CINA
Cellular Imaging
and Nano Analytics

Systems Biology Synthetic Biology

D-BSSE

SystemsX
The Swiss Initiative in Systems Biology

Projective Constraint Optimization: Filling the missing cone

C-CINA: Center for Cellular Imaging and Nano Analytics

Henning Stahlberg

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Filling the Missing Cone

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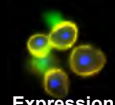
John Spence, U. Arizona, TX
Kaoru Mitsuoka, Tokyo
Dan Masiel, UC Davis
Mike Sarahan, UC Davis
Nigel Browning, UC Davis
Wanda Kukulski, EMBL

Now with Lydia Kavraki, Rice University

Electron Crystallography Bottlenecks


Expression


Purification

Samples: "PHS" (Pure, Homogeneous, Stable).
"Is a gel filtration profile still perfect after 1 week at 4°C ?"


2D Crystallization

2D crystals: Automation.


Sample Prep

Grid preparation: New sample supports (TiSi? Graphene?).


Imaging

Images: Automation? Phase contrast STEM?

E-diff: Automated electron diffraction in TEM.


Image Processing

MRC, 2dx, IPLT: Throughput, User-friendliness, Automation,
Maximum Likelihood for badly ordered 2D crystals.


Model Building

Missing Cone: PCO (Projective Constraint Optimization)

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Hybrid Input Output Algorithm
Iterative Fienup-Gerchberg-Saxton - Algorithm

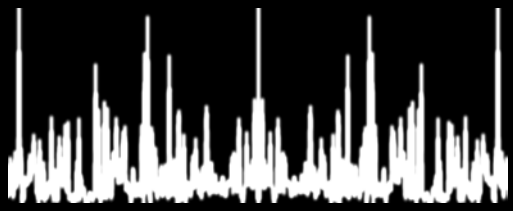
Opt. Lett. 3, 27-29 (1978)	Reconstruction of an object from the modulus of its Fourier transformation J. R. Fienup
Biophys. J. 25, 495-512 (1979)	Structure determination of asymmetric membrane profiles using an iterative Fourier method R.M. Stroud, D. A. Agard
Biophys. J. 37, 589-602 (1982)	Linking regions between helices in Bacteriorhodopsin revealed D. A. Agard , R.M. Stroud
Ultramicroscopy 31, 365-378 (1989)	Approximation of missing-cone data in 3D electron microscopy M. Barth, R.K. Bryan, R. Hegerl
J. Struct. Biol. 144, 209-218 (2003)	Three-dimensional diffractive imaging for crystalline monolayers with one-dimensional compact support J.C.H. Spence, U. Weierstall, T.T. Fricke, R.M. Glaeser, K.H. Downing

