

MX automation: few perspectives from ESRF

Present

Future

Unique interface (user/BL scientist)

Manual puck assignment

Auto puck assignment

SHIPMENT ASSIGNMENT

Shipment is always linked to a session with date

Immutable session date is part of the folders structure

89 shipments candidates for FX89

Display only shipments scheduled for future sessions or in processing status

Shipment			Experiment				
Name	Status	Created on	Start on	beamline			
ESRF FX23 15...	PROCESSING	08-11-2024	15-11-2024	ID30A-1	1 parcels / 2 containers (25 samples)	+	⊗
ESRF FX23 8 ...	SENT TO USER	01-11-2024	08-11-2024	ID30A-1	1 parcels / 4 containers (62 samples)	+	⊗
ESRF FX23 25...	SENT TO USER	17-10-2024	25-10-2024	ID30A-1	1 parcels / 3 containers (38 samples)	+	⊗
ESRF FX23 4 ...	AT_ESRF	26-09-2024	04-10-2024	ID30A-1	2 parcels / 3 containers (16 samples)	+	⊗
ESRF FX23 13...	SENT TO USER	05-09-2024	13-09-2024	ID30A-1	1 parcels / 2 containers (20 samples)	+	⊗
ESRF FX23 6 ...	SENT TO USER	29-08-2024	06-09-2024	ID30A-1	1 parcels / 2 containers (32 samples)	+	⊗
ESRF FX23 30...	SENT TO USER	22-08-2024	30-08-2024	ID30A-1	1 parcels / 5 containers (64 samples)	+	⊗
ESRF FX23 26...	SENT TO USER	18-07-2024	26-07-2024	ID30A-1	1 parcels / 1 containers (6 samples)	+	⊗
ESRF FX23 28...	SENT TO USER	20-06-2024	28-06-2024	ID30A-1	1 parcels / 3 containers (69 samples)	+	⊗
ESRF FX23 21...	SENT TO USER	13-06-2024	21-06-2024	ID30A-1	1 parcels / 2 containers (32 samples)	+	⊗
ESRF FX23 14...	SENT TO USER	06-06-2024	14-06-2024	ID30A-1	1 parcels / 3 containers (39 samples)	+	⊗
ESRF FX23 7 ...	SENT TO USER	30-05-2024	07-06-2024	ID30A-1	1 parcels / 7 containers (112 samples)	+	⊗
ESRF FX23 12...	SENT TO USER	04-04-2024	12-04-2024	ID30A-1	1 parcels / 3 containers (33 samples)	+	⊗
ESRF FX23 5 ...	SENT TO USER	26-03-2024	06-04-2024	ID30A-1	1 parcels / 2 containers (18 samples)	+	⊗
ESRF FX23 28...	SENT TO USER	22-03-2024	29-03-2024	ID30A-1	1 parcels / 6 containers (81 samples)	+	⊗
ESRF FX23 4 ...	SENT TO USER	27-02-2024	08-03-2024	ID30A-1	1 parcels / 3 containers (48 samples)	+	⊗
ESRF FX23 16...	SENT TO USER	09-02-2024	16-02-2024	ID30A-1	1 parcels / 1 containers (12 samples)	+	⊗
ESRF FX23 5 ...	SENT TO USER	01-02-2024	09-02-2024	ID30A-1	1 parcels / 3 containers (48 samples)	+	⊗

PUCK ASSIGNMENT

89 shipments candidates for FX89

Display only shipments scheduled for future sessions or in processing status

Shipment			Experiment		
Name	Status	Created on	Start on	beamline	
ESRF FX23 15...	PROCESSING	08-11-2024	15-11-2024	ID30A-1	1 parcels / 2 containers (25 samples)
ESRF FX23 8 ...	SENT TO USER	01-11-2024	08-11-2024	ID30A-1	
ESRF FX23 25...	SENT TO USER	17-10-2024	25-10-2024	ID30A-1	
ESRF FX23 4 ...	AT_ESRF	28-09-2024	04-10-2024	ID30A-1	
ESRF FX23 13...	SENT TO USER	05-09-2024	13-09-2024	ID30A-1	
ESRF FX23 6 ...	SENT TO USER	29-08-2024	06-09-2024	ID30A-1	
ESRF FX23 30...	SENT TO USER	22-08-2024	30-08-2024	ID30A-1	
ESRF FX23 26...	SENT TO USER	18-07-2024	26-07-2024	ID30A-1	
ESRF FX23 28...	SENT TO USER	20-06-2024	28-06-2024	ID30A-1	
ESRF FX23 21...	SENT TO USER	13-06-2024	21-06-2024	ID30A-1	
ESRF FX23 14...	SENT TO USER	06-06-2024	14-06-2024	ID30A-1	
ESRF FX23 7 ...	SENT TO USER	30-05-2024	07-06-2024	ID30A-1	
ESRF FX23 12...	SENT TO USER	04-04-2024	12-04-2024	ID30A-1	
ESRF FX23 5 ...	SENT TO USER	26-03-2024	06-04-2024	ID30A-1	
ESRF FX23 28...	SENT TO USER	22-03-2024	29-03-2024	ID30A-1	
ESRF FX23 4 ...	SENT TO USER	27-02-2024	08-03-2024	ID30A-1	
ESRF FX23 16...	SENT TO USER	09-02-2024	16-02-2024	ID30A-1	
ESRF FX23 5 ...	SENT TO USER	01-02-2024	09-02-2024	ID30A-1	

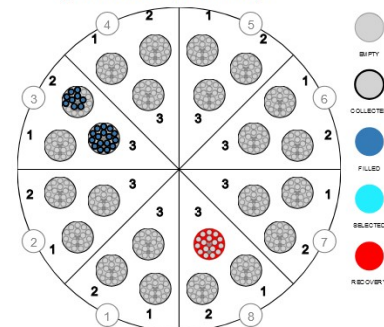
Loaded or to be Loaded on MxCube

Shipment	Parcel	Container	Container type	Beamline	Cell	Position
ESRF FX23 15 Nov 2024	dewer1	gsk1007 (16 samples)	UNIPUCK	ID30A-1	3	3
ESRF FX23 15 Nov 2024	dewer1	cps0801 (9 samples)	UNIPUCK	ID30A-1	3	2

Unload all

ID30A-1 (FlexHCDUnipuckPlate)

Unload SC





AUTOMATED SESSION/PUCK ASSIGNMENT

Barcode reader for the dewar

Connection between sample changer and LIMS

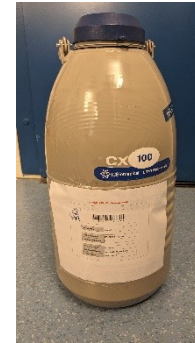
Puck detection





* E S R F 0 3 2 4 4 6 8 *

Parcel label	Dewar1
Shipment name	UG-BIOCAT-101124
Number of parcels	1
Proposal number	MX-2649
Laboratory name	University of Groningen
Local contact	NURIZZO D



Barcode reader for the dewar

Puck detection

Connection between sample changer and DRAC

Position	Type	Barcode
1.1	Unipuck	
1.2	Unipuck	
1.3	Unipuck	
2.1	Unipuck	
2.2	Unipuck	
2.3	Unipuck	
3.1	Unipuck	
3.2	Unipuck	
3.3	Unipuck	
4.1	Unipuck	
4.2	Unipuck	
4.3	Unipuck	
5.1	Unipuck	
5.2	Unipuck	
5.3	Unipuck	
6.1	Unipuck	
6.2	Unipuck	
6.3	Unipuck	
7.1	Unipuck	
7.2	Unipuck	
7.3	Unipuck	
8.1	Unipuck	
8.2	Unipuck	
8.3	Unipuck	

CPS 1231

CPS 1232

CPS 1233

CPS 1234

CPS 1235

CPS 1236

CPS 1237

Puck Barcode:

V

CPS 1234

Puck number: Add puck to list

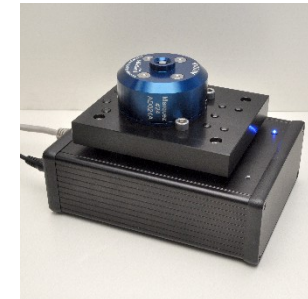
User Port

Open

Ln level

92%

■ Unipuck segment
■ SC3 Puck segment



Present

Future

Unique interface (user/BL scientist)

Manual puck assignment

Auto puck assignment

Single user

Unique operator (SSO)

SINGLE SIGN-ON

On-time and early/late session selection

The screenshot displays the MXCuBE-Web (OSC) interface. At the top, the status bar shows 'Samples', 'Data collection', 'Equipment', and 'System log'. The main panel is divided into several sections:

- Beamline Actions:** Energy: 12.8420 KeV, Resolution: 2.000 Å, Transmission: 100.0 %, Cryo: 100.02 k, Wavelength: 0.97 Å, Detector: 388.6 mm, Flux: 2.71e+11 ph/s.
- Phase Control:** DataCollection, Beam size (A30), Omega (200.00), Kappa (0.0), Kappa Phi (0.0), Sample alignment, and Show motors.
- Sample Information:** Sample: AP2B2 - CD041773_C10-3, Queued Samples (104).
- Session Selection Dialog:** A modal window titled 'Select a session' is open, showing a table of sessions. The 'Scheduled (5)' tab is active, and session 'FX-23' is highlighted.

ID	Description	Start	End	Link 1	Link 2	Link 3
MX-2644	ID30A-1 Swiss consortium in macromolecular X-ray crystallography structure determination for academic researchers: EPFL, UNIL, UNIGE, Zurich and SL	29-11-2024 09:30:00	29-11-2024 17:00:00	🔗	🔗	🔗
FX-75	ID30A-1 Mail-in	20-11-2024 09:30:00	21-11-2024 01:00:00	🔗	🔗	🔗
MX-2647	ID30A-1 Coordinated user access to HTX lab – MASSIF-1 facilities for fragment screening and new structures. With funding through INEXT Discovery, In	19-11-2024 09:30:00	20-11-2024 01:00:00	🔗	🔗	🔗
FX-23	ID30A-1 MXpress for GSK	15-11-2024 09:30:00	15-11-2024 17:00:00	🔗	🔗	🔗
FX-66	ID30A-1 Mail-in Ligand screening	14-11-2024 09:30:00	15-11-2024 01:00:00	🔗	🔗	🔗
MX-2647	ID30A-1 Coordinated user access to HTX lab – MASSIF-1 facilities for fragment screening and new structures. With funding through INEXT Discovery, In	13-11-2024 17:00:00	14-11-2024 01:00:00	🔗	🔗	🔗
MX-2654	ID30A-1 Structural Biology at EMBL Grenoble, IBS-CIBB and IAB	13-11-2024 17:00:00	14-11-2024 01:00:00	🔗	🔗	🔗

At the bottom of the dialog, there are buttons for 'Reschedule', 'Cancel', and 'Select FX-23'. A status bar at the very bottom indicates: '16:32:14: Diffractometer phase changed to DataCollection'.

Present	Future
Unique interface (user/BL scientist)	
Manual puck assignment	Auto puck assignment
Single user	Unique operator (SSO)
Single session	Multiple sessions

MULTIPLE SESSIONS

A single operator runs multiple session in one go

MX-CUBE UI

Present	Future
Unique interface (user/BL scientist)	
Manual puck assignment	Auto puck assignment
Single user	Unique operator (SSO)
Single session	Multiple sessions
Immutable queue	On-the-fly mutable (+/-) queue order

MX-CUBE QUEUING

Diffraction Plan including the processing plan in LIMS

MXCuBE-Web (OSC) interface showing a diffraction plan for four packs (RUGX-008 to RUGX-012). The interface includes a 'Collect Queue?' dialog box with options for auto loop centring, auto mount next sample, and crystal snapshots. A table lists sample details including MXPressA, sample ID, path, and number of images.

Type	Sample	Path	# Images
MXPressA	W598pyAla_b10p1 - TmmS3 (2.3.01)	...data/Visitor/mx2649/id30a1/20241110/RAW_DATA/TmmS3/TmmS3-W598pyAla_b10p1/TmmS3-W598pyAla_b10p1_0_004d.cbf	100
MXPressA	b12p10 - C8BE (3.1.10)	...data/Visitor/mx2649/id30a1/20241110/RAW_DATA/C8BE/C8BE-b12p10C8BE-b12p10_0_004d.cbf	100
MXPressA	b12p11 - C8BE (3.1.11)	...data/Visitor/mx2649/id30a1/20241110/RAW_DATA/C8BE/C8BE-b12p11C8BE-b12p11_0_004d.cbf	100
MXPressA	b12p12 - C8BE (3.1.12)	...data/Visitor/mx2649/id30a1/20241110/RAW_DATA/C8BE/C8BE-b12p12C8BE-b12p12_0_004d.cbf	100
MXPressA	b12p13 - C8BE (3.1.13)	...data/Visitor/mx2649/id30a1/20241110/RAW_DATA/C8BE/C8BE-b12p13C8BE-b12p13_0_004d.cbf	100
MXPressA	b12p14 - C8BE (3.1.14)	...data/Visitor/mx2649/id30a1/20241110/RAW_DATA/C8BE/C8BE-b12p14C8BE-b12p14_0_004d.cbf	100
MXPressA	b12p15 - C8BE (3.1.15)	...data/Visitor/mx2649/id30a1/20241110/RAW_DATA/C8BE/C8BE-b12p15C8BE-b12p15_0_004d.cbf	100
MXPressA	b12p16 - C8BE (3.1.16)	...data/Visitor/mx2649/id30a1/20241110/RAW_DATA/C8BE/C8BE-b12p16C8BE-b12p16_0_004d.cbf	100

MXCuBE-Web (OSC) interface showing a diffraction image and a table of sample details.

Sample	Path	# Images
MXPressA	W598pyAla_b10p1 - TmmS3 (2.3.01)	100
MXPressA	b12p10 - C8BE (3.1.10)	100
MXPressA	b12p11 - C8BE (3.1.11)	100
MXPressA	b12p12 - C8BE (3.1.12)	100
MXPressA	b12p13 - C8BE (3.1.13)	100
MXPressA	b12p14 - C8BE (3.1.14)	100
MXPressA	b12p15 - C8BE (3.1.15)	100
MXPressA	b12p16 - C8BE (3.1.16)	100

MX-CUBE UI

Present	Future
Unique interface (user/BL scientist)	
Manual puck assignment	Auto puck assignment
Single user	Unique operator (SSO)
Single session	Multiple sessions
Immutable queue	On-the-fly mutable (+/-) queue order
Per sample queuing list	Sample changer / Puck queuing overview

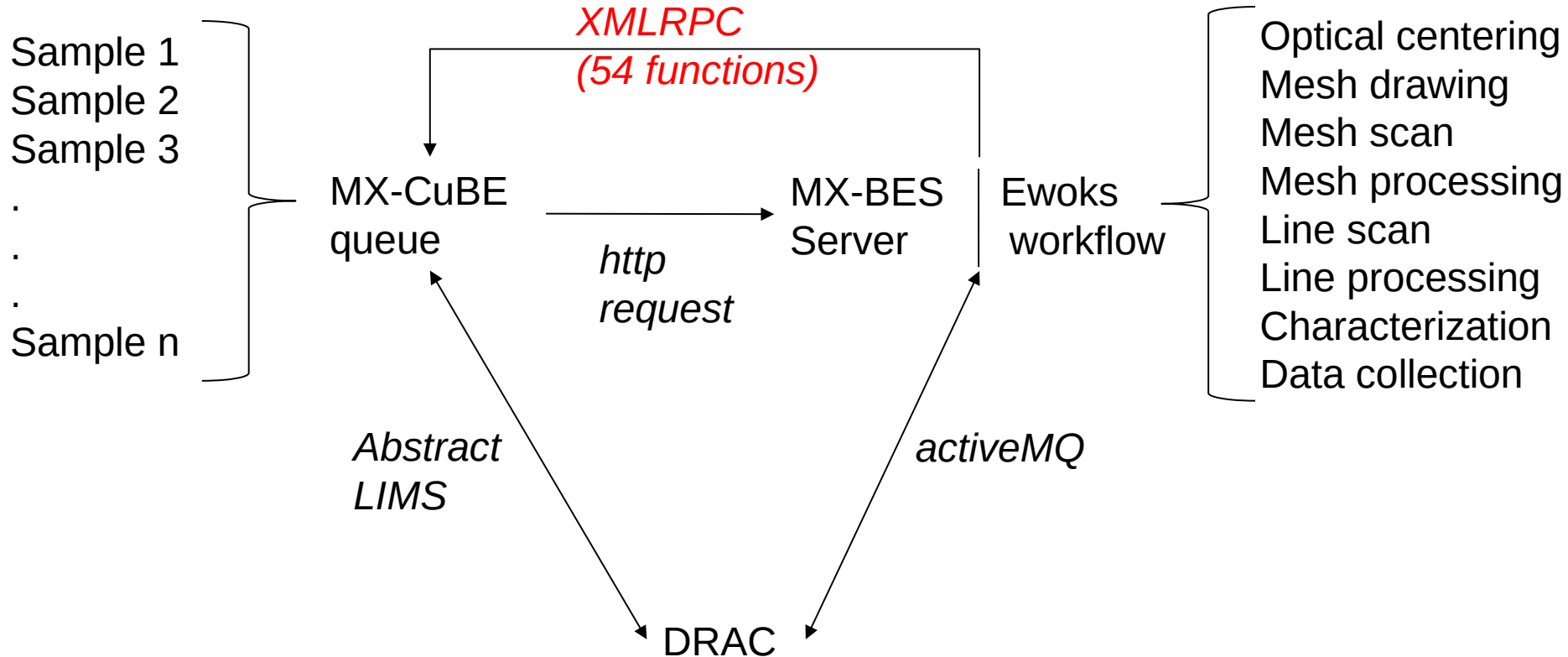
MX-CUBE QUEUING

Design a Sample Changer overview / Puck overview

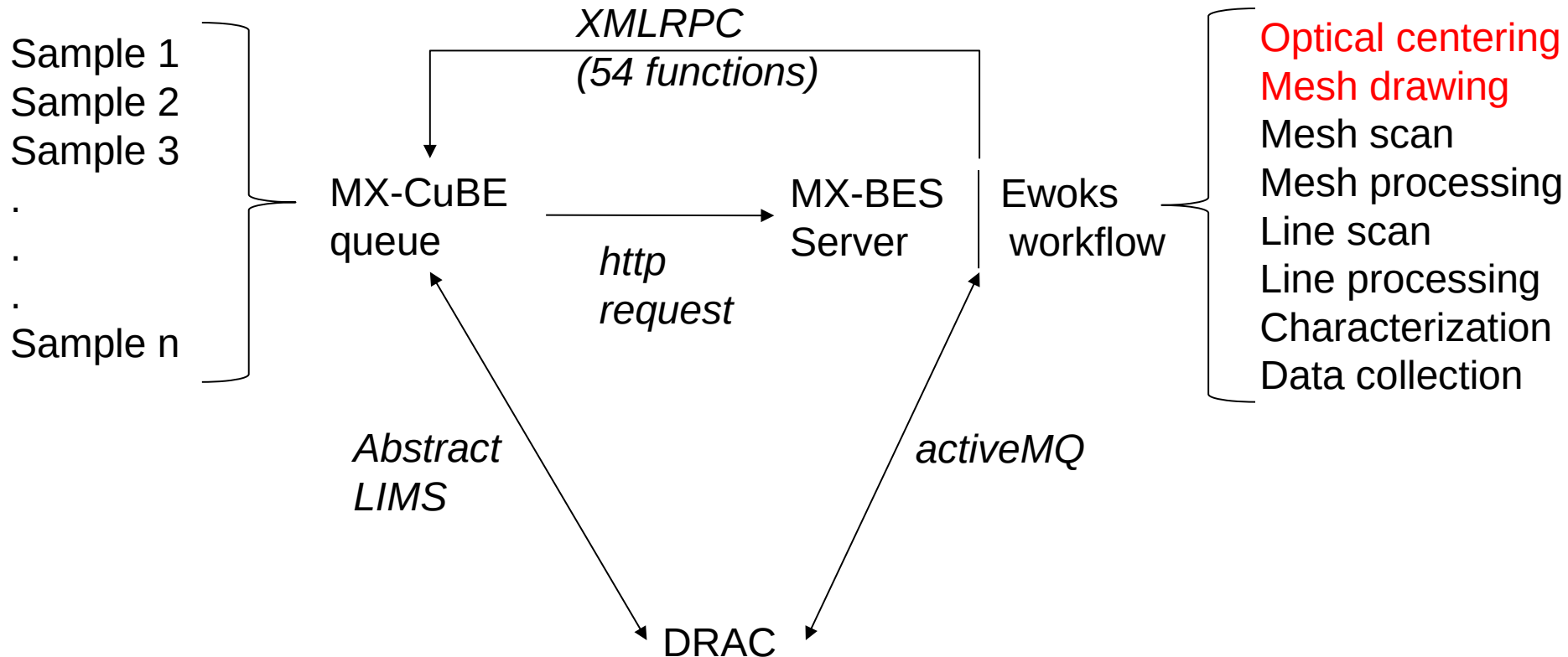
The screenshot displays the MXCuBE-Web (OSG) interface. At the top, there are navigation tabs for 'Samples', 'Data collection', 'Equipment', and 'System log'. A search bar and a 'Filter' dropdown are visible. Below the navigation, there are buttons for 'Synchronize with', 'Create new sample', 'Clear sample list', and 'View Mode'. A 'Filter:' input field and a 'Filter options' dropdown are also present. A 'Stop queue' button is located in the top right corner. The main area is divided into four columns representing different cells: Cell 2 (Puck 1, Code: RUGX-008), Cell 2 (Puck 2, Code: RUGX-009), Cell 2 (Puck 3, Code: RUGX-010), and Cell 1 (Puck 1, Code: RUGX-012). Each puck contains a grid of sample entries, each with a checkbox, a sample code (e.g., 'Linh1-V35C-R79H-RK-Au1'), and a status indicator (green or red circle).

This screenshot shows a detailed view of a sample changer puck. On the left, there is a video feed showing the internal mechanism of the sample changer. On the right, there is a 'Sample changer' section with a 'Stop queue' button. Below this, there is a list of samples with their codes and status indicators. The interface also includes a 'Sample changer' section with a 'Stop queue' button and a list of samples with their codes and status indicators.

WORKFLOW / MXCUBE REFURBISHMENT



SOFTWARE REFURBISHMENT

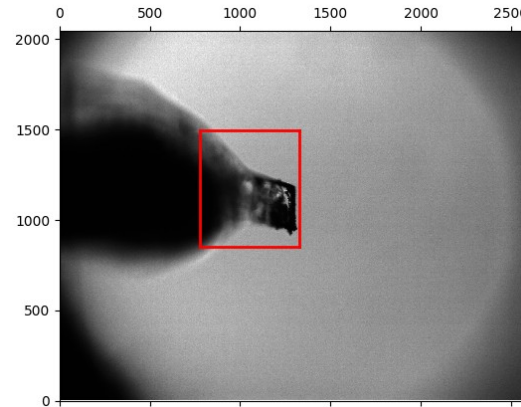
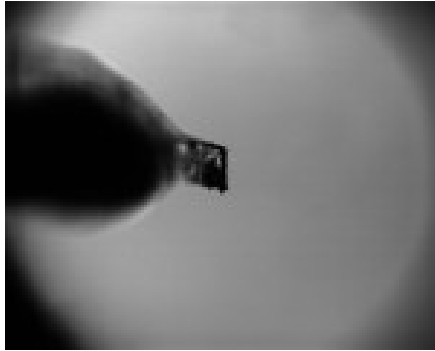


OPTICAL ANALYSIS

A set of optical images

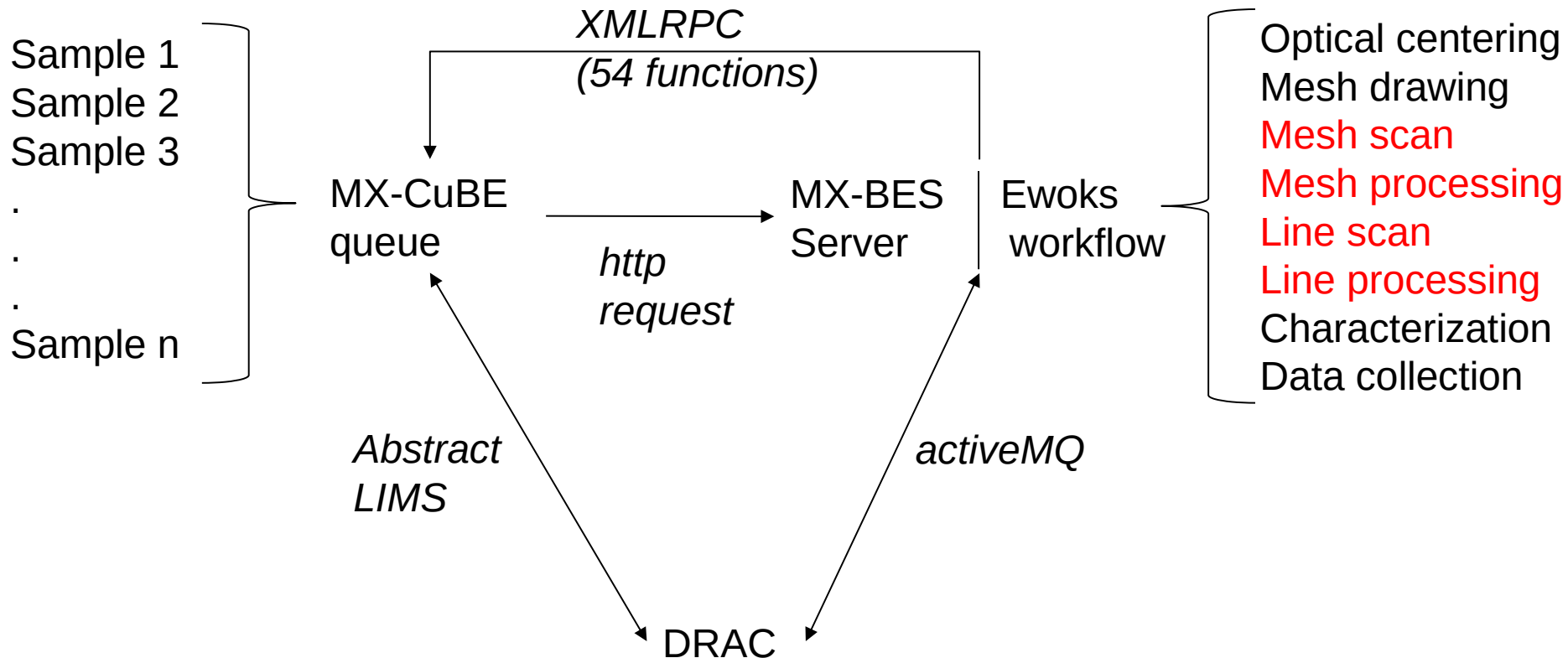
optical centering
➔
MURKO / LUCID

- Loop centering
- Crystal recognition
- Mesh drawing



Single object returning the optical features of the crystal inside the loop
(center of mass, position of the crystal, size of the loop, etc...)

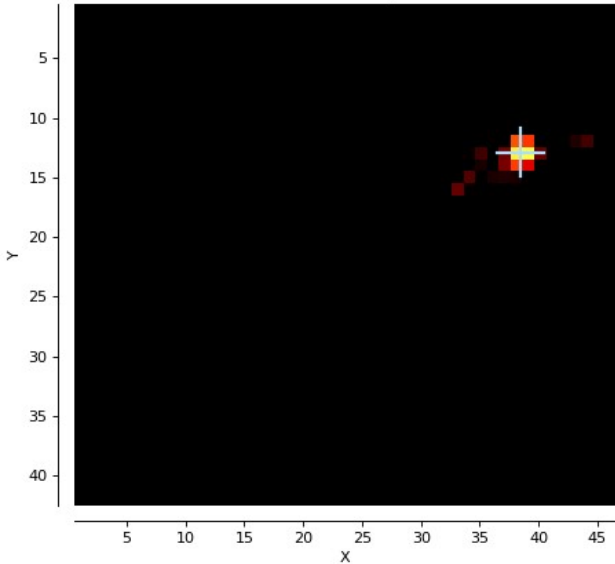
SOFTWARE REFURBISHMENT



X-RAY CENTERING

A set of diffraction images

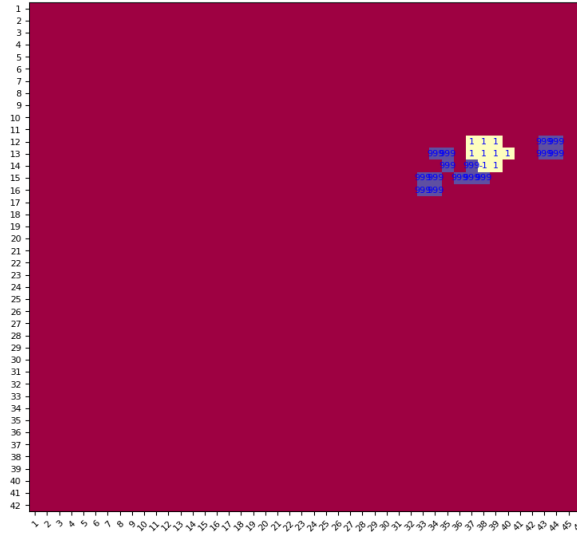
Diffraction signal



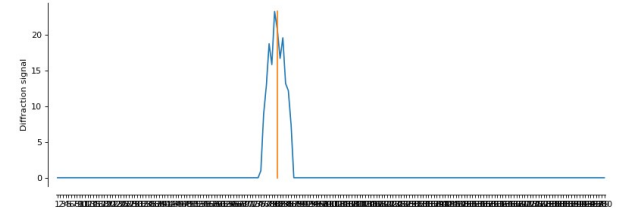
Diffraction alignment



Crystal map



- Best diffraction centering
- Crystal size
- Crystal homogeneity
- Resolution limit



Single object returning the diffraction features of the crystal
(best diffracting volume and position, size of the crystal, , etc...)

