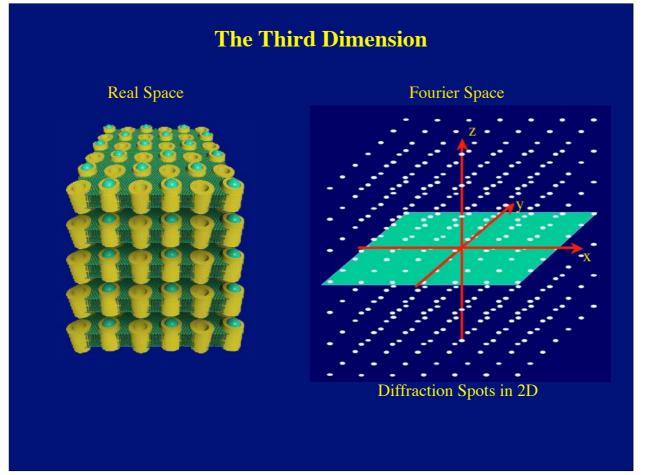
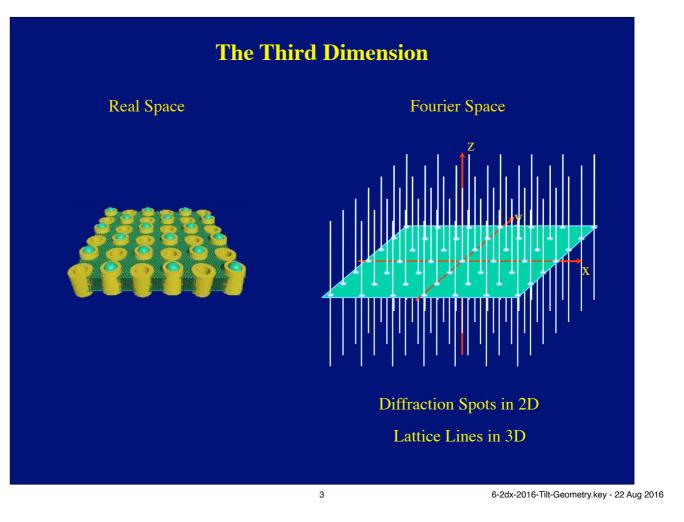
# Tilt Geometry

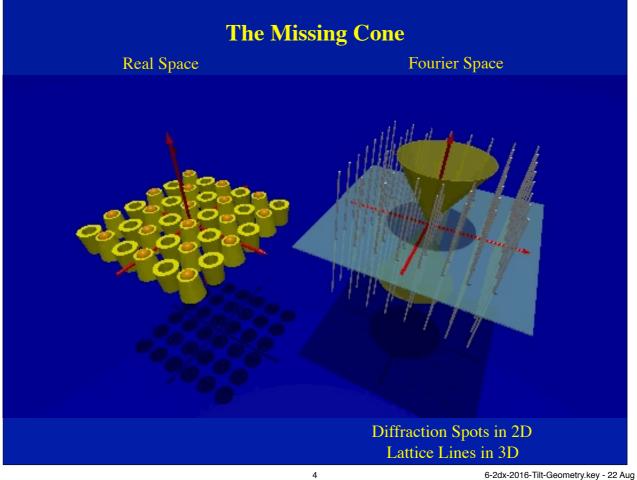
Henning Stahlberg, Biozentrum, Uni Basel, Switzerland c-cina.org

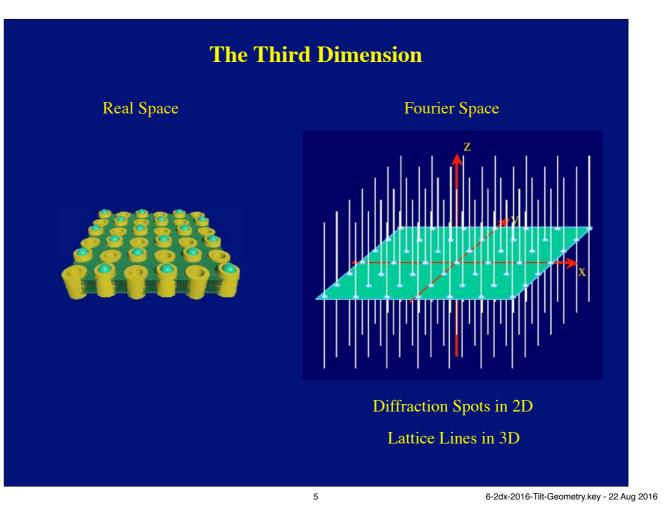
> 2dx Workshop Basel, August 23-26, 2016