Real Space

Fourier Space

f

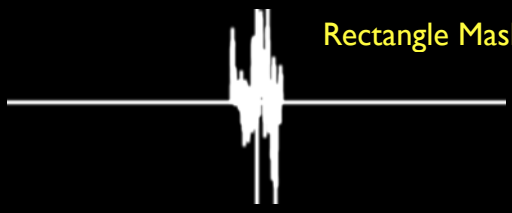
F



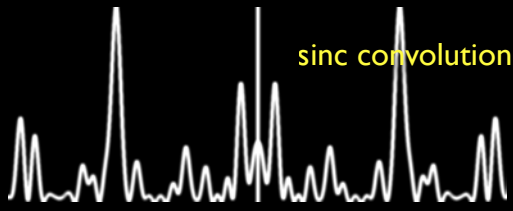
multiplication with g

convolution with G

Rectangle Mask

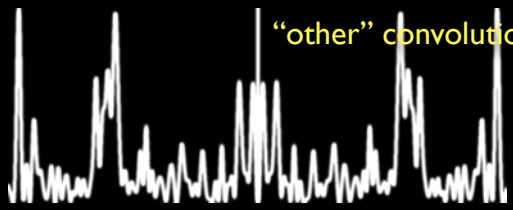


sinc convolution



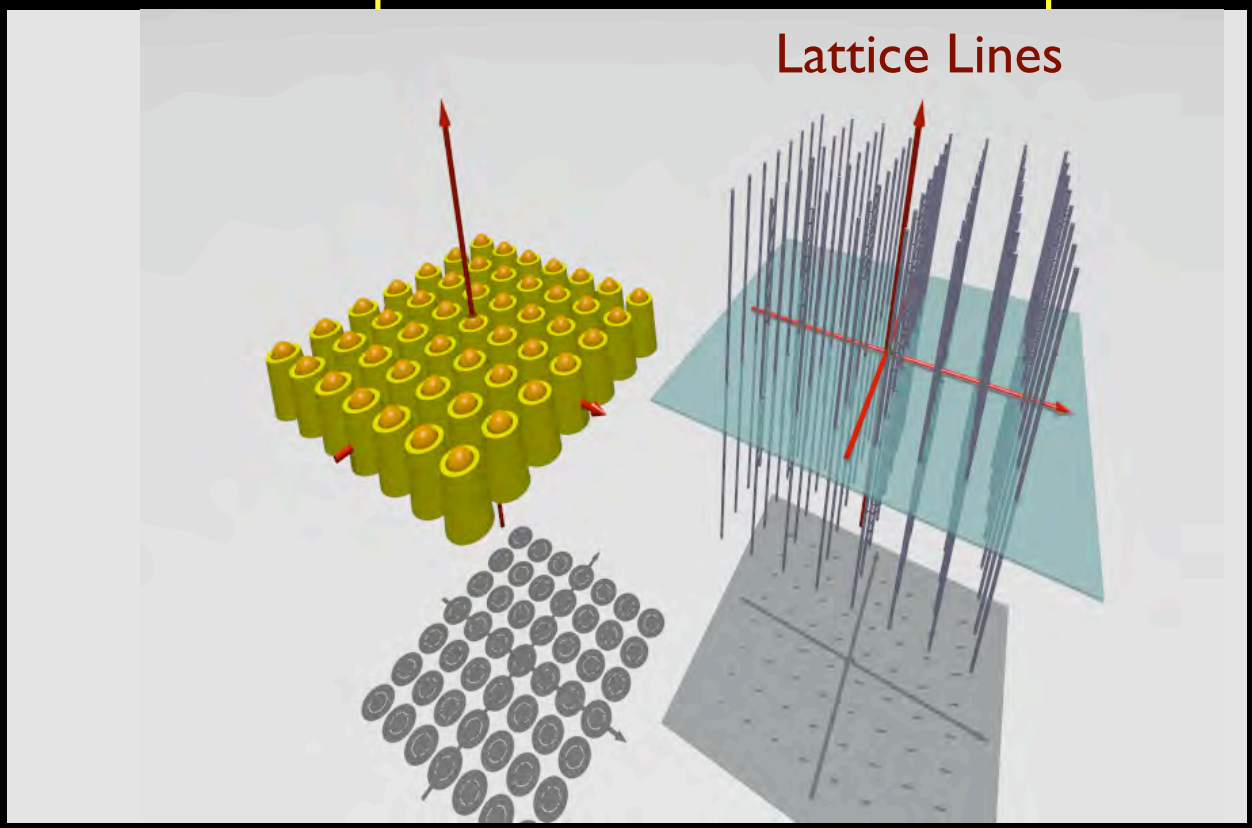
Fermi Mask

"other" convolution



Real Space

Fourier Space



convolution with g

multiplication with G

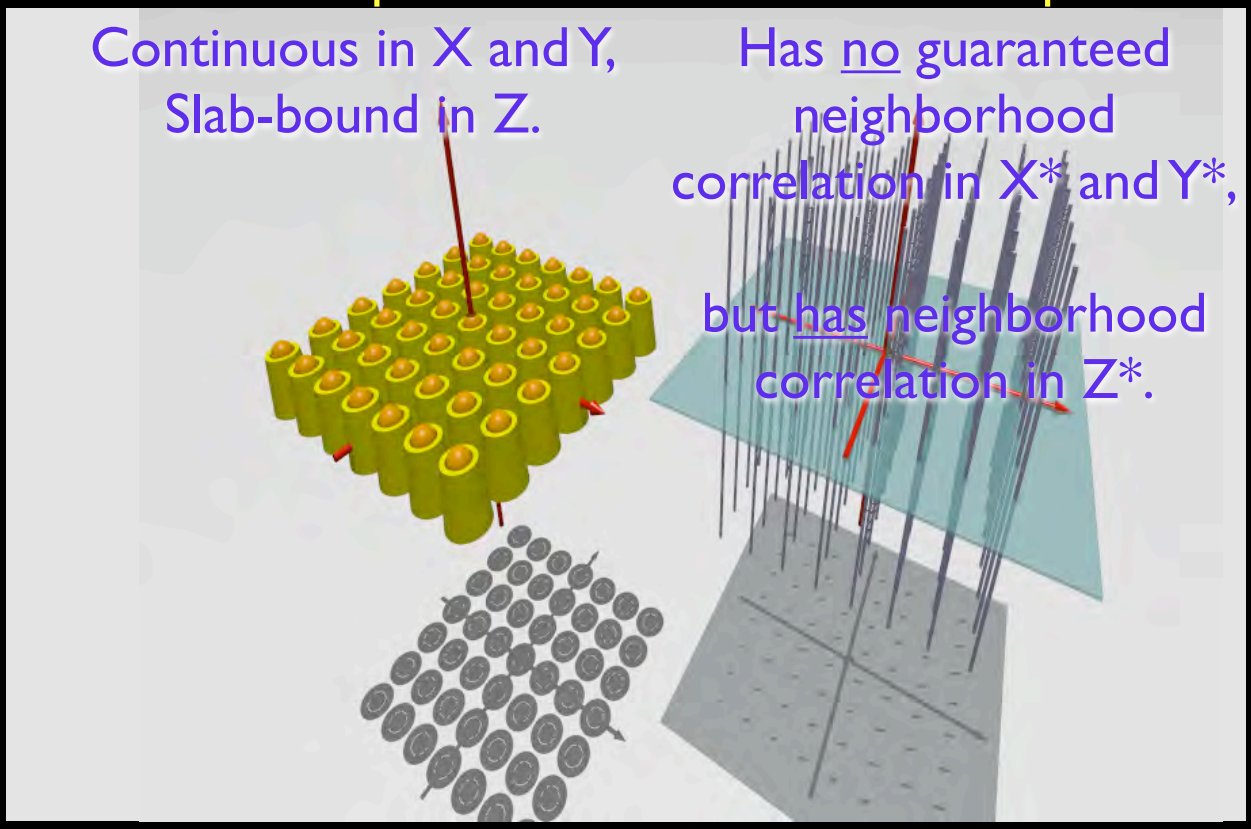
Real Space

Fourier Space

Continuous in X and Y,
Slab-bound in Z.

Has no guaranteed neighborhood correlation in X* and Y*,

but has neighborhood correlation in Z*.