ACKNOWLEDGEMENTS

Thoroughly discussed :
During our bi-weekly Automation Task
Force meetings
and
In our MXCuBE automation WG:

Matthew Bowler
Antonia Beteva
Marcus Oskarsson
Estelle Mossou
Daniele de Sanctis
Andrew McCarthy

Johannes
Kamps
Max Nanao
Ludovic Broche
Romain Talon
Wout de Nolf
Yan Walesh

And many
more

Questions still under discussion:

- Common definition of automated/unattended DC
- Long term perspective for the DC model
- Granularity of the abstraction

Macromolecular Crystallography And Structural Biology at synchrotron

MASSIF-1 AUTOMATED DATA COLLECTION

Automation for Room Temperature experiments

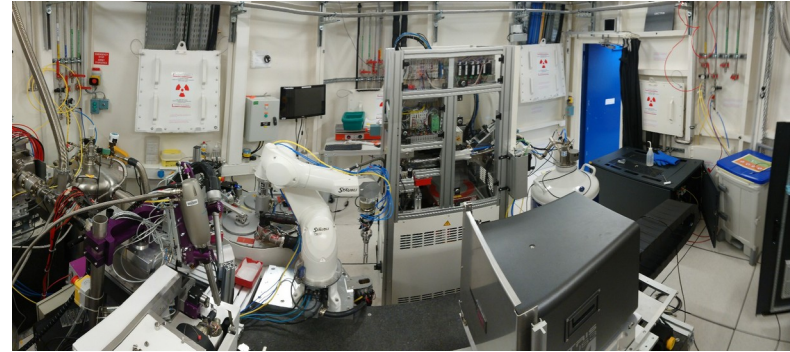
- Crystal Direct Harvester
- In-Situ data collection



MASSIF-1 AUTOMATED DATA COLLECTION

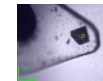
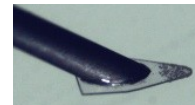
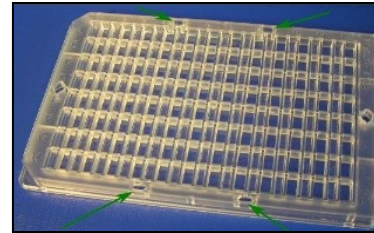
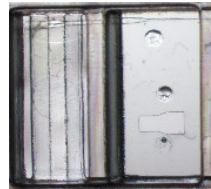
Automation for Room Temperature experiments

- Crystal Direct Harvester



- In-Situ data collection

MiTeGen

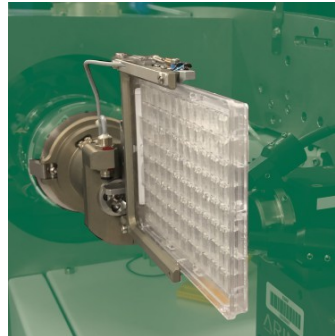
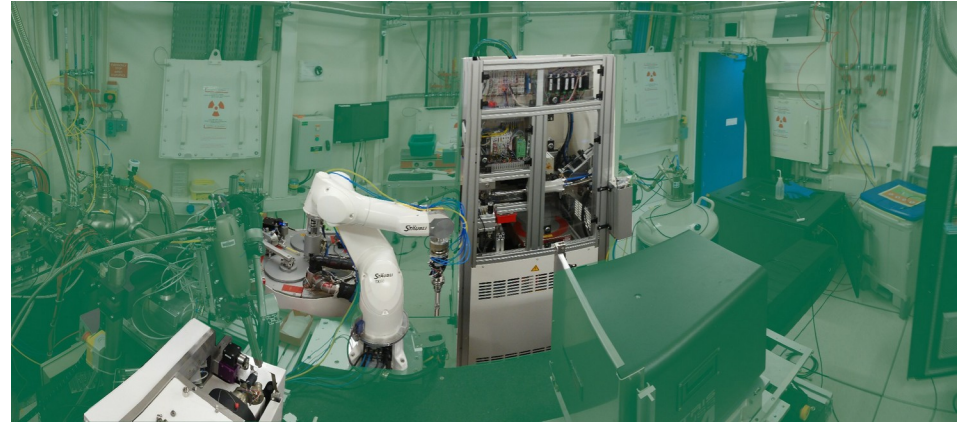


Crystal Direct technology

MASSIF-1 AUTOMATED DATA COLLECTION

Automation for Room Temperature experiments

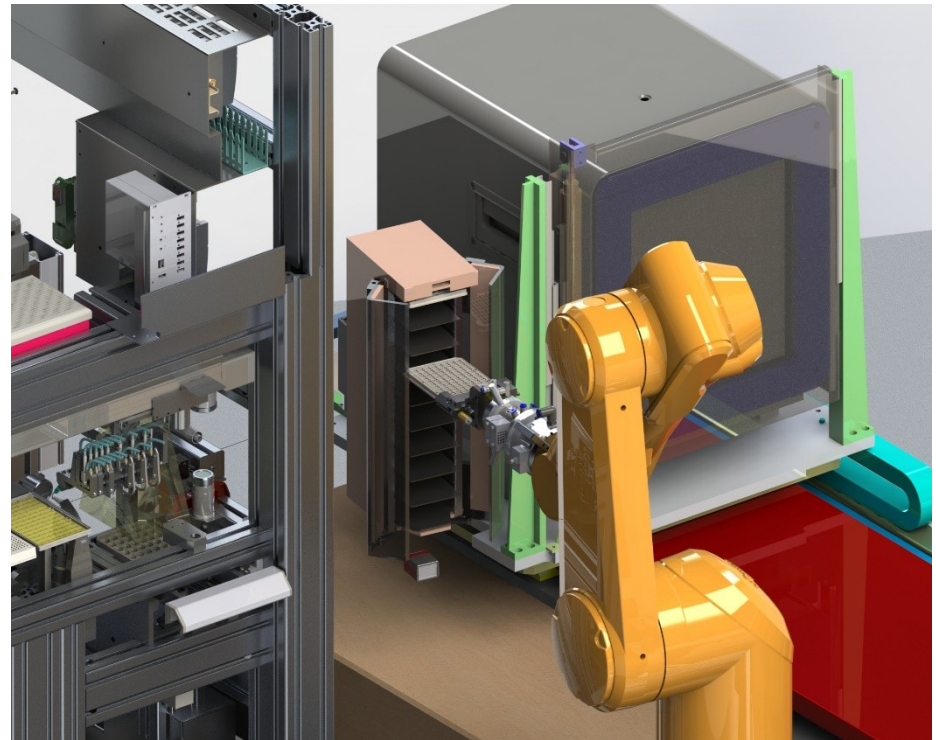
- Crystal Direct Harvester
- In-Situ data collection



MASSIF-1 AUTOMATED DATA COLLECTION

Automation for Room Temperature experiments

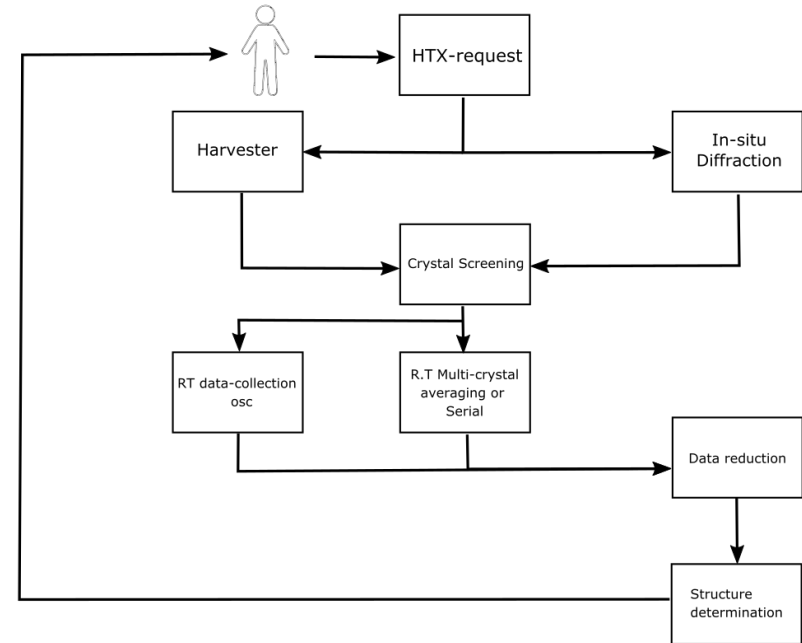
- Crystal Direct Harvester
- In-Situ data collection



MASSIF-1 AUTOMATED DATA COLLECTION

Automation for Room Temperature experiments

- Crystal Direct Harvester
- In-Situ data collection



CURRENT FEATURES

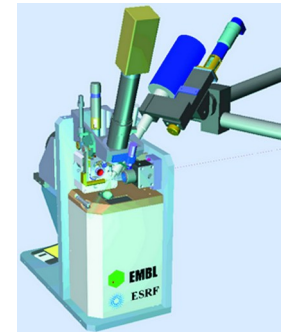
Main:

- Ω axis (SOC < 1 μm)
- Centering table
- Alignment table
- Apertures
- Capillary
- Beamstop
- smartMagnet
- OAV camera
- Scintillator
- Fluo det translation



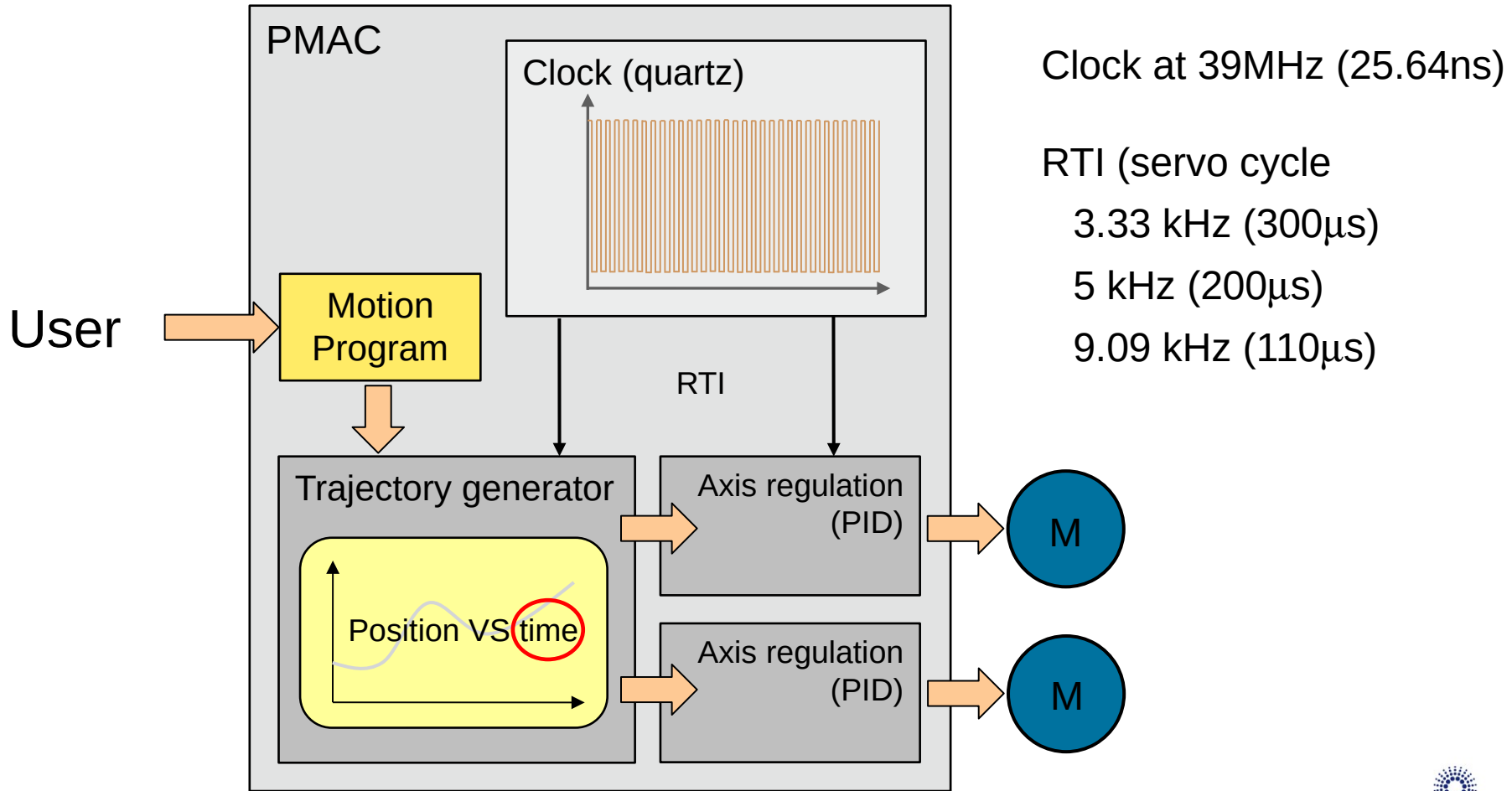
Ancillary:

- Mini-Kappa
- Plate manipulator
- Fluo det translation
- Luciole control
- REX control
- Shutter control
- Flex communication

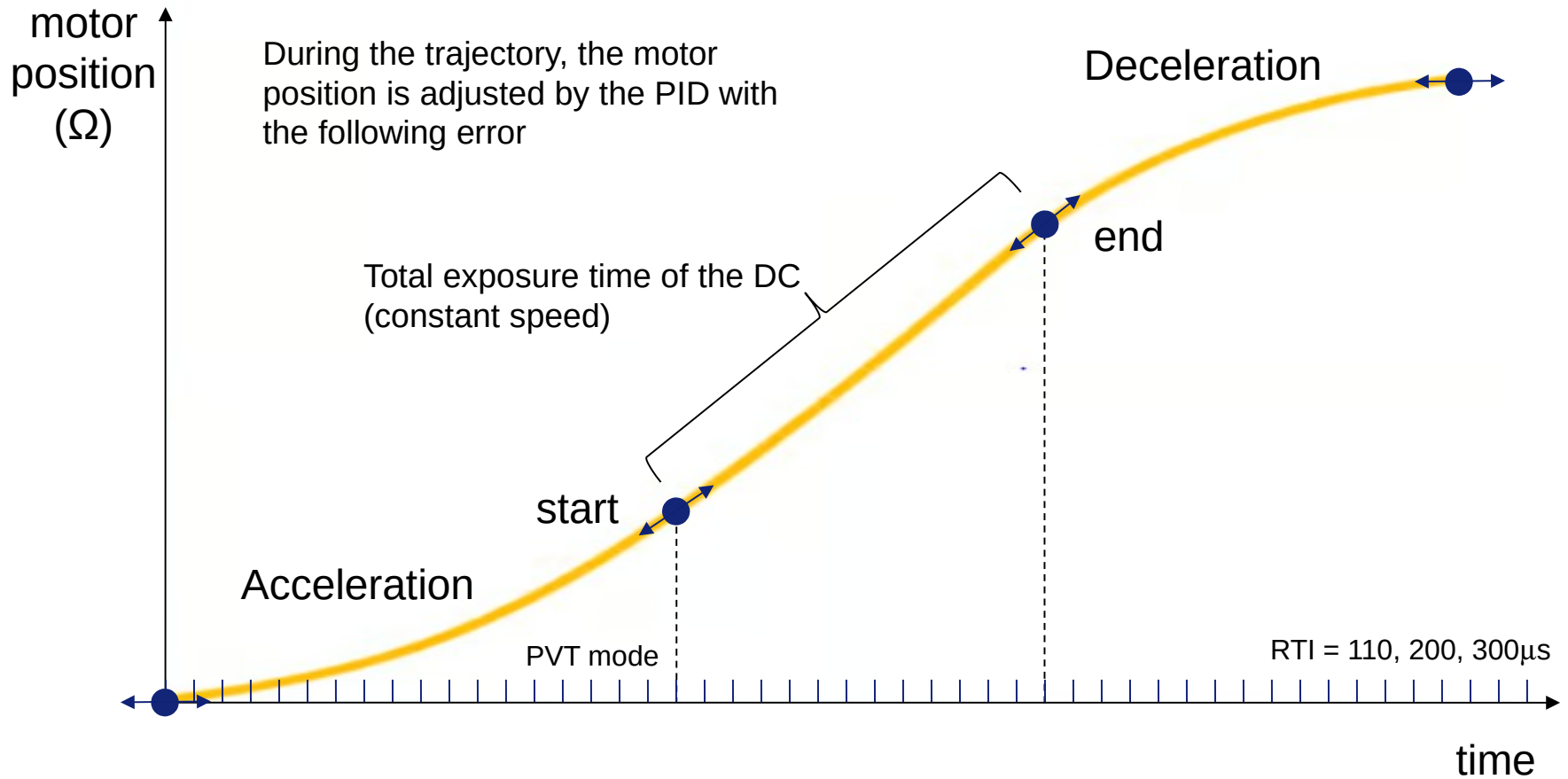


Protein microcrystals and the design of a microdiffractometer: current experience and plans at EMBL and ESRF/ID13
Perrakis, A., Cipriani, F., Castagna, J.-C., Claustre, L., Burghammer, M., Riekkel, C. & Cusack, S. (1999). Acta Cryst. D55, 1765-1770.

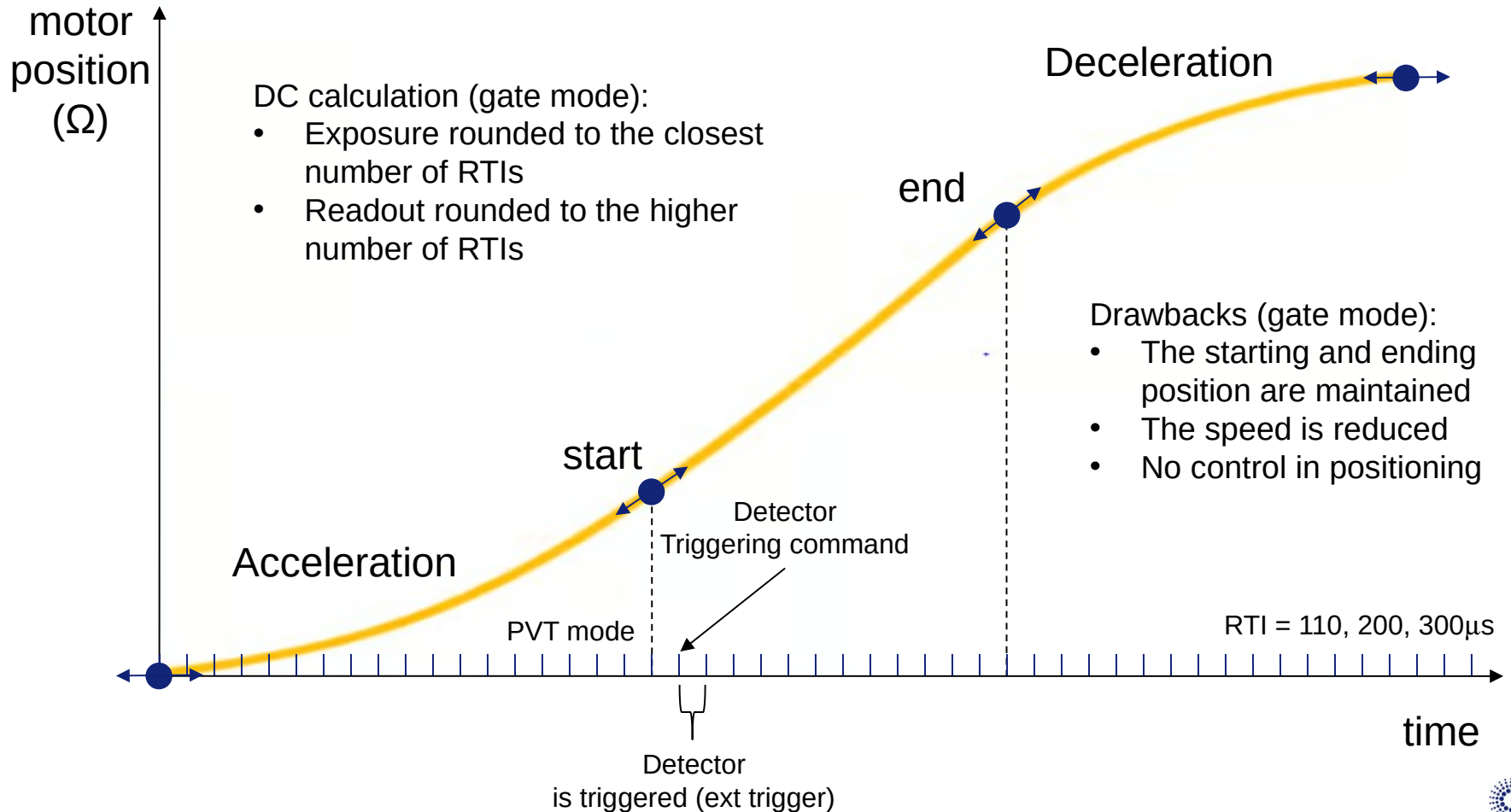
PMAC – DELTA TAU



MOTION PROGRAMME

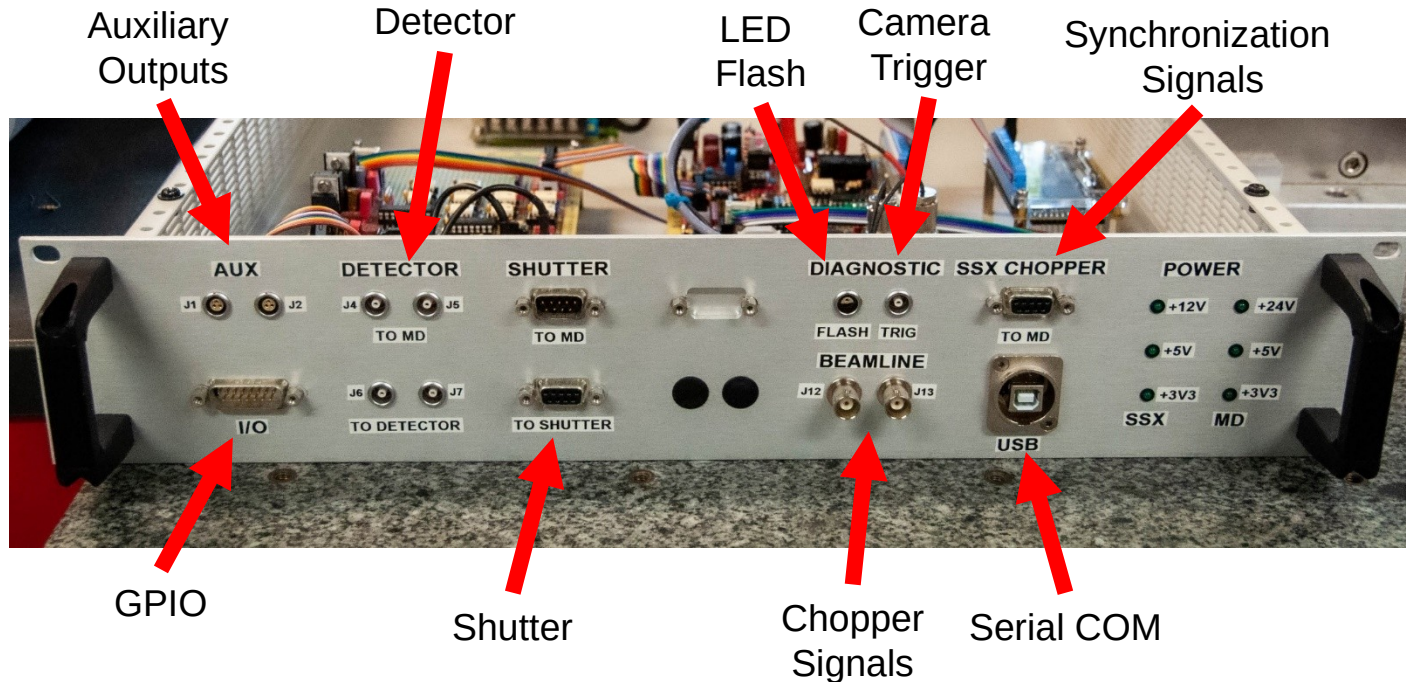


MOTION PROGRAMME



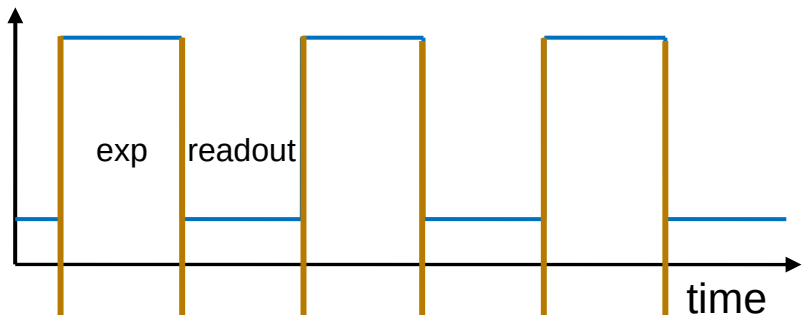
SSX CONTROL UNIT ON ID29 (FPGA)

Clock at 50MHz (20-ns)

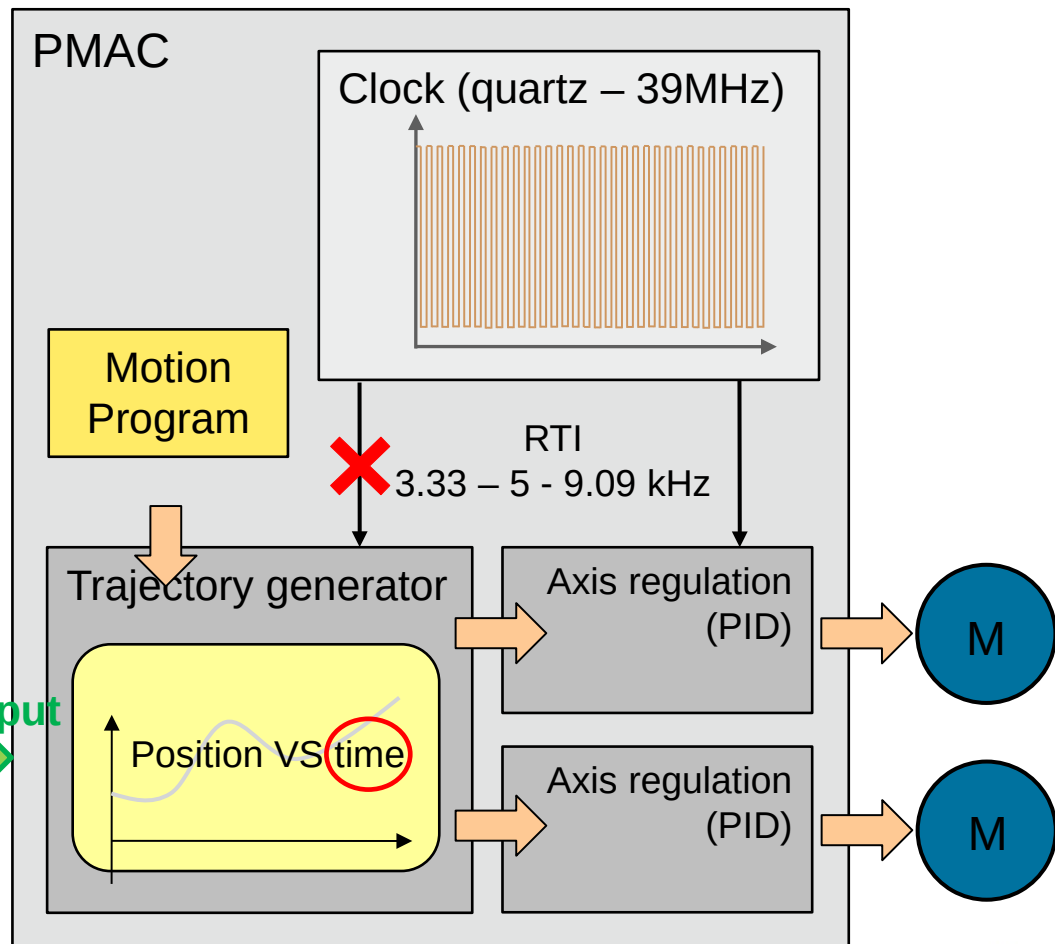
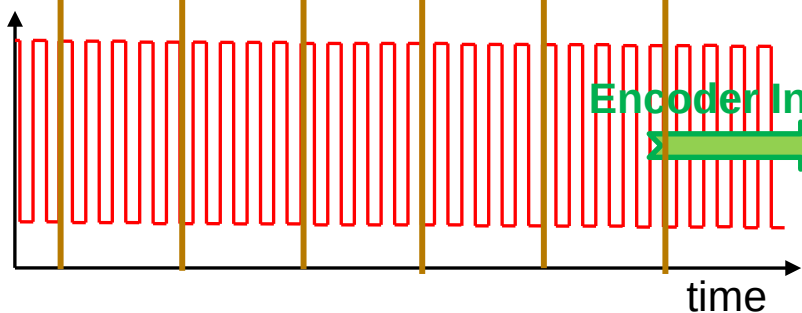


