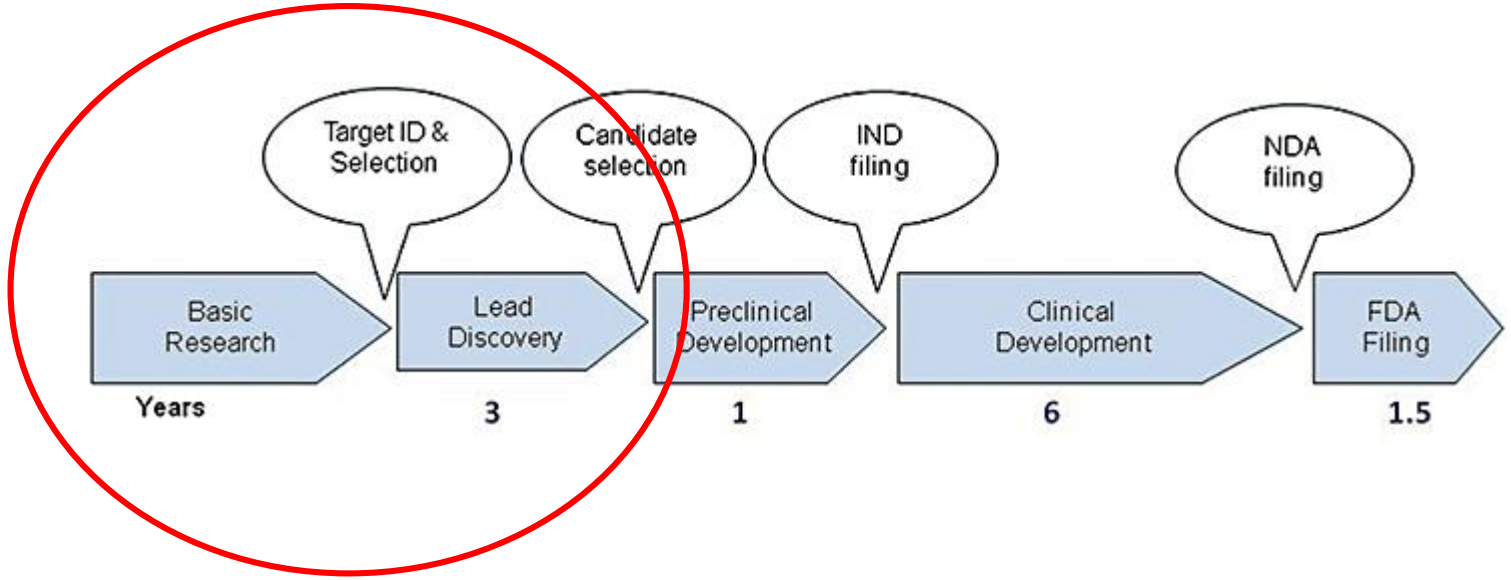


What is MX used for in industry?

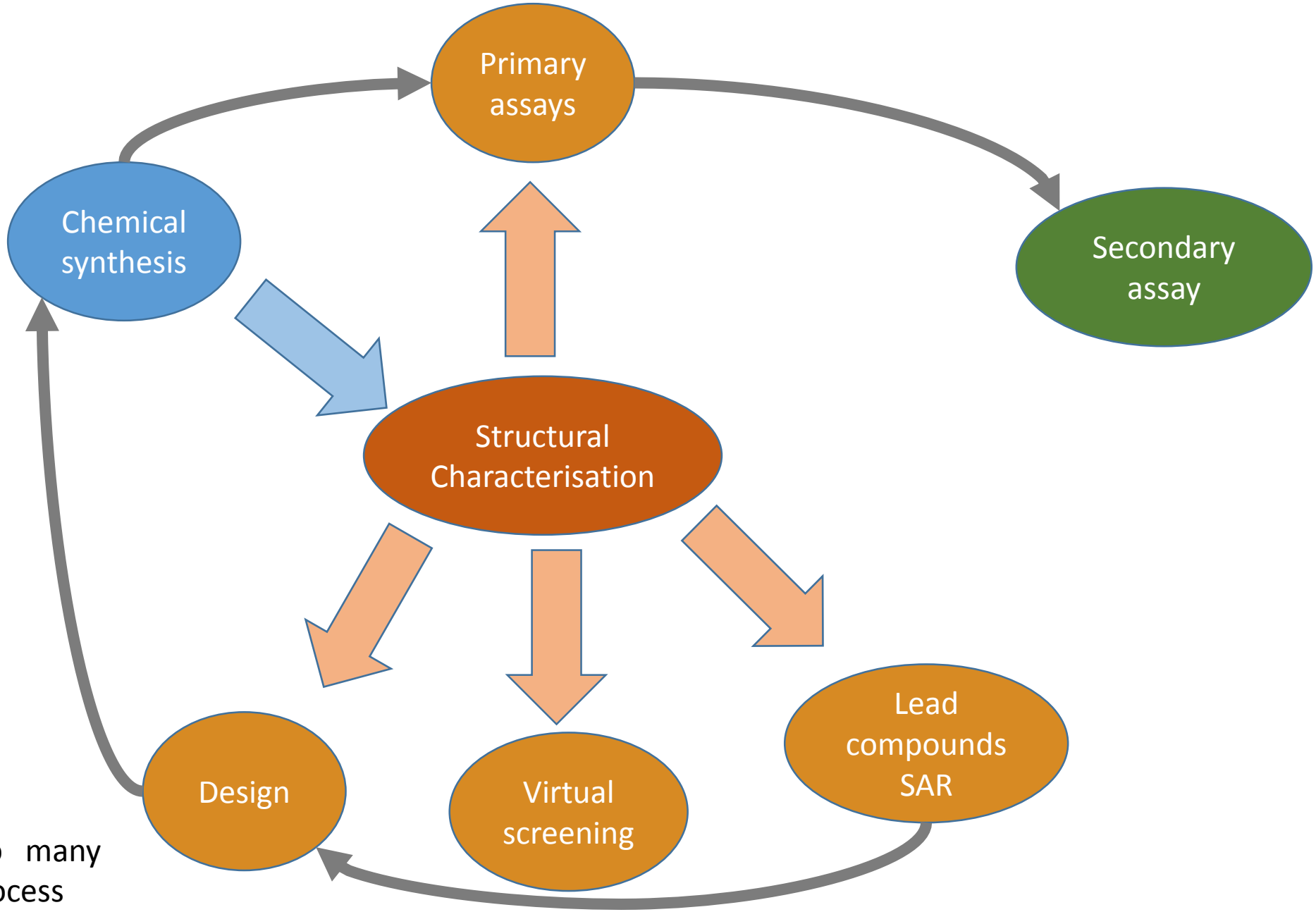
- Mainly related to drug discovery
- Also technology development

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)



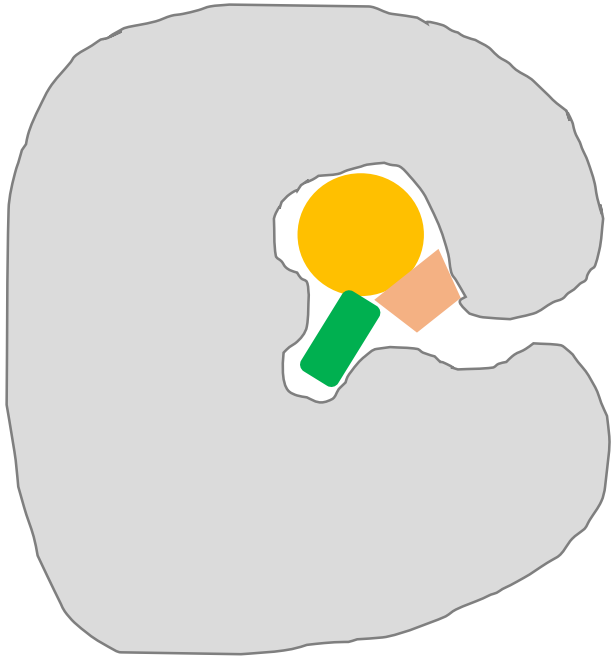
Structure is integral to many steps of the discovery process