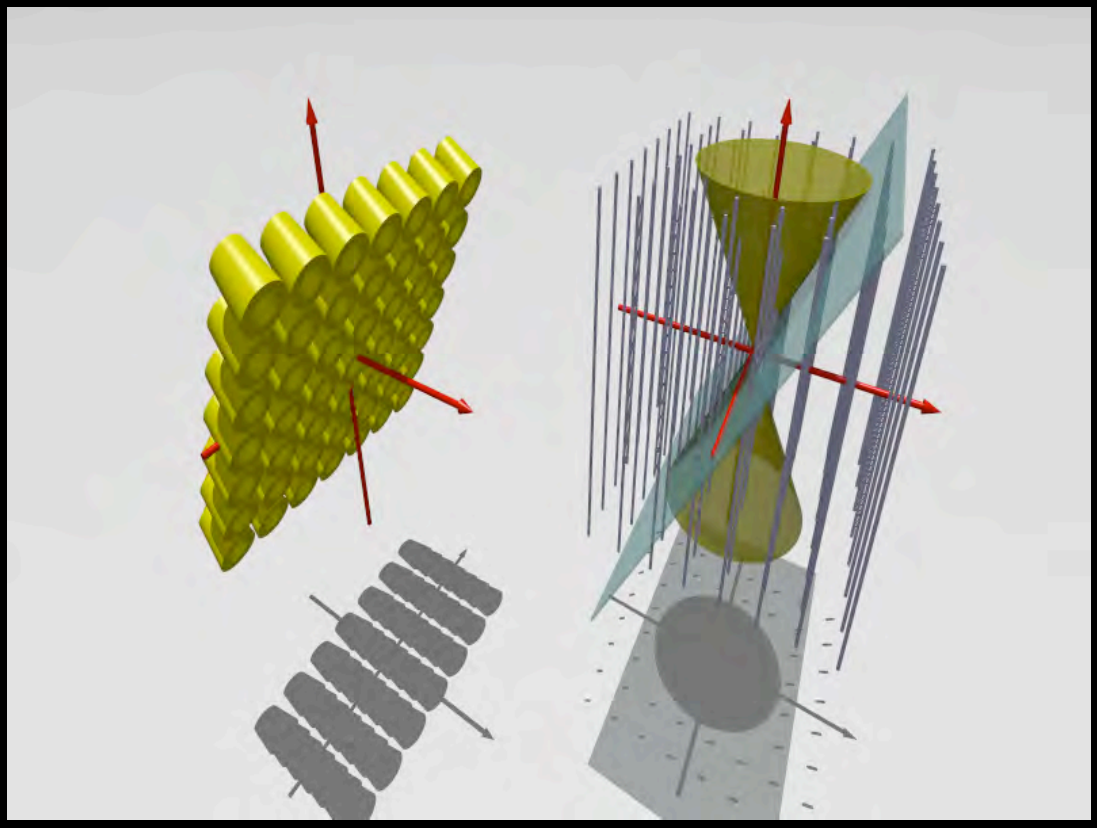


convolution with g

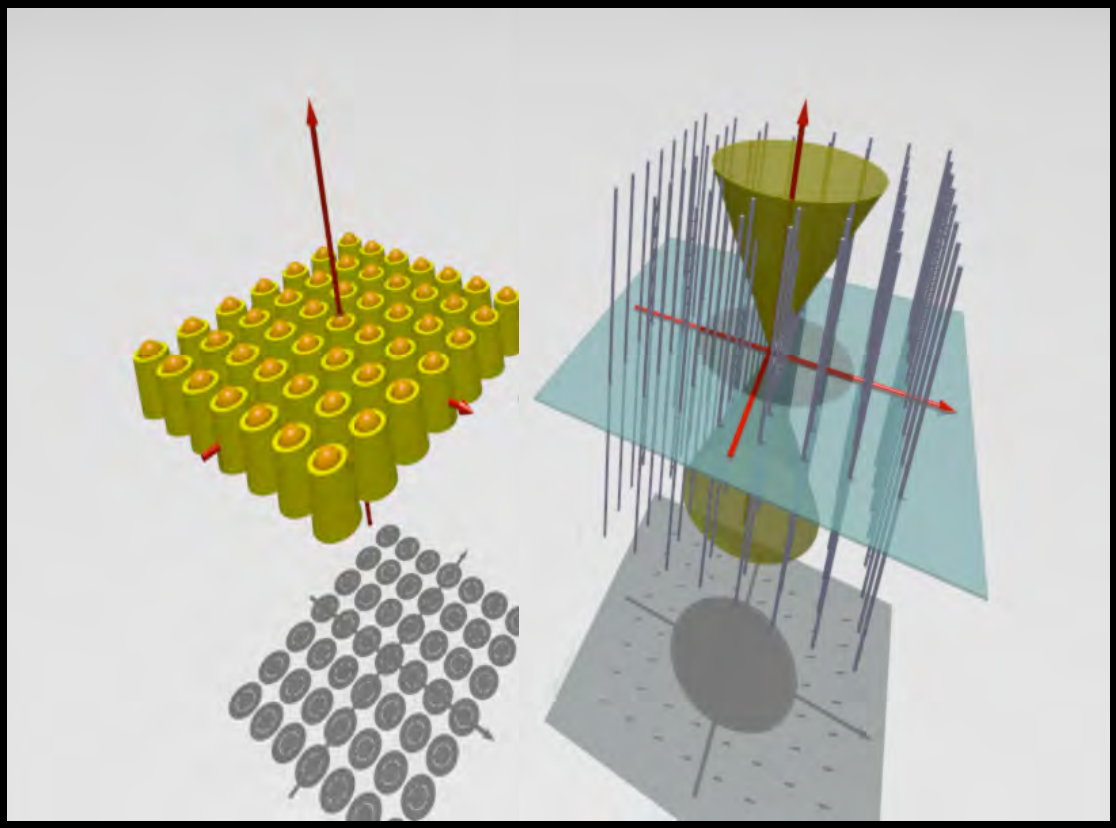
multiplication with G

Real Space

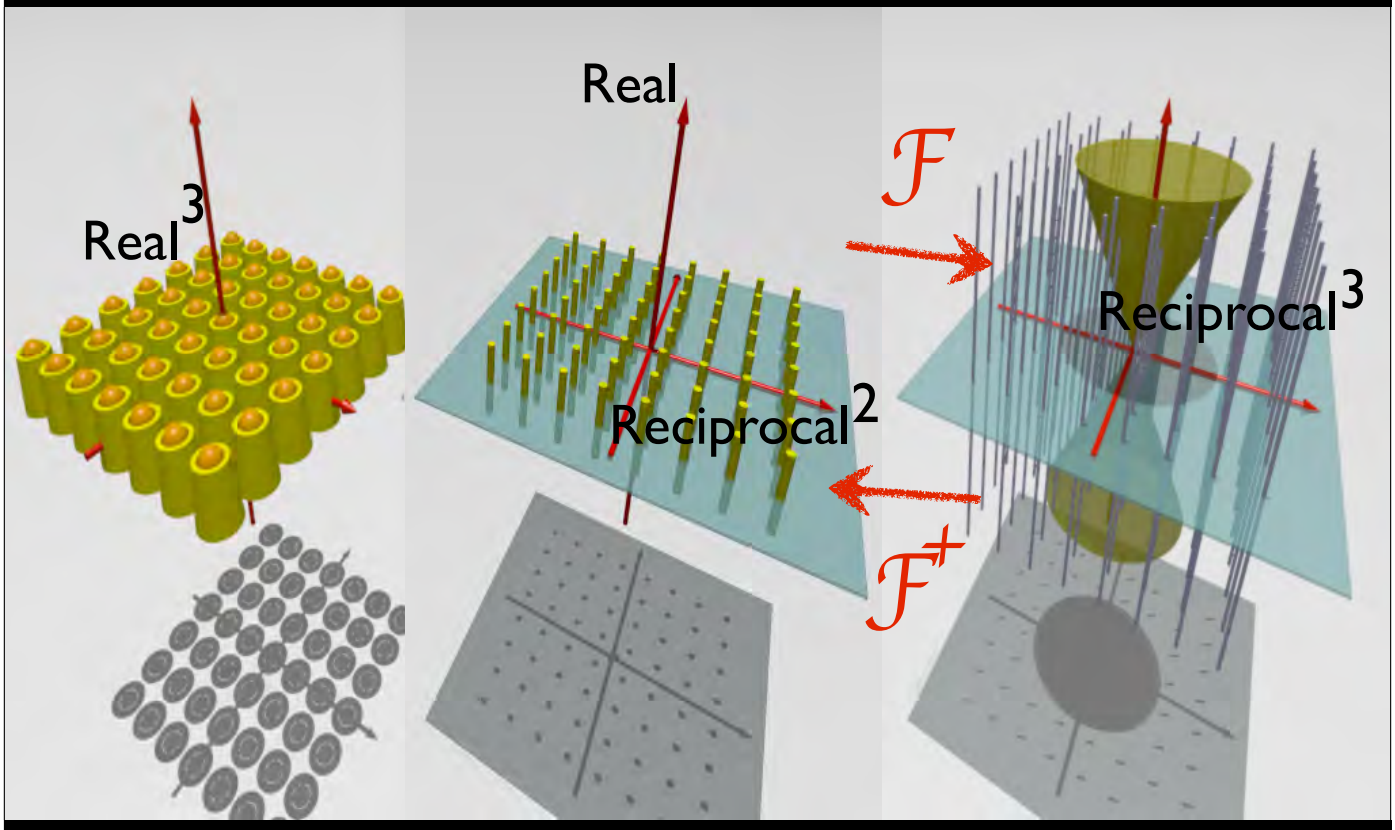
Fourier Space



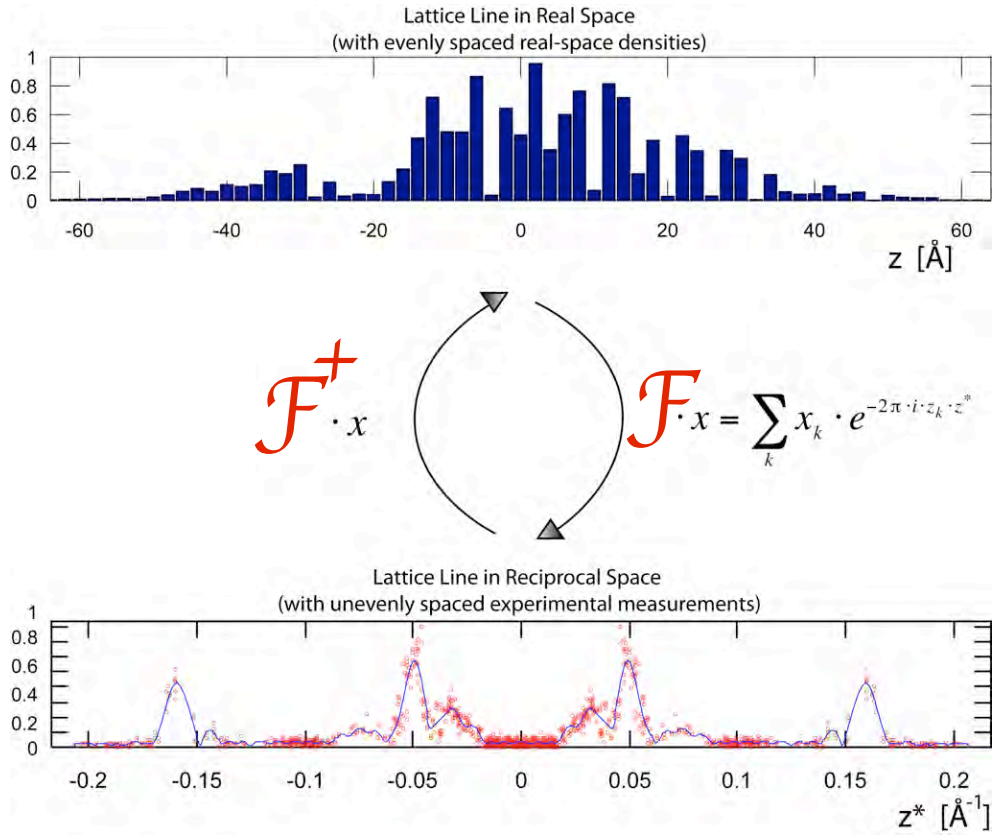
Real Space Fourier Space



Real Space Hybrid Space Fourier Space



Matrix *F* translates one real-space lattice line into Fourier space lattice line measurements



Inversion of Matrix *F* by Singular Value Decomposition

$$\mathcal{F}_{z,z^*} \; x_z = \hat{x}_{z^*}$$

Truncated Singular Value Decomposition:
Matrix *F* is represented by a quasi-diagonal matrix *S*, surrounded by V^T and *U*.

$$\mathcal{F}_{z,z^*} = U \cdot S \cdot V^T$$

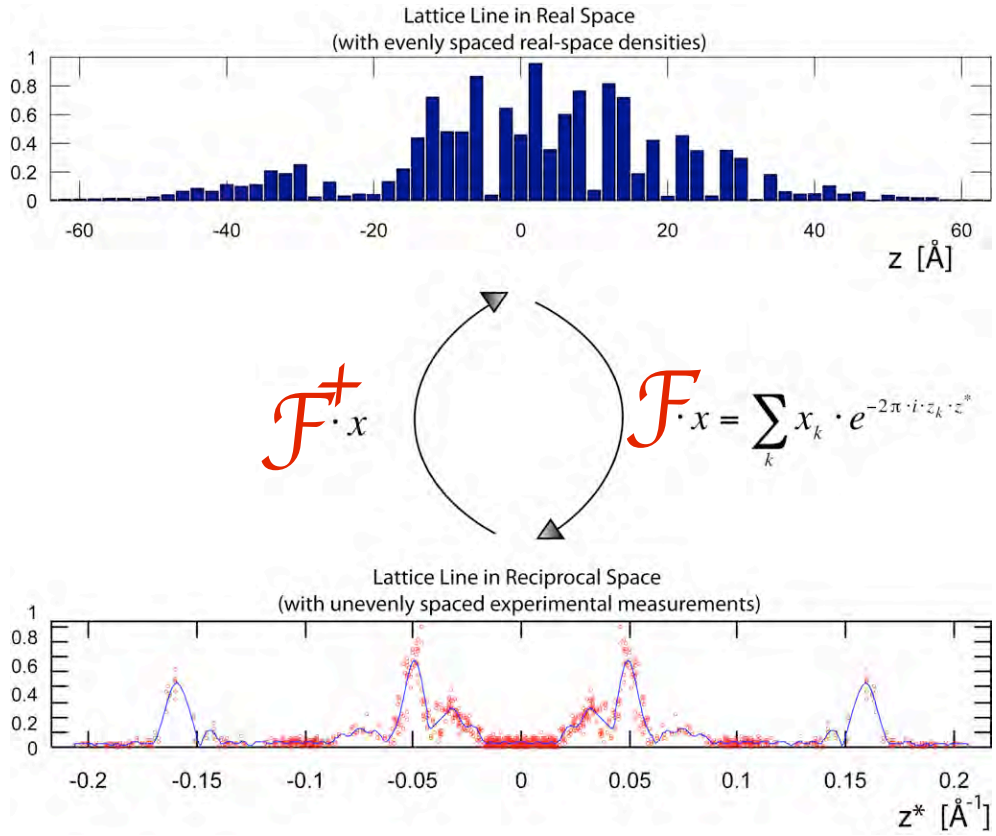
Inversion is by transposition of V^T, *U*, and inversion of *S*.