TRIGGERED TIME BASE

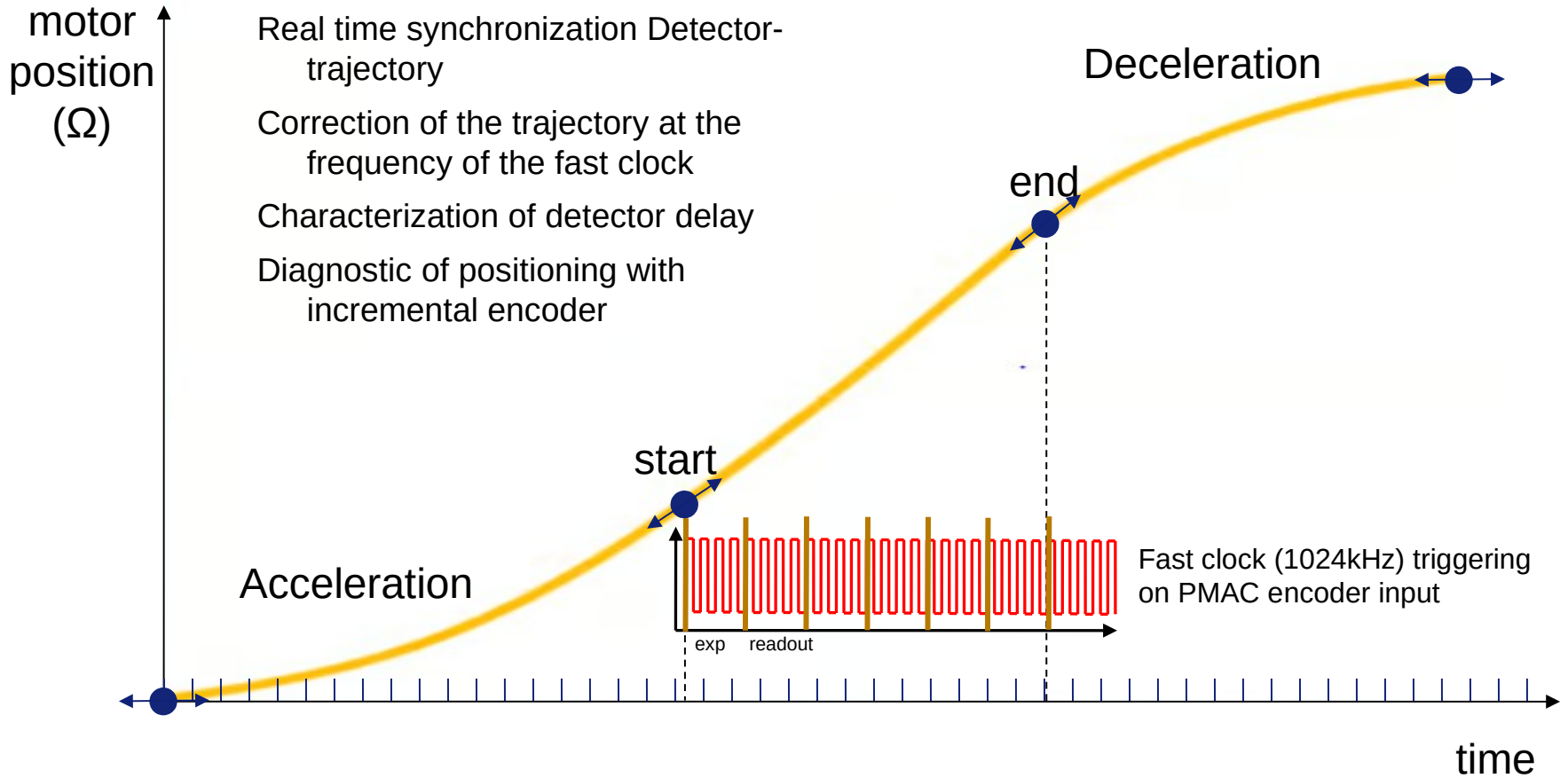
Detector signal



Fast clock



TRIGGERED TIME BASE



Macromolecular Crystallography And Structural Biology at synchrotron

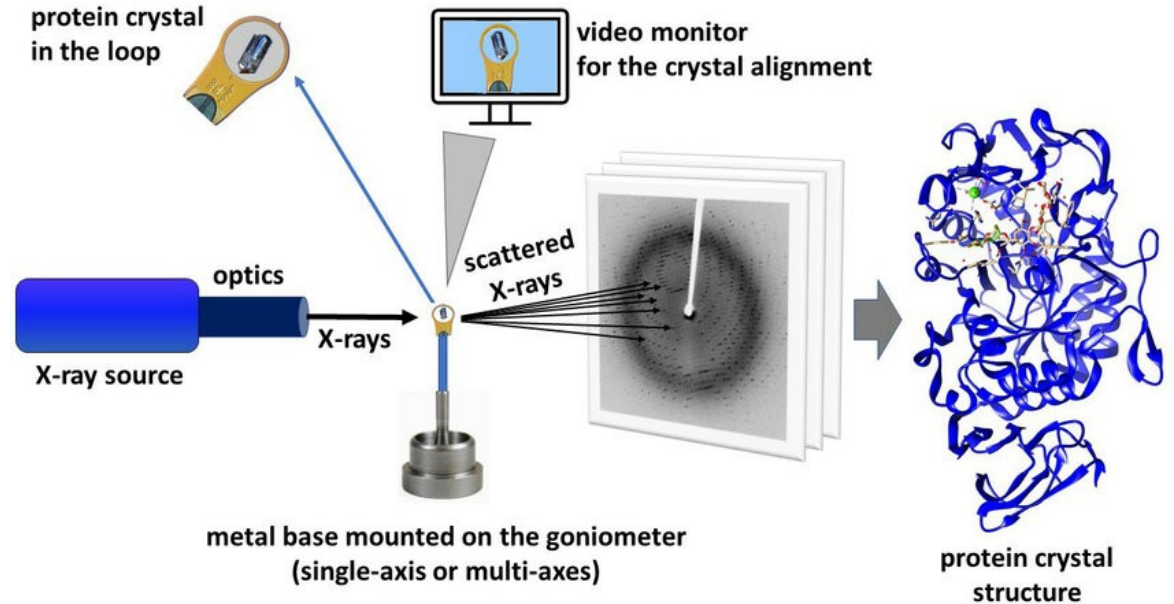
CONTENTS

Theory

1. Crystallogenesi*s*
2. Crystal symmetri*e*s
3. X-rays
4. Diffraction
5. Phase problem

Practice

6. Crystal harvesting
7. Data collection
8. Data processing
9. Solving structures
10. Examples



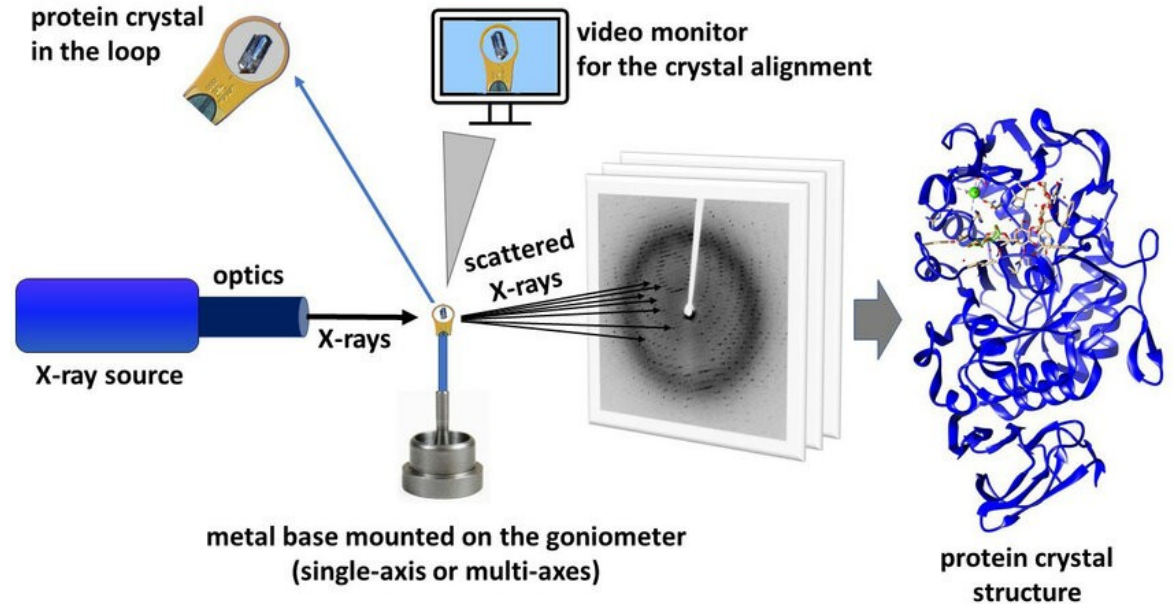
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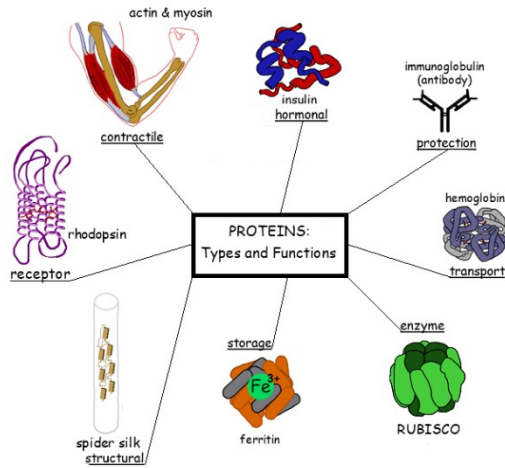
1. *Crystallogenesi*s
2. *Crystal symmetries*
3. *X-rays*
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5. *Phase problem*

Practice

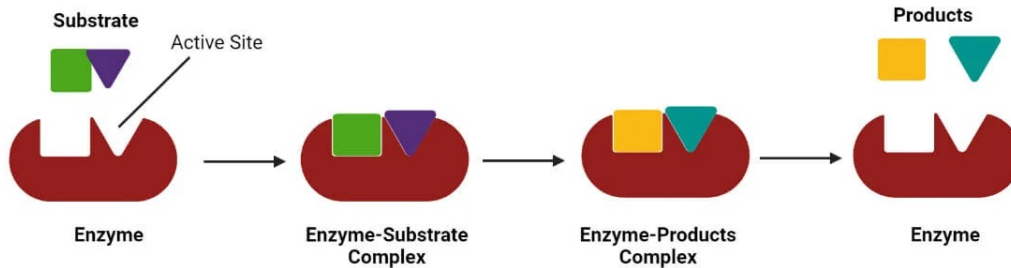
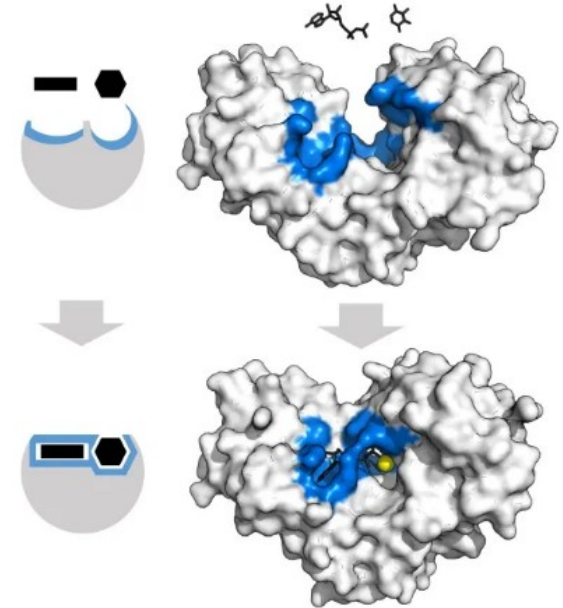
6. *Crystal harvesting*
7. *Data collection*
8. *Data processing*
9. *Solving structures*
10. *Examples*



LOCK-AND-KEY MODEL (1894)

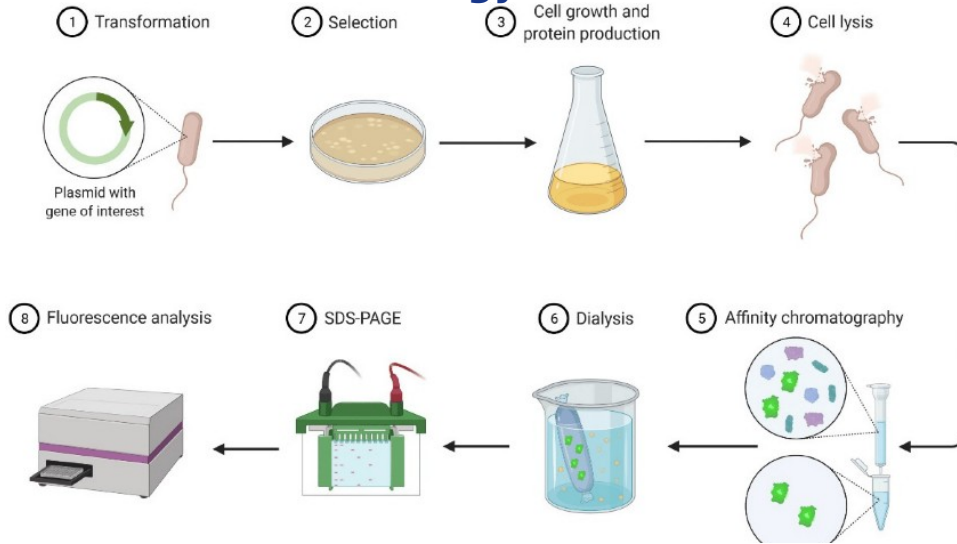


Emil Fischer (1852-1919)
Nobel prize in Chemistry(1902)

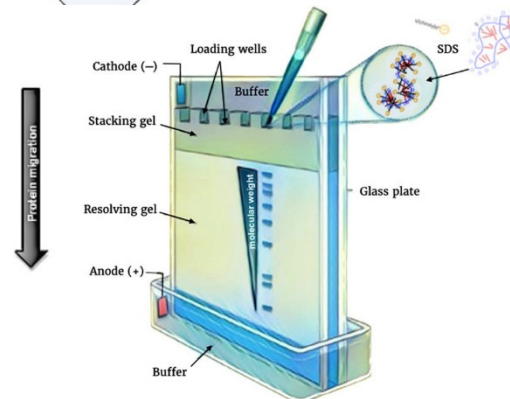


PURIFICATION

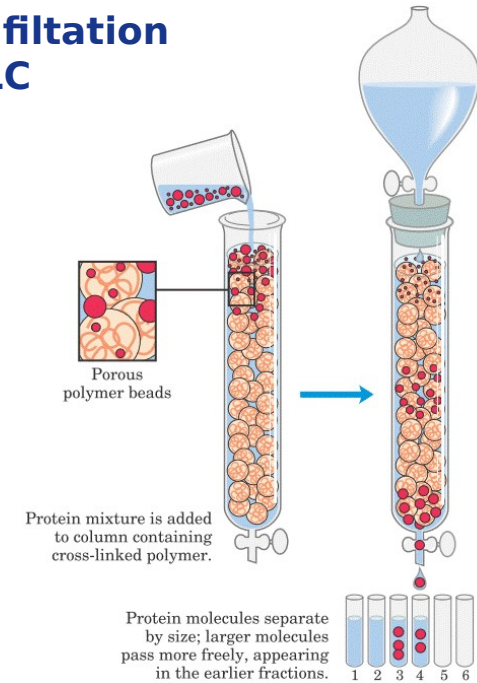
Molecular biology



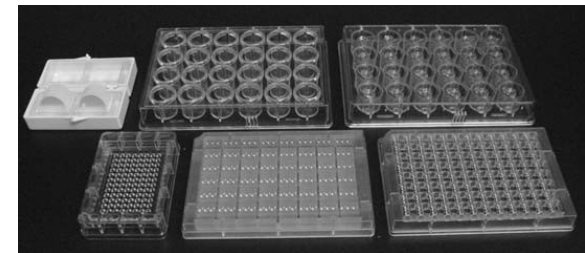
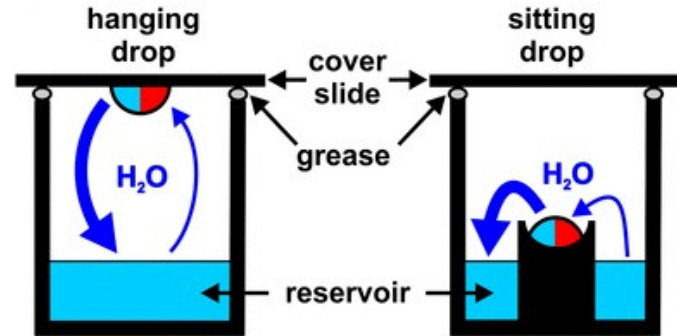
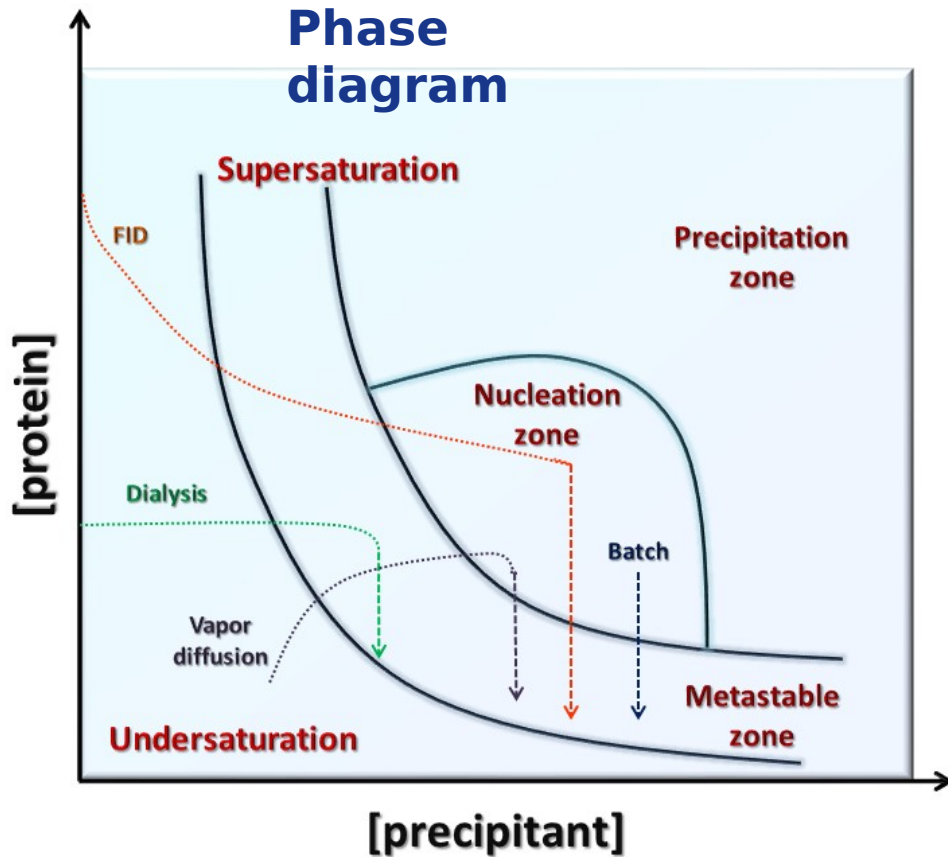
SDS-PAGE electrophoresis



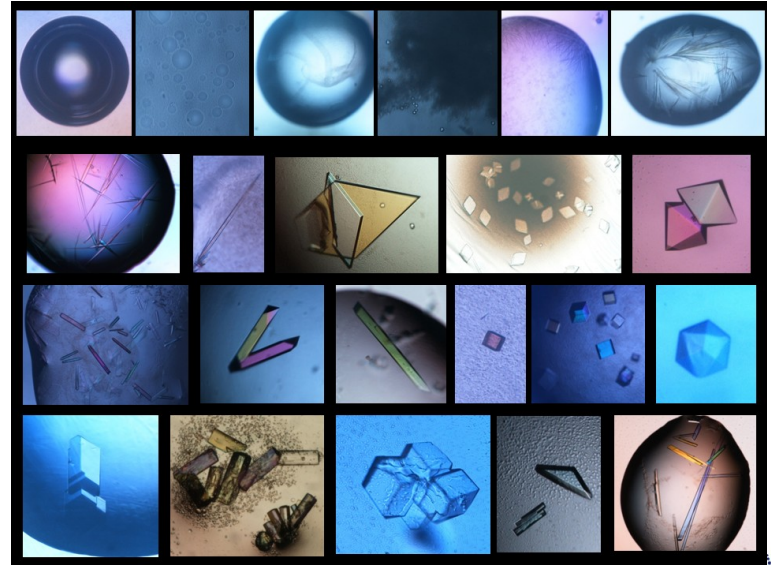
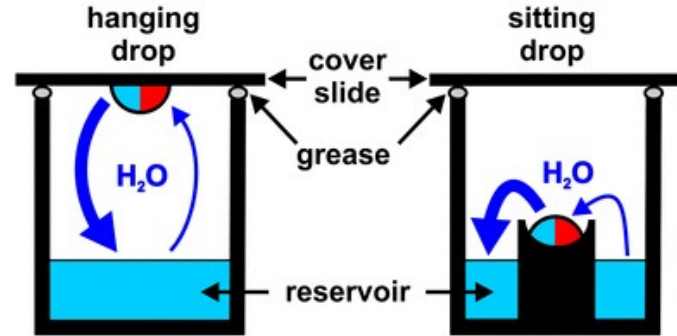
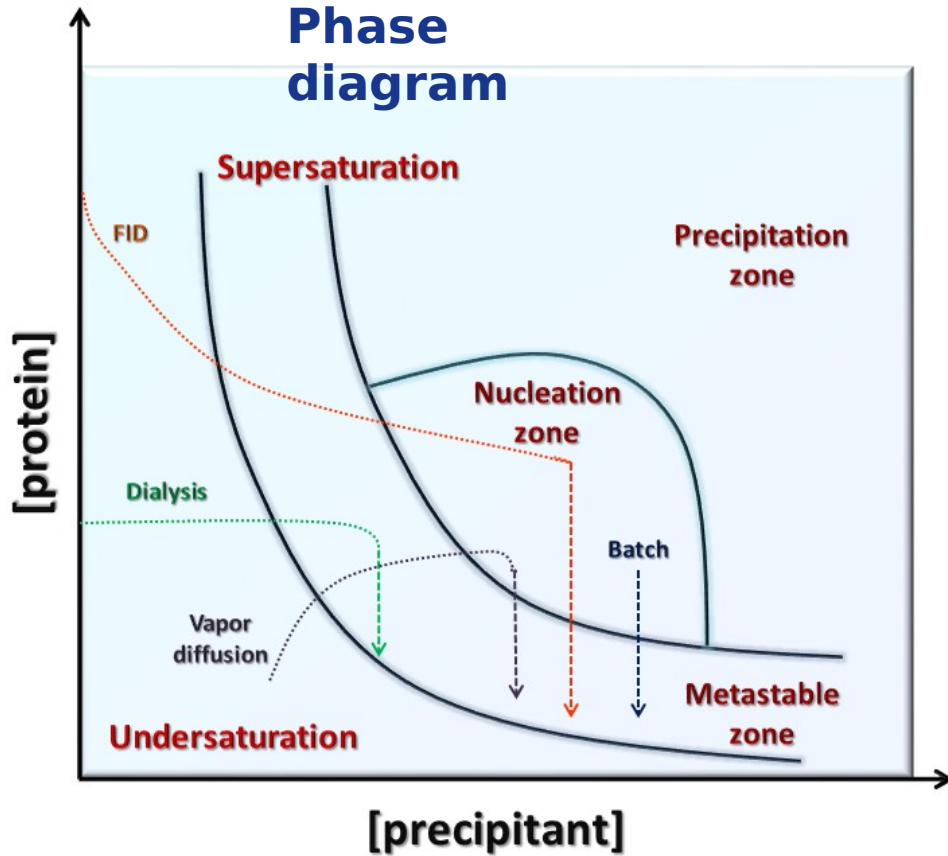
Gel filtration HPLC



CRYSTALLOGENESIS



CRYSTALLOGENESIS



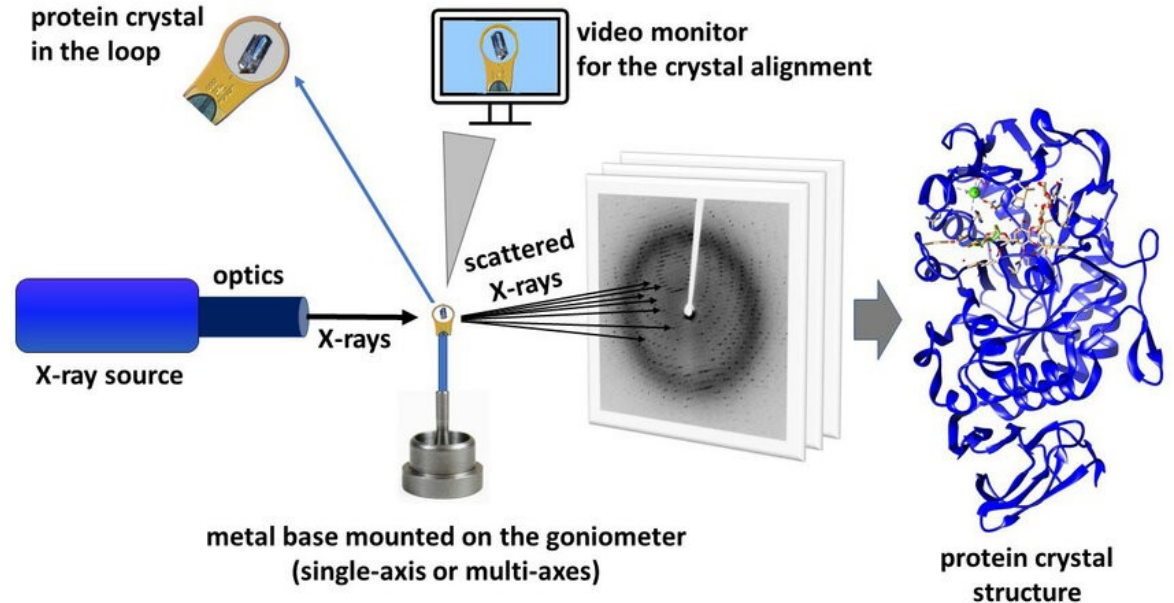
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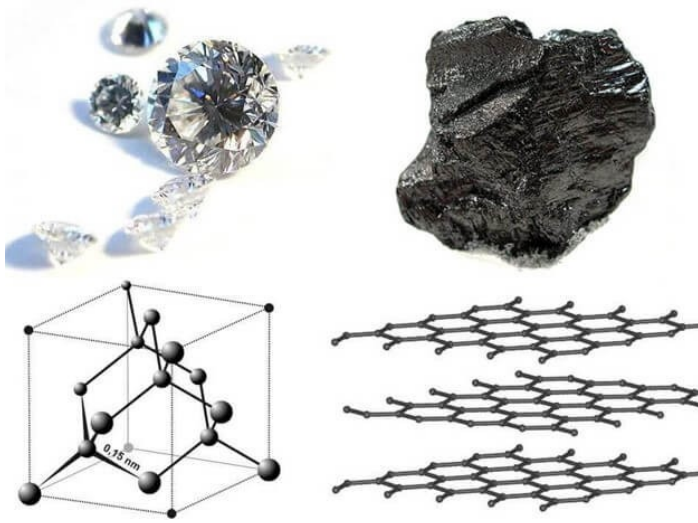


CRYSTALS


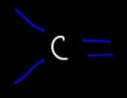
Steno's Law (1669)

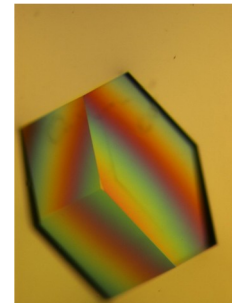
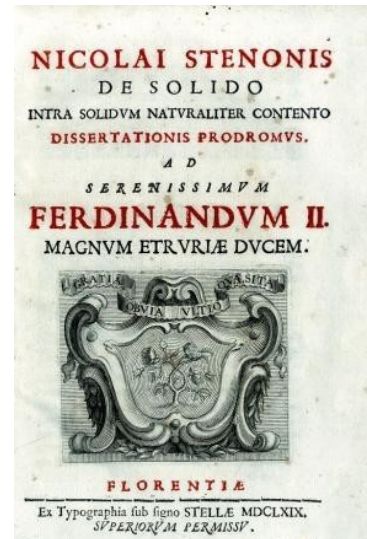
The **angles between corresponding faces** on crystals of any solid chemical or mineral species **are constant** and are characteristic of the species. The interfacial angle is measured between face normals.

The law constant of interfacial angles **holds for any crystals of a given species**, whether they are natural or hand-made, regardless of size or provenance.