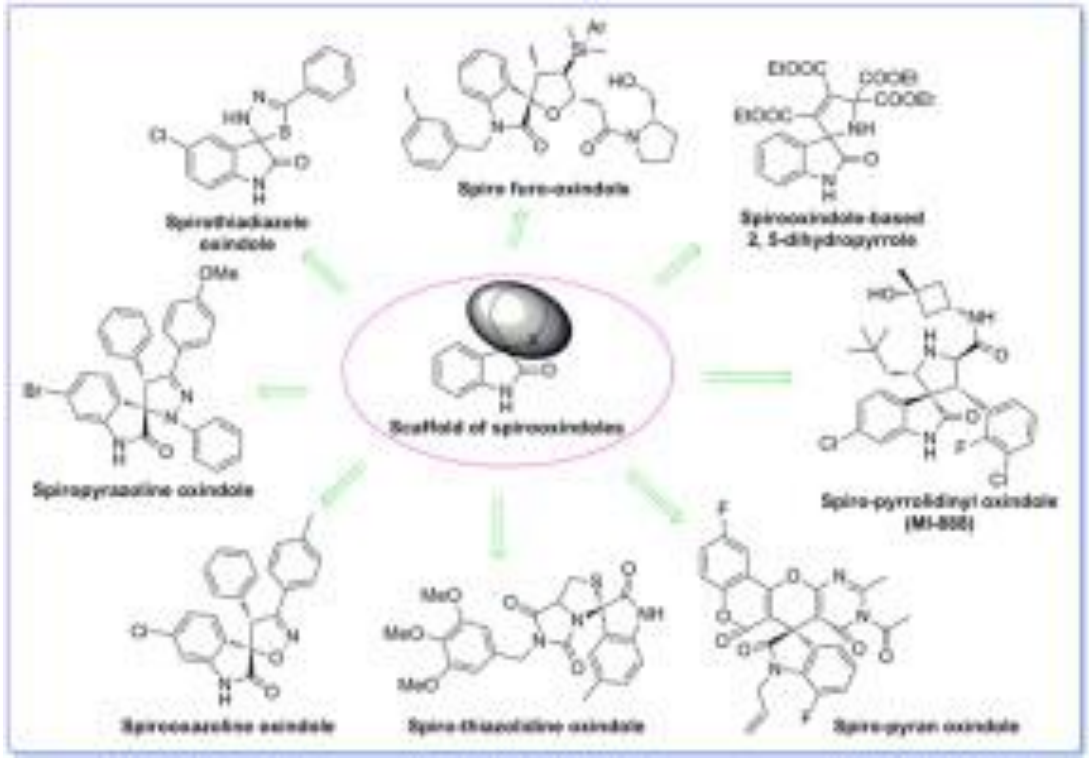
How structures contribute to screening

Structural aided drug design	• Use of crystal structures to help design molecules	• Have docked a compound into the crystal and use this 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	• Join selected fragments together to fit into the chemical space to increase potency • Requires a crystal structure to be available

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

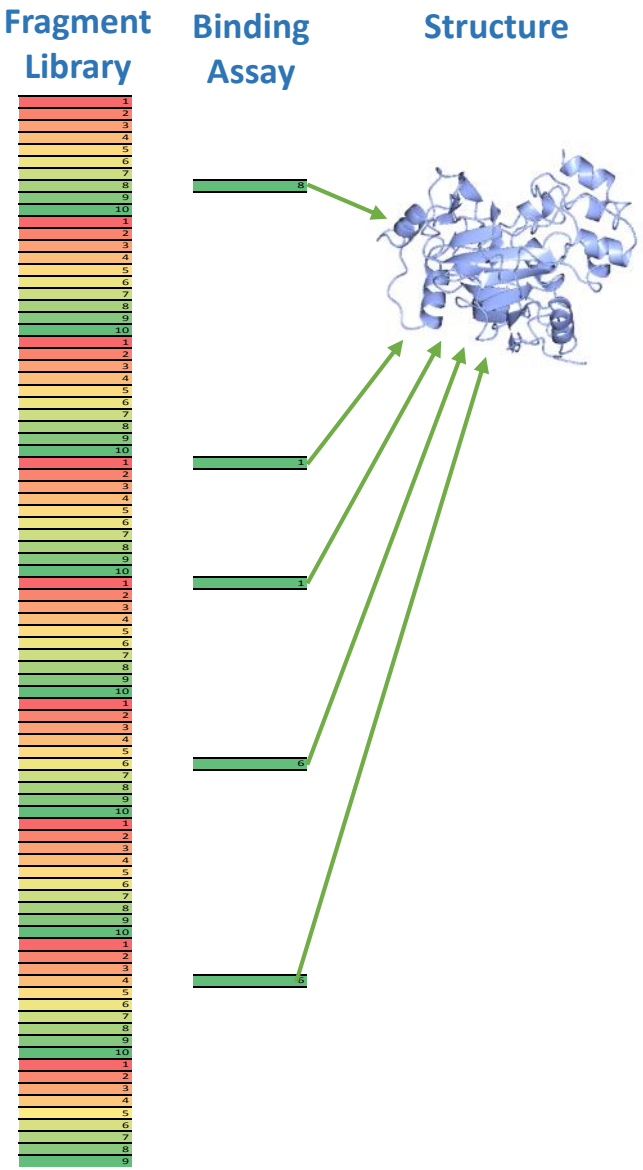




Spirooxindoles: Promising scaffolds for anticancer agents

European Journal of Medicinal Chemistry. Volume 97, 5 June 2015, Pages 673-698

Screening for fragments: conventional

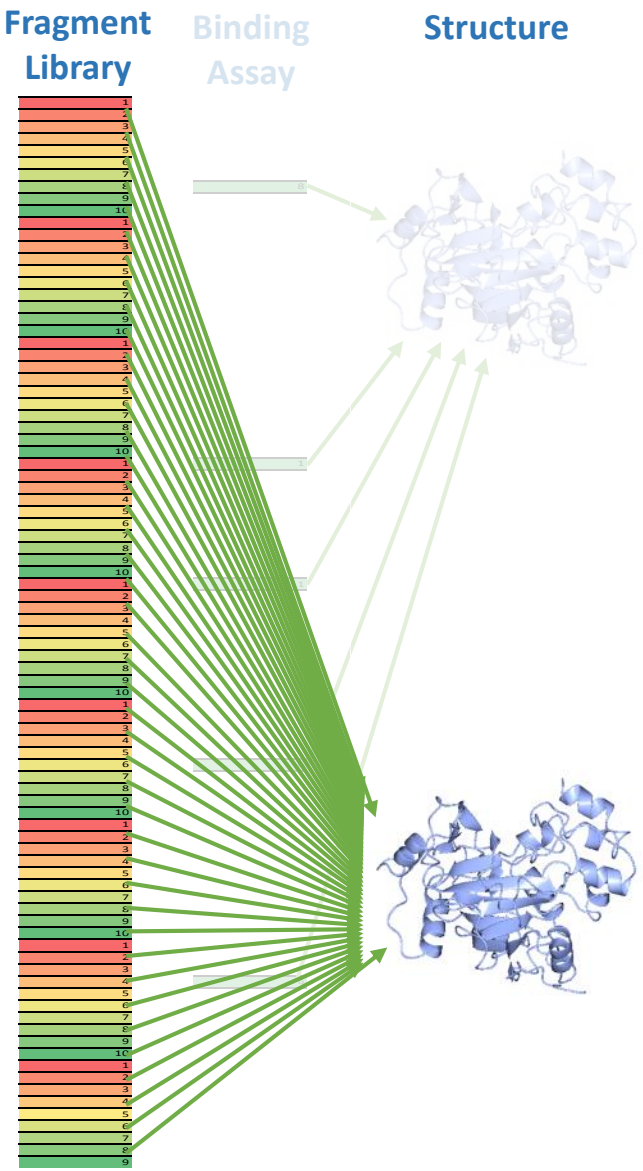


Current practice: pre-select by biophysics

Limited sensitivity!

Still need crystal structure

Screening for fragments: XChem



Current practice: pre-select by biophysics

Limited sensitivity!

Still need crystal structure

Better: screen by crystal structure

Very sensitive! High compound conc.