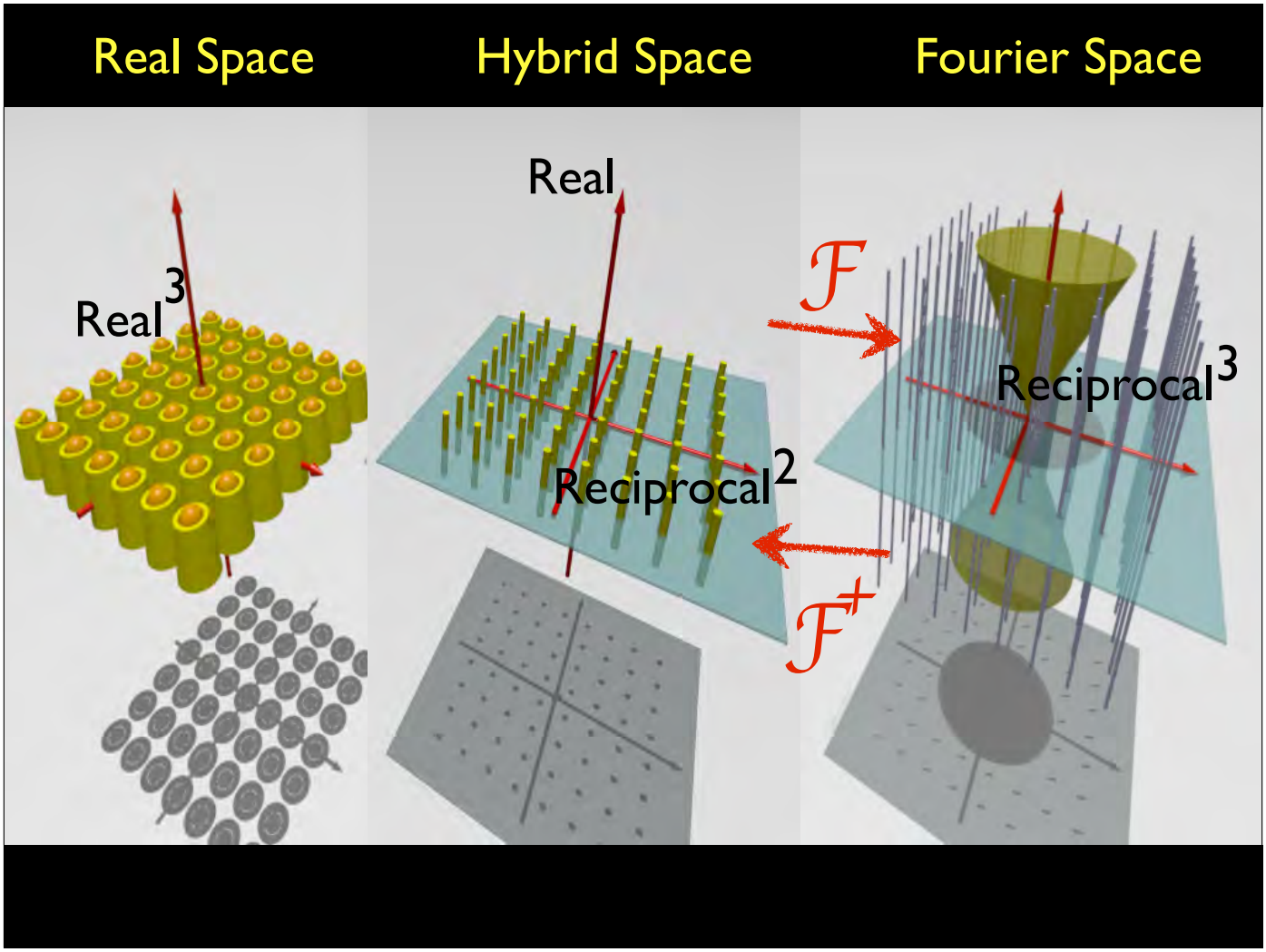
$$\mathcal{F}^{+}_{z,z^*} = V \cdot S^{+} \cdot U^T$$

$$x_z = \mathcal{F}^{+}_{z,z^*} \cdot \hat{x}_{z^*}$$

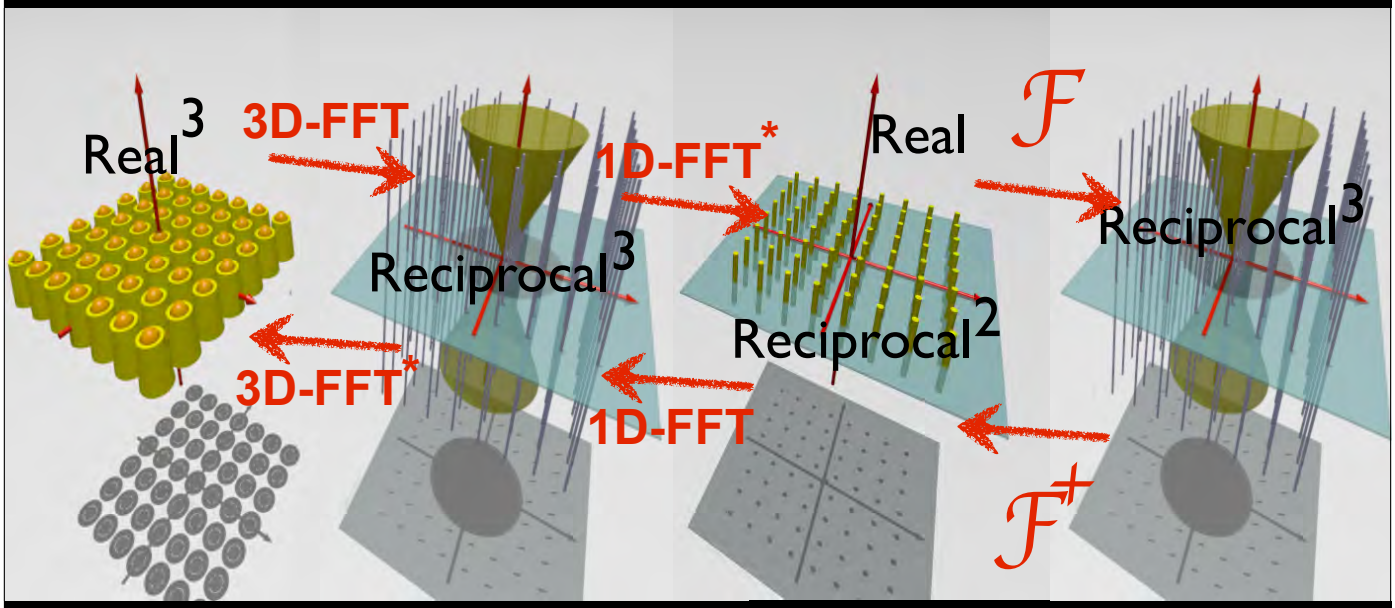
F is a least squares solution with lowest energy.
It is not a unique solution.

Matrix *F* translates one real-space lattice line into Fourier space lattice line measurements





Real Space | Evenly spaced Fourier Space | Hybrid Space | Non-evenly spaced Fourier Space

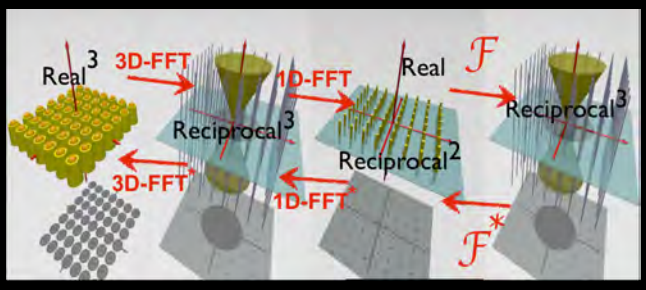


Real Space | Fourier Space

Apply Boundary Conditions

Enforce Constraints

Enforce Knowns



Real Space Constraints:
- Real Valued
- Non-negative densities
- Symmetric
- Membrane Slab Bounded