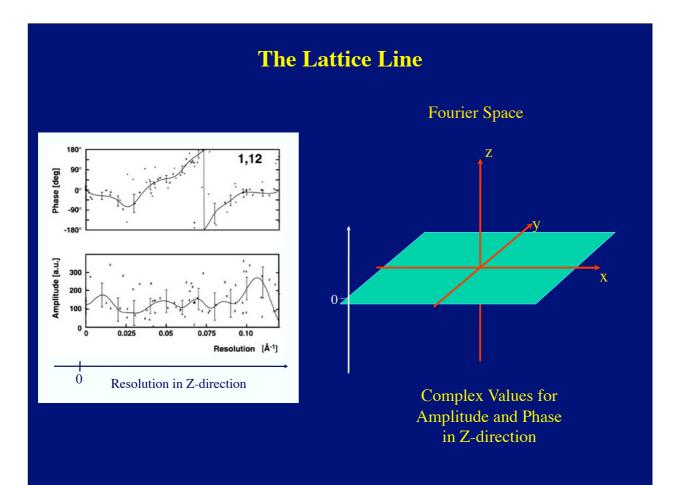
> > 1

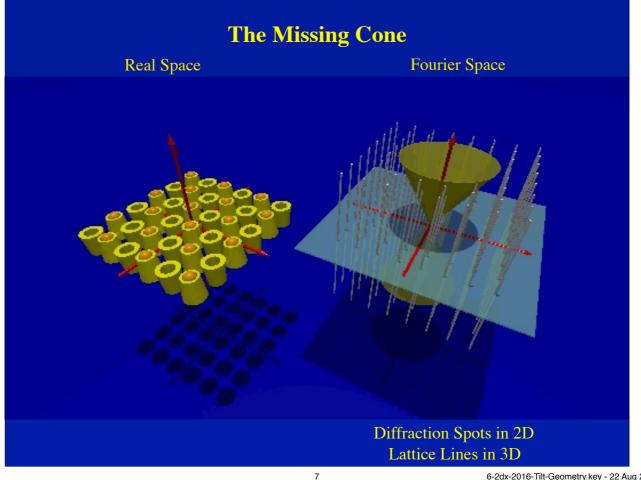


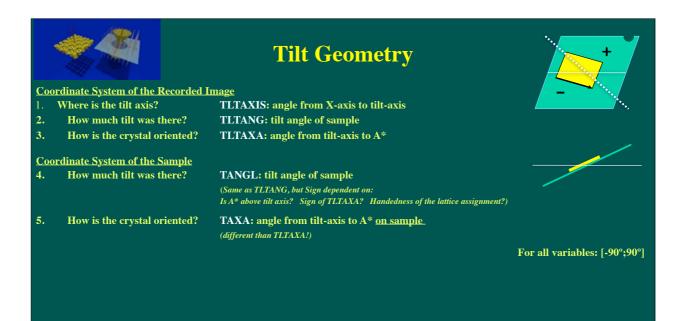












	Tilt Ge	eometı	<b>·y</b>								
Coordinate System of the Recorded	i <u>mage</u>						/*•.	•••			
1. Where is the tilt axis?         TLTAXIS: angle from X-axis to tilt-axis											
2. How much tilt was there?	TLTANG: tilt angle of sa	mple									
3. How is the crystal oriented?	TLTAXA: angle from tilt	-axis to A*									
<b>Coordinate System of the Sample</b>											
4. How much tilt was there?	TANGL: tilt angle of sam (Same as TLTANG, but Sign dep	endent on:				/					
	Is A* above tilt axis? Sign of TL			lattice assign	ement?)						
5. How is the crystal oriented?	<b>TAXA: angle from tilt-ax</b> (different than TLTAXA!)	tis to A* <u>on sa</u>	mple_								
Four ways to determine/refine tilt g <ul> <li>From defocus of negative</li> </ul>	eometry: (ctfsearch3)	 12780 12610 11770 11400 11090 10590 10580	Defoc 11990 10920 11230 10190 9630 9540 9460	us values 10960 10800 10190 9480 9360 9260 8330	in 49 pc 9900 9480 8890 <b>8685.35</b> 8590 7850 7310	9510 9330 8460	on imag 8430 8070 7390 6790 6730 6060 5930	e 7500 7090 6890 6590 5930 5670 5200			