Worse: far too difficult

100s of crystal structures

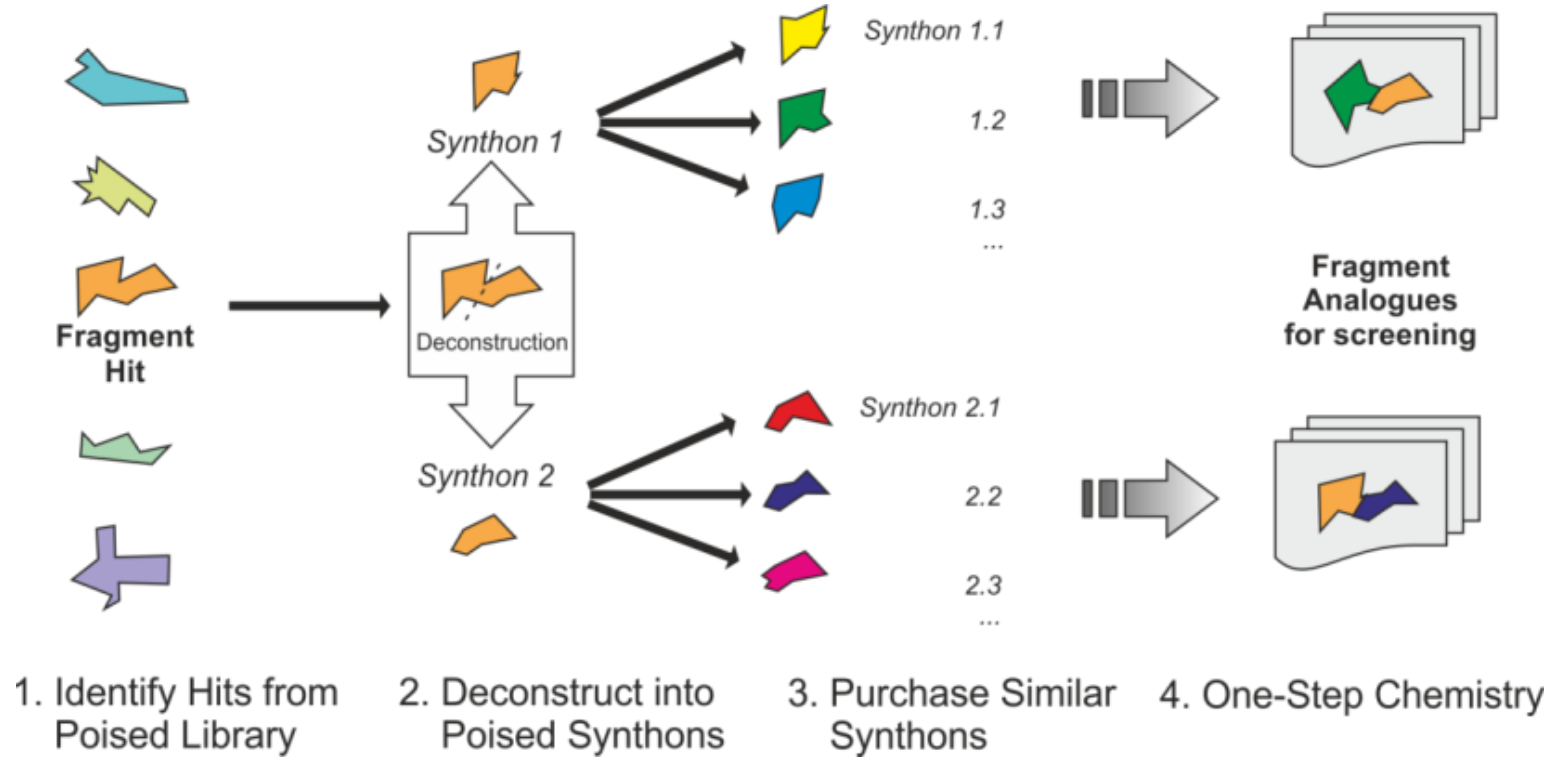
Meanwhile, beamlines got much faster…

Job for a facility

Make it fast

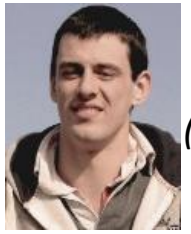
and *accessible*

Poised fragments: cheap single-step expansion



Poised reactions are:

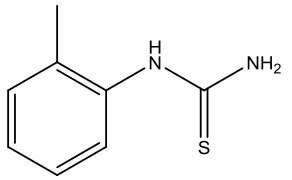
- robust and reliable;
- make drug-like products;
- be possible using commercially available starting points;
- compatible with a range of substrates.



Oakley Cox
(Paul Brennan)

Proof of concept: tough target

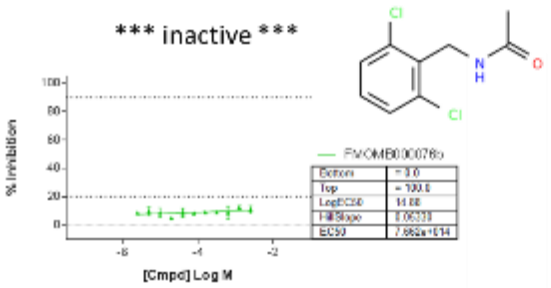
- PHIP 2nd bromodomain
- No hits from biophysics (~10k)
- 9 hits from 1000 soaks – 2 poised



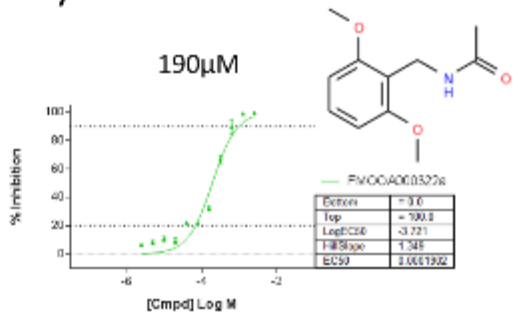
Original fragment hit

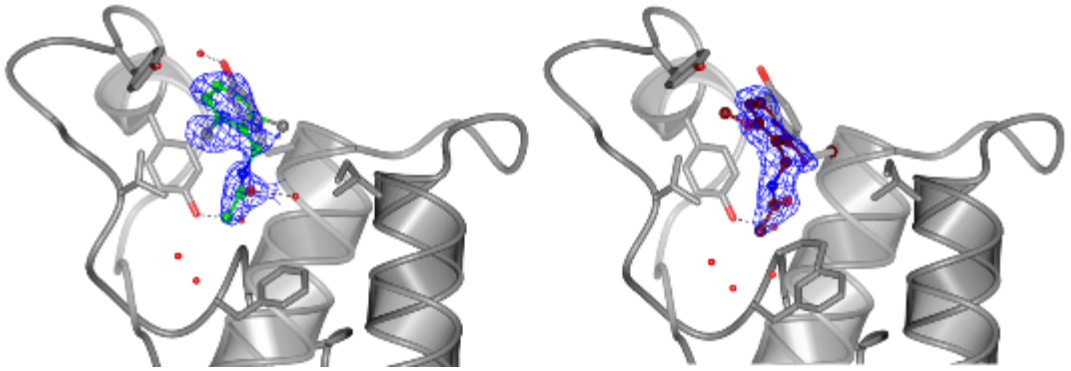
ID	IC_{50} (µM)	LE
FMOOA463	68	0.45
FMOOA365	128	0.45
FMOOA473	128	0.29
FMOOA315	140	0.36
FMOOA314	143	0.45
FMOOA470	194	0.35
FMOOA312	256	0.42
FMOOA476	276	0.45
FMOOA475	294	0.41
FMOOA471	295	0.35
FMOOA474	347	0.24
FMOOA316	414	0.39
FMOOA472	415	0.47
FMOOA465	423	0.34
FMOOA310	482	0.39
FMOOA466	536	0.31
FMOOA313	565	0.32
FMOOA468	566	0.38
FMOOA309	747	0.36
XST00942	768	0.4
FMOOA462	791	0.31
FMOOA477	1021	0.28
FMOOA308	1200	0.34
FMOOA467	1291	0.34
FMOOA363	1500	0.32
FMOOA357	1700	0.35
FMOOA321	1900	0.35
FMOOA369	2100	0.29
FMOOA319	2100	0.27
FMOOA358	2200	0.29
FMOOA362	2600	0.31
FMOOA367	2600	0.26
FMOOA311	3200	0.29
FMOOA359	3200	0.29
FMOOA361	4800	0.27
FMOOA360	>5000	<0.29
FMOOA461	>5000	<0.29
FMOOA460	>5000	<0.27
FMCAC421	>5000	<0.25
FMOOA368	>5000	<0.25
FMOOA364	>5000	<0.25
FMOOA318	>5000	<0.25
FMOOA317	>5000	<0.25
FMOOA366	>5000	<0.23
FMOOA320	>5000	<0.23
FMOOA469	>5000	<0.23
FMOOA464	>5000	<0.21