Diamond vs Graphite

	
Tetrahedral	Trigonal Planar
$sp^3 \rightarrow 3D$	$sp^2 \rightarrow 2D$
Colorless	Black



7 LATTICE SYSTEMS

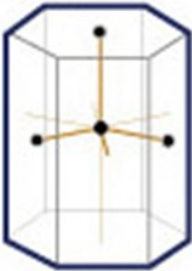
© AllAboutGemstones.com



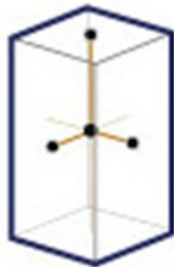
Cubic



Hexagonal



Tetragonal



Rhombohedral



Orthorhombic



Monoclinic

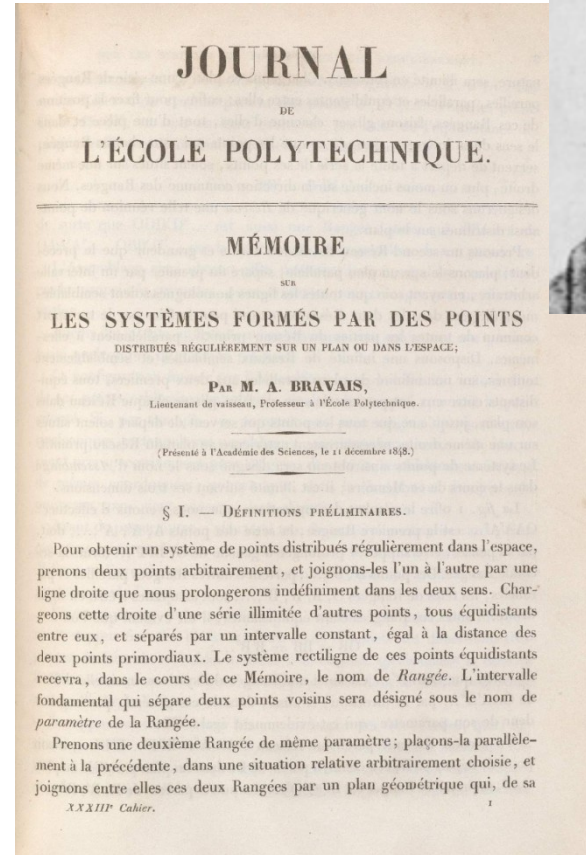


Triclinic



14 BRAVAIS LATTICES

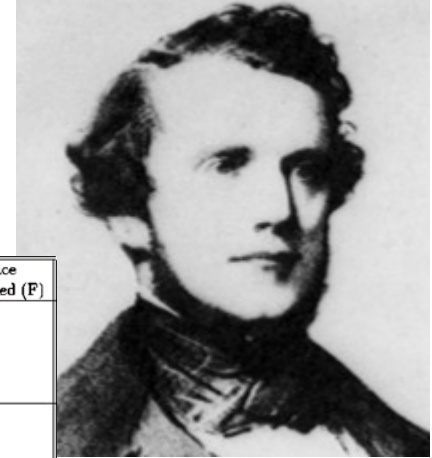
Crystal family	Lattice system	Point group (Schönflies notation)	14 Bravais lattices			
			Primitive (P)	Base-centered (S)	Body-centered (I)	Face-centered (F)
Triclinic (a)		C_1				
Monoclinic (m)		C_{2h}				
			mP	mS		
Orthorhombic (o)		D_{2h}				
			oP	oS	oI	oF
Tetragonal (t)		D_{4h}				
			tP	tI		
Hexagonal (h)	Rhombohedral	D_{3d}				
	Hexagonal	D_{6h}				
Cubic (c)		O_h				
			cP	cI	cF	



Auguste Bravais(1811-1863)

14 BRAVAIS LATTICES

$$\mathbf{R} = n_1 \mathbf{a}_1 + n_2 \mathbf{a}_2 + n_3 \mathbf{a}_3$$



Auguste Bravais(1811-1863)

Crystal family	Lattice system	Point group (Schönflies notation)	14 Bravais lattices					
			Primitive (P)	Base-centered (S)	Body-centered (I)	Face-centered (F)		
Triclinic (a)		C_1						
Monoclinic (m)		C_{2h}						
			mP	mS				
Orthorhombic (o)		D_{2h}						
			oP	oS	oI	oF		
			Tetragonal (t)	D_{4h}				
					tP		tI	
Hexagonal (h)	Rhombohedral	D_{3d}						
	hR							
	Hexagonal	D_{6h}						
	hP							
Cubic (c)		O_h						
			cP		cI	cF		

Bravais lattice	Parameters	Simple (P)	Volume centered (I)	Base centered (C)	Face centered (F)
Triclinic	$a_1 \neq a_2 \neq a_3$ $\alpha_{12} \neq \alpha_{23} \neq \alpha_{31}$				
Monoclinic	$a_1 \neq a_2 \neq a_3$ $\alpha_{23} = \alpha_{31} = 90^\circ$ $\alpha_{12} \neq 90^\circ$				
Orthorhombic	$a_1 \neq a_2 \neq a_3$ $\alpha_{12} = \alpha_{23} = \alpha_{31} = 90^\circ$				
Tetragonal	$a_1 = a_2 \neq a_3$ $\alpha_{12} = \alpha_{23} = \alpha_{31} = 90^\circ$				
Trigonal	$a_1 = a_2 = a_3$ $\alpha_{12} = \alpha_{23} = \alpha_{31} < 120^\circ$				
Cubic	$a_1 = a_2 = a_3$ $\alpha_{12} = \alpha_{23} = \alpha_{31} = 90^\circ$				
Hexagonal	$a_1 = a_2 \neq a_3$ $\alpha_{12} = 120^\circ$ $\alpha_{23} = \alpha_{31} = 90^\circ$				

32 POINT GROUPS

A group of point symmetry operations leave at least one point unmoved. Lattice translation is not considered in point group.



Johann Hessel (1796-1872)

Crystal family	Crystal system	Group names						
Cubic		23	$m\bar{3}$		432	$\bar{4}3m$	$m\bar{3}m$	
Hexagonal	Hexagonal	6	$\bar{6}$	$6/m$	622	6mm	$\bar{6}m2$	6/mmm
	Trigonal	3	$\bar{3}$		32	3m	$\bar{3}m$	
Tetragonal		4	$\bar{4}$	$4/m$	422	4mm	$\bar{4}2m$	4/mmm
Orthorhombic					222		mm2	mmm
Monoclinic		2		$2/m$		m		
Triclinic		1	$\bar{1}$					

230 SPACE GROUPS

Hermann-Mauguin symbol (internat. Table vol. A)

$P2_12_12_1$

D_2^4

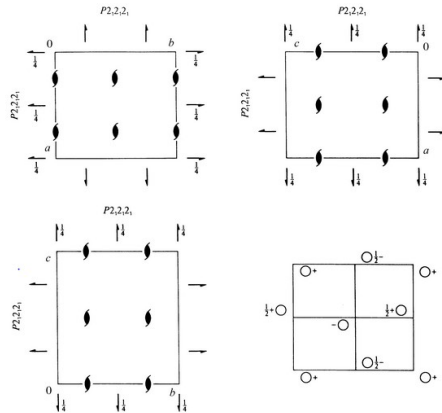
222

No. 19

$P2_12_12_1$

Orthorhombic

Patterson symmetry $Pmmm$



Origin at midpoint of three non-intersecting pairs of parallel 2_1 axes

Asymmetric unit $0 \leq x \leq 1/2; 0 \leq y \leq 1/2; 0 \leq z \leq 1$

Symmetry operations

(1) 1 (2) $2(0, 0, 1/2)$ $1/4, 0, z$ (3) $2(0, 1/2, 0)$ $0, y, 1/4$ (4) $2(1/2, 0, 0)$ $x, 1/4, 0$

Generators selected (1); $\kappa(1, 0, 0)$; $\kappa(0, 1, 0)$; $\kappa(0, 0, 1)$; (2); (3)

Positions

Multiplicity, Wyckoff letter, Site symmetry	Coordinates				Reflection conditions
4 a 1	(1) x, y, z	(2) $x+1/2, -y, z+1/2$	(3) $-x, y+1/2, -z+1/2$	(4) $x+1/2, -y+1/2, -z$	General: $h0l: h=2n$ $0k0: k=2n$ $00l: l=2n$



Evgraf Fedorov (1853-1919)

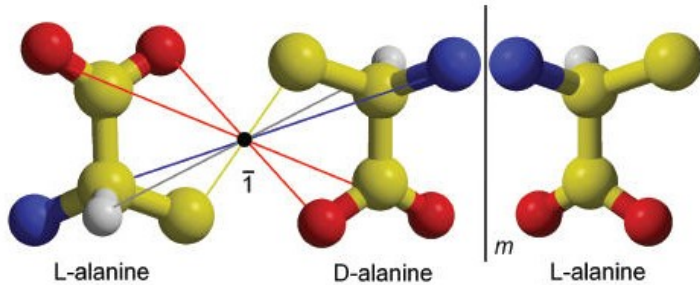
Triclinic	None		
Monoclinic*	[010] ('unique axis b') [001] ('unique axis c')		
Orthorhombic	[100]	[010]	[001]
Tetragonal	[001]	$\begin{Bmatrix} [100] \\ [010] \end{Bmatrix}$	$\begin{Bmatrix} [1\bar{1}0] \\ [110] \end{Bmatrix}$
Hexagonal	[001]	$\begin{Bmatrix} [100] \\ [010] \\ [\bar{1}10] \end{Bmatrix}$	$\begin{Bmatrix} [1\bar{1}0] \\ [120] \\ [2\bar{1}0] \end{Bmatrix}$
Rhombohedral (hexagonal axes)	[001]	$\begin{Bmatrix} [100] \\ [010] \\ [\bar{1}10] \end{Bmatrix}$	
Rhombohedral (rhombohedral axes)	[111]	$\begin{Bmatrix} [1\bar{1}0] \\ [01\bar{1}] \\ [\bar{1}01] \end{Bmatrix}$	
Cubic	$\begin{Bmatrix} [100] \\ [010] \\ [001] \end{Bmatrix}$	$\begin{Bmatrix} [111] \\ [1\bar{1}\bar{1}] \\ [\bar{1}\bar{1}1] \\ [\bar{1}11] \end{Bmatrix}$	$\begin{Bmatrix} [1\bar{1}0] & [110] \\ [01\bar{1}] & [011] \\ [\bar{1}01] & [101] \end{Bmatrix}$

SPACE GROUPS IN MX

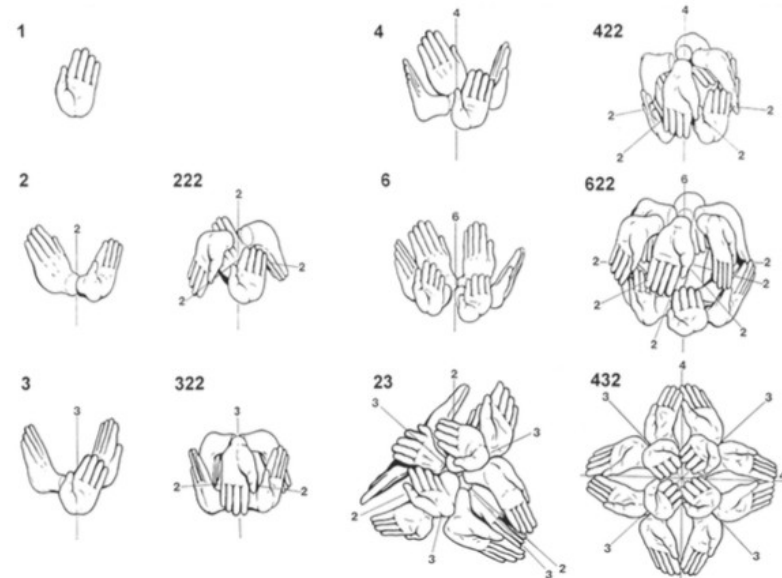
11 point group in proteins

Crystal family	Crystal system	Group names						
Cubic		23	$m\bar{3}$	432	$\bar{4}3m$	$m\bar{3}m$		
	Hexagonal	6	$\bar{6}$	$\frac{6}{m}$	622	6mm	$\bar{6}m2$	6/mmm
Hexagonal	Trigonal	3	$\bar{3}$		32	3m	$\bar{3}m$	
		4	$\bar{4}$	$\frac{4}{m}$	422	4mm	$\bar{4}2m$	4/mmm
Tetragonal					222		mm2	mmm
Orthorhombic		2		$\frac{2}{m}$		m		
Monoclinic		1	$\bar{1}$					
Triclinic								

Combination of point groups and Bravais lattices leaves **65 space groups** for protein crystals (chiral objects)

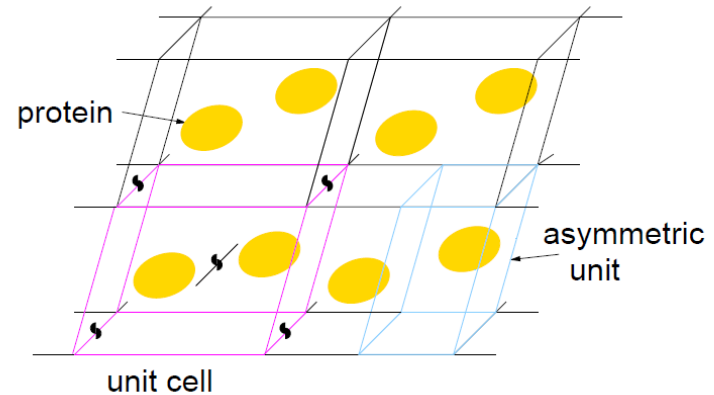
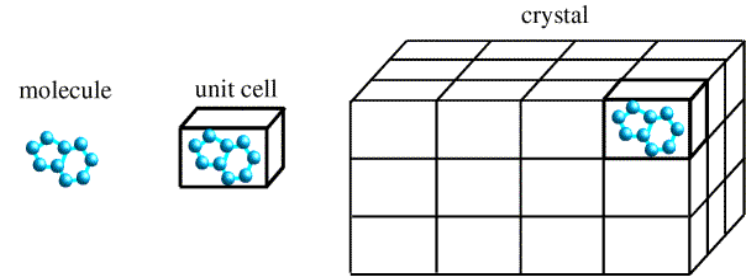
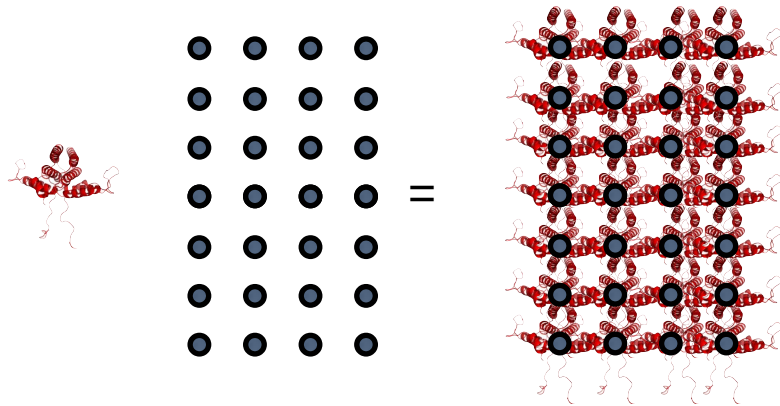


Combination of all point symmetry operations generates **32 point groups** but for proteins, only **11** are allowed



PROTEIN CRYSTAL

Crystal is a convolution of the molecule over the lattice



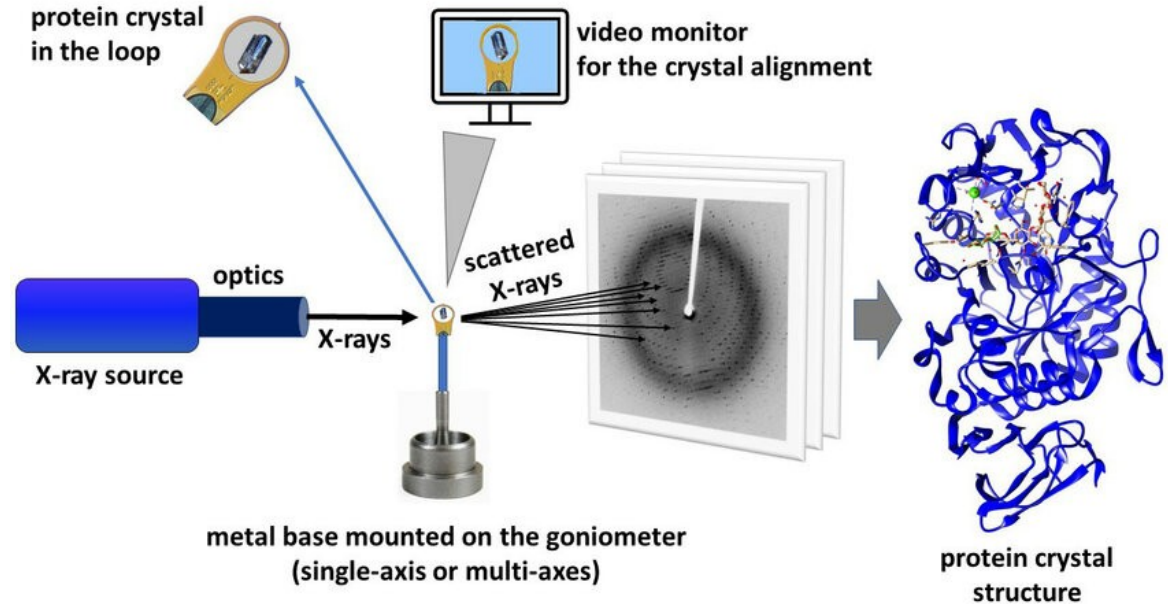
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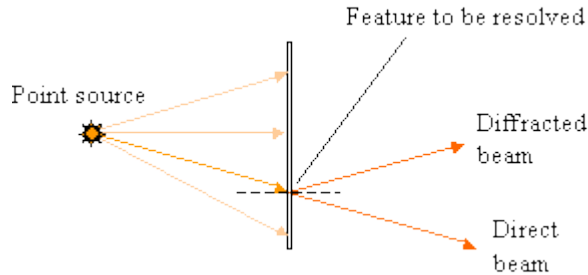
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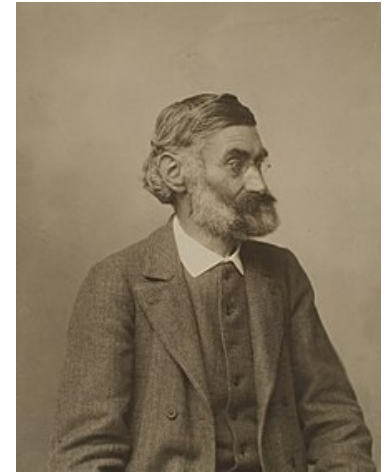
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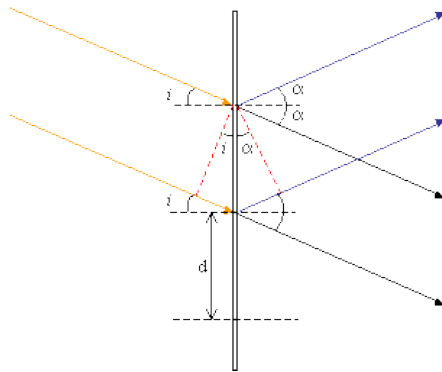
HOW TO PROBE A CRYSTAL?



$$d_{min}$$



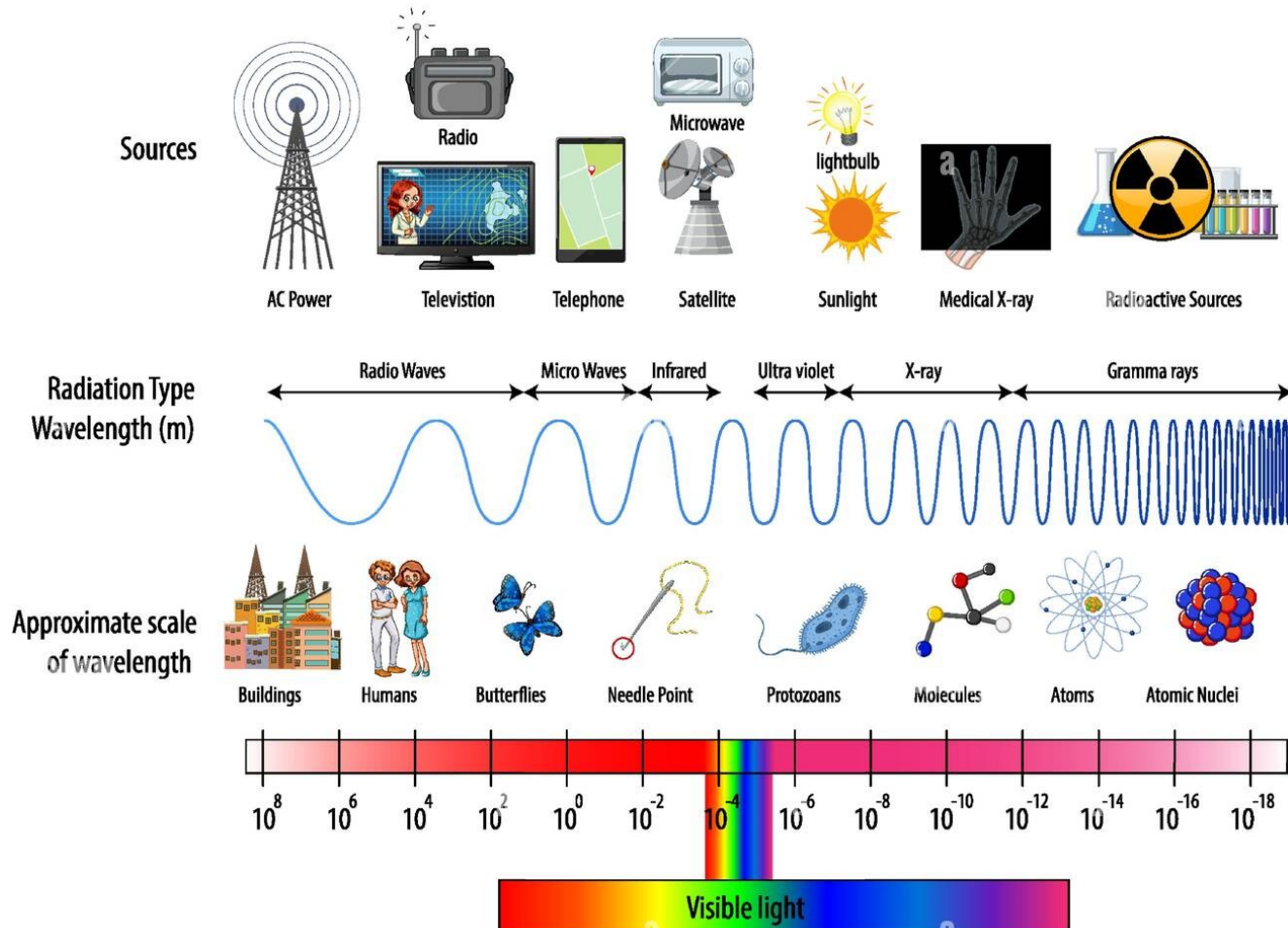
Ernst Abbe (1840-1905)



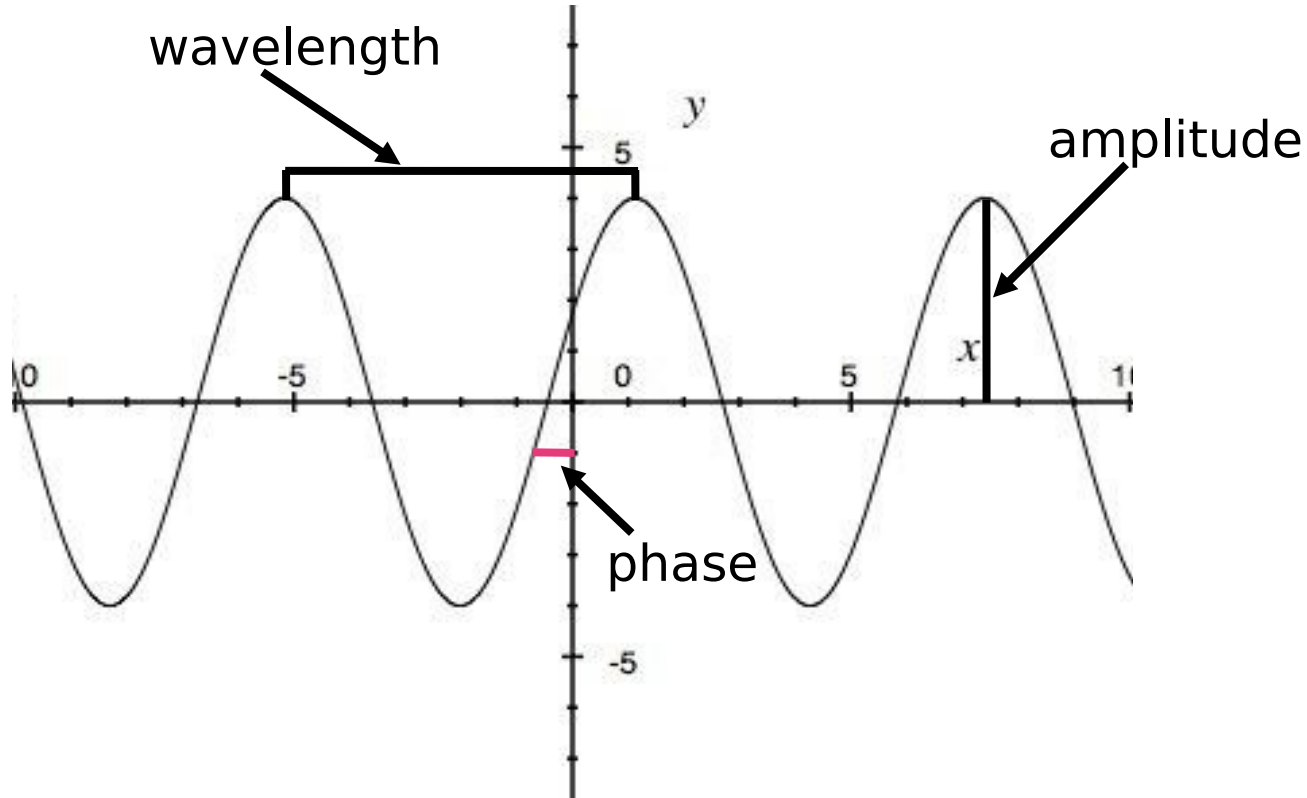
The limit of resolution (or resolving power) is a measure of the ability of the objective lens to separate in the image, adjacent details of the object.

n is the refractive index.

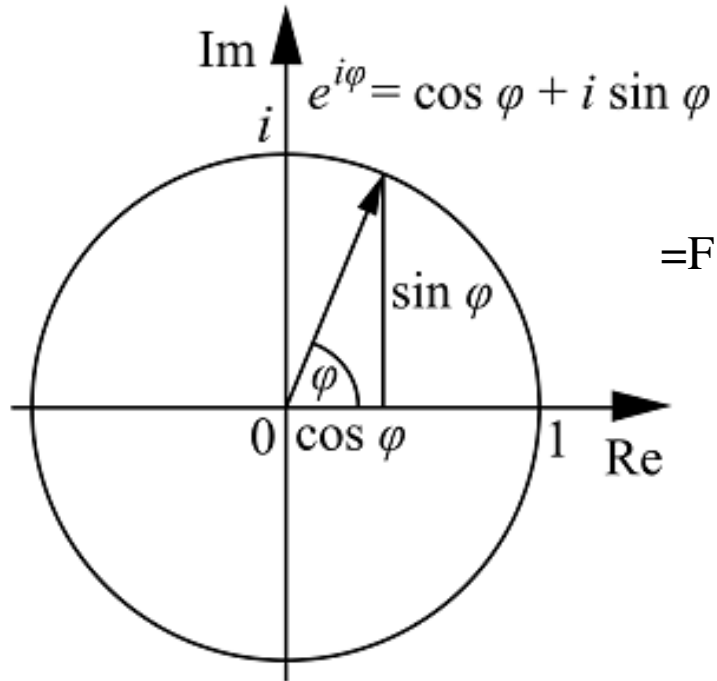
HOW TO PROBE A CRYSTAL?