Determination and Refinement of the Tilt Geometry in 2dx

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6-2dx-2016-Tilt-Geometry.key - 22 Aug 2016

## Determination and Refinement of the Tilt Geometry in 2dx

# Defocus Gradient accross image

Rough Tilt Geometry, but correct sign of tilt angle (TLTAXIS, TLTANGL for grid) No clue about crystal orientation (TAXA, TANGL for crystal)

#### Lattice Distortion

Precise Tilt Geometry if tilt larger than 25°, but no clue about sign of tilt angle (sign taken from above)

# SpotSplitting

Precise Tilt Geometry if tilt larger than 25°, but no clue about sign of tilt angle (sign taken from above)

## 3D Merging

Precise Tilt Geometry for sample (TAXA, TANGL for crystal), can calculate back to carbon film orientation (TLTAXIS, TLTANGL for grid)

11

•					MloK1-fili	m   2dx (4.0.0	)								
					<b>1</b>	<b>I</b>							e d		
loK1-	K2-2014-11-17_MloK1_cAMP/2014-11	1-17_12-05-02	🛯 🛋 MloK1	00/MIoK1009	9001200										
	Init Program	Get Defoc	us & Tilt									0			
	Init Files and Parameters	_								Results					
	Citylate Fins	Setup expected til	Setup Gracient in the accross image								37741.16,37573.21,22				
	Defocus	Graa	1en		crc	)SS%/	ma	01 <i>0///</i> /	111	defocus TLTAXIS	37741.		39.6767		
								9		TLTANG			16.3541		
	Get Lattice & Tilt Refine Lattic Rough Tilt				rect s	(TLTA	XIS, TL	TANGL	for gr	$d_{10}^{89.577}$					
			out crystal orientation (TAXA, TANGL for cry								TAU 25.88				
	CTF Correction (Part 1)	The Geometry	Determine Tilt Geometry Yes No										16.350		
	Get SpotList	Determine T											1 89.677		
	Unbend I	Which algor	Which algorithm to use for Tilt Geometry from Defocus?							DEFOCUS_TLTAXIS					
		0=CTFF								DEFOCUS_TLTANG					
	Get SpotList (or refine)	Grid Tilt Axis	Grid Tilt Axis (TLTAXIS: X-axis -> Tilt axis, on image)												
	Unbend II Unbend MovieA1 Unbend MovieA2	Output (Dauble	Output (Double click for legbrowser)								0		25.881		
		Output (Double click for logbrowser) Verbosity Level: Low Harm Mark Inon-ring Tit panels into one overview image ====================================								mages FFT of Downsam	pled Image				
		37247.2	37267.2		37507.2	37617.2	37987.2 37957.2	37967.2		Periodogram					
	Unbend MovieB	37307.2	37397.2		37567.2	37737.2		37947.2		TXT: Defocus T TXT: Positions					
	CTF correction (Part 2)	37277.2	37337.2	37467.2	37457.2	0.0	0.0	0.0		TXT: runfile f					
		37307.2	37327.2	37507.2	37657.2	37737.2	37657.2	0.0		Tile 4,4	or writebouring				
		37287.2	37297.2	37527.2	37617.2	37877.2	37697.2	37757.2		Gallery of Tho					
		37307.2	37317.2	37507.2	37577.2	37777.2	37887.2	37937.2		TXT: runfile f	or tiltgeom				
		37267.2	37337.2	37437.2	37617.2	37697.2	37837.2	37867.2		TXT: result of	runfile for	tiltgeom			
	0	Defocus = 377		3.21,22.46						TXT: runfile f					
	DEPENDENT SCRIPTS 2dx_deftilt_sub.com	TLTAXIS = $89$ . TLTANG = $-16$	.3541			TXT: result of			narki						
45 45	initialize	TLTAXA = 26.	<pre>==== 2dx tiltgeom2: Determine other tilt angles with respect to lattice. ====================================</pre>								Gallery of Marked Thonring Fits				
		Status	991/							Gallerv of Mar	Preview	FIUS			
		Calc. Mag=			505.6			I	ast QVAL=		Manager and Annual College				
		Unbend Unb.I		<i>IQ1</i> 28	IQ2 46	1Q3 47	IQ4 52	1Q5 47	1Q6 54	QVI 774					
		Unb.II		29	60	51	59	63	49	909		1.1			
		MovieA		-		-	-	-	-						
		MovieB PowerBins		-	-	[A]	- 15	- 12	9	7 5 3					
		(noise=7)				#	808	81	56	14 8 7					
		Tilt Info Grid TAxis				Defoc. 89.7	Latt. 86.3		SpSpli	t Mer - 87		Ser Se	file (4)		
		Grid TAxis Grid TAngle				-16.4	-23.3			28					
		Xst. TAxis				25.9	28.2			- 25					
100		Xst. TAngle				16.4	23.3			- 28	· · · · · · · · · · · · · · · · · · ·	State Party			