Original hit



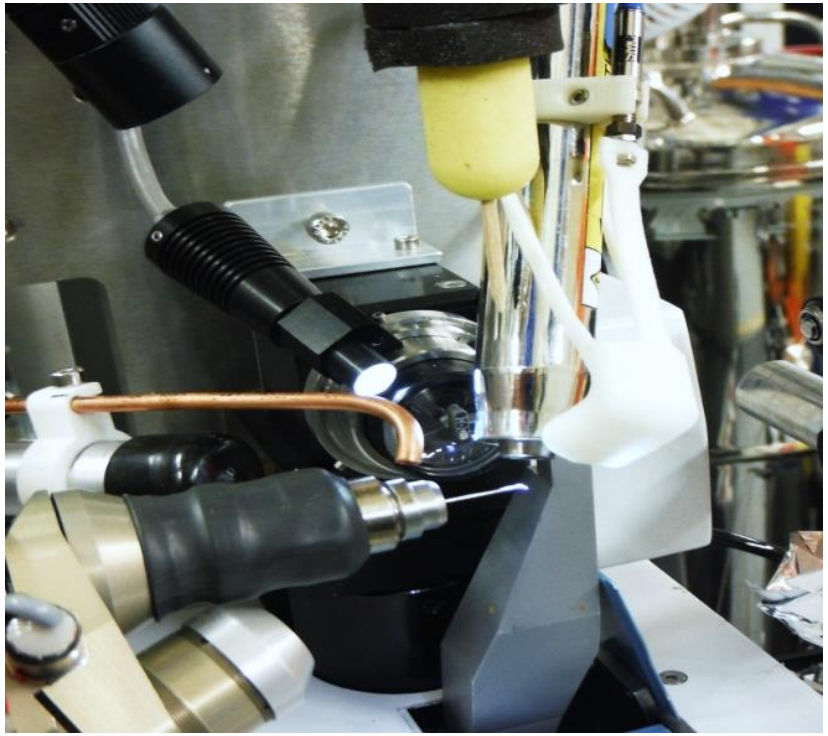
Synthesis round 1





Tobias Krojer
Oakley Cox
Anthony Bradley
Lin Mei

Automated and unattended data collection



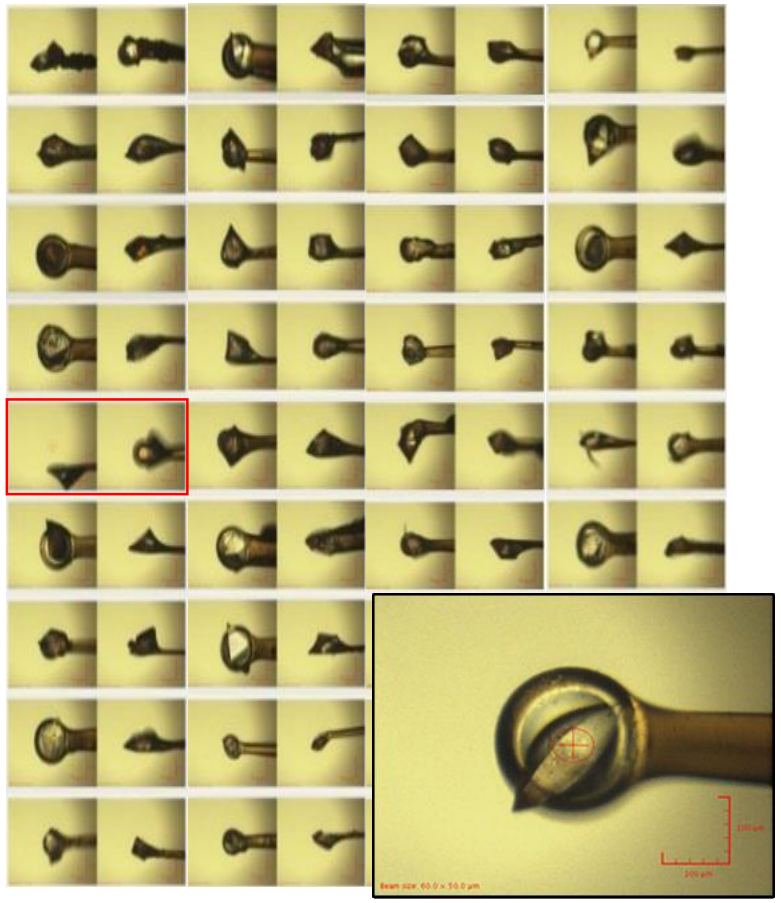
Stable reliable beamline

Automated loop centring (~30 samples /hour)

José Brandaõ-Neto

Alice Douangamath

Mark Williams

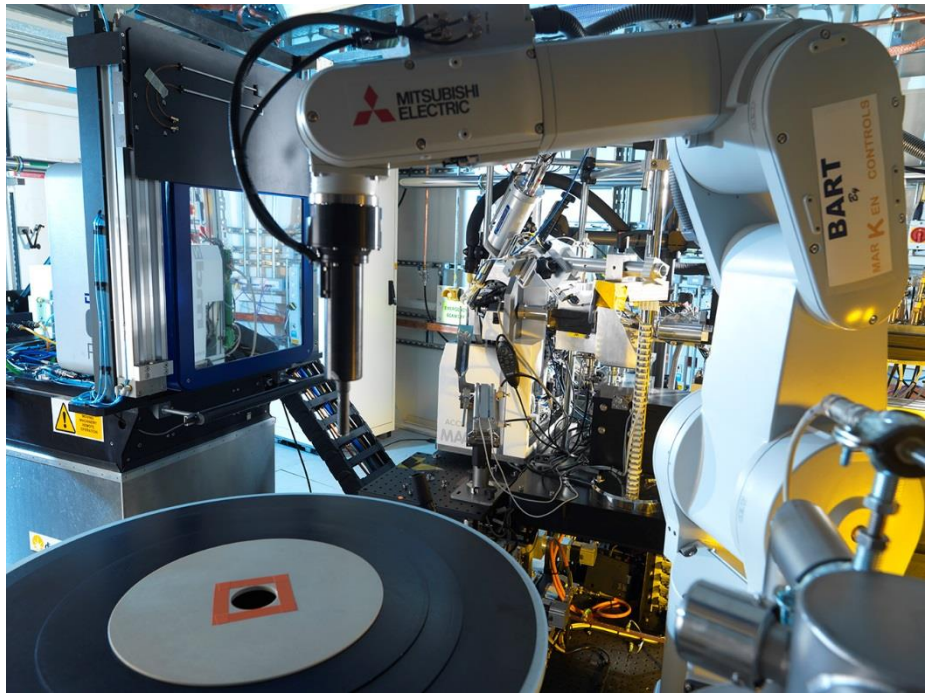


Automatic loop centring, > 97% accuracy

BART robot (I03 team) - transformative

In-house control system:

- **VERY robust**
- Fast: <20sec exchange



37 pucks, 592 samples

Ken Jones

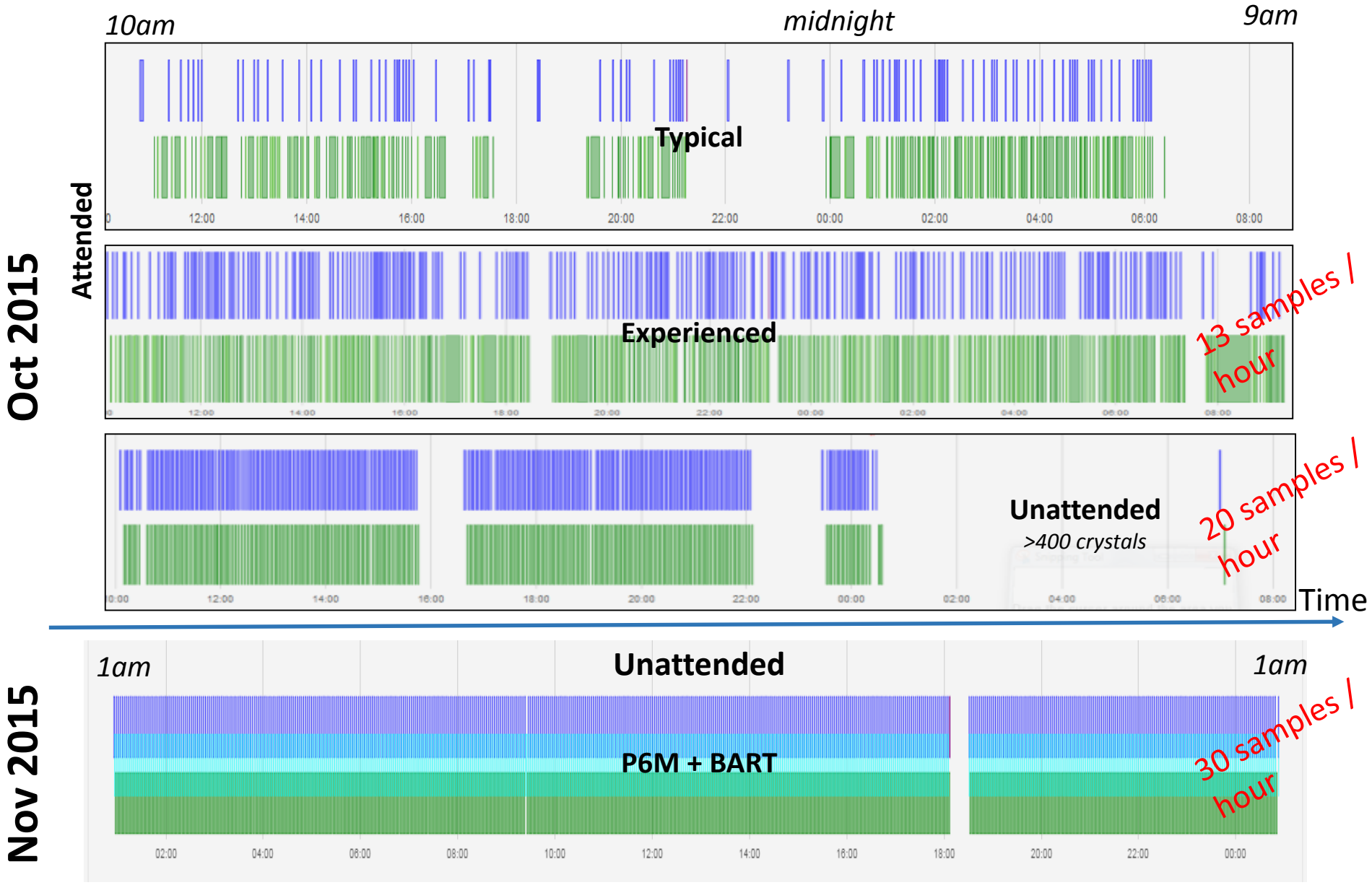
James O'Hea

Stuart Fisher

Mark Williams

Katherine McAuley

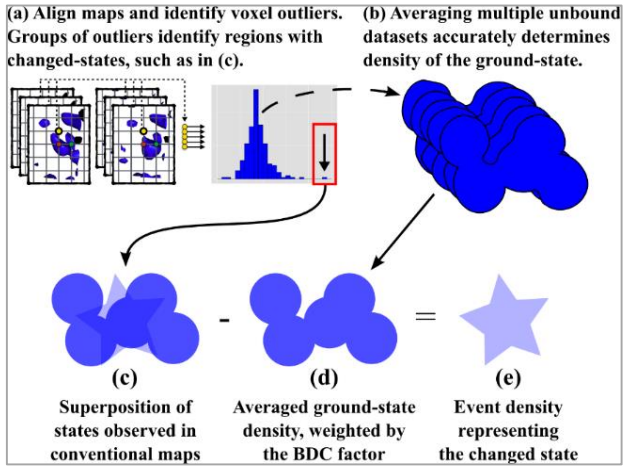
A truly automated beamline

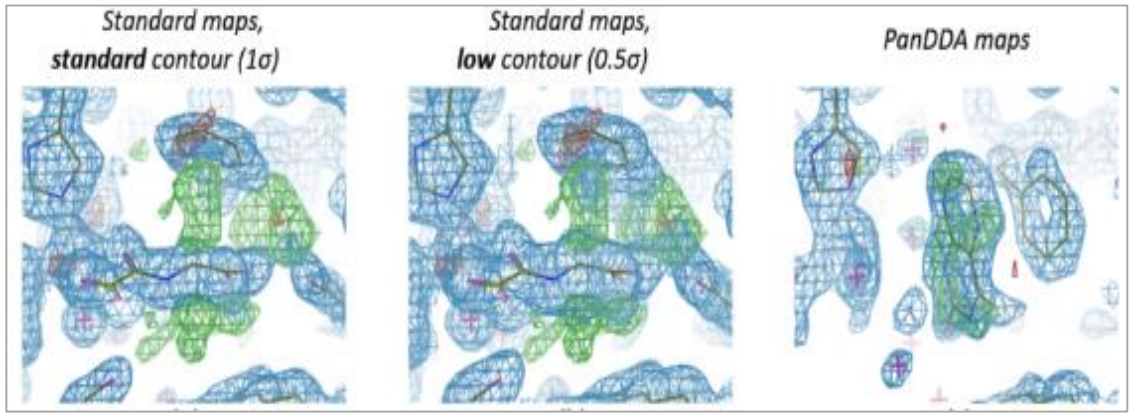


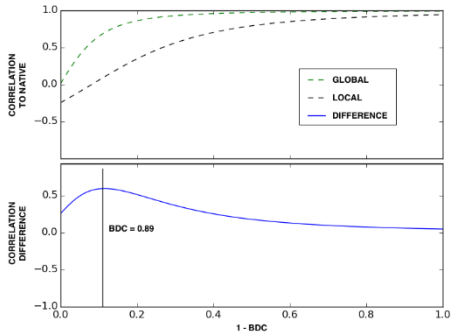
Finding hits – PanDDA

- Used through the XChem explorer interface

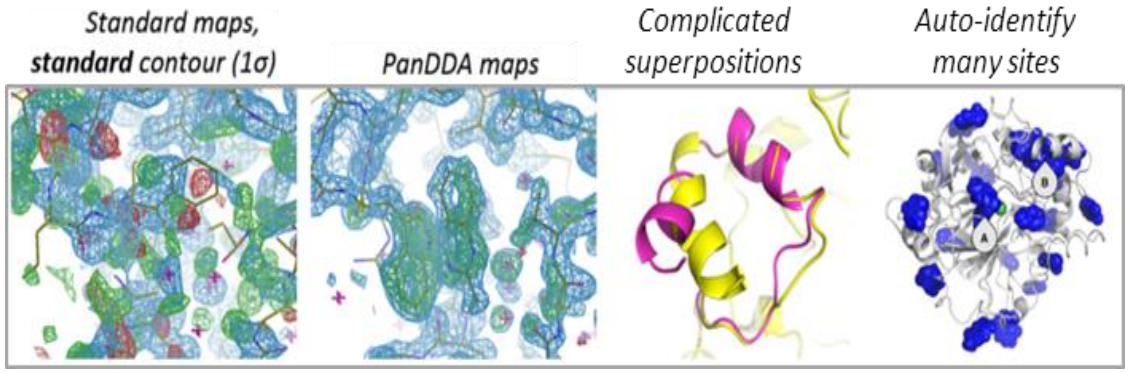