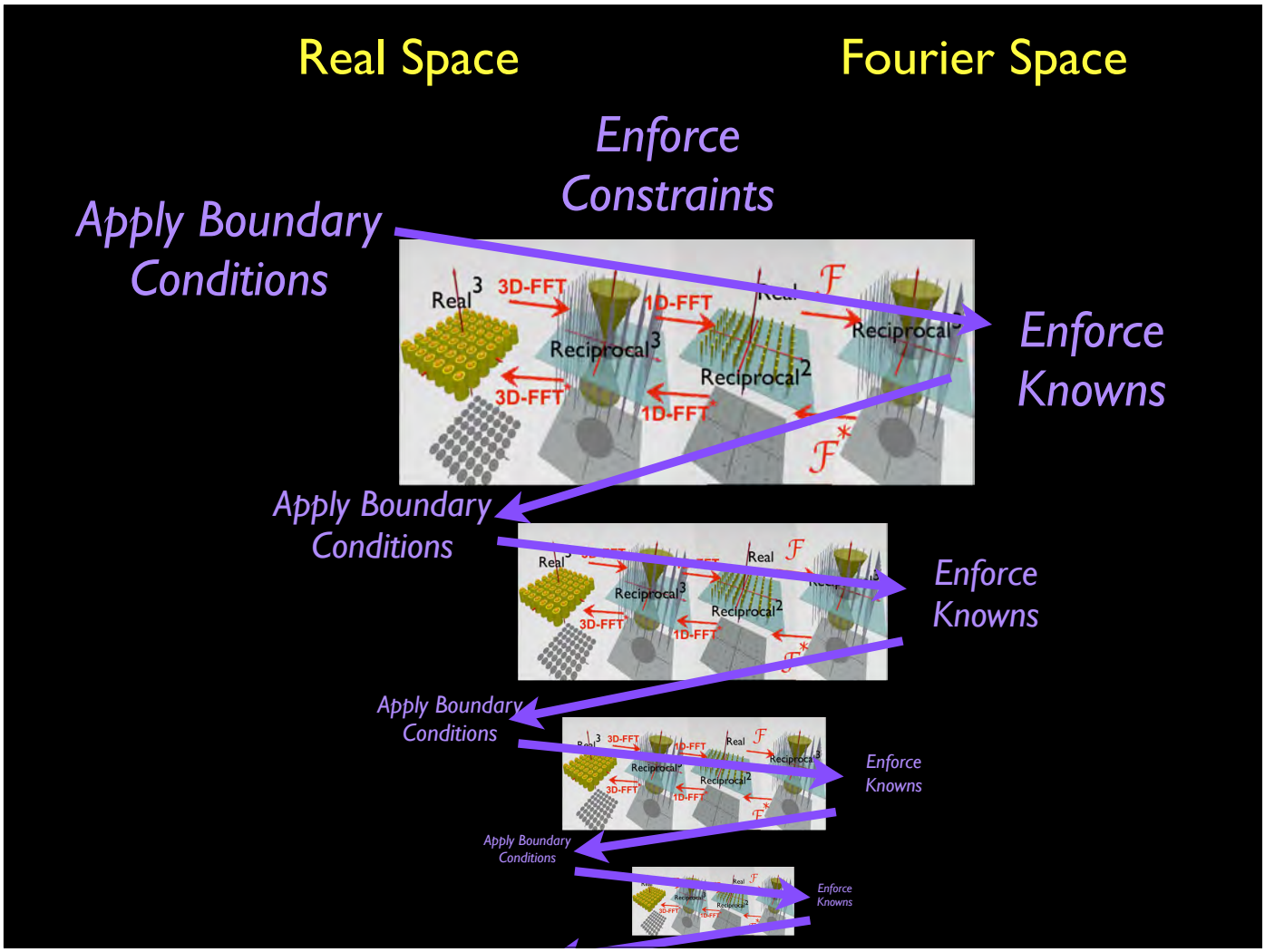
Real Space Constraints at later stages:
- Mostly Contiguous Density: Shrink-Wrap
- Protein surface well defined: Edge Detection, Noise Suppression

Fourier Constraints in missing cone:
- Known resolution range
- Scattering Profile

Fourier Constraints outside missing cone:
- Known Amplitudes
- Known Phases

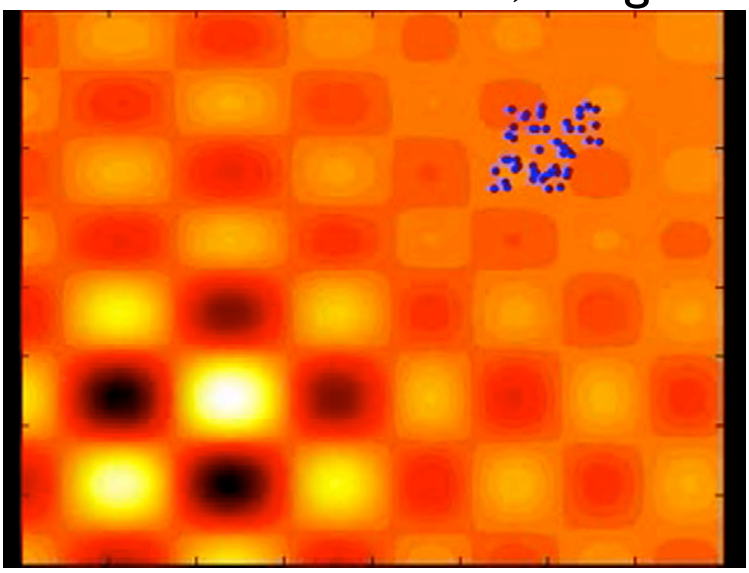
- Use individual weights for all constraints

- Apply constraints only in fractions



Particle Swarm Optimization

- Start with ZEROs in missing cone
- Get model for missing cone (= position)
- Refine model, using swarm optimization:

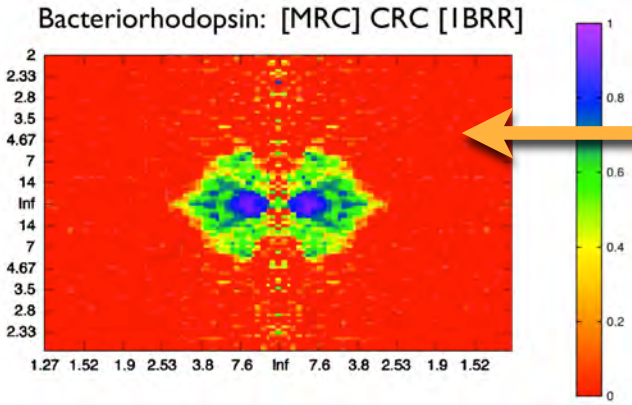


(new position) = (old position)
+ a*(random)
+ b*(personal best - old position)
+ c*(global best - old position)

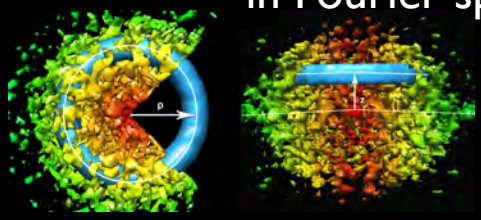
with a,b,c being variable parameters relative to the amount of exploration, memory and collaboration the solutions should have

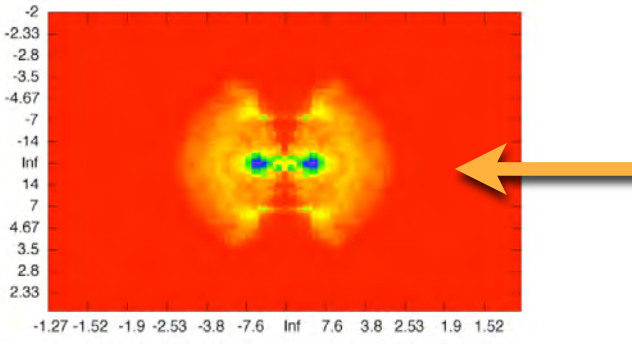
http://www.youtube.com/watch?v=b1zkbVRaguo