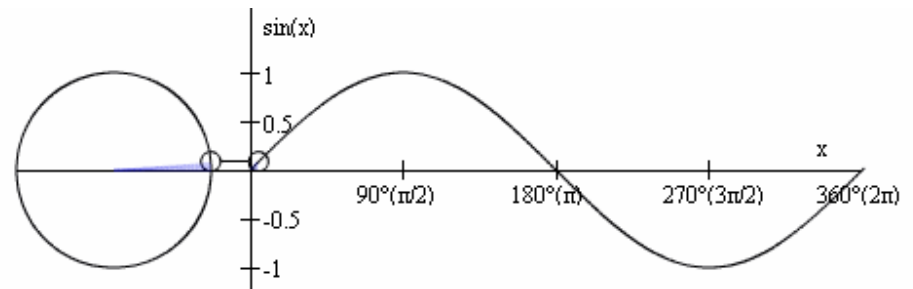
X-RAY AS A WAVE



XRAY AS A WAVE



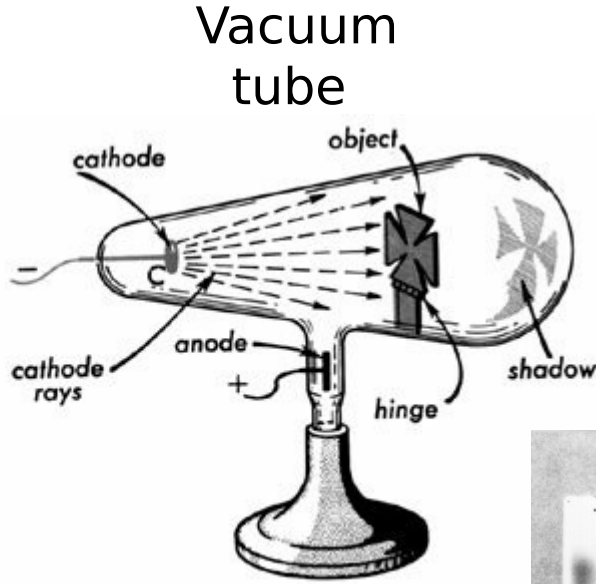
A complex number may represent a wave, defined by an Amplitude and a Phase (Euler and trigonometric functions)



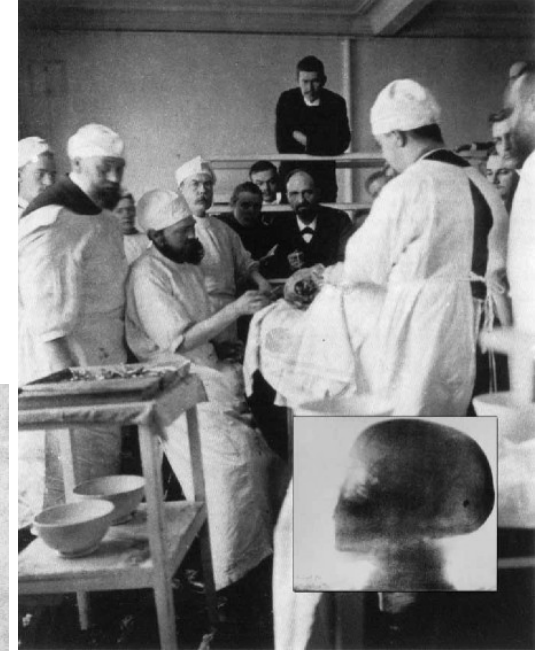
X-RAY PRODUCTION



William Crookes
(1832-1919)



Wilhelm Röntgen is usually credited as the discoverer of X-rays in 1895

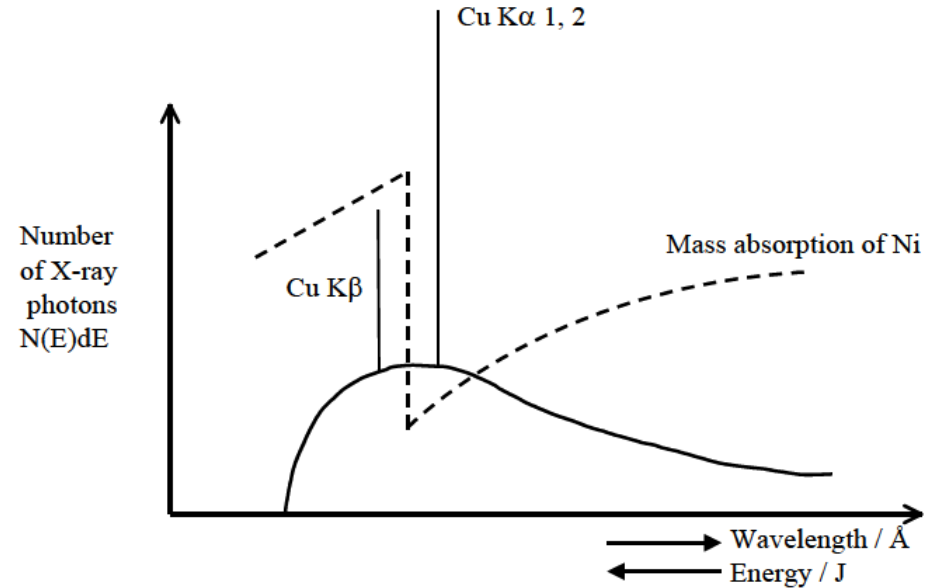
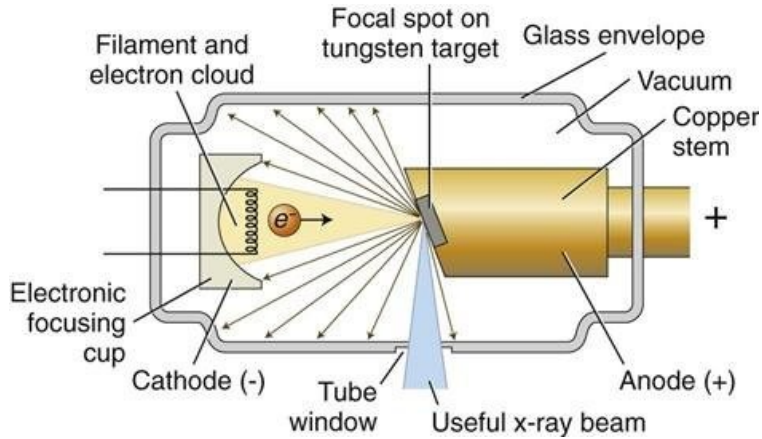


Medical applications of X-rays

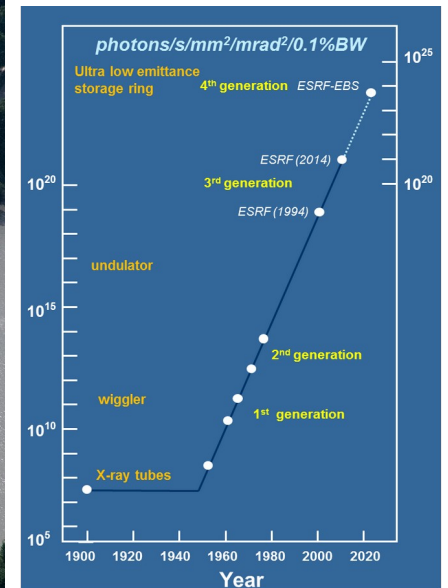
MONOCHROMATIC BEAM

Rotating anode

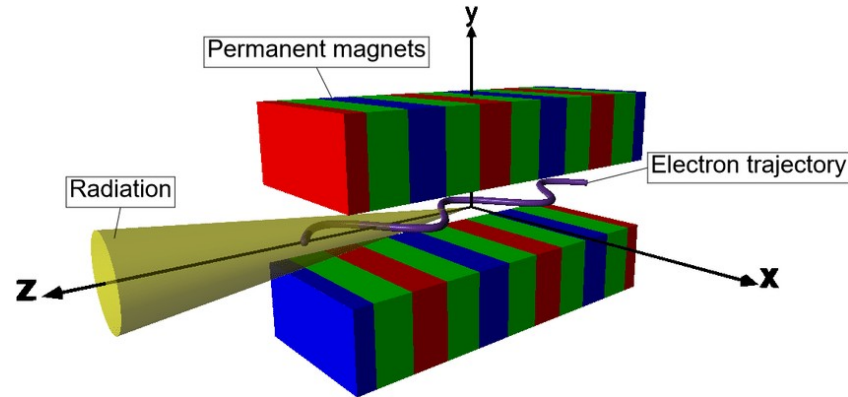
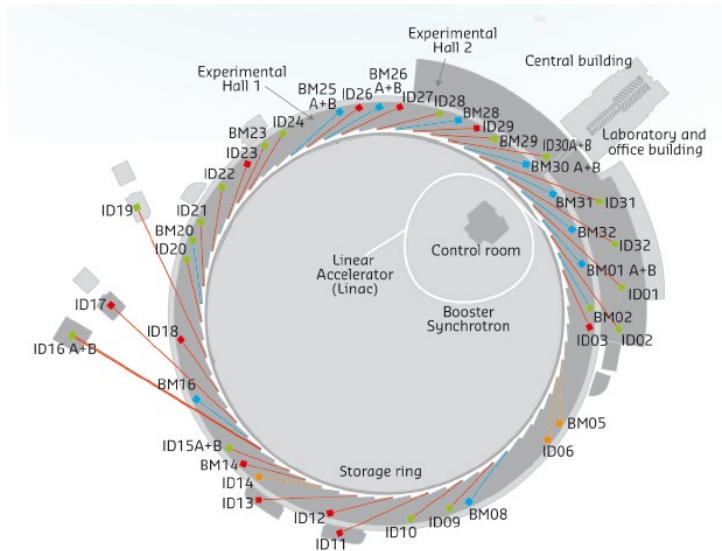
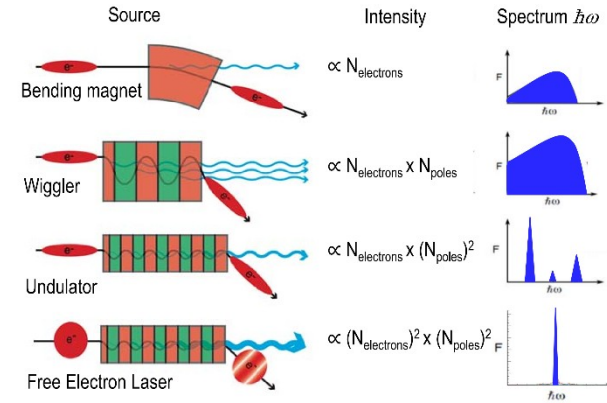
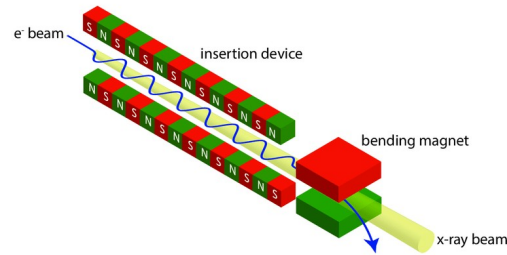
~50% of the the Cu $K\alpha$ is transmitted
($\sim 10^{10}$ ph/s/mm²)



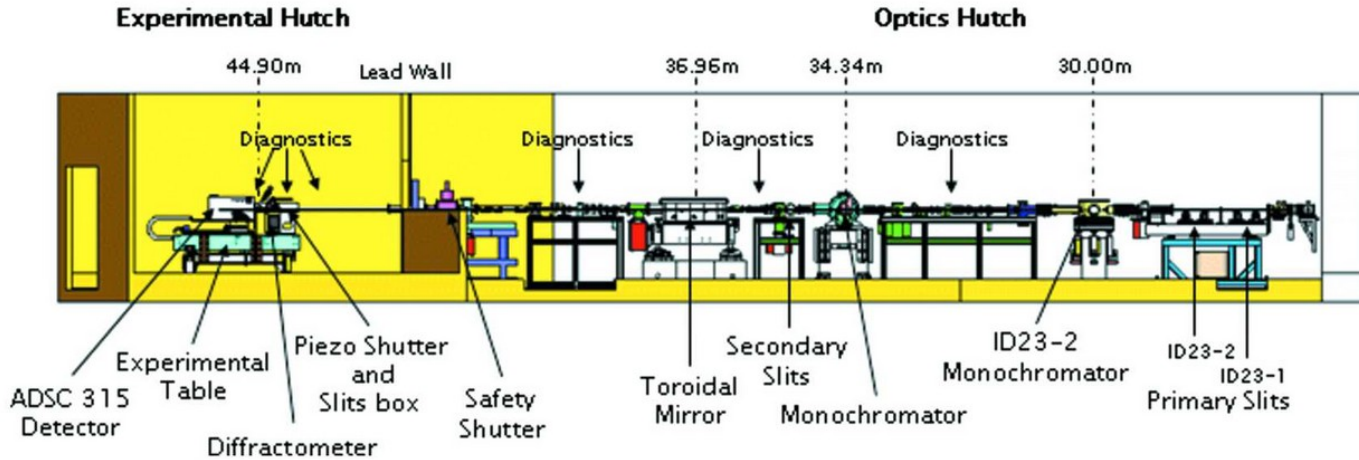
WHAT IS A SYNCHROTRON ?



HOW DOES IT WORK?



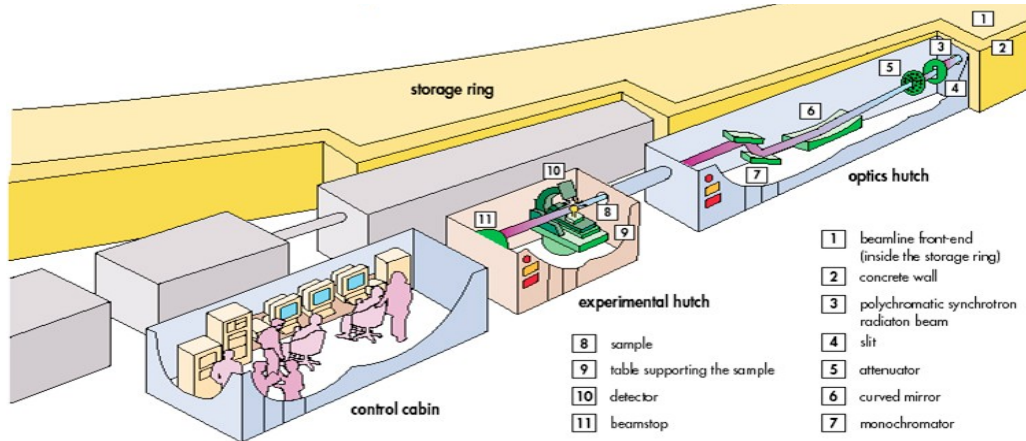
WHAT IS A BEAMLINE?



Size
$30\mu\text{m}$ \varnothing

Flux
> 10^{12} ph/s

Energy resolution



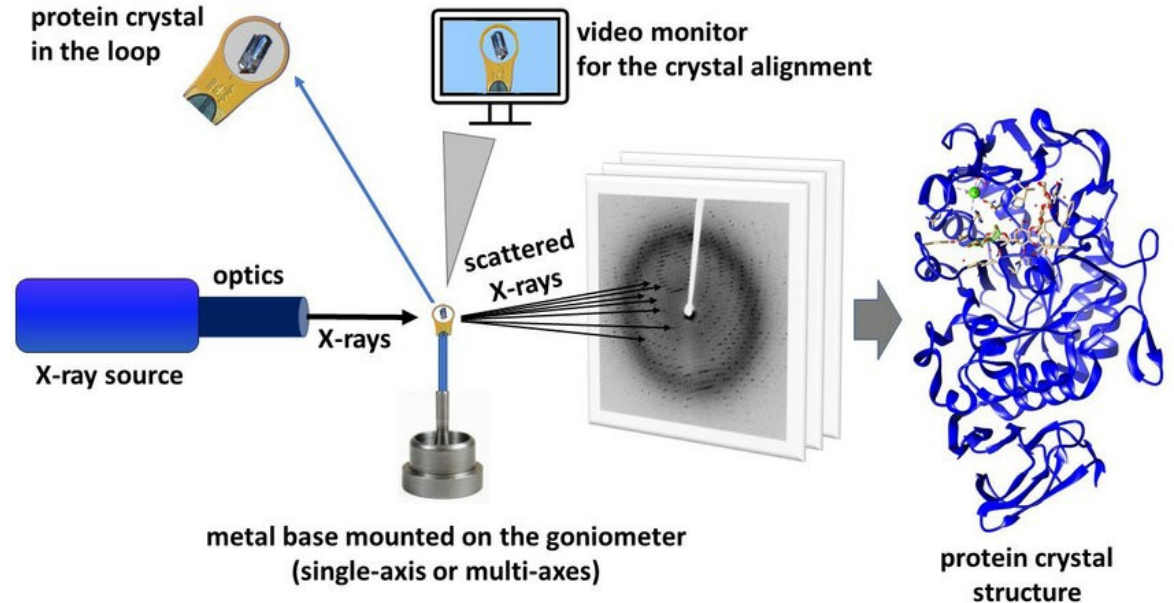
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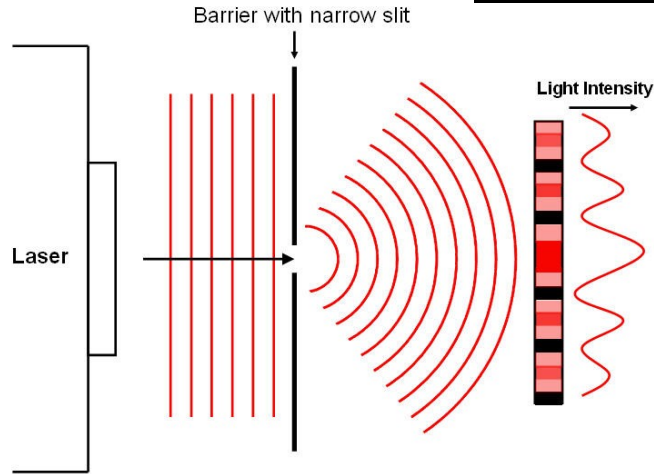


DIFFRACTION



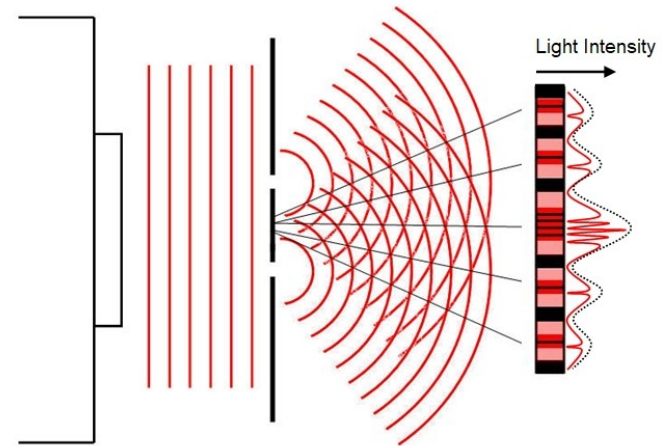
Waves interfere constructively in specific directions

Single Slit Diffraction



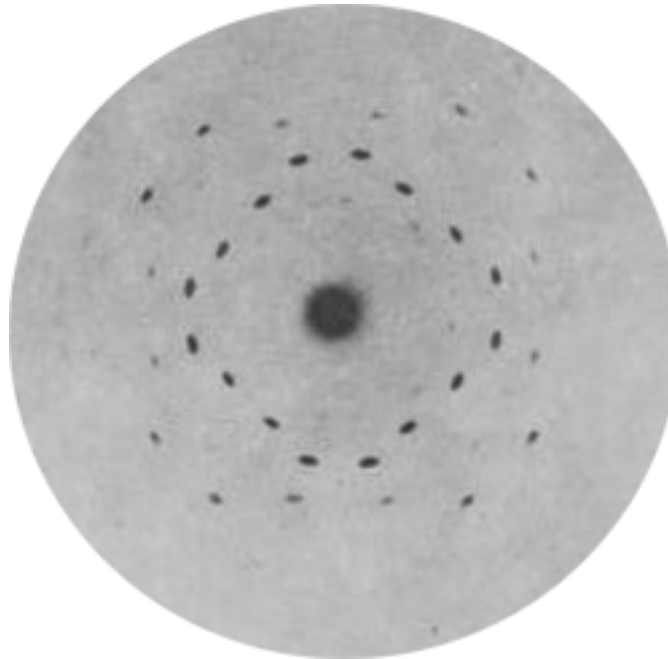
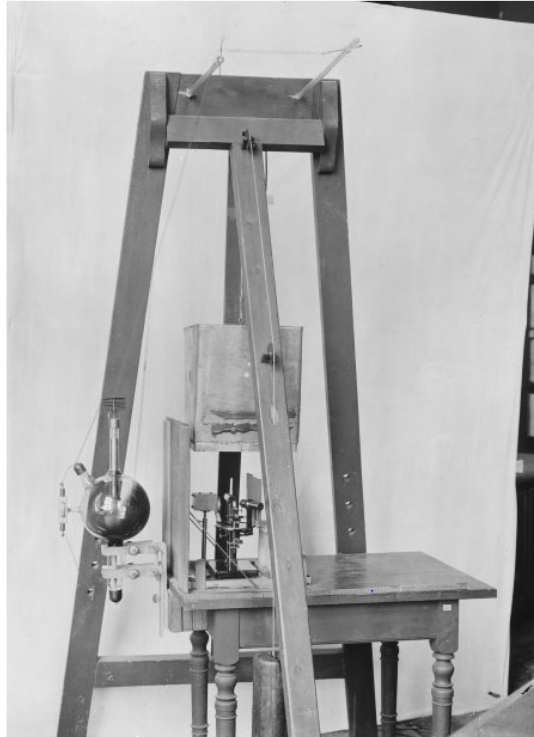
Plane waves through a slit - slit becomes source of spherical waves

Double Slit Diffraction

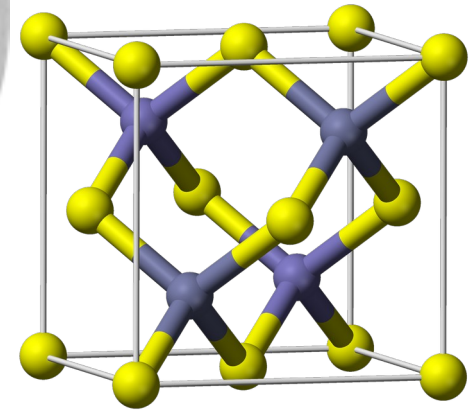


X-RAY DIFFRACTION

Max Laue's photo of X-ray diffraction from Zinc blende ZnS (Zn sulphide) cubic face centered (F3m)

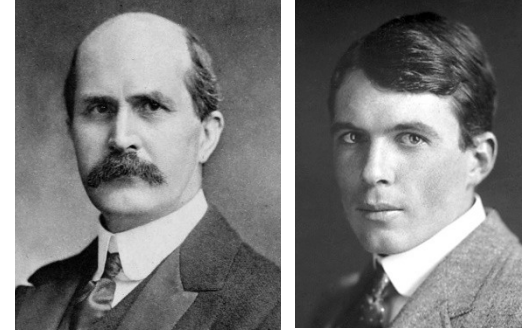


Max von Laue (1879-1960)
Nobel Prize 1914

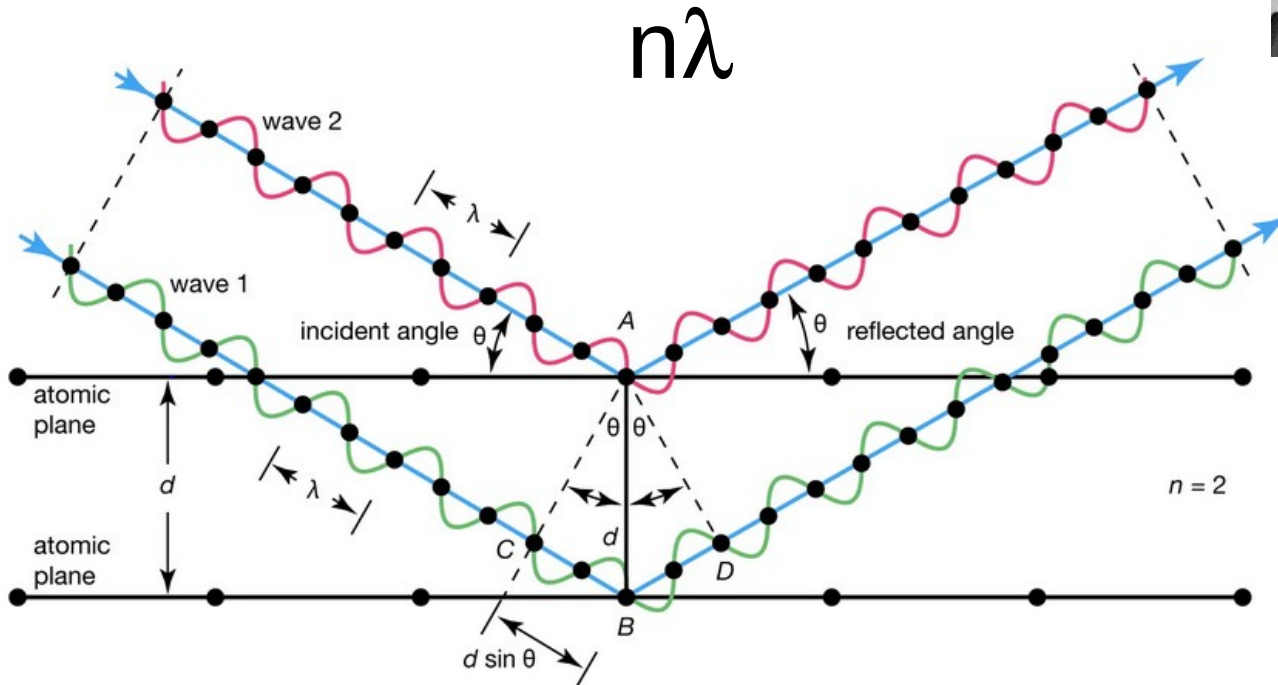


BRAGG'S LAW

Bragg diffraction describes the condition for constructive interference from **monochromatic** waves, with amplitude and phase, reflected by planes in the crystalline material



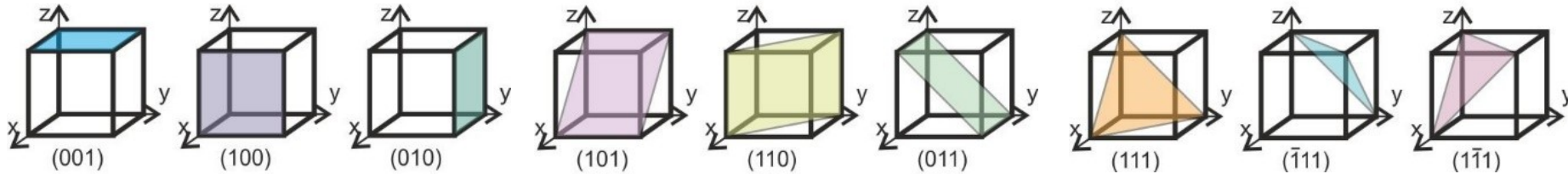
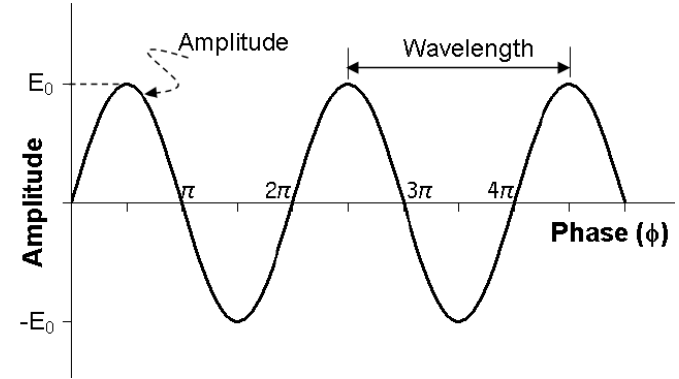
W. H & W. L Bragg
(1862-1942) – (1890-1971)
Nobel prize 1915



BRAGG'S LAW

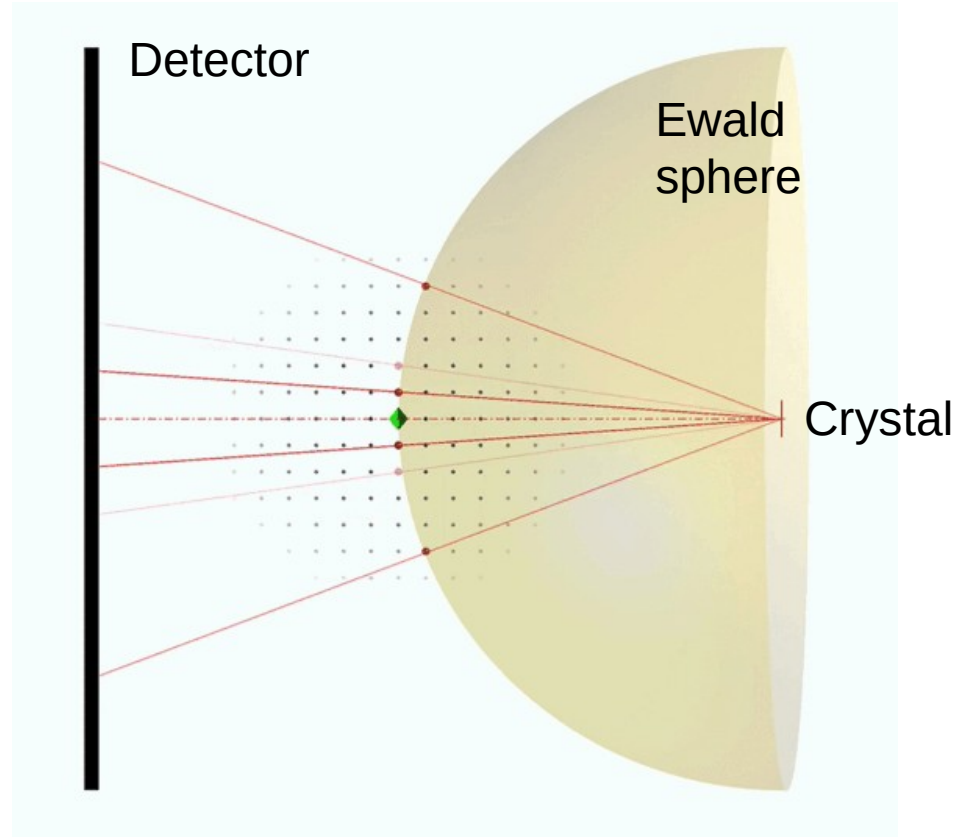
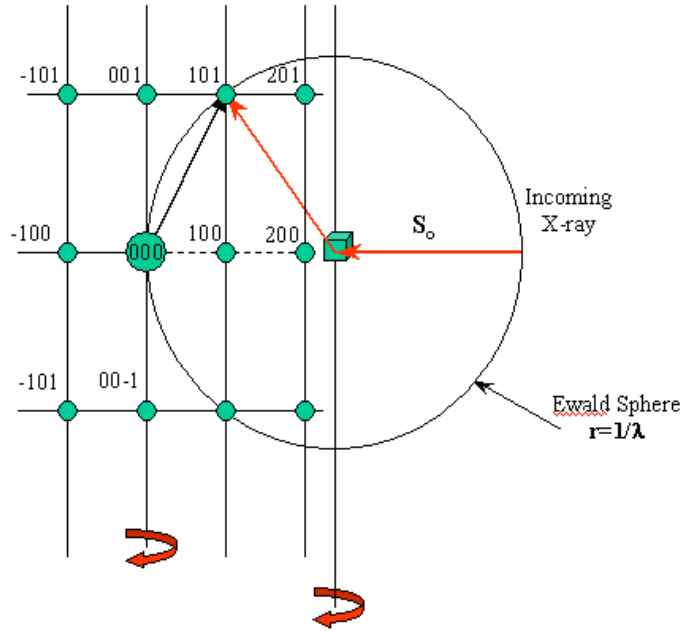
Bragg diffraction describes the condition for constructive interference from **monochromatic** waves, with amplitude and phase, reflected by planes in the crystalline material

$$n\lambda$$



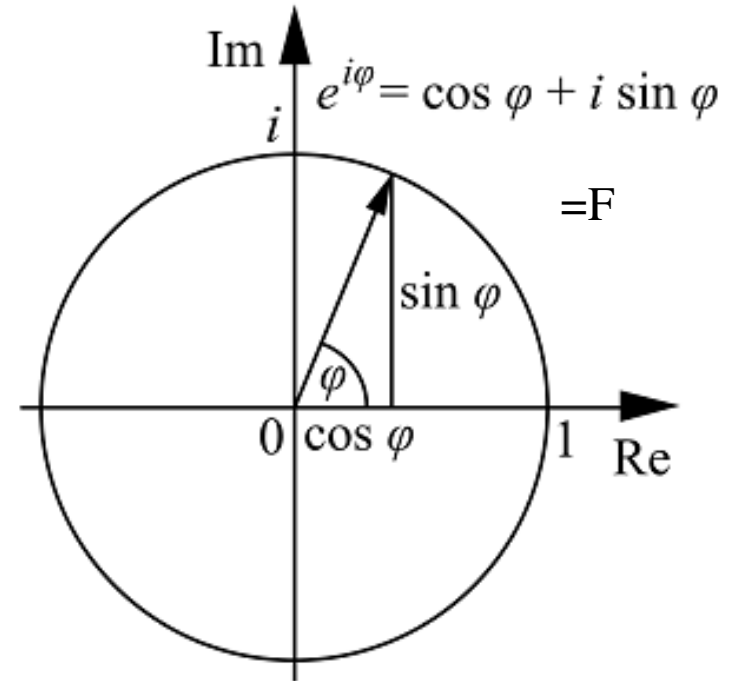
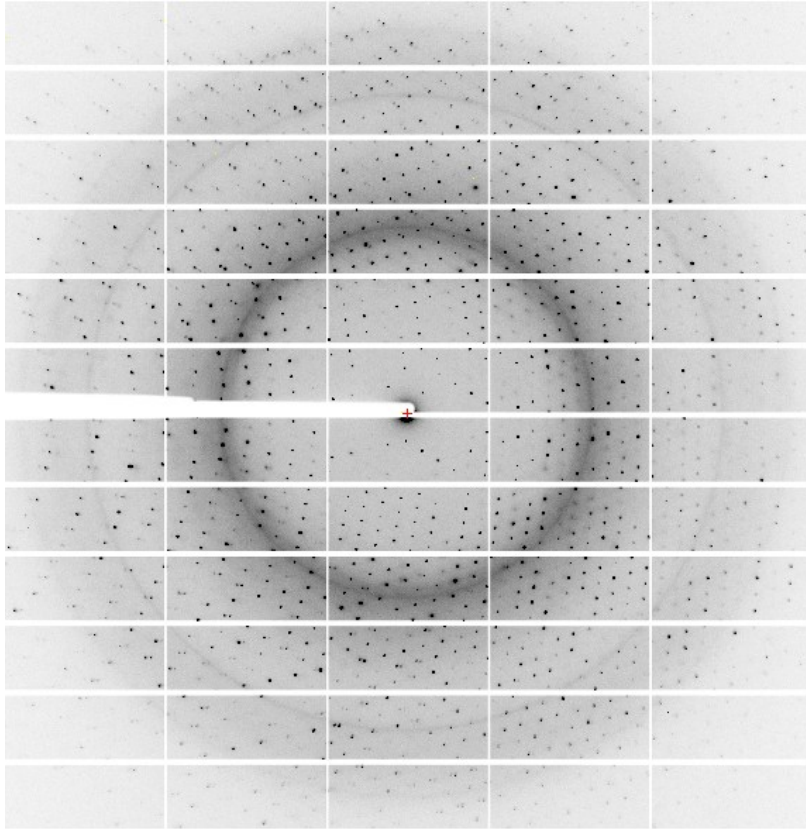
Each plane gives a spot of diffraction and this spot is defined by a triplet of integer hkl (Miller indices) in the reciprocal space