Pan-Dataset Density Analysis



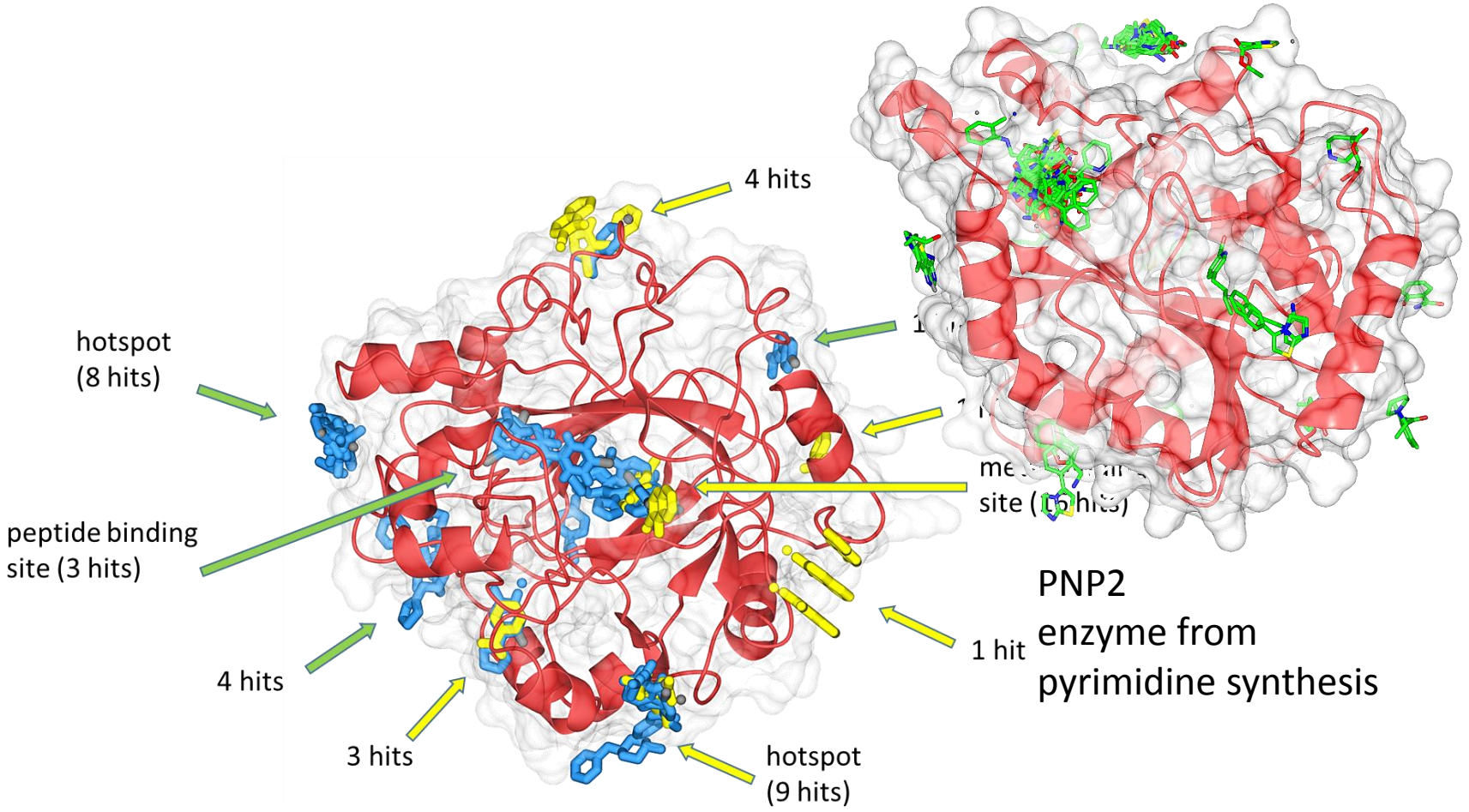




Pearce et al, Nat Comms, 2017

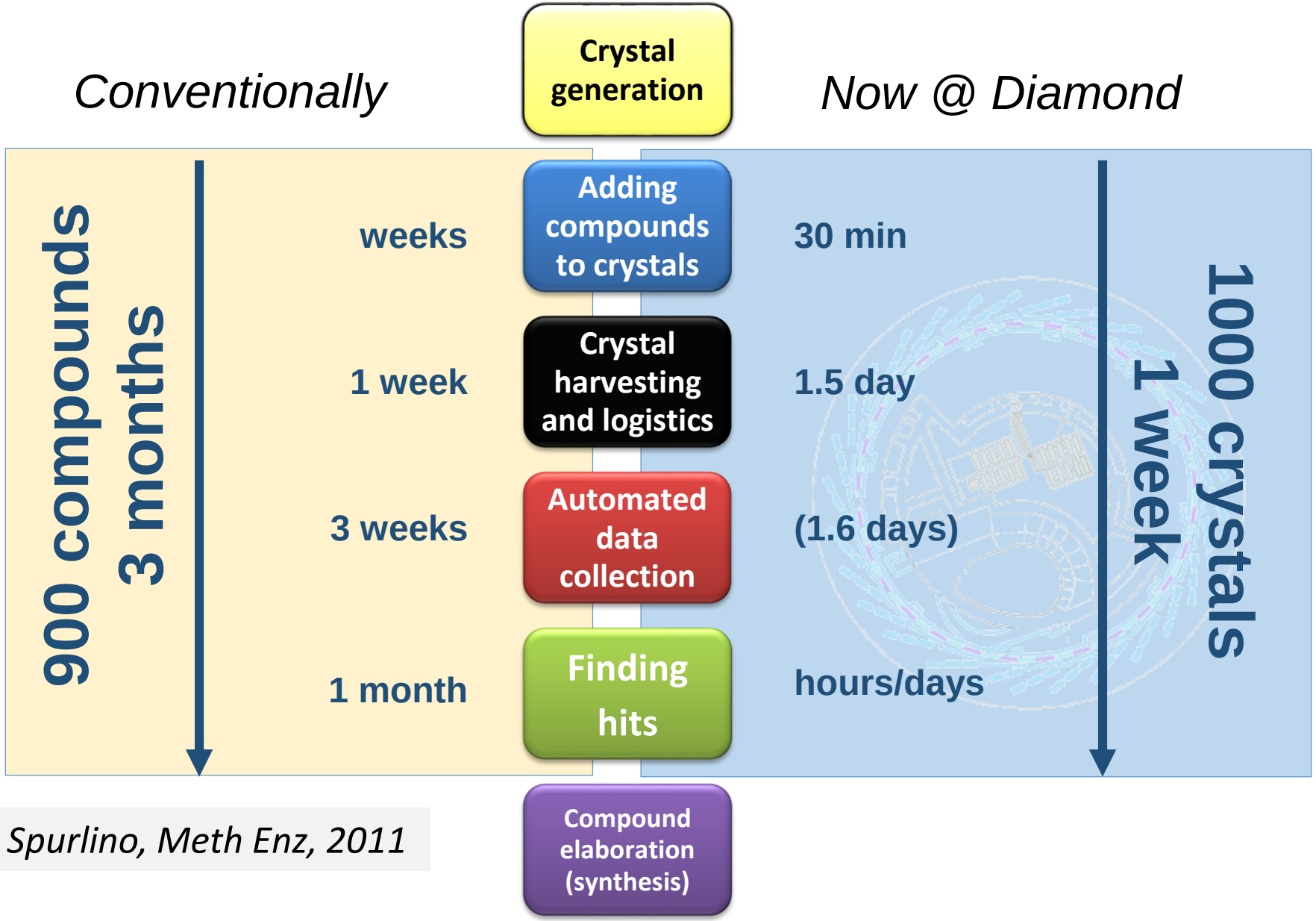


Fragment binding results



JMJD2D, a histone lysine demethylase

XChem: order of magnitude speedup



Spurlino, Meth Enz, 2011

CRYSALIN LIMITED

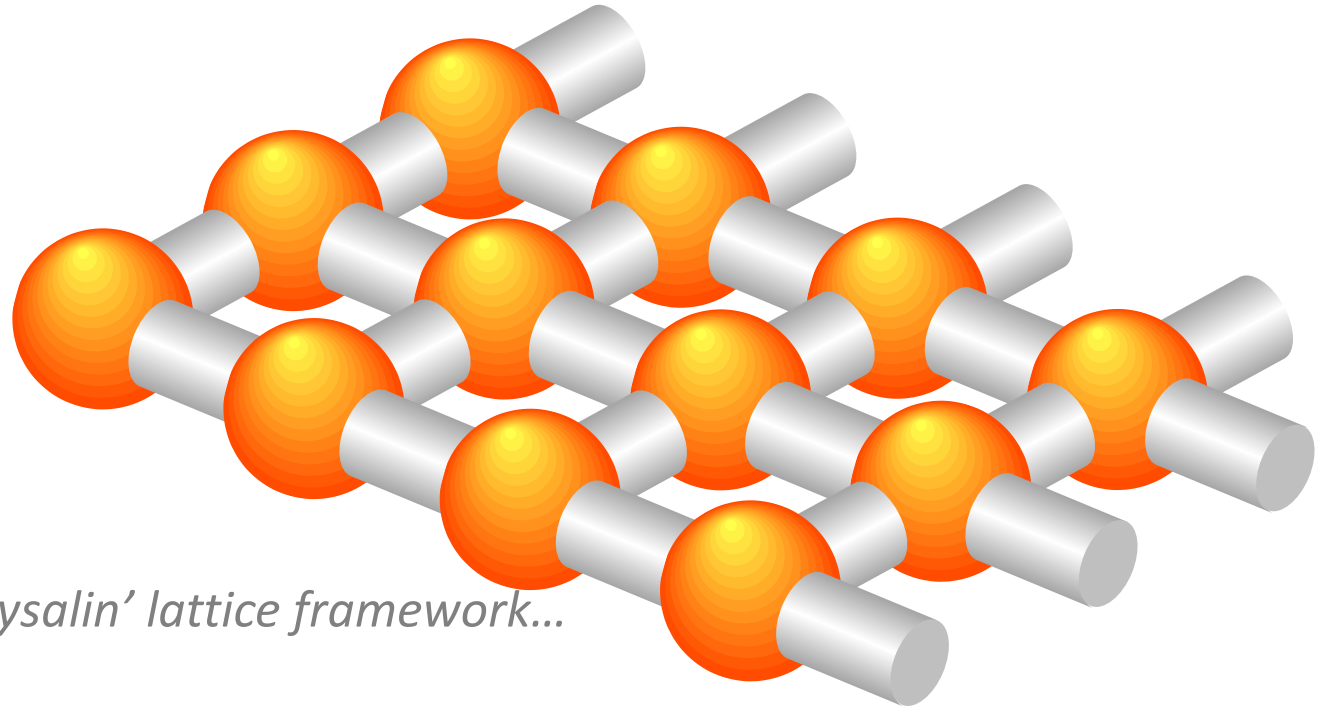
Aim: to provide rapid determination of protein structures, irrespective of target class, at sub 3Å resolution.