Resolution Plot



Cylindrical Ring Correlation (CRC) in x/z plane in Fourier space



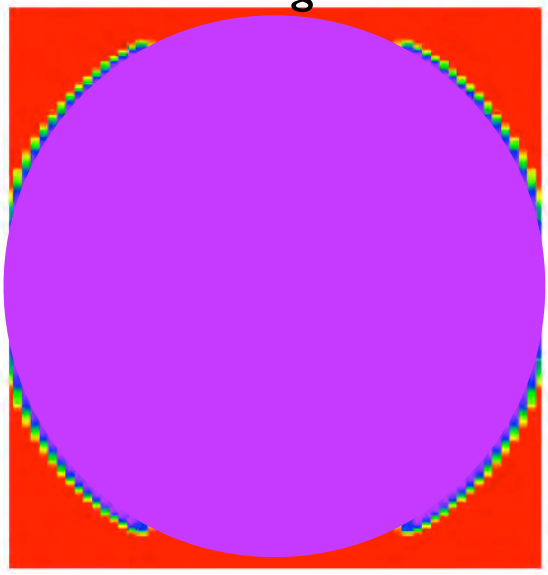


Cynlindrically averaged amplitudes in x/z plane in Fourier space

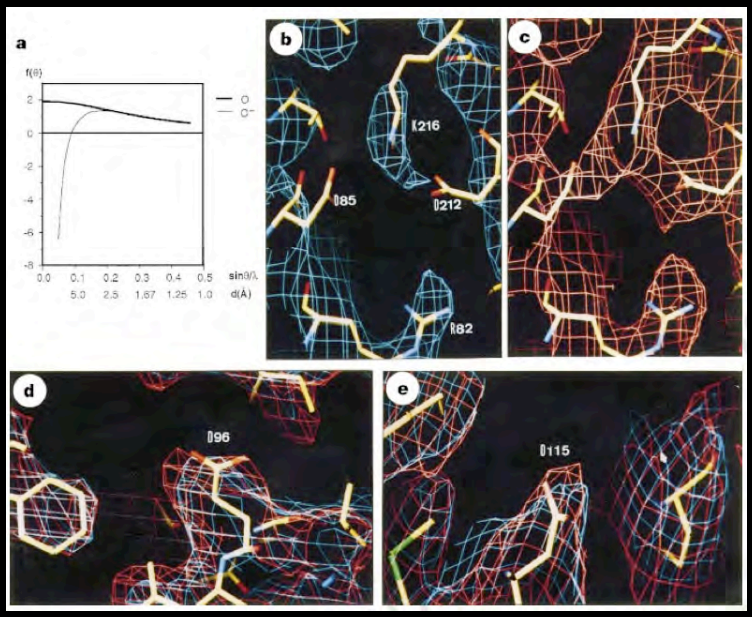
Simulated Dataset (without noise)

Start with Random Values

Zeroes in Missing Cone



Bacteriorhodopsin
(data from Kaoru Mitsuoka / Yoshinori Fujiyoshi)

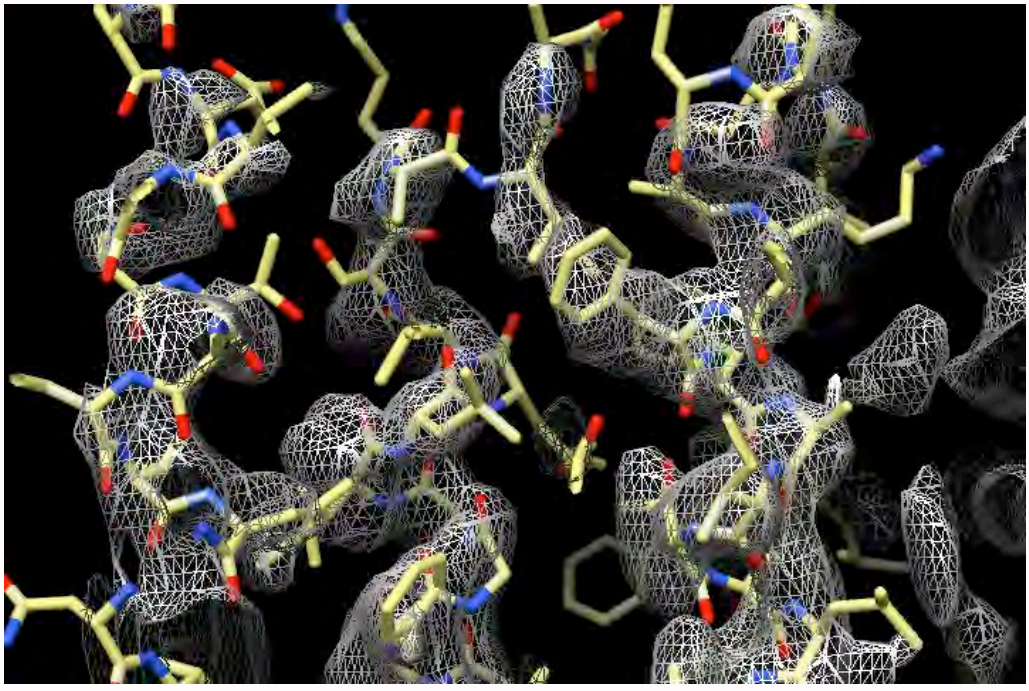


MRC EM Density
Kimura et al, 1997
(3.0 Å, electron crystallography)

1BRR
Essen et al 1998
(2.8 Å, XRD with lipids)

Projective Constraint Optimization

Bacteriorhodopsin
(data from Mitsuoka / Fujiyoshi)



PCO: 100 Initial Rounds, followed by 30 rounds Edge Detection

Real Space Constraints:
- Real Valued
- Non-negative densities
- Symmetric
- Membrane Bounded
- Shrink Wrap
- Edge Detection, Noise Suppression

Fourier Constraints:
- Known Amplitudes
- Known Phases
- Known resolution range
- Scattering Profile

Projective Constraint Optimization

Bacteriorhodopsin
(data from Mitsuoka / Fujiyoshi)

Schiffbase region

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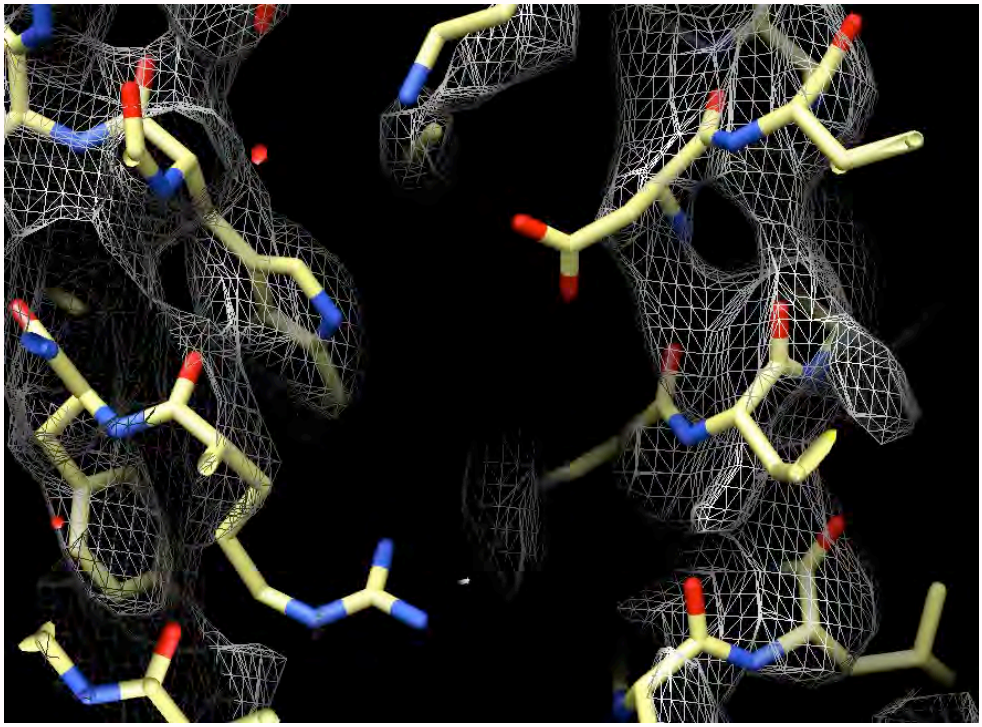

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Projective Constraint Optimization

Bacteriorhodopsin
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Lipid region

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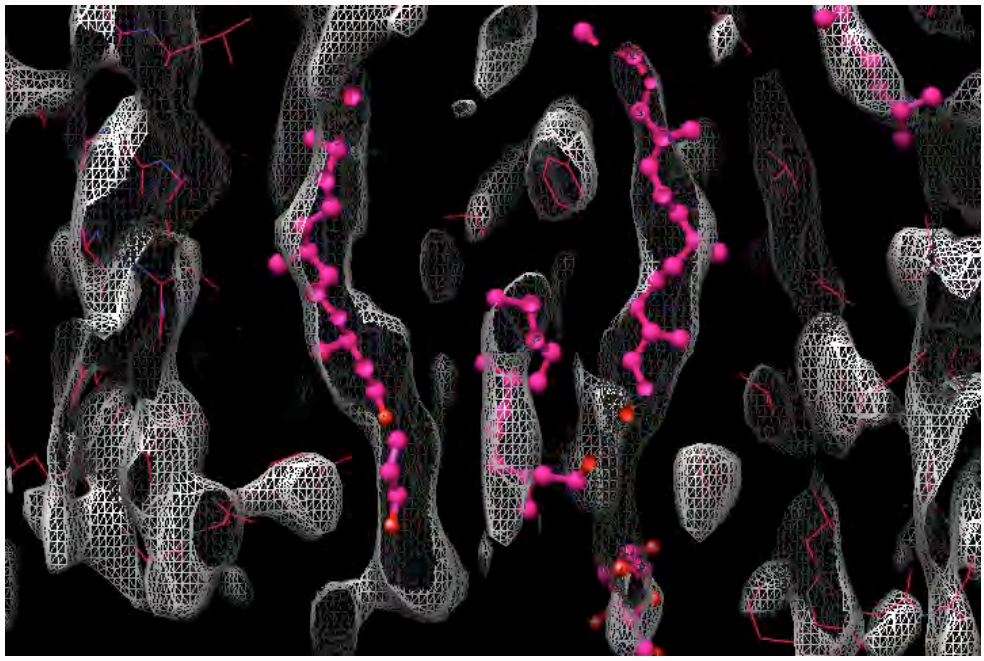
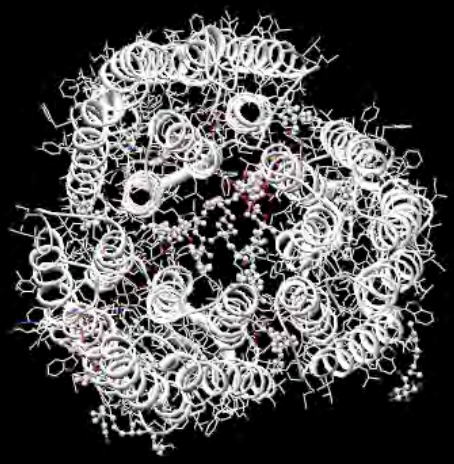


