Iterative Helical Real Space Reconstruction cycle



asymmetric reconstruction

most novel part

imposed symmetry



symmetry search





Summary of steps in IHRSR procedure

- Box stretches of filaments from EM
- cut into overlapping segments of fixed length
 - optimal length dictated by disorder, Signal/Noise
 - extent of overlap generated by axial rise per subunit
- Do initial alignment to center segments avoid interpolation
- Generate an initial reference featureless cylinder works!
- Make guess about approximate symmetry
 - Can use information from power spectrum, AP SR, 2₁ screw...
- Azimuthal increment (number of reference projections) determined by diameter of filament, resolution desired
- Check convergence, check AP NQ statistics, check agreement between power spectrum and Fourier Transform of reconstruction

generator creates IHRSR script

~	IHRSR Script Generator 🗕 🗙
	Select input image stack file Number of images to use?
	Image size (pixels) 100 🔶
	Scale (A/pixel) 4.0000
	radial minimum (Angstroms) 0.0000 🗧 🔷
	radial limit (Angstroms) 50.000
	Angular increment of reference projections (degrees) 4.0000
	In-plane angular deviation allowed from 0 or 180 degrees 10.000
Г	Point group?
	Click if point group symmetry exists in (collational symmetry) = 2
	Number of cycles 50
	Symmetry search Center of Search:
	Rotation per subunit (degrees) 0.0000 🐥 Rise (Angstroms) 0.10000 🖨
	Search increments: Delta Phi 0.10000 🔶 Delta z 0.10000 🜩 Finish
	Search range (pixels) in AP NQ 2 Quit

hplotn is utility for displaying alignment parameters

<	helixplots		////// - ×
	Histograms	Number of Records= 31	23
	X-shifts	V-shifts Total Shift	
	Coef. of Corr.	In-plane Angles Reference Freq	uency
	Scatter Plots		
	Xshift vs Vshift	Xshift vs referencecc vs reference	e
	Xshift vs angle If Print button pressed, next plot will Print be printed		
	angle vs cc	Osiect Pile	Exit

reference frequency distribution is important reality check



no correlation should exist between shifts and rotations



"weird" rotations have poor coefficients of correlation



xhelicals is graphical symmetry search



filrecon is utility for filament reconstitution

✓ 2D filament reconstitution		×
Scale (A/pixel) 1.7800		
Angle (degrees) 22.067 🖕 Axial rise (Angstroms) 1.4490 🖕		
Length of new image (pixels)		
Length of segments (pixels)		
Celest Angles Pile Maximum angular deviation	000	
Generate SPIDER script Exit		

parameters are from TMV...

Y-Shift shifts for segments 120.0 100.0-80.0 Frequency 60.0 40.0 20.0-Û 75.0 275.0 475.0 675.0 875.0 shift (pixels)

filrecon is utility for filament reconstitution

filrecon is utility for filament reconstitution



What is to be done (today)?

 create solid cylinder as starting model ../spider_linux_mp MO 3 volume001 100,100,100

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run generator to create