EWALD SPHERE



Each diffraction plane is represented as node

CRYSTAL DIFFRACTION



The 2D detector measures
Intensities of the diffraction peaks
BUT

DATASET STATISTICS

	Overall	Inner shell	Outer shell
Resolution (Å)	50-1.40	50 - 5.7	1.47 – 1.40
R_{merge}	0.052	0.022	1.047
R_{meas}	0.065	0.027	1.314
R_{pim}	0.038	0.015	0.785
$CC_{1/2}$	0.997	0.995	0.474
# observations	571512	9792	43402
# unique	127131	2224	9827
$\langle I \rangle / \sigma(I)$	12.8	50.8	1.2
Completeness	0.929	0.88	0.745
Multiplicity	4.5	4.4	4.4

$CC_{1/2}$ the intensities of two randomly sets of reflections correlates

$\langle I \rangle / \sigma(I)$ Intensity of the reflection over the error

Completeness, ratio between the # of unique observations and the total # theoretical reflections

Multiplicity, ratio between # of observed and unique reflections

$$R_{\text{merge}} = \frac{\sum_{hkl} \sum_{i=1}^n |I_i(hkl) - \bar{I}(hkl)|}{\sum_{hkl} \sum_{i=1}^n I_i(hkl)}$$

$$R_{\text{meas}} = \frac{\sum_{hkl} \sqrt{\frac{n}{n-1}} \sum_{i=1}^n |I_i(hkl) - \bar{I}(hkl)|}{\sum_{hkl} \sum_{i=1}^n I_i(hkl)}$$

$$R_{\text{pim}} = \frac{\sum_{hkl} \sqrt{1/n-1} \sum_{i=1}^n |I_i(hkl) - \bar{I}(hkl)|}{\sum_{hkl} \sum_{i=1}^n I_i(hkl)}$$

merged data

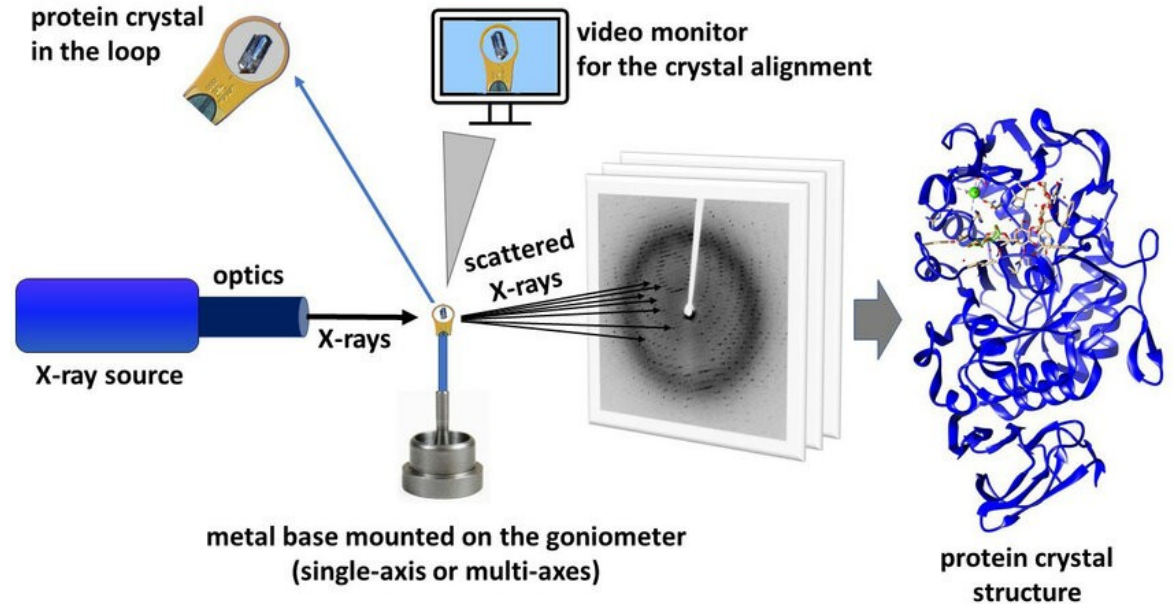
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Theory

1. Crystallogenesi
2. Crystal symmetries
3. X-rays
4. Diffraction
5. Phase problem
 1. SAD
 2. MR

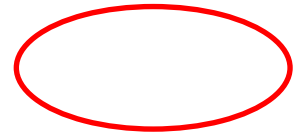
Practice

6. Crystal harvesting
7. Data collection
8. Data processing
9. Solving structures
10. Examples

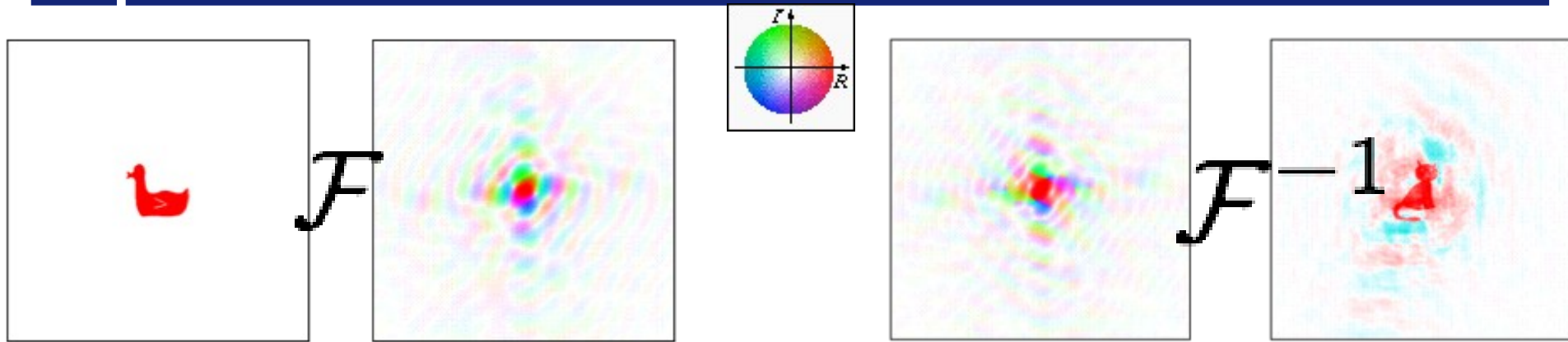


WHY RETRIEVING PHASES

the electron density over the volume of the cell
(Fourier transform)



WHY RETRIEVING PHASES



Phases are the most important information to obtain



THE PHASE PROBLEM

The electron density over the volume of the cell:



How to retrieve the phase?

Method

Direct (< 1000 non hydrogen / ua)

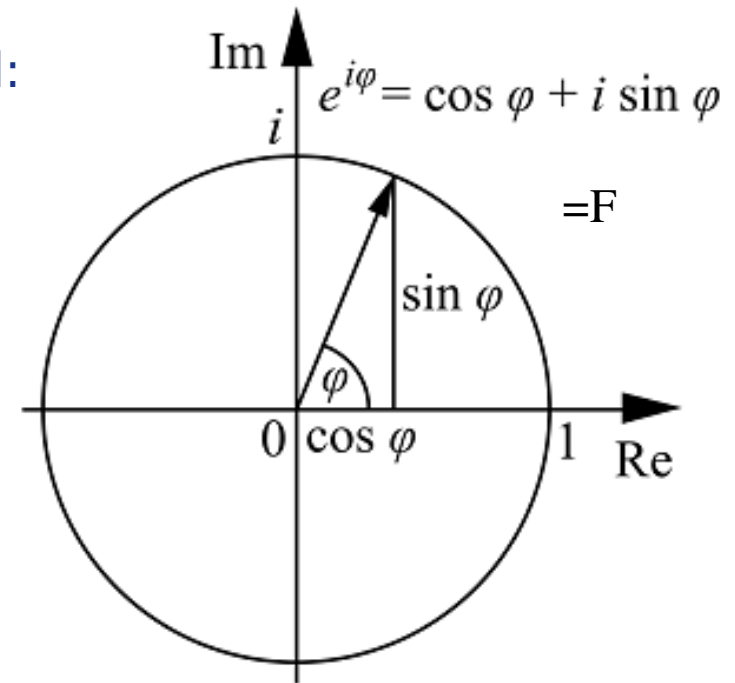
SIR, MIR

SIRAS, MIRAS

MAD, SAD

Molecular Replacement

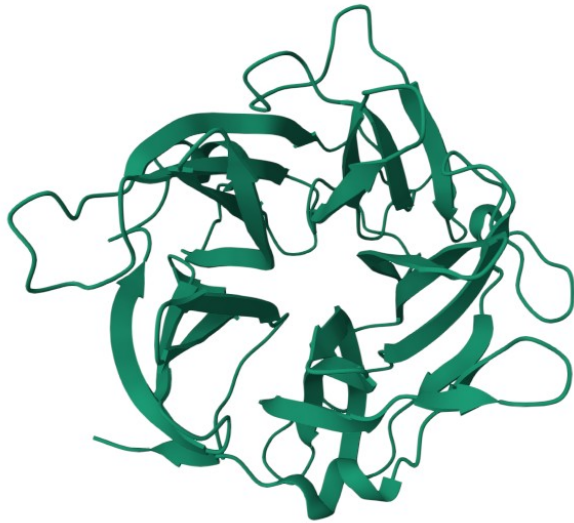
Isomorphous Replacement



The 2D detector measures Intensities of the diffraction peaks BUT



HEAVY ATOM DERIVATIVE

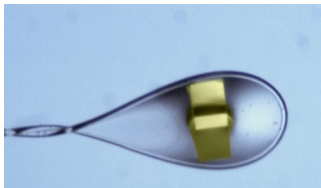


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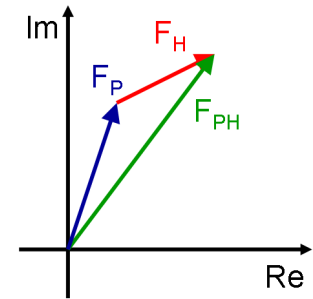


F_{ph}

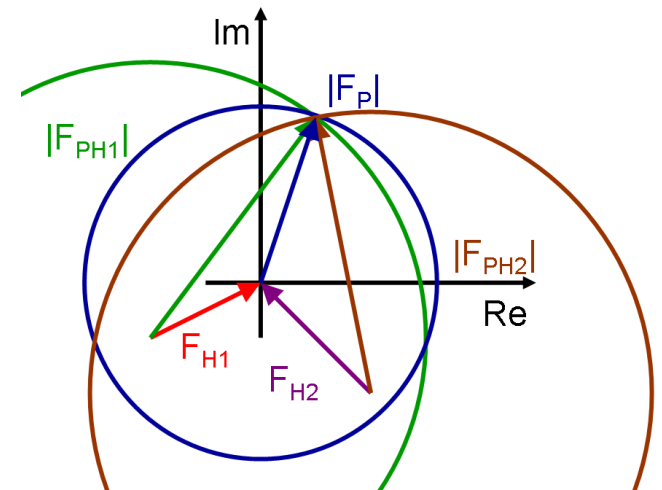
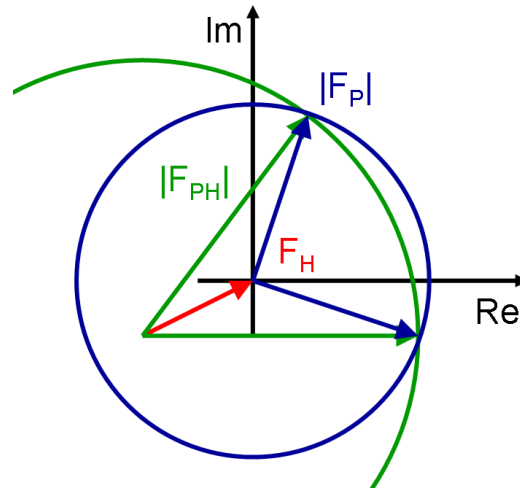
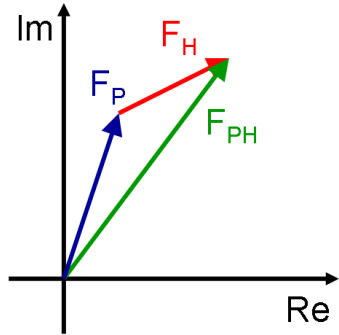
+



Soaking, noble gases
co-crystallization, Se-Met derivative,



HARKER CONSTRUCTION



THE PHASE PROBLEM

The electron density over the volume of the cell:



How to retrieve the phase?

Method

Direct (< 1000 non hydrogen / ua)

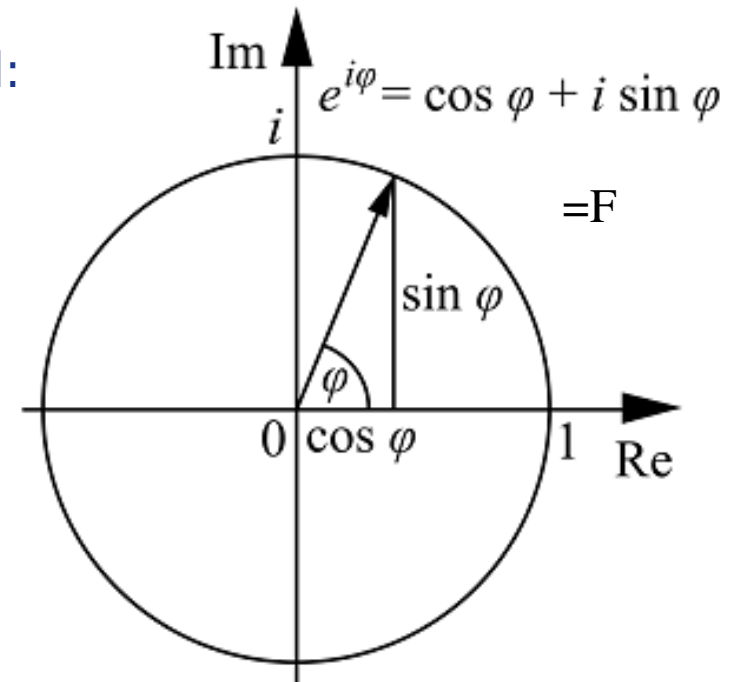
SIR, MIR

SIRAS, MIRAS

MAD, SAD

Molecular Replacement

*Isomorphous
Replacement*



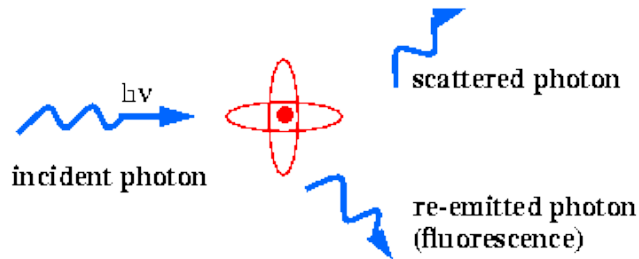
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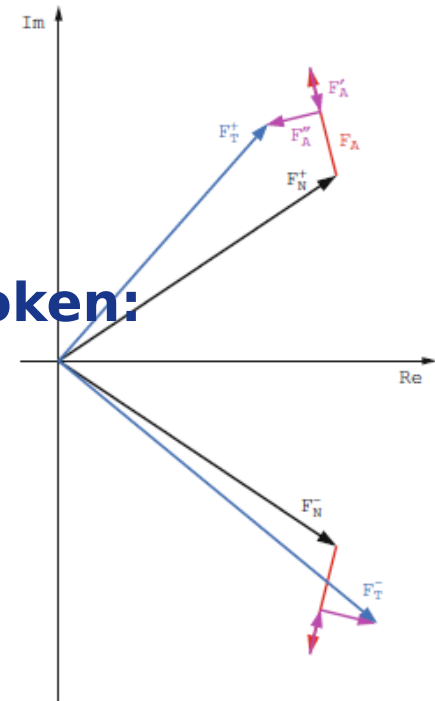
FRIEDEL'S LAW

When incident X-ray energy matches the binding energy of the electrons of the heavy atom, the absorption of X-rays increases.

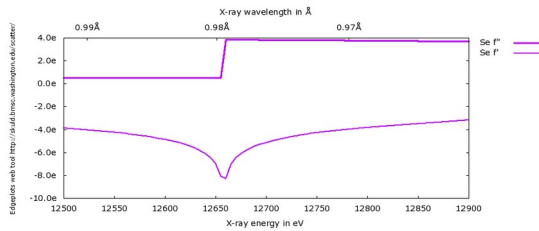
The scattering factor of the HA takes the form of :



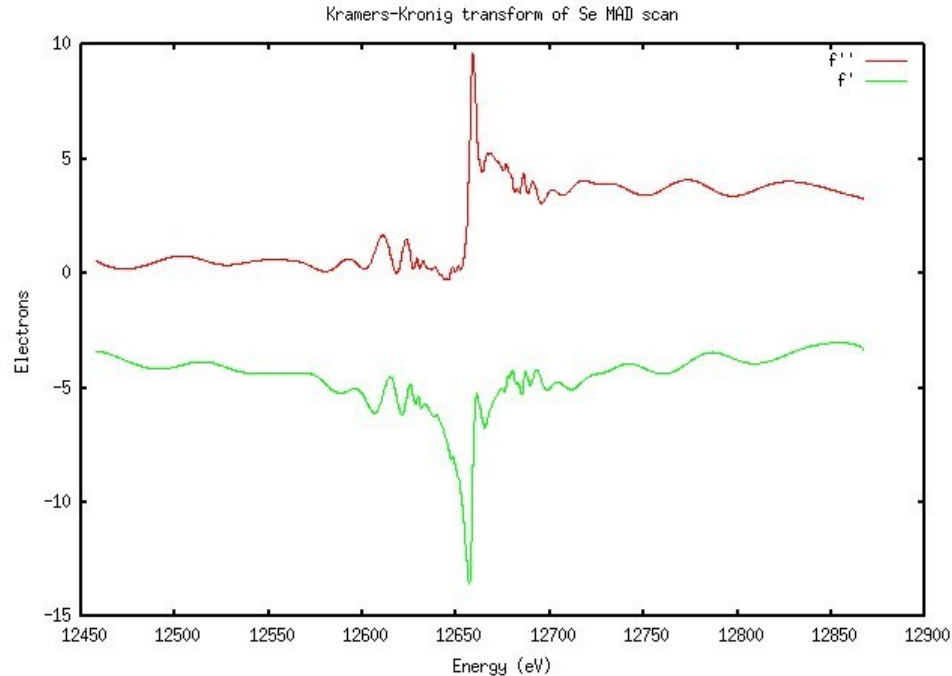
Friedel's law is broken:
 $F(hkl) \neq F(-h-k-l)$



ENERGY SCAN



SAD is a single wavelength data collection (high E remote, peak or inflection)
MAD is a 3 or 4 wavelength data collection

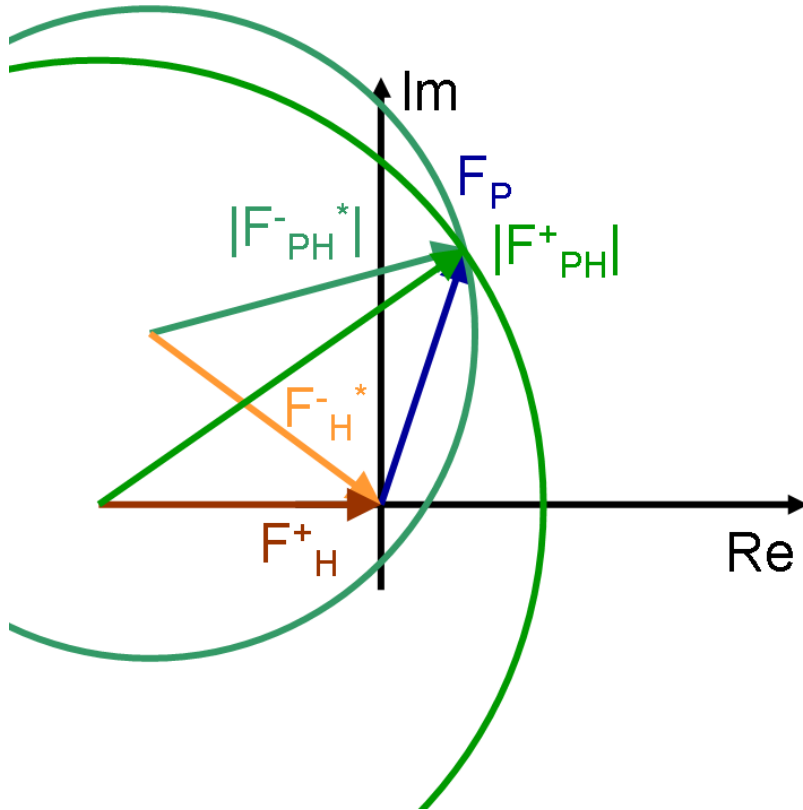


SOLVING THE PHASE PROBLEM IN SAD/MAD

Anomalous differences are smaller than for HA.

However, the great advantage of anomalous phasing is that we can use a single crystal (no need for a native data set).

Seleno-methionine has been the silver bullet for the last two decades



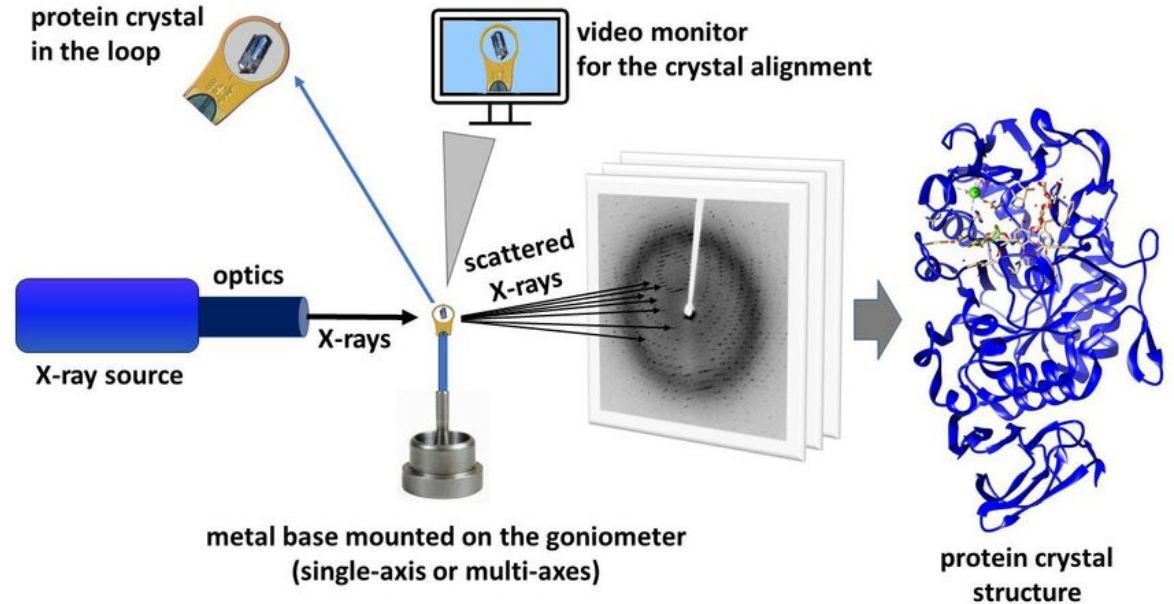
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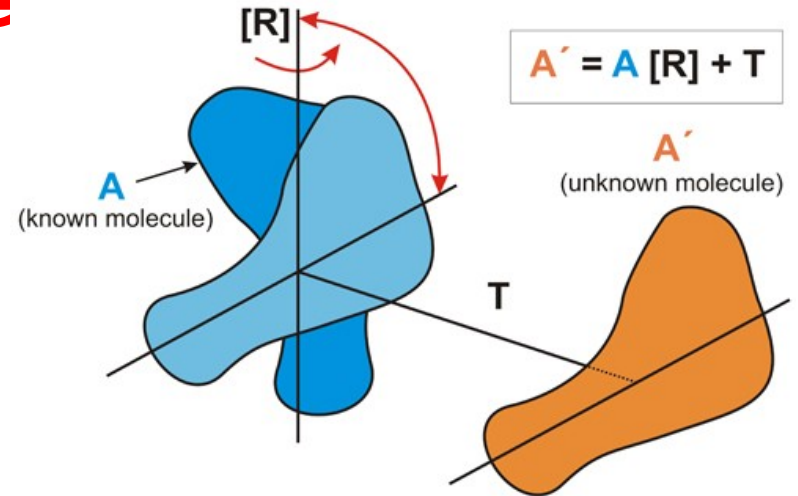
SIR, MIR

SIRAS, MIRAS

MAD, SAD

Isomorphous Replacement

Molecular Replacement

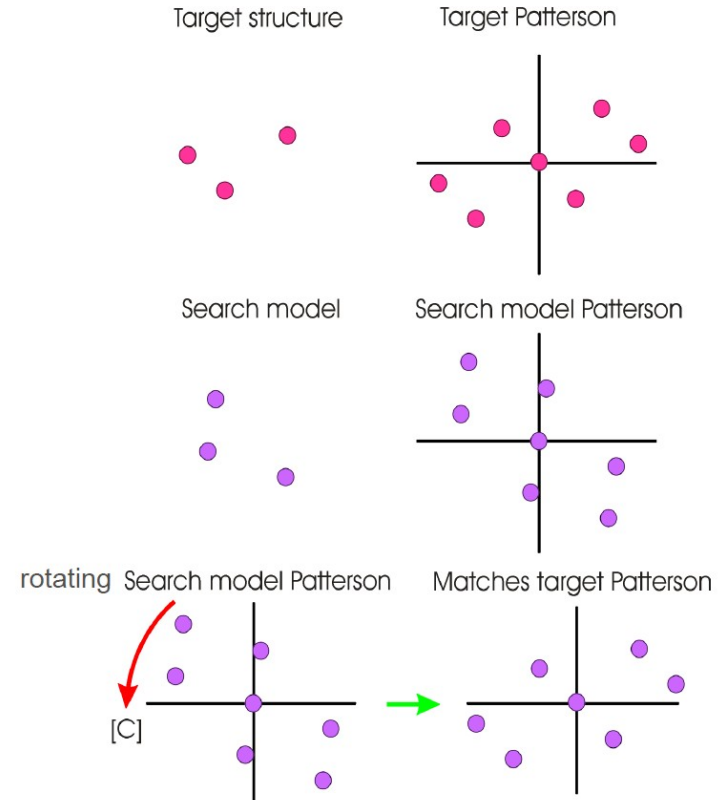


THE ROTATION-TRANSLATION FUNCTION

The Patterson function
(phase zero inverse Fourier):

P

R-factor for the translation function:



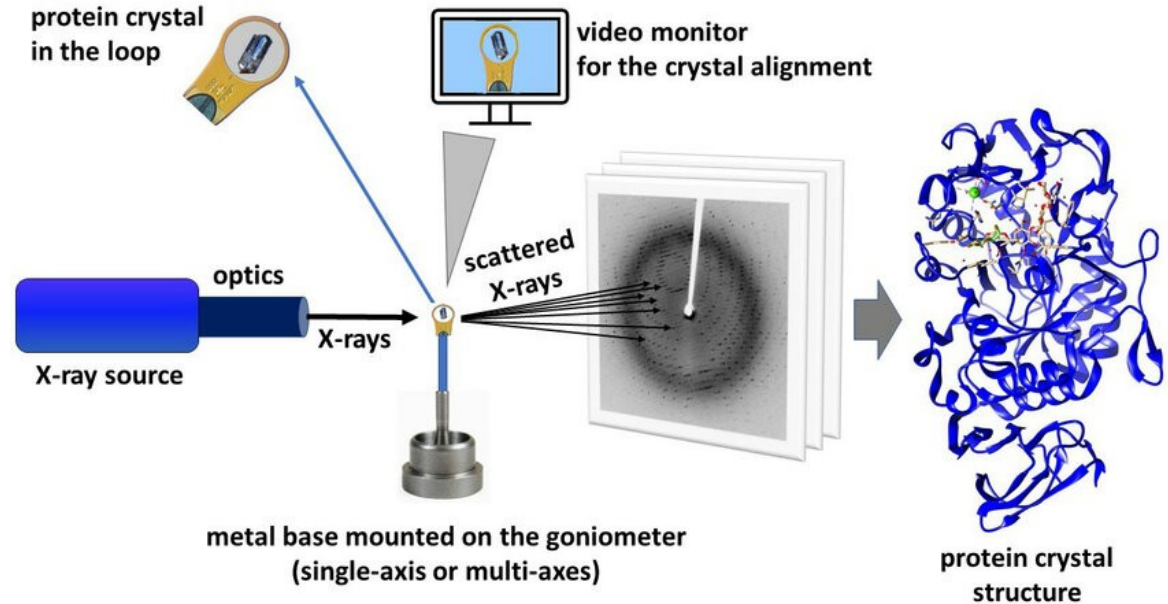
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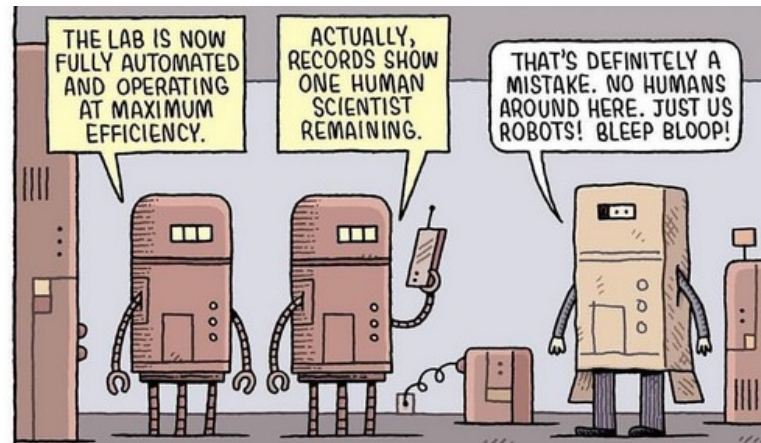
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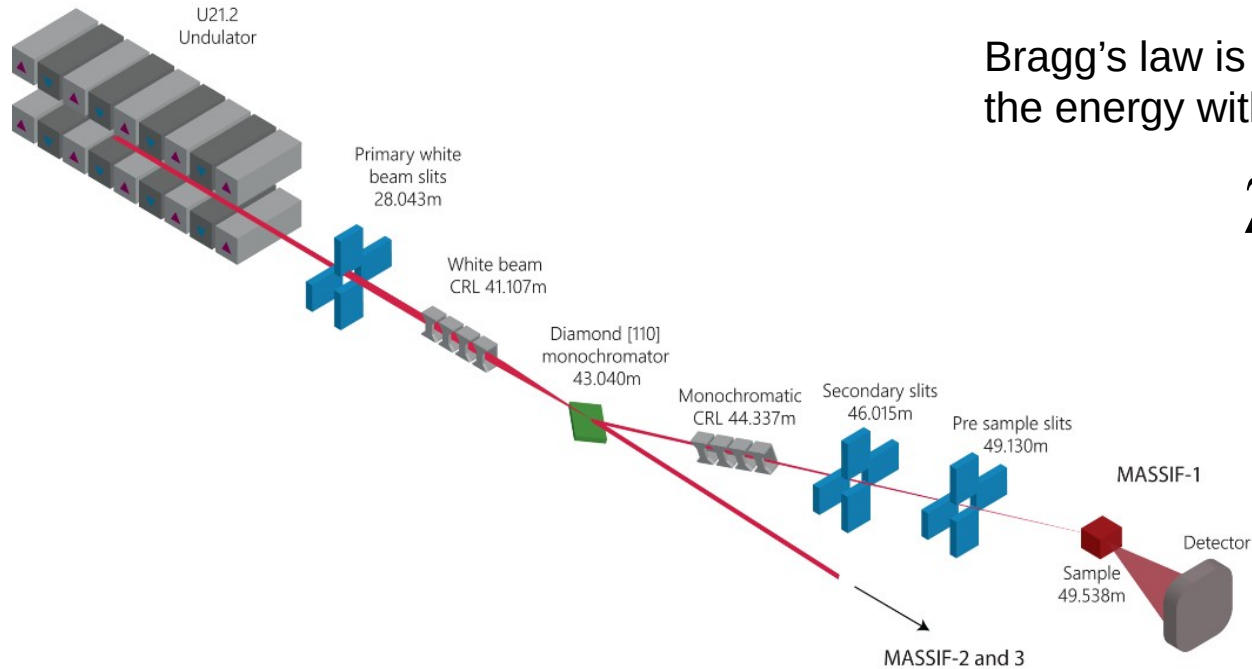
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Automated synchrotron STRUCTURAL BIOLOGY at MASSIF-1



MASSIF-1 LAYOUT

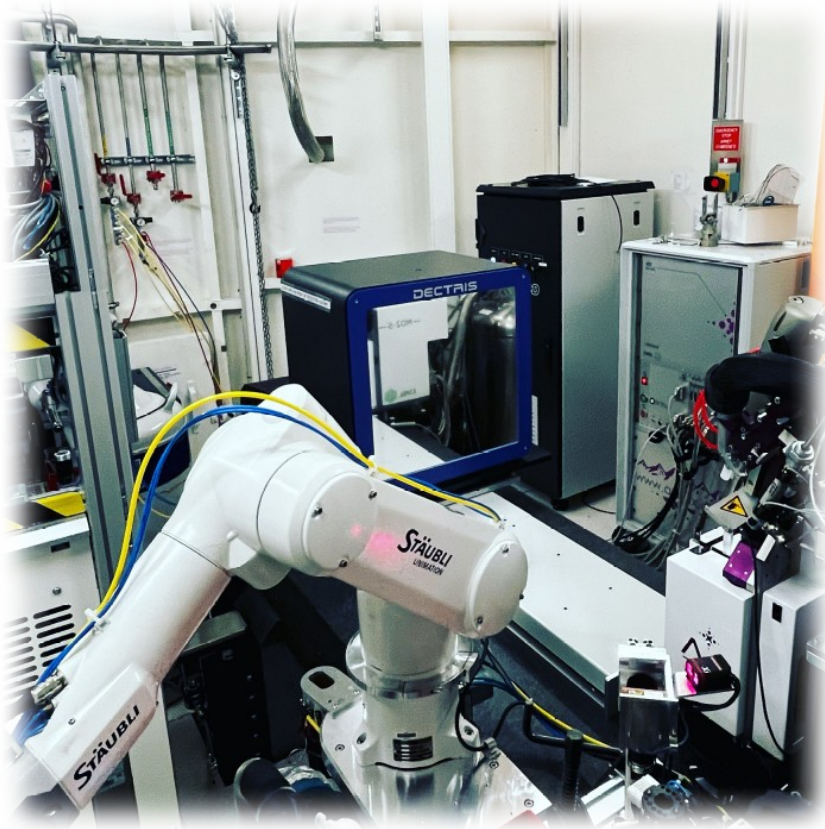


Bragg's law is used also for defining the energy with the monochromator:

λ

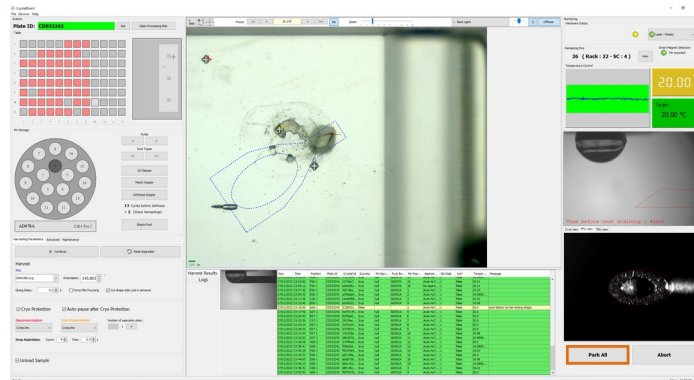
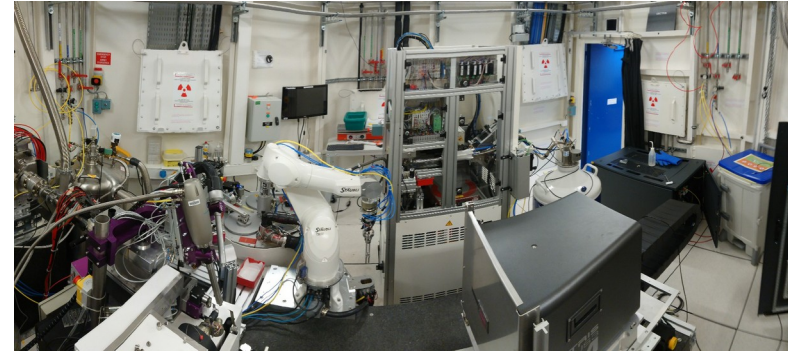
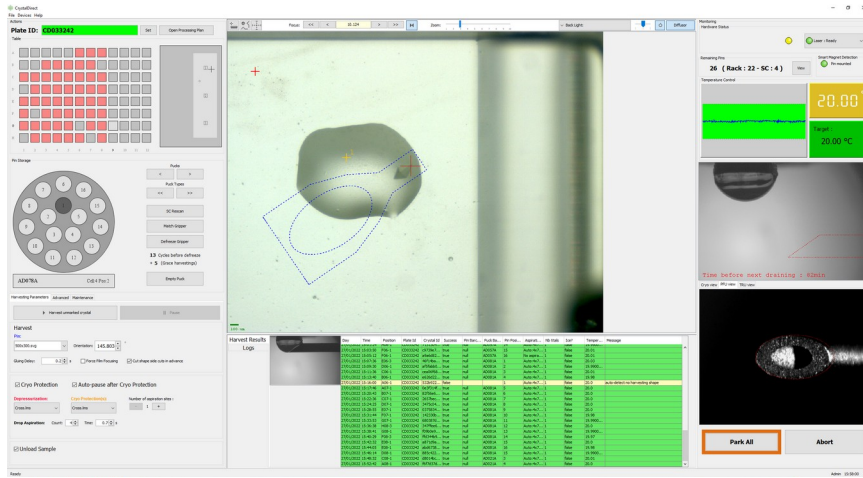
Bowler, MW, Nurizzo, D *et al.* JSR, Volume 22 | Part 6 | November 2015 | Pages 1540–1547 |

EXPERIMENTAL HUTCH

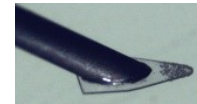
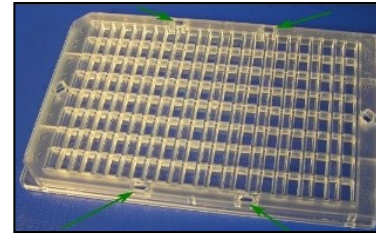
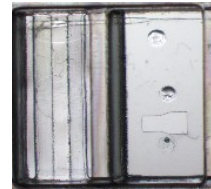


- Fully autonomous beamline
 - no user control
 - data collection optimized for every sample
- Flexible booking, queuing system
- Fully automated data collection from any sample either room or cryogenic temperatures with complex strategies and optimized parameters for each sample
- **Flex HCD Sample changer – 396 samples capacity**
- **Pilatus3 6M**
- **Arinax MD2S diffractometer with mini-Kappa**
- **CrystalDirect Harvester**

AUTOMATED CRYSTAL HARVESTING

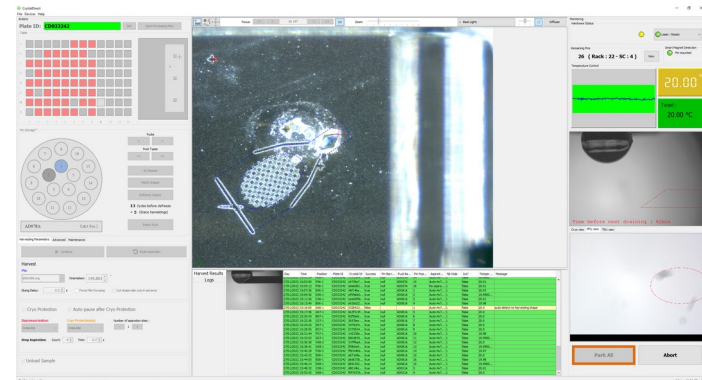
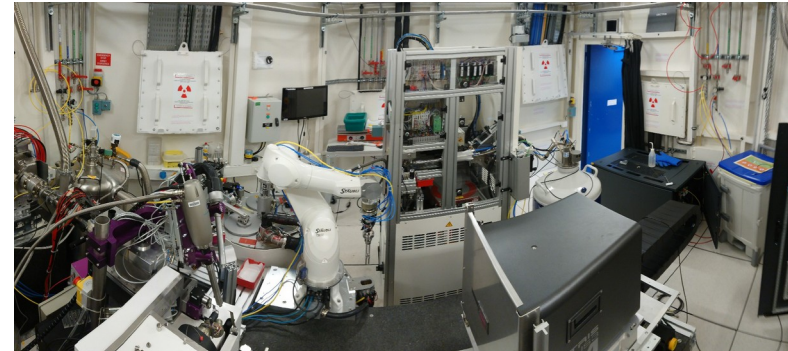
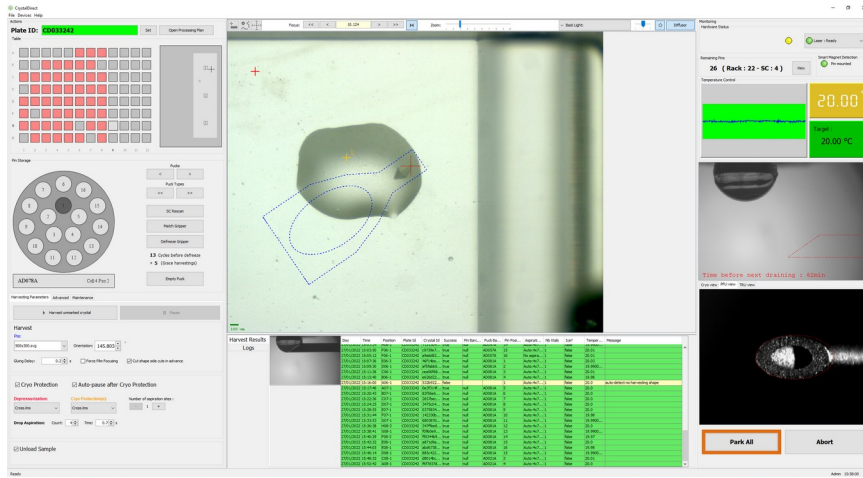


MiTeGen

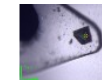
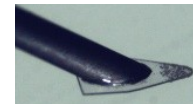
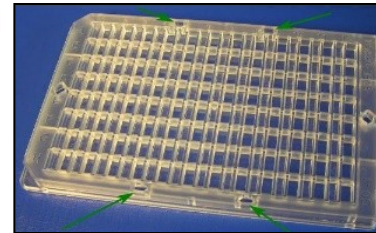
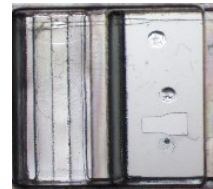


Crystal Direct technology

AUTOMATED CRYSTAL HARVESTING

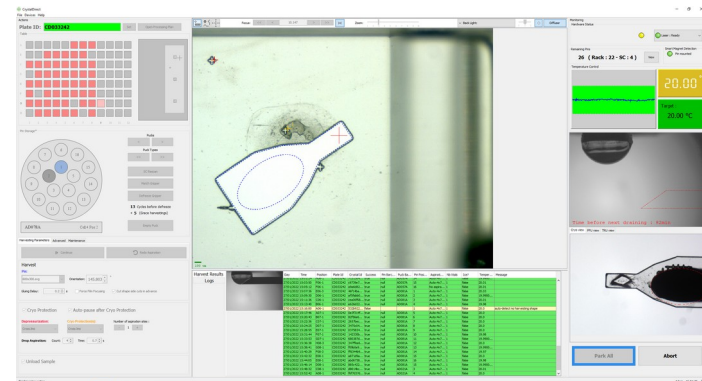
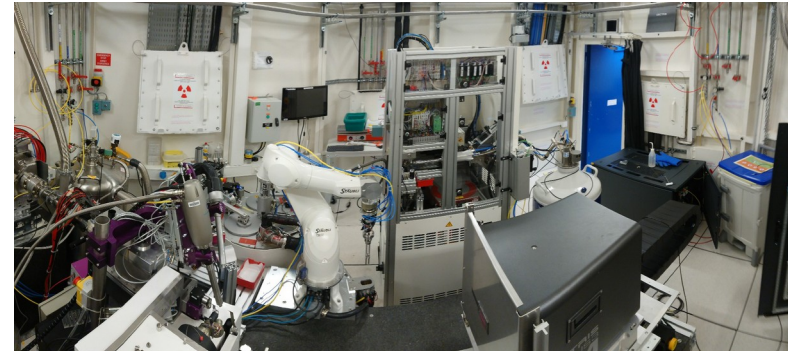
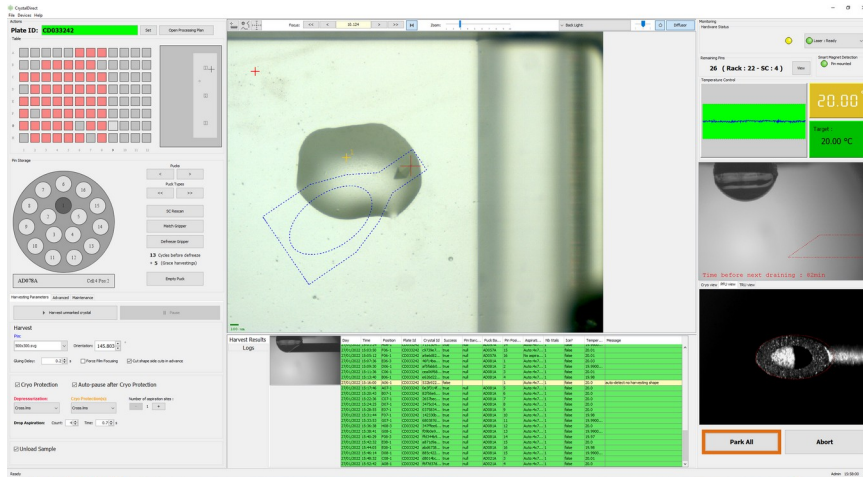


MiTeGen

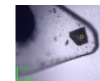
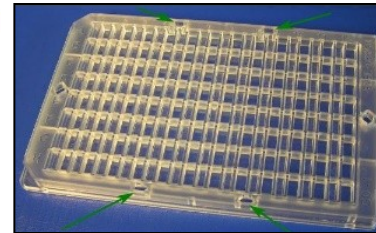
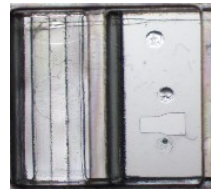


Crystal Direct technology

AUTOMATED CRYSTAL HARVESTING



MiTeGen



Crystal Direct technology

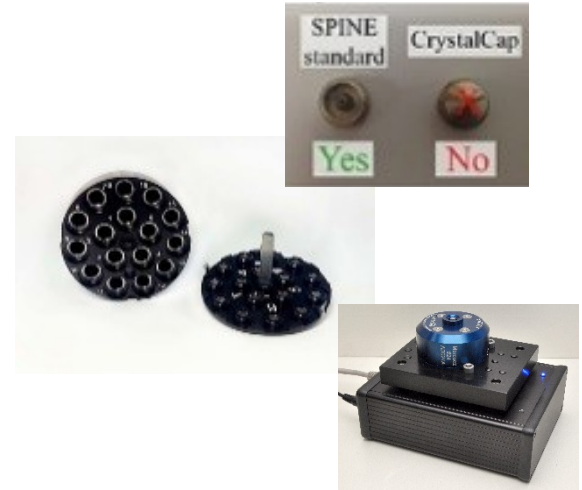
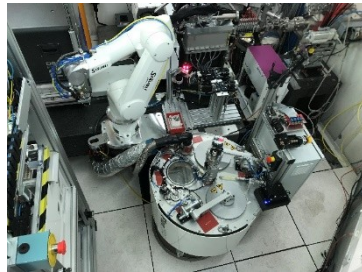
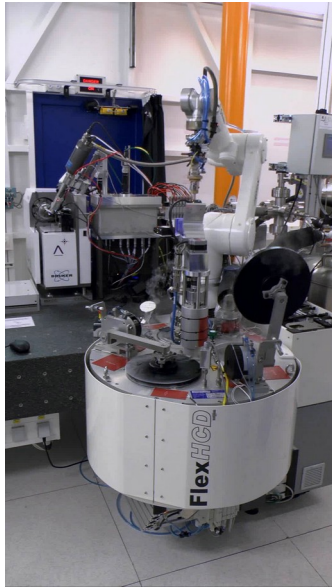
COURIER DELIVERY

Dry shippers are delivered daily at the beamline as part of our queuing system

Delivery is free of charge for Academic Users



CRYSTAL TRANSFER



Activities: Flex Sample Changer

File Desktop Help

Queue	Type	Name
1.0	UfBlock	CP5385
1.1	UfBlock	CP5386
1.2	UfBlock	CP5387
1.3	UfBlock	CP5388
1.4	UfBlock	CP5389
1.5	UfBlock	CP5390
1.6	UfBlock	CP5391
1.7	UfBlock	CP5392
1.8	UfBlock	CP5393
1.9	UfBlock	CP5394
1.10	UfBlock	CP5395
1.11	UfBlock	CP5396
1.12	UfBlock	CP5397
1.13	UfBlock	CP5398
1.14	UfBlock	CP5399
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1.33	UfBlock	CP5418
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1.36	UfBlock	CP5421
1.37	UfBlock	CP5422
1.38	UfBlock	CP5423
1.39	UfBlock	CP5424
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1.44	UfBlock	CP5429
1.45	UfBlock	CP5430
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1.50	UfBlock	CP5435
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1.73	UfBlock	CP5458
1.74	UfBlock	CP5459
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1.97	UfBlock	CP5482
1.98	UfBlock	CP5483
1.99	UfBlock	CP5484
1.100	UfBlock	CP5485

Port: Bypass

IV

CP57387

Port number: Add port to list

Lock:

Aug 19 12:07

Flex Sample Changer

Color Test

Open

Unblock segment

Lock Block segment

77%

Asim: 123728

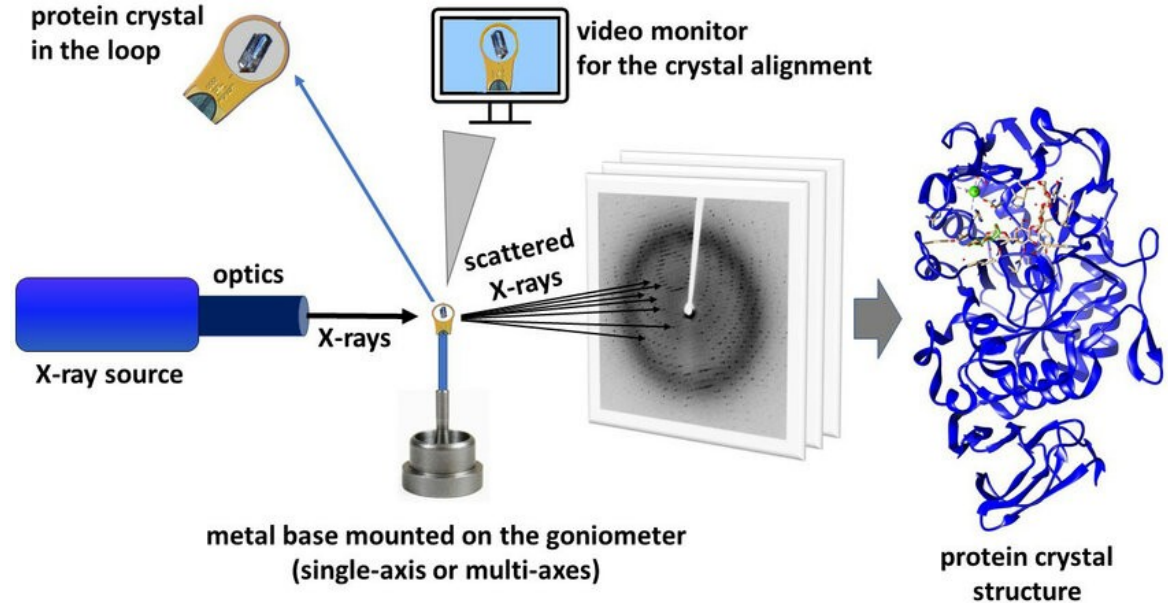
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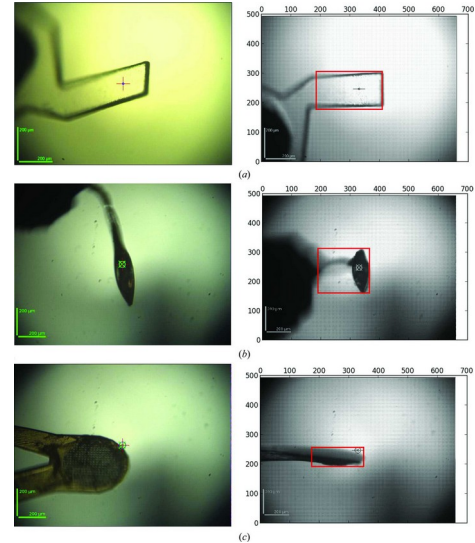
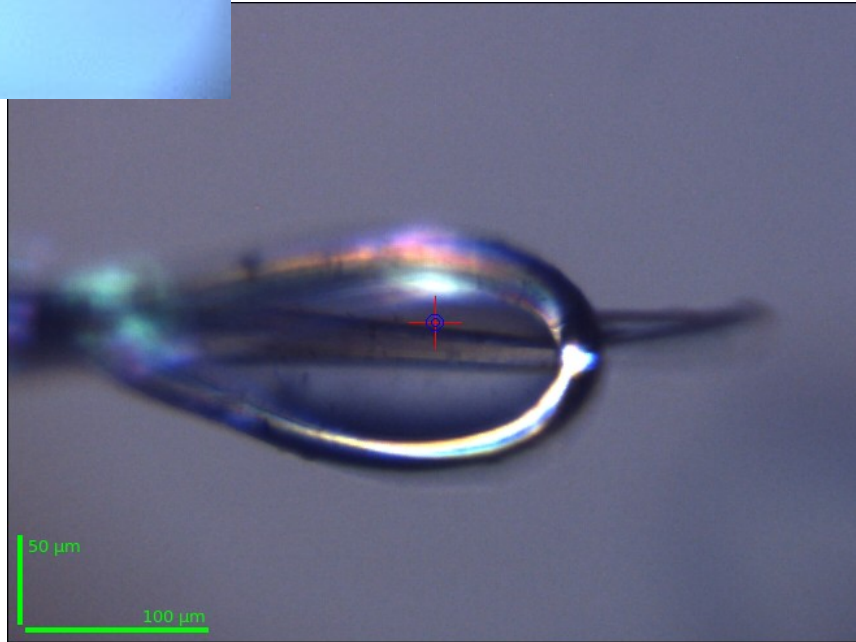
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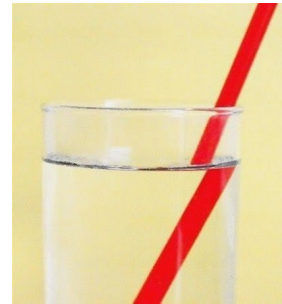
OPTICAL LOOP ALIGNMENT



No restrictions on the mounting system

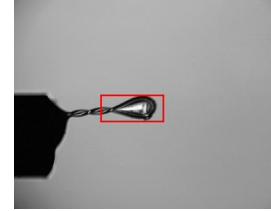


Optical defects might be disastrous



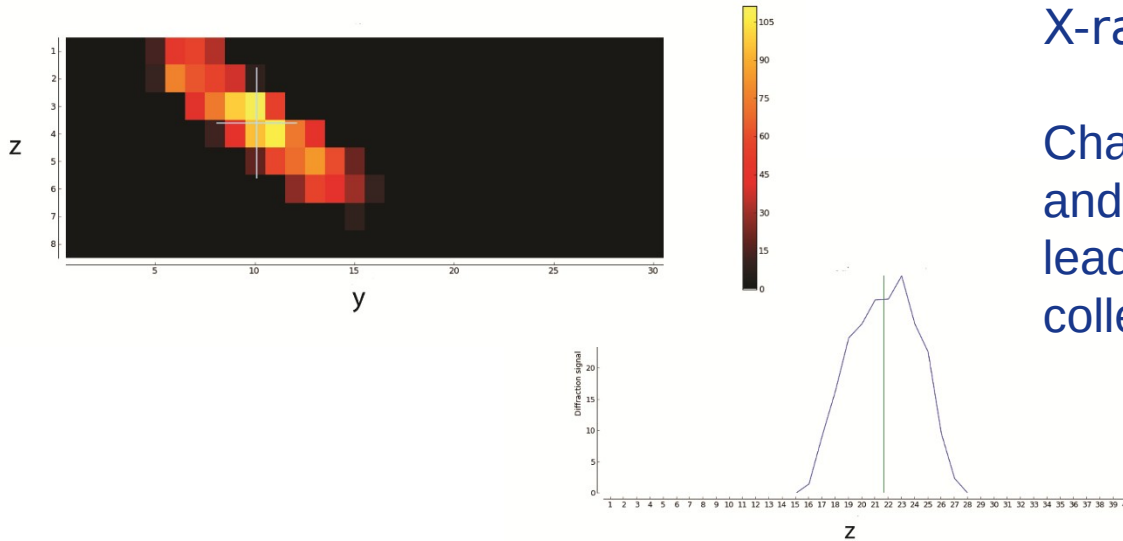
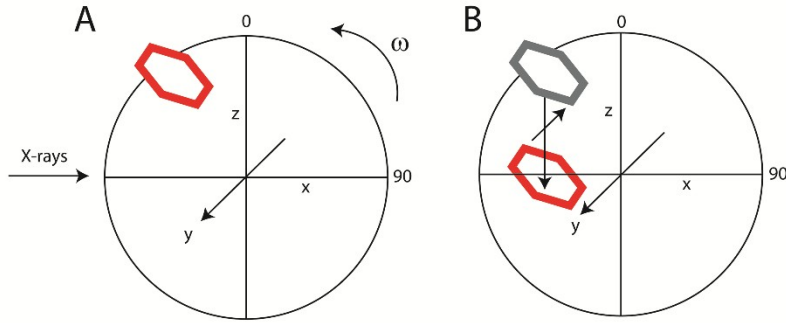
Svensson, O., Malbet-Monaco, S., Popov, A., Nurizzo, D. & Bowler, M. W. (2015). Acta Cryst. D71, 1757-1767.