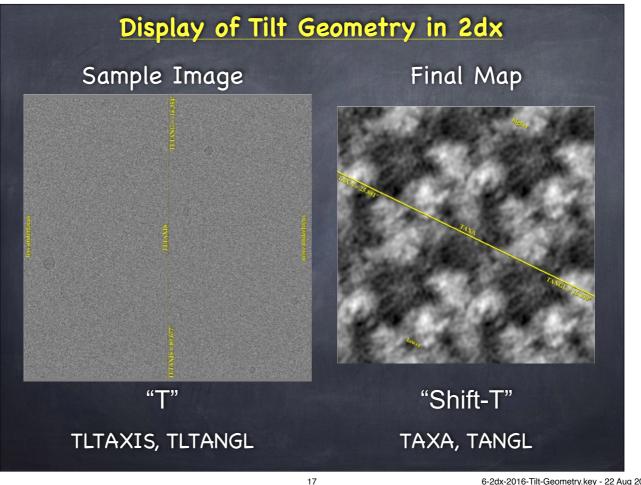
•				MIoK1-fil	lm   2dx (4.0.0	)								
					<b>I</b>									
lloK1-l	K2-2014-11-17_MloK1_cAMP/2014-11	-17_12-05-02 🛛 🛛 🖾 Mlo	K100/MIoK100990	01200										
	Init Program	Get Lattice & Tilt								O B C				
	Init Files and Parameters													
	0.1.1.1.555	Setup		🔁 🔀 Re	sults									
	Calculate FFTs	Crystal Tilt Angle (TAN	GL =TLTANG*AisA	bove*Sign	TLTAXA*Hand	16.350		14	attice	"17.205,-34.515,				
	Get Defocus & Tilt			1.1	LATTICE_TLTAXIS "86.34									
R	Get Lattice & Tilt							1.1	ATTICE_TLTANG	"-23.3142"				
-		Lattice Determination	attice Determination							"30.1453				
*	Refine Lattice								LATTICE_TAXA "28.07180					
	Calculate Crystal Tilt from Carb	Determine Lattice Yes No							LATTICE_TANGL "23.31427					
		Lattice Determination Algorithm to run							CALCULATEDMAG 22007.93					
	CTF Correction (Part 1)	0=FindLattice (sea	rch for one or two e	14	laterror_abserror 51.62									
	let Satterico D	Riel Unit Cell length (for entire project) Still @ mail (n) piect) 131.0 131.0 131.0 131.0 131.0 131.0 131.0 131.0						14	aterror_rmsderror	1.2754				
-	Laince D							14	aterror_peaksused	140 of 14				
	Unbendi	Geometry 'ff' tilt's larger than 25°,							aterror_nodepeakdensi	ty 1.650				
	Get SpotList Preceise Til	t Geometry	if filt lo	1.1	LATTICE_done									
	Unbend II but no olu	<b>a</b> 17 205	-34 515	SI	POTS done									
	but no clu	e about sigr	bgOdwse <sup>†</sup> III†	n abov	(e)									
	Unbend MovieA1	New 2nd lattice would	ld have been 0.0	FI	FFT of Image									
	Unbend MovieA2	Due to the bad score	e it will not be generate a firs	e used. st spotli	st			FI	FFT of Downsampled Image Edge-Tapered Image					
		==== 2dx_lencalc: To c ==== emtilt: To calcu	calculate some v	values fo	or the tilt of	cometry dete	erminatio	on. =====						
	Unbend MovieB	1. TILTAXIS =	86.34969 = 2	ALPHA - F	PHITLT	410001 010110			Powerspectrum					
	CTF correction (Part 2)	2. TILTAXIS = Taking first option.	-33.35969 = 1	ALPHA + F	HITLT			Pi	Pre Band-Pass Powerspectrum					
		==== NOT saving tilt of	meometry from 1a	attice, b	because of 10	w tilt. ====		Po						
	Generate Map	==== 2dx_calcmag - to												
		==== Theoretical magn:	ification is 220	007.938,	given magni:	ication is 2	22500 ===		Origin-Shifted Average Powerspectrum					
		==== Angle in reciproc			•									
		==== 2dx_laterror to : Absolute Error: 51.62		current	lattice. ===									
	DEPENDENT SCRIPTS	RMSDn ERROR: 1.27549 Peaks Used: 140 of 140												
418	2dx_makedirs	Nodes/Peak Density: 1.	.65014											
10	initialize	==== 2dx getLattice - ###################################	######2dx getLat	##										
		Status							Preview	1				
		Calc. Mag=	22007					Last QVAL=	909.5					
		Unbend Unb.I	IQ1 28	1Q2 46	IQ3 47	IQ4 52	1Q5 47	<i>IQ6</i> 54	QVAL 774.7					
		Unb.II	28	60	51	52	63	49	909.5					
		MovieA		-	-	-	-	-						
		MovieB		-	-	-	-	-						
		(noise=7)			[A] #	15 808	12 81	9 56	7 5 3	•				
		Tilt Info			Defoc.	Latt.	01	SpSplit	100 March 100 Ma					
		Grid TAxis			89.7	86.3			87.8					
		Grid TAngle			-16.4	-23.3			-28.0					
		Xst. TAxis			25.9	28.1			25.8	A DESCRIPTION OF THE OWNER OF THE				