- 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…

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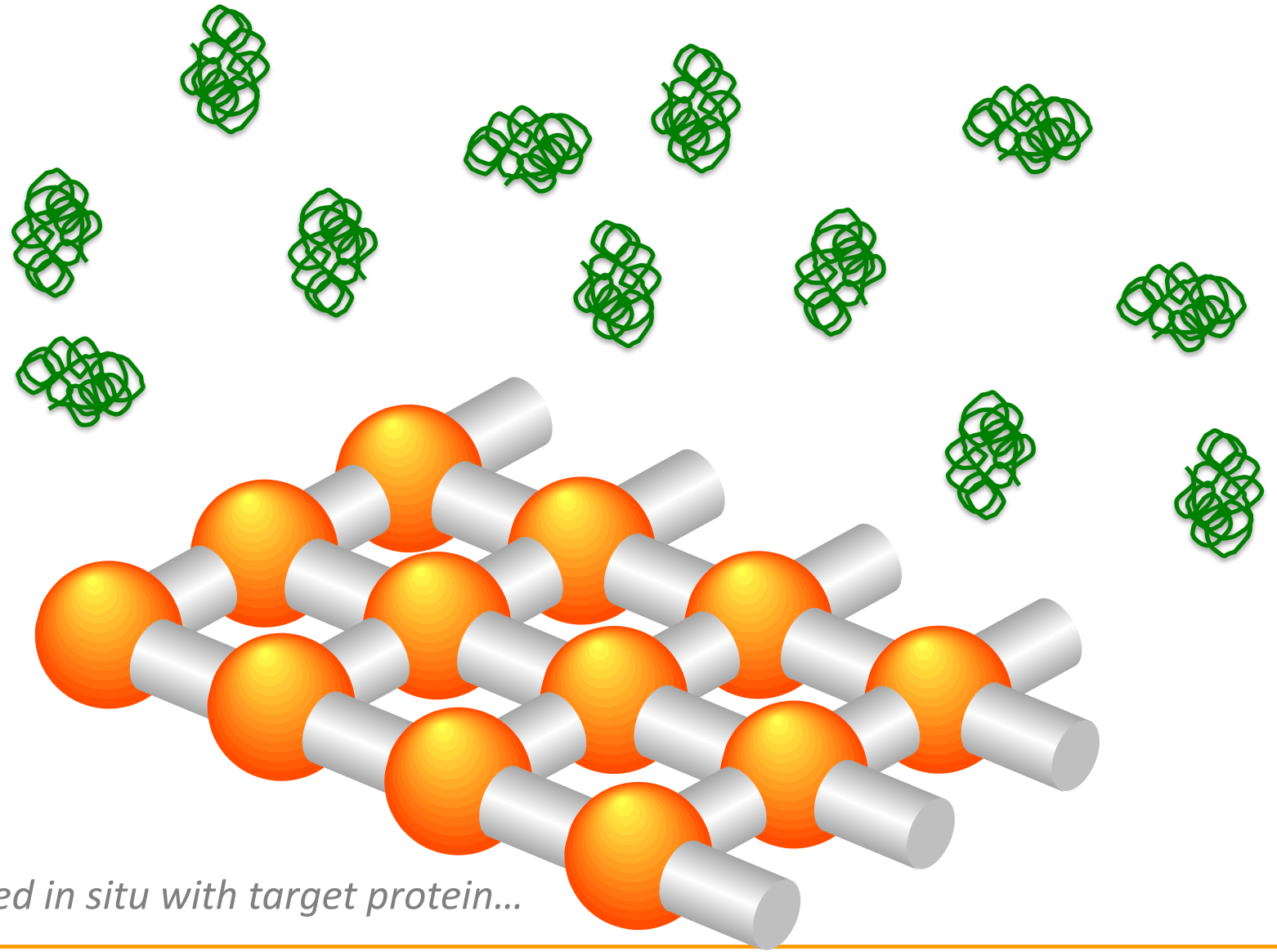
TECHNICAL CONCEPT 2D



…placed in situ with target protein…

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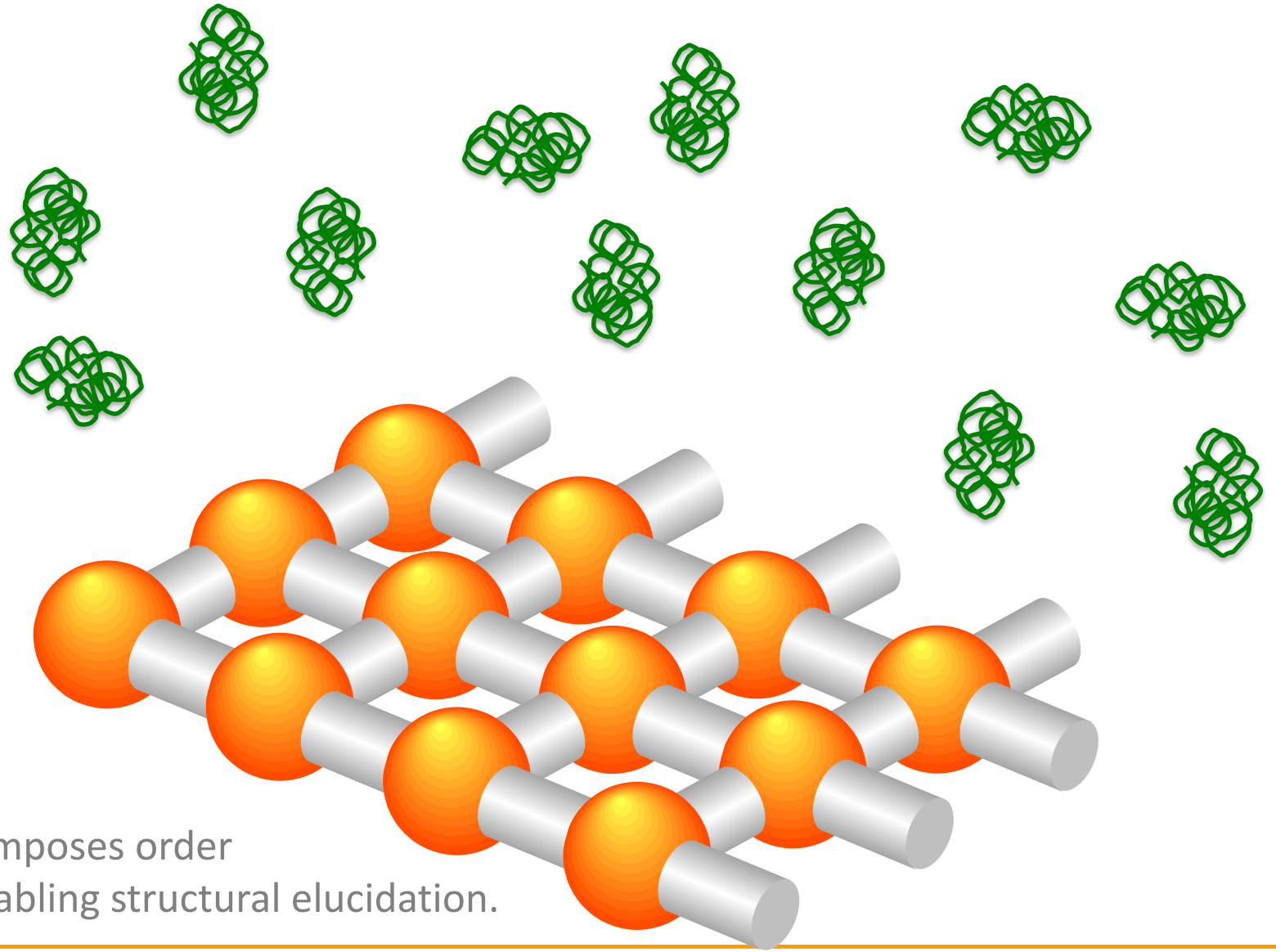
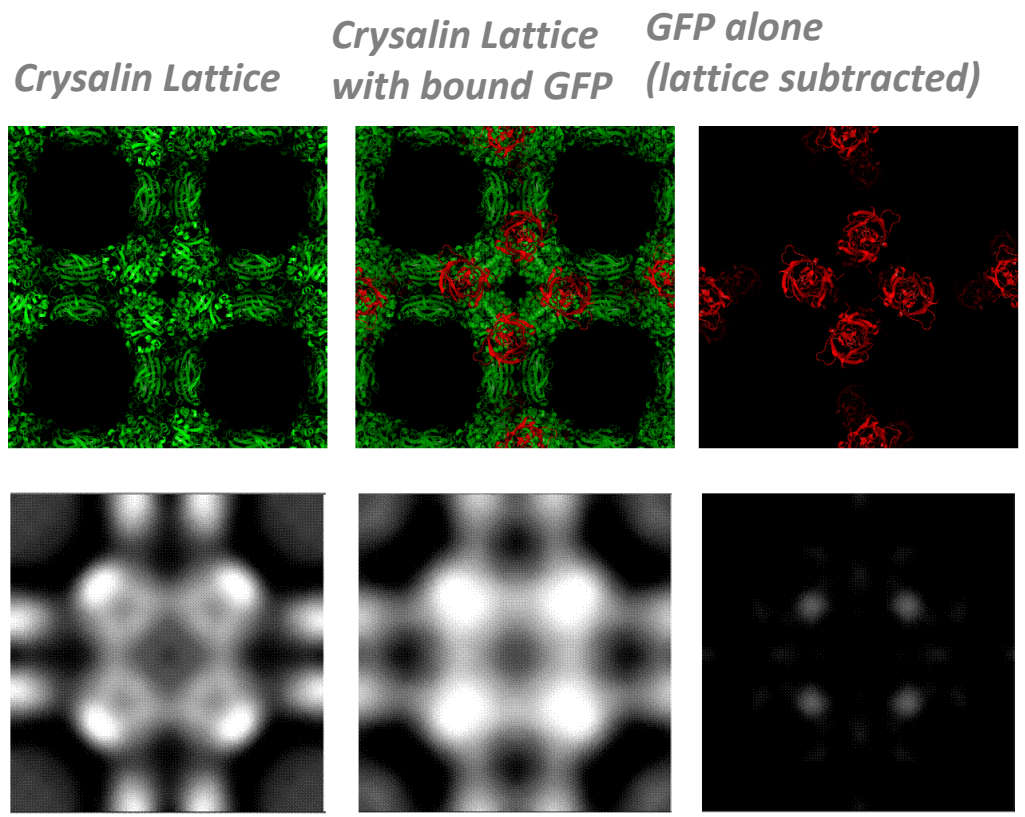
TECHNICAL CONCEPT 2D



…imposes order
enabling structural elucidation.