X-RAY ALIGNMENT

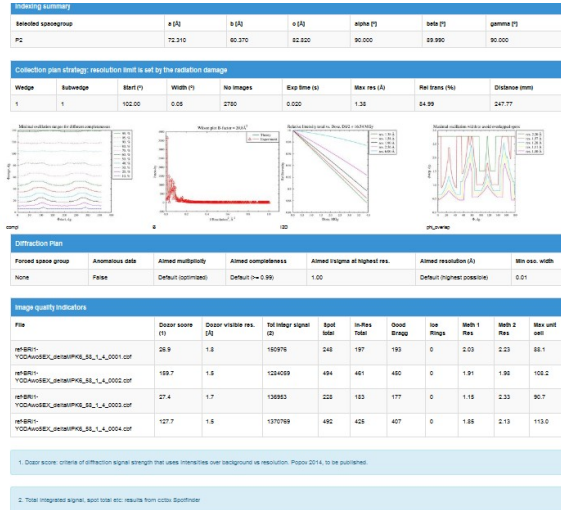


The centering table fitted on the ω axis brings the sample at the center of the camera and in the X-ray beam

Characteristics such as beam size and flux as well as crystal volume lead to highly optimised data collection



CHARACTERIZATION



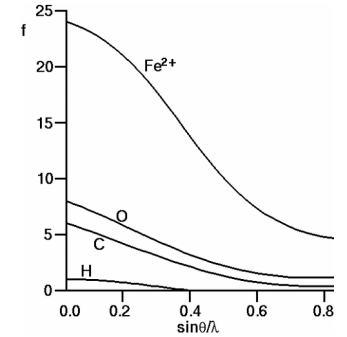
Cell parameters $a, b, c, \alpha, \beta, \gamma$

Point group (Bravais type)

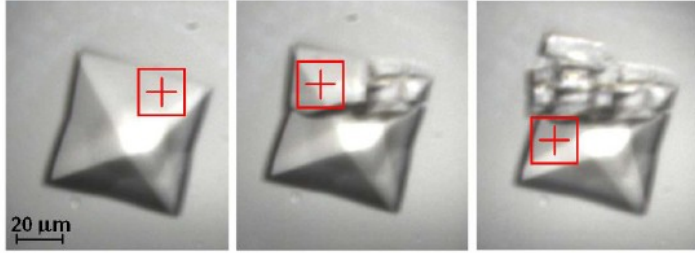
The total rotation angle to collect the entire reciprocal space

The expected resolution according to the crystal size and beam size

Check the quality of X-ray centering (feedback loop)



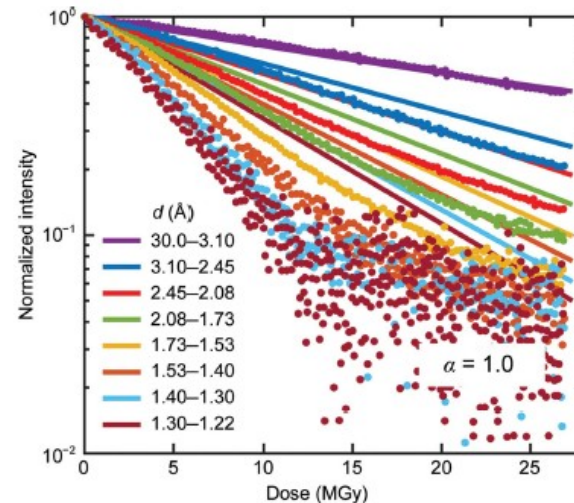
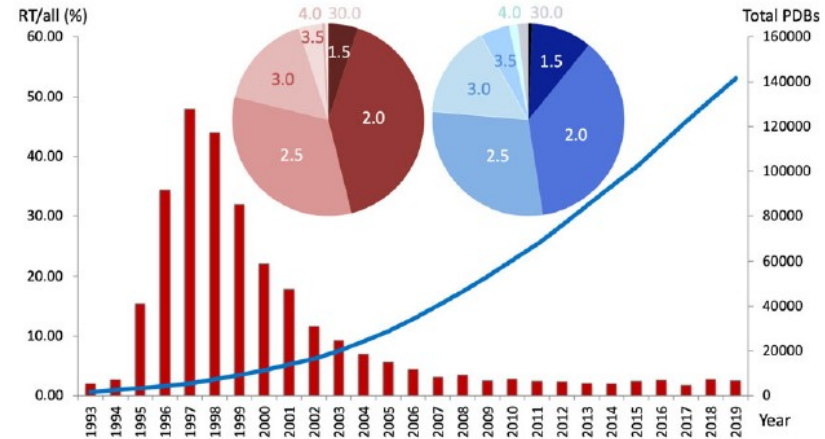
DC STRATEGY – RADIATION DAMAGE



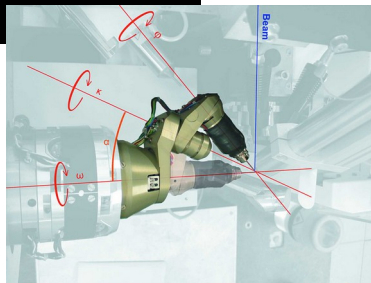
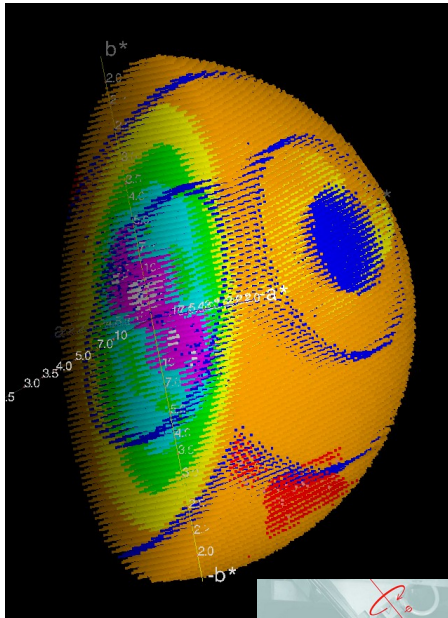
DC is mainly done (>90%) at cryo-temperature

RD is visible even at cryo-temperature

Exposure time / transmission is adjusted to collect a **complete** data set without **theoretical** RC (Henderson limit = 20MGy)



CUSP IN P1



OSC 17-05-2021 13:59:40
data\id30a1\inhouse\topid30a1\20210517\RAW_DATA\TYTH\TYTH-CD031809_009-2

Snapshots Automesh Mesh Line Characterisation Characterisation Mesh Line

Success Success Success Success Success Success Success Success

OSC 13-09-2021 07:43:25
data\vector\75\id30a1\20210911\RAW_DATA\NRAS\NRAS-20210902-VVA077-16

Prefix	Run	Images	Exposure Time	Rec. (corner)	Wavelength	Transmission	Directory and Image Template	Time	Run status	Indicators	View Results	Plotting	Comments																								
map-NRAS-20210902-VVA077-16	0	300	0.05 s	1.3 Å (11.0 Å)	0.9855 Å	100%		07:43:30	Data collection successful																												
int-NRAS-20210902-VVA077-16	1	100	0.05 s	1.3 Å (11.0 Å)	0.9855 Å	100%		07:45:09	Data collection successful																												
ref-NRAS-20210902-VVA077-16	1	4	0.1 s	1.3 Å (11.0 Å)	0.9855 Å	100%		07:46:13	Data collection successful																												
NRAS-20210902-VVA077-16	1	1800	0.02 s	1.5 Å (11.1 Å)	0.9855 Å	85.10%		07:47:55	Data collection successful				<table border="1"> <thead> <tr> <th>P 1</th> <th>Res.</th> <th>Completeness</th> <th>Range</th> <th>I/sigma</th> <th>CC1/2</th> </tr> </thead> <tbody> <tr> <td>Overall</td> <td>47.76-1.42</td> <td>95.3%</td> <td>5.0</td> <td>9.3</td> <td>99.8</td> </tr> <tr> <td>Inner</td> <td>47.76-2.77</td> <td>93.3%</td> <td>1.9</td> <td>6.1</td> <td>99.7</td> </tr> <tr> <td>Outer</td> <td>1.44-1.42</td> <td>97.4%</td> <td>250.1</td> <td>0.3</td> <td>15.2</td> </tr> </tbody> </table>	P 1	Res.	Completeness	Range	I/sigma	CC1/2	Overall	47.76-1.42	95.3%	5.0	9.3	99.8	Inner	47.76-2.77	93.3%	1.9	6.1	99.7	Outer	1.44-1.42	97.4%	250.1	0.3	15.2
P 1	Res.	Completeness	Range	I/sigma	CC1/2																																
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Outer	1.44-1.42	97.4%	250.1	0.3	15.2																																
map-NRAS-20210902-VVA077-16	1	64	0.05 s	1.3 Å (11.0 Å)	0.9855 Å	100%		07:50:18	Data collection successful																												
int-NRAS-20210902-VVA077-16	2	15	0.1 s	1.3 Å (11.0 Å)	0.9855 Å	50.24%		07:51:35	Data collection successful																												
NRAS-20210902-VVA077-16	2	1800	0.02 s	1.5 Å (11.1 Å)	0.9855 Å	88.10%		07:52:30	Data collection successful				<table border="1"> <thead> <tr> <th>P 1</th> <th>Res.</th> <th>Completeness</th> <th>Range</th> <th>I/sigma</th> <th>CC1/2</th> </tr> </thead> <tbody> <tr> <td>Overall</td> <td>47.50-1.91</td> <td>95.3%</td> <td>4.5</td> <td>11.4</td> <td>99.9</td> </tr> <tr> <td>Inner</td> <td>47.50-2.25</td> <td>93.3%</td> <td>1.5</td> <td>6.8</td> <td>99.9</td> </tr> <tr> <td>Outer</td> <td>1.63-1.91</td> <td>96.0%</td> <td>245.2</td> <td>0.4</td> <td>15.2</td> </tr> </tbody> </table>	P 1	Res.	Completeness	Range	I/sigma	CC1/2	Overall	47.50-1.91	95.3%	4.5	11.4	99.9	Inner	47.50-2.25	93.3%	1.5	6.8	99.9	Outer	1.63-1.91	96.0%	245.2	0.4	15.2
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Comments: From 1SPY0: total rotation = 360.0 degrees, min osc width = 0.20 degrees, Dynamic aperture set to 100 um

In triclinic and monoclinic SG some volume of reciprocal space is not accessible with a rotation around a single axis (mini- κ)

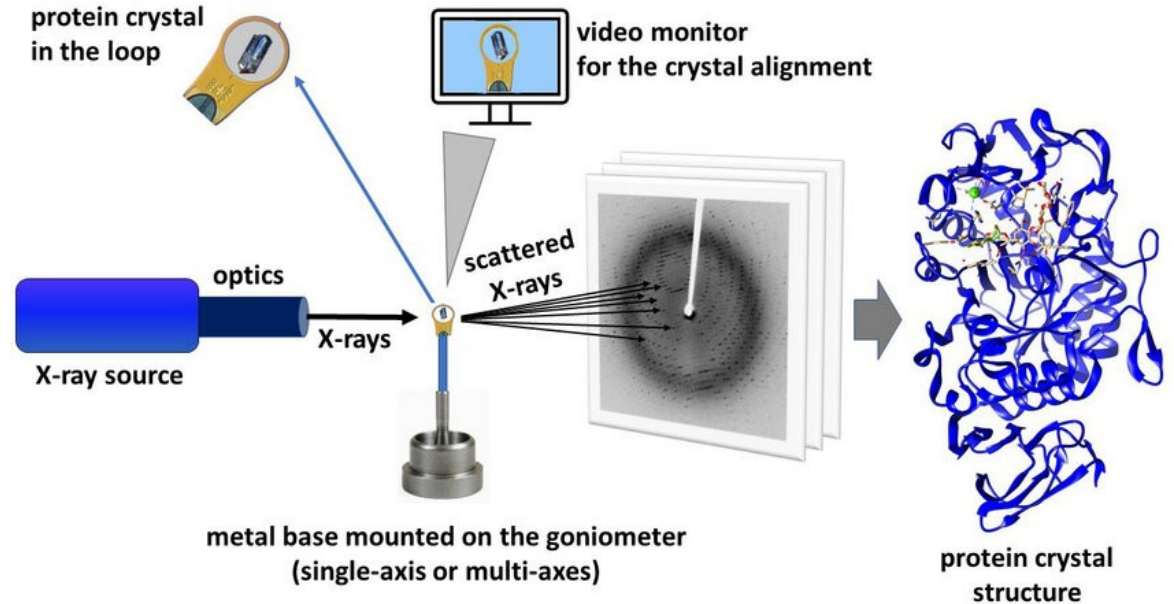
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DATA PROCESSING

15-06-2024 21:02:02
data/vision-ft79/g20a120240615/RAW_DiD/aya001/aya001-2024-06-10_RLY0008_06/run_01_MXPress/run_01_04_datacollection

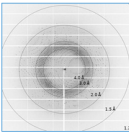

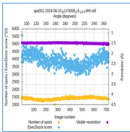
Summary Examine Parameters Data Collectors 3 Sample Last Collect Results 1 Workflow 1

Workflow: MXPressF
Protein: aya001
Sample: 2024-06-10_RLY0008_06
Pfx: aya001-2024-06-10_RLY0008_06_1
Run #: 4
Images (Total): 720 (4048)
Transmission: 100.0 %

Res. (corner): 1.56 Å (1.19 Å)
En. (Wave): 12.442 keV (0.9655 Å)
Omega range: 0.25°
Omega start (total): 90.00° (180°)
Exposure Time: 0.0348 s
Flux start: 2.55e+12 ph/sec
Flux end: 2.58e+12 ph/sec

P 42 21 2 Compl. Res. Rmerge
Overall: 97.7% 42.6-1.3 4.8
Inner: 99.6% 42.6-7.0 2.3
Outer: 79.6% 1.31-1.29 310.0

a	b	c
85.23 Å	85.23 Å	98.66 Å
α	β	γ
90°	90°	90°

Comments: Predefined parameters: total rotation = 180.0 degrees, aimed resolution = 1.90 Å, (ISP/B); Dynamic aperture set to 100 μm Fbest data collection: 180.0 degrees, exposure time 0.035 s, resolution 1.56 Å, transmission 100.00 %

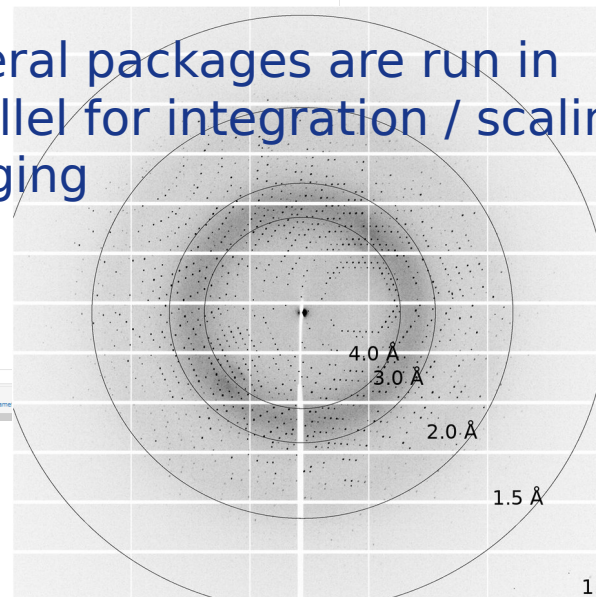
15-06-2024 20:58:52
data/vision-ft79/g20a120240615/RAW_DiD/aya001/aya001-2024-06-10_RLY0008_05/run_01_MXPress/run_01_04_datacollection

Summary Examine Parameters Data Collectors 3 Sample Last Collect Results 1 Workflow 1

Pipeline	SpaceGroup	x.b.c (Å)	α,β,γ (°)	Shell	Resolution (Å)	Multiplicity	Completeness %	Anomalous multiplicity	Anomalous completeness %	<I>Sigma	R _{meas}	R _{merge}	R _{pim}	cc(1/2)	cc3no	sigino	ISA	Download
ANOM XDSAPP	P 43 21 2	84.9 90.0	90.0	Overall	49.5-1.3	9.8	99.8	4.8	90.0	15.7	6.5	5.9	2.5	100				
TEST		84.9 90.0	90.0	Inner	49.5-7.1	10.3	99.7	0.7	99.7	0.7	3.3	1.0	100					
		84.9 90.0	90.0	Outer	1.3-1.3	2.1	69.2	1.4	33.3	0.1	285.6	300.1	321.0	10				
ANOM XIA2_DIALS	P 41 21 2	84.9 90.0	90.0	Overall	84.8-1.4	10.8	99.8	5.7	97.4	16.3	6.5	5.9	2.5	100				
		84.9 90.0	90.0	Inner	85.1-2.7	12.1	100.0	6.9	100.0	78.0	3.7	2.4	1.4	100				
		84.9 90.0	90.0	Outer	1.4-1.4	3.7	95.5	2.1	74.1	0.3	216.1	195.4	127.8	40				
ANOM EDNA_proc	P 41 21 2	84.9 90.0	90.0	Overall	49.5-1.4	12.5	100.0	6.3	100.0	20.7	6.2	6.6	2.3	100				
		84.9 90.0	90.0	Inner	49.5-5.3	10.8	99.8	6.4	100.0	63.4	3.9	3.5	1.5	100				
		84.9 90.0	90.0	Outer	1.5-1.5	10.0	99.9	4.9	99.8	0.9	286.7	222.9	192.1	60				
ANOM grenades_parallelproc	P 41 21 2	84.9 90.0	90.0	Overall	99.0-1.6	12.8	99.7	6.4	99.7	23.4	5.9	5.4	2.2	100				
		84.9 90.0	90.0	Inner	99.0-8.5	9.7	99.9	6.4	100.0	67.7	4.2	3.8	1.5	100				
		84.9 90.0	90.0	Outer	1.5-1.6	11.1	97.3	5.5	97.2	1.0	240.7	210.0	97.7	60				
ANOM grenades_fastproc	P 4 2 2	84.9 90.0	90.0	Overall	99.0-1.5	12.8	99.9	6.3	99.8	22.2	6.1	5.6	2.3	100				
		84.9 90.0	90.0	Inner	99.0-8.3	9.9	99.7	6.4	100.0	69.9	4.1	3.6	1.5	100				
		84.9 90.0	90.0	Outer	1.5-1.5	9.5	97.4	4.7	97.2	0.8	272.2	243.2	199.4	60				
ANOM grenades_parallelproc	P 4 2 2	84.9 90.0	90.0	Overall	99.0-1.6	12.8	99.7	6.4	99.7	23.1	6.0	5.5	2.3	100				
		84.9 90.0	90.0	Inner	99.0-8.5	9.7	99.9	6.4	100.0	66.3	4.2	3.7	1.5	100				
		84.9 90.0	90.0	Outer	1.5-1.6	11.1	97.3	5.5	97.3	1.0	256.4	228.8	103.9	60				
ANOM grenades_parallelproc	P 4	84.9 90.0	90.0	Overall	99.0-1.5	6.7	99.8	3.3	99.1	18.7	6.7	4.7	3.0	100				
		84.9 90.0	90.0	Inner	99.0-8.7	6.0	99.9	3.1	95.8	57.1	4.0	3.1	2.0	100				
		84.9 90.0	90.0	Outer	1.5-1.6	6.7	99.4	3.3	95.3	1.1	162.3	155.2	95.1	60				
ANOM grenades_parallelproc	P 1	84.9 90.0	90.0	Overall	99.0-1.7	1.8	90.0	0.8	80.3	11.8	5.5	4.4	4.4	100				
		84.9 90.0	90.0	Inner	99.0-8.2	1.8	88.0	1.0	72.8	33.4	4.7	2.4	2.4	100				
		84.9 90.0	90.0	Outer	1.7-1.7	1.8	91.4	1.0	54.1	1.0	109.8	102.3	102.3	50				

Comments: Predefined parameters: total rotation = 180.0 degrees, aimed resolution = 1.90 Å, (ISP/B); Dynamic aperture set to 100 μm Fbest data collection: 180.0 degrees, exposure time 0.037 s, resolution 1.63 Å, transmission 100.00 %

Several packages are run in parallel for integration / scaling / merging



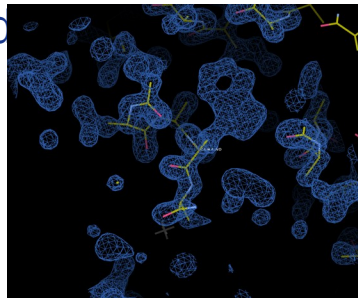
SOLVING THE PHASE PROBLEM

SAD

look for anomalous signal
comparing $F(hkl)$ and $F(-h-k-l)$ in
XDS

Find the heavy atom site(s) with
SHELXD

Run density modification (NCS,
solvent flattening, histogram
matching) in both hands

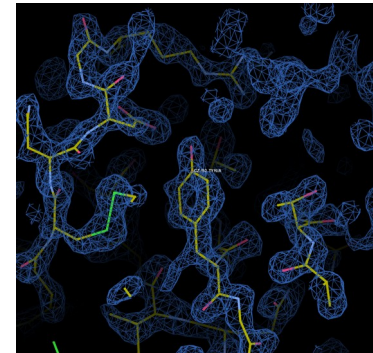


MR

look for a model in the PDB with
same cell or use the model
provided by the user

Run the molecular replacement
(Rotation and translation function)

Refine the MR model



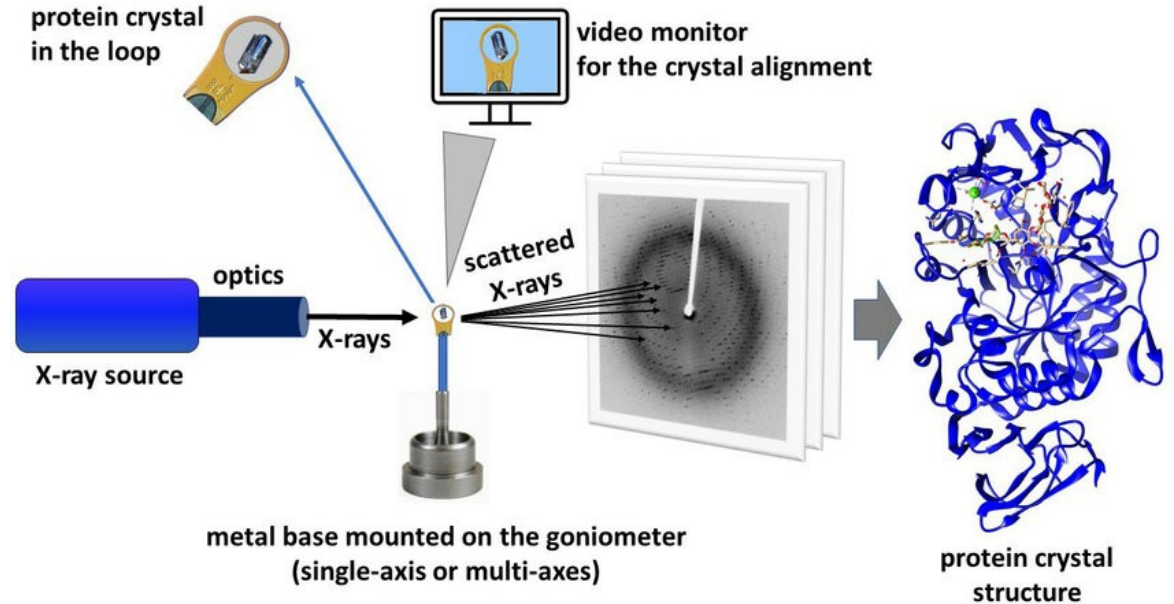
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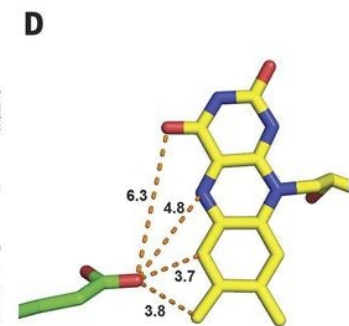
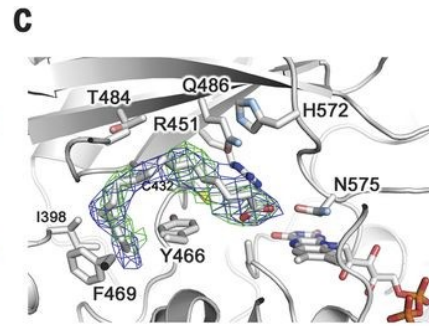
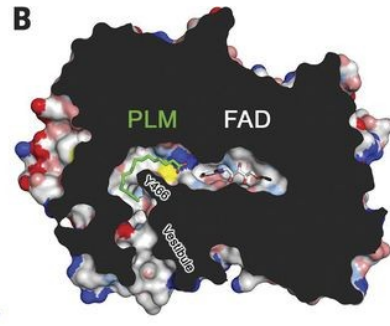
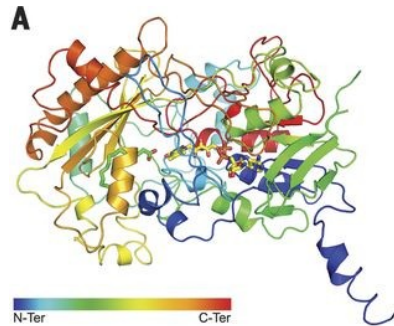
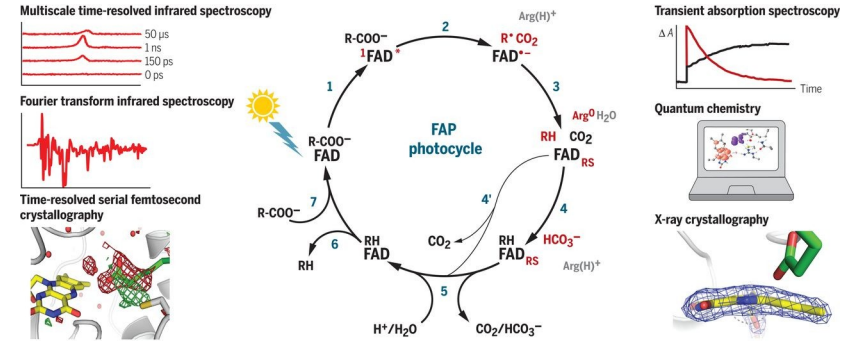
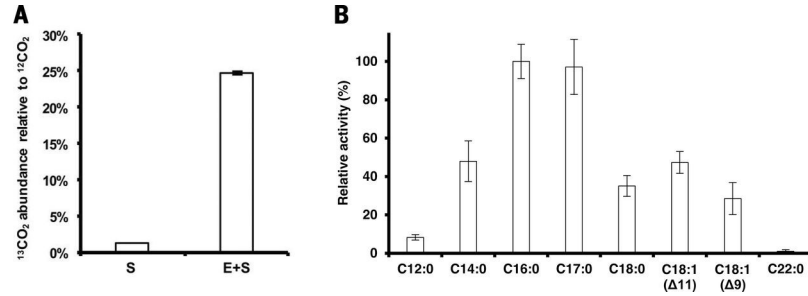
6. Crystal harvesting
7. Data collection
8. Data processing
9. Examples



DESIGN OF MEMBRANE PROTEINS ANALOGUES

Computational design of soluble and functional membrane protein analogues. Casper et al. Nature DOI: 10.1038/s41586-024-07601-y

FATTY ACID PHOTODECARBOXYLASE



An algal photoenzyme converts fatty acids to hydrocarbons. Sorrigué et al. Science. DOI: 10.1126/science.aan6349

During a protein crystallography experiments, most of the X-ray photons that interact with the sample...

- a. damage the sample, and only a small part contributes to the diffraction pattern
- b. contribute to the diffraction pattern, and only a small part damages the sample
- c. cause fluorescence in the sample

How can synchrotron radiation be advantageous for protein crystallography studies?

- a. Because of its high level of automation
- b. Because of its high intensity
- c. Because of its high stability

Why do we collect diffraction data at 100K?

- a. Crystal quality is higher at 100K than at room temperature
- b. To reduce the size of the cell
- c. To reduce radiation damage
- d. Crystals are easier to carry at 100K

Why proteins crystalize only in 65 space groups out of the 230?

- a. Because of the chirality of the amino-acid $C\alpha$
- b. Because their molecular weight being too high
- c. Because of the water molecules surrounding the protein in the mother liquor

What is mathematical relation between the electron density and the amplitudes?

- a. A linear regression
- b. A Fourier transform
- c. The square root