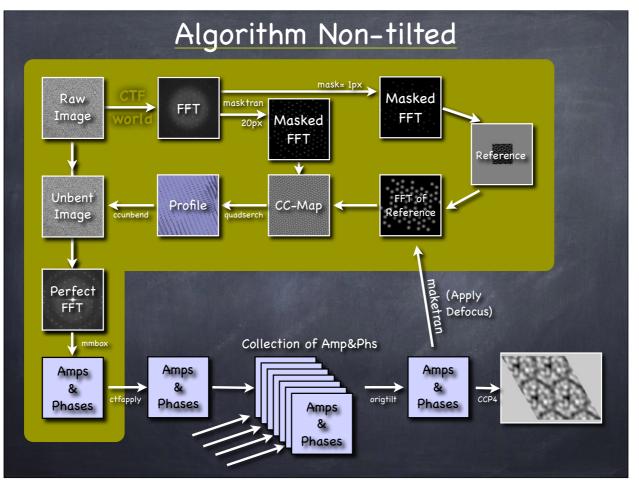
13

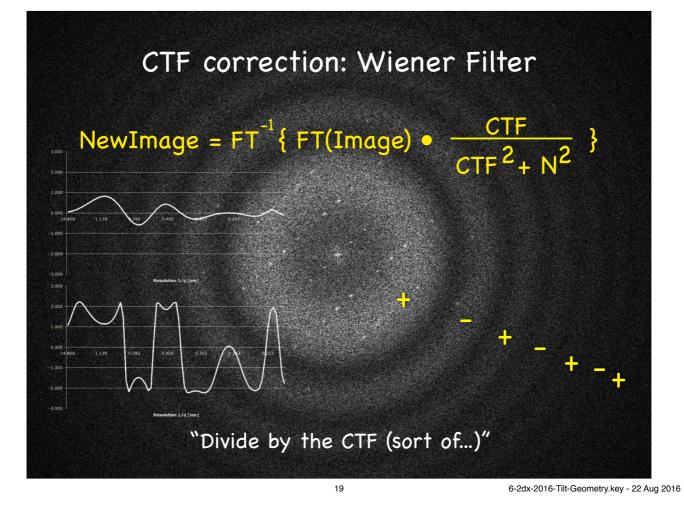
				MloK	1-film   2dx (4.0.	D)				
4				1	1 🐷 1					¢°
⊗ Mic	K1-I	(2-2014-11-17_MIoK1_cAMP/2014-11-	17_12-05-02 🛛 🐼 🖬 MloK1	00/MIoK10099001200						
0		Evaluate Lattice	Refine Tilt from Spo	otSplitting						
•	-	Get Spacegroup & PhaseOrigin								
-			Setup			Level:	Simplified	Results		
\$	1	Set PhaseOrigin Manually						TTREFI	NE_TLTAXIS	" 89.6770"
	D	Generate SymMap						TTREFI	NE_TLTANG	" -16.3540"
	-		Common Image Processin	a				TTREFI	NE_TLTAXA	" 26.8183"
	1	Mask Crystal from Polygon		<b>X</b>				TTREFI	NE_TAXA	" 25.8773"
	1	Refine Params Unbend I	Lower Resolution Limit (R	ESMIN)	200			TTREFI	NE_TANGL	" 16.3540"
	D	Refine Params Unbend II	Upper Resolution Limit (R	ESMAX, in Angstroem	s) <b>6</b> 5.0					
			ALAT (Z-dimension of unit		200					
		Refine Params Unbend I (Coars								
	1	Refine Params Unbend II (Coars								
	Þ	Refine SpotList	Switches For Algorithm Se	lection						
		Refine Tilt from SpotSplitting		0		_				
	D	Refine Defocus (Not Recommen	Mode of operation	0=nothing	0	<u></u>				0
	-		Output (Double click for log	browser)	Verbo	sity Level: Low	/	🖸 🥫 🔀 Images		*