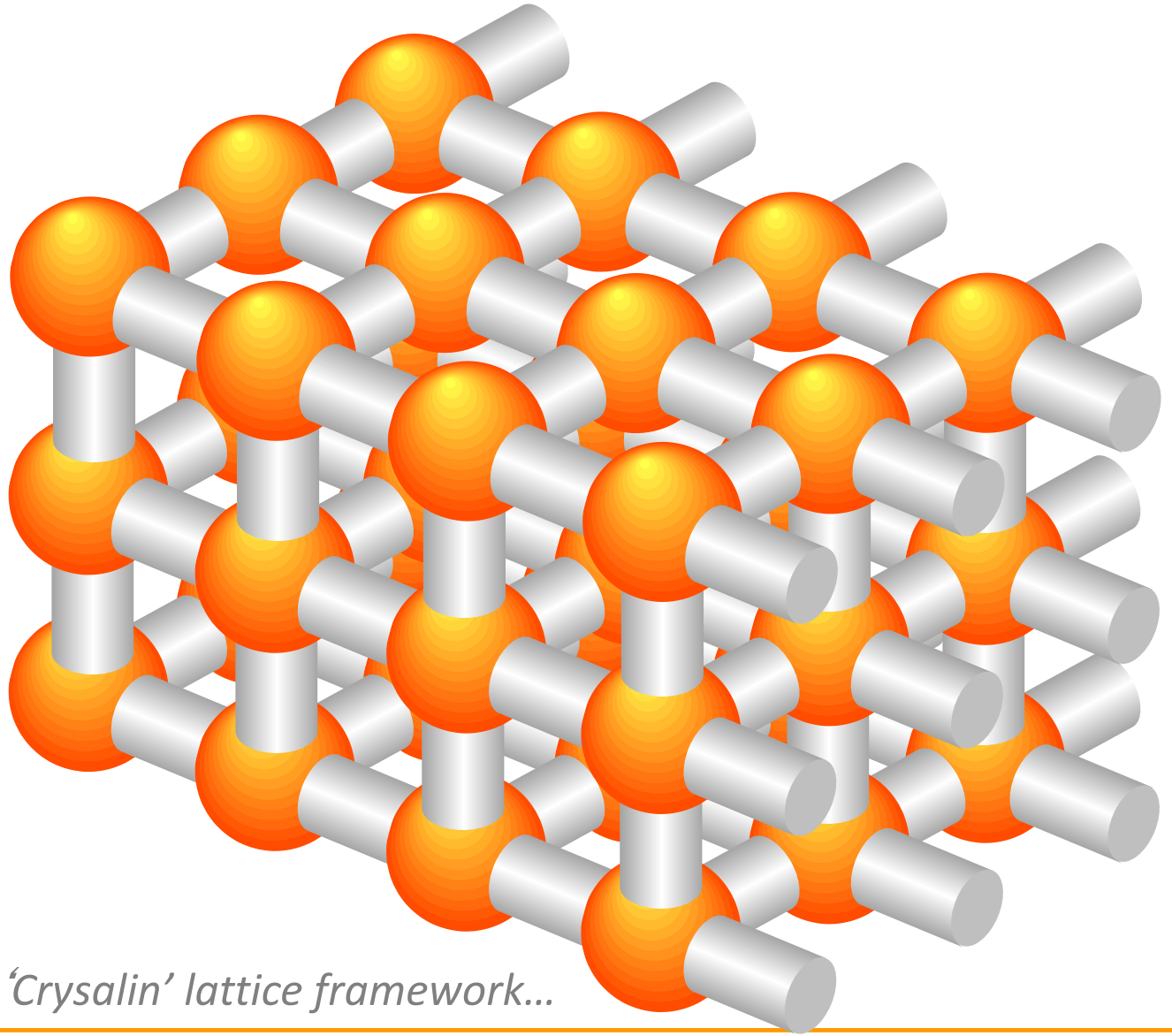
2D SYSTEMS



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

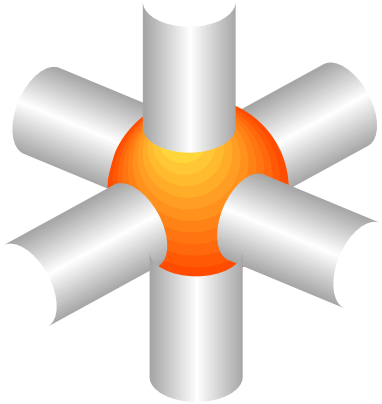
* *Generation of protein lattices by fusing proteins with matching rotational symmetry*, John C. Sinclair, Karen M. Davies, Catherine Vénien-Bryan & Martin E. M. Noble, Nature Nanotechnology 6, 558–562 (2011)

TECHNICAL CONCEPT 3D

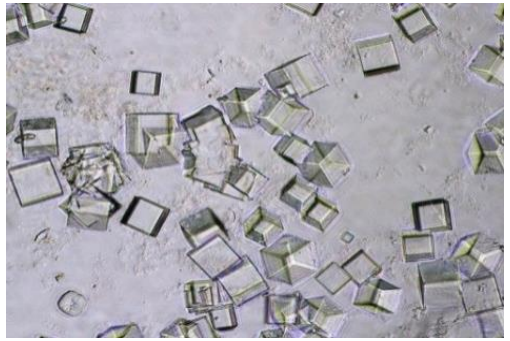


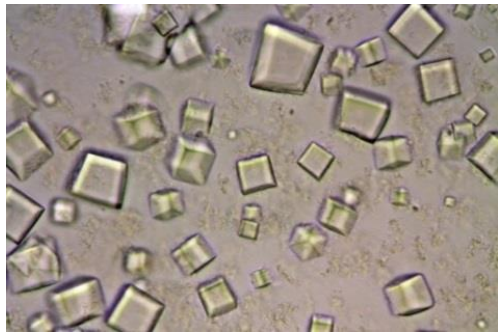
The 3D ‘Crysalin’ lattice framework…

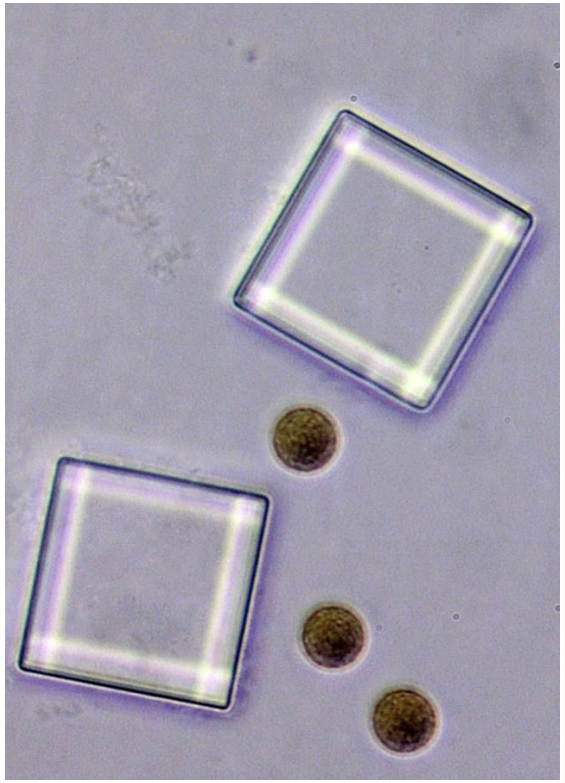
Crysalin formation



- 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)







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

"I don't want to be a robot"

Definitely not a robot!

Any questions?