PCO: 100 Initial Rounds, followed by 30 rounds Edge Detection

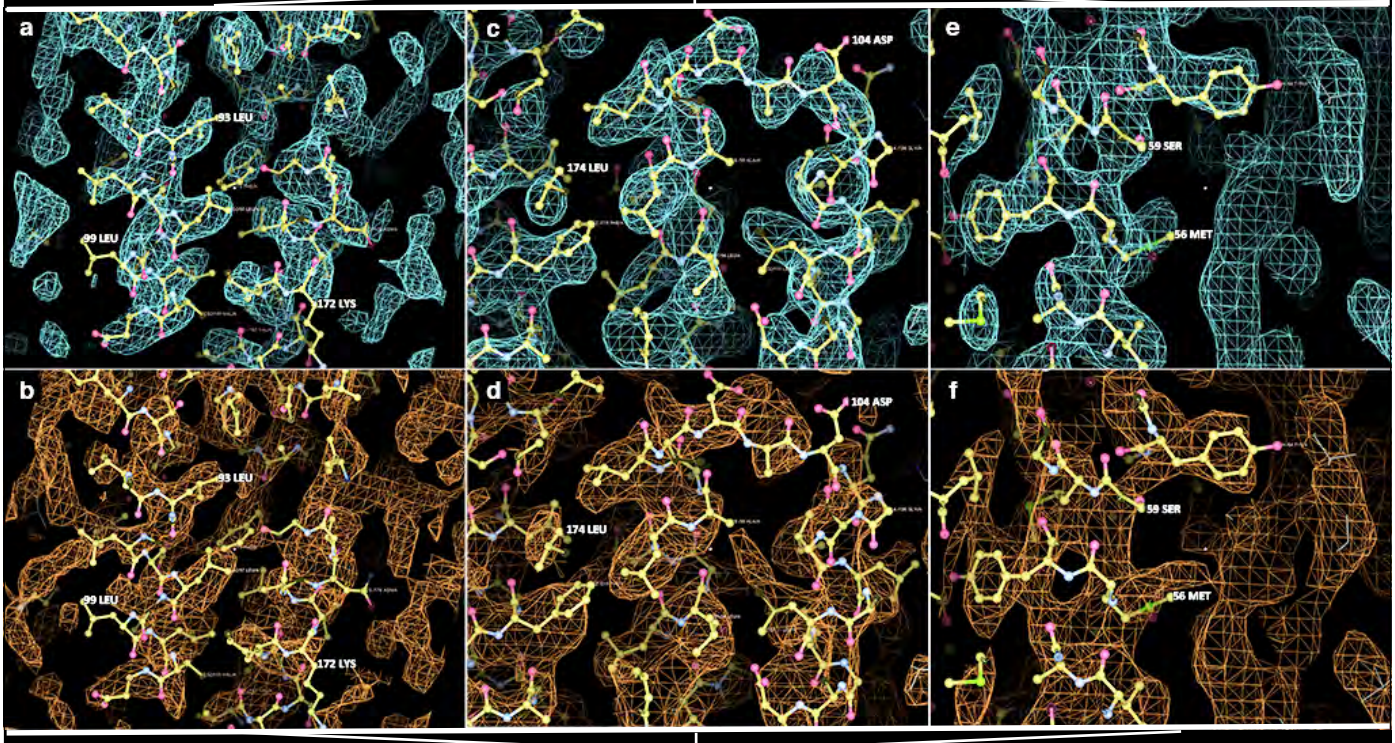
Bacteriorhodopsin
(data from Mitsuoka / Fujiyoshi)

Model: 1BRR (Essen *et al.*, PNAS 1998):
lipid in center of trimer



Structural Comparison

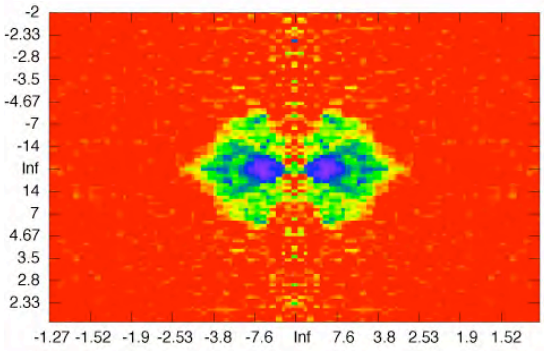
PCO Refined Data



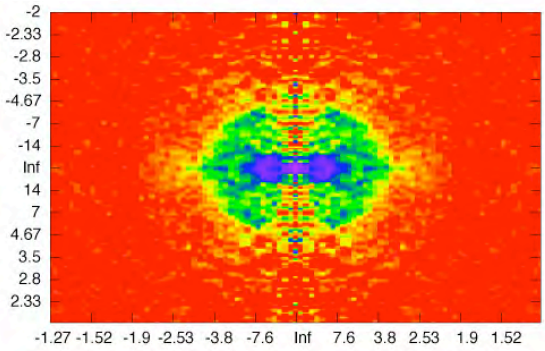
Kimura et al, 1997

Comparison - Cylindrical Ring Correlation (CRC)

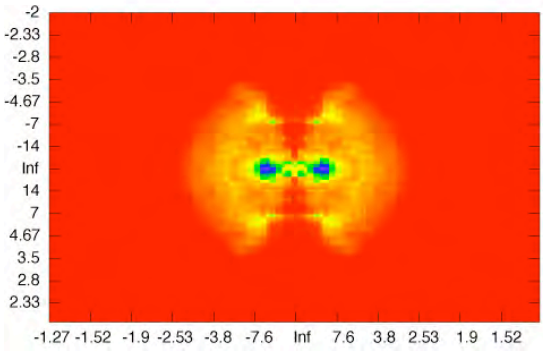
[MRC] CRC [1BRR]

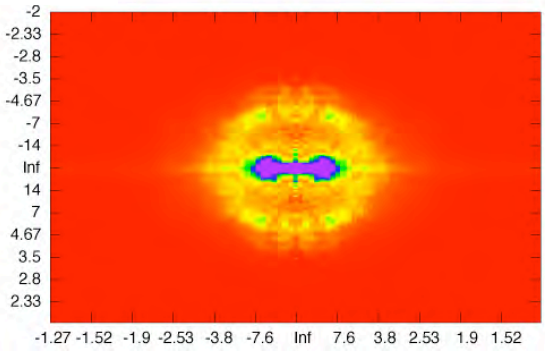


[PCO Refined] CRC [1BRR]



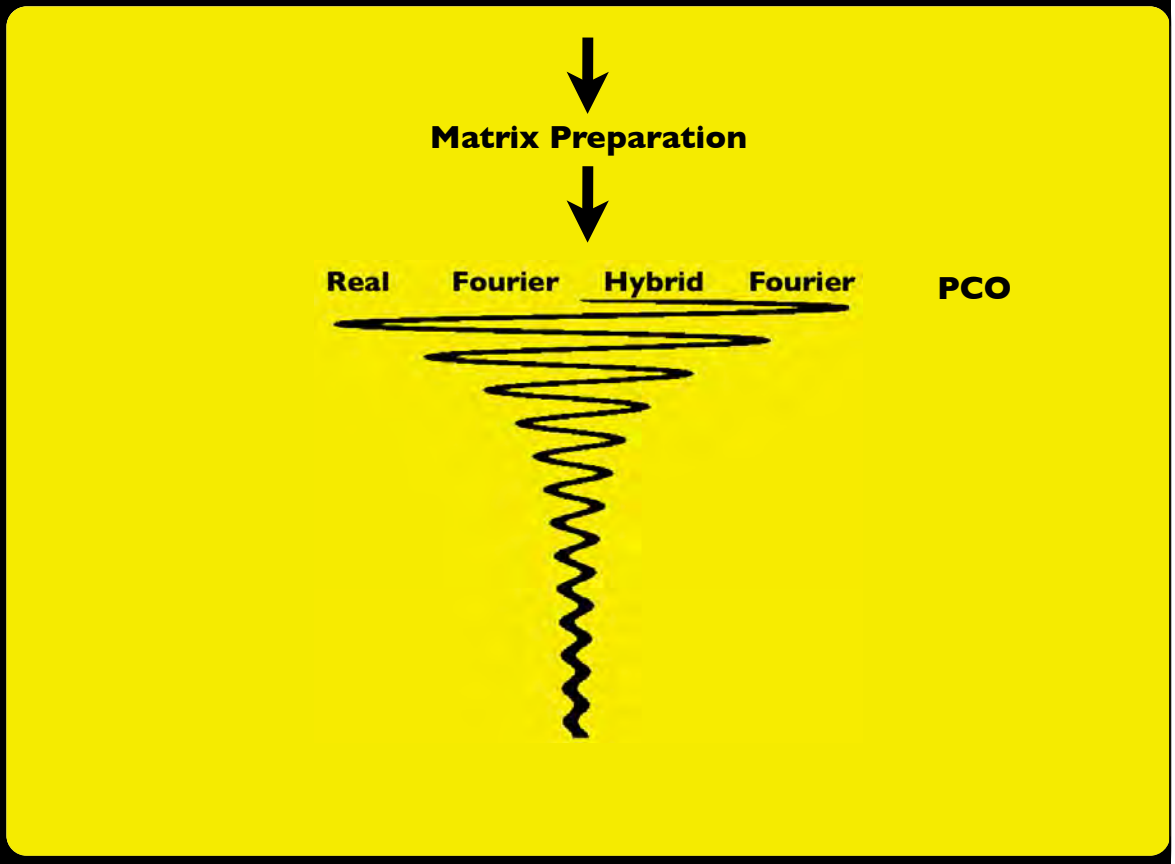
Cylindrically Averaged Amplitudes





2dx

PCO will become available within 2dx



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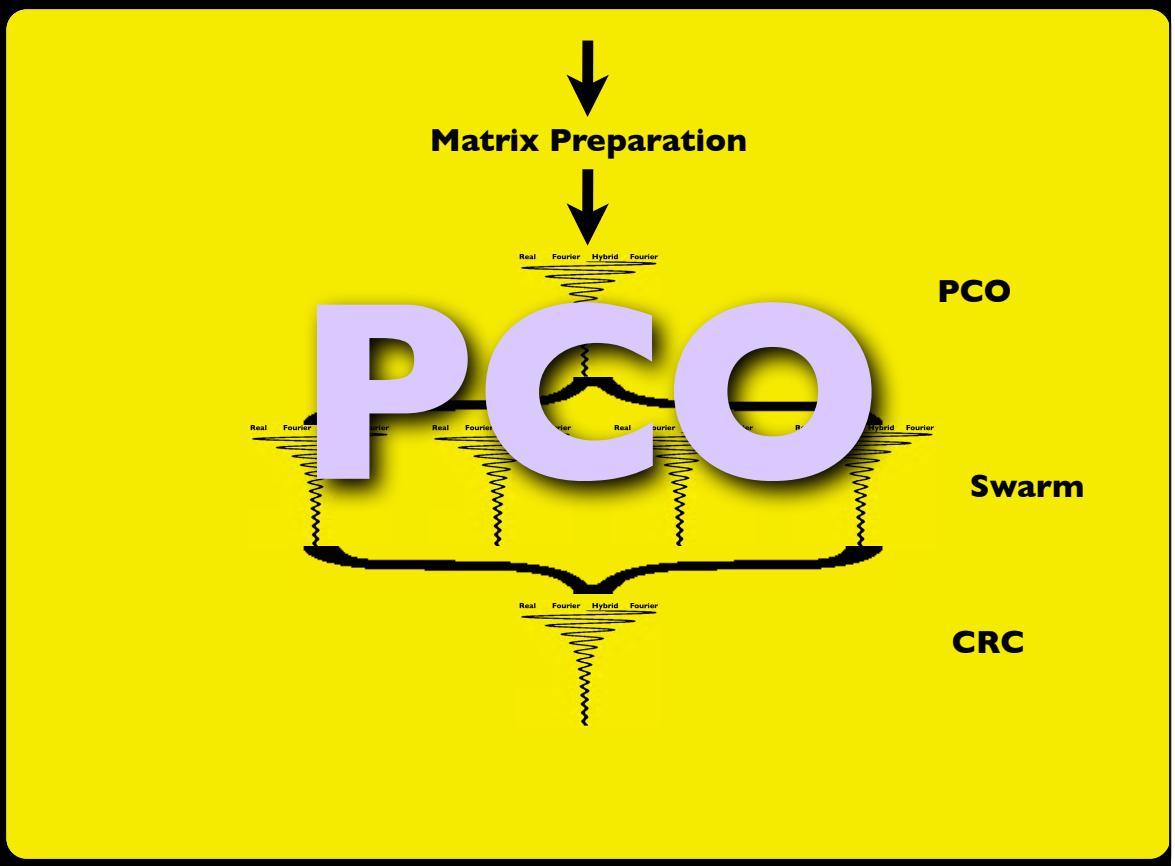
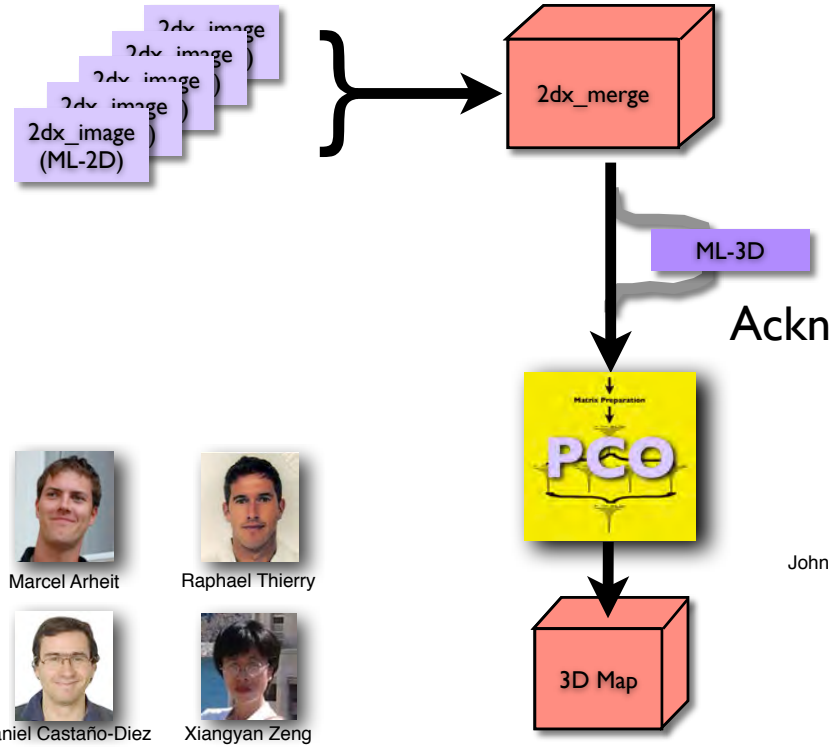


PCO will become available within 2dx

2dx.org



Acknowledgement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John Spence, University Arizona, TX
Kaoru Mitsuoka, Tokyo
Dan Masiel, UC Davis
Mike Sarahan, UC Davis
Nigel Browning, UC Davis
Tilman Schirmer, Uni Basel

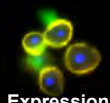
Marcel Arheit

Raphael Thierry

Daniel Castaño-Diez

Xiangyan Zeng

Electron Crystallography Bottlenecks

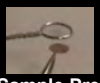

Expression


Purification

Samples: "PHS" (Pure, Homogeneous, Stable).
"Is a gel filtration profile still perfect after 1 week at 4°C ?"


2D Crystallization

2D crystals: Automation.


Sample Prep

Grid preparation: New sample supports (TiSi? Graphene?).


Imaging

Images: Automation? Phase contrast STEM?

E-diff: Automated electron diffraction in TEM.


Image Processing

MRC, 2dx, IPLT: Throughput, User-friendliness, Automation,
Maximum Likelihood for badly ordered 2D crystals.


Model Building

Missing Cone: PCO (Projective Constraint Optimization)