		Refine Params (Only Tilted Imag	*****	########2dx_refine	tilt###########	*****	******	#		
	1	Modify Image Amplitude	==== Anaconda Python exe	cutable not found.	Is it install	ed ??? ======				
		SpotSplit	opt/anaconda/bin/p Cation of Ana	conda Python is de	fined in Prefe	rences. =====				
		Image Inventory	==== 2dx T finetilt ==== 2dx ttrefine.exe -	To refine values.						
		Precise Til	t Geometry I	t filt larg	er than	25º,				
	-	but no du	TLTAXA : old 26.823	of w tilt 35 and	alo (siar	takan	fron	n above)		
		Cleanup Dur no clu	==== 2dx refinetilt - no	rmal end. ======						
		All Parameters	######################################	###2dx_refinetilt ting-point excepti	finished.##### ons are signal	ling: IEEE_IN	VALID_FL	# AG		
		0								
	-	DEPENDENT SCRIPTS								
	42	2dx_sym2spcgrp_sub.com 2dx_makedirs								
	42	initialize	Status						Previ	ew
			Calc. Mag=	22007.9				Last QVAL=	460.4	
			Unbend	101 10		104	105	106	QVAL	
			Unb.I Unb.II	35 7 39 8	4 66 3 65	96 113	168 161	219 228	415.3	
			MovieA	-		-	-	-		
			MovieB	-		-	-	-	200	
			PowerBins		[A]	15	12	9 7	5 3	
			(noise=7)		#	818	77	59 15	7 7	
			Tilt Info		Defoc.	Latt.		SpSplit	Merge	
			Grid TAxis Grid TAngle		89.7 -16.4	86.3		89.7 -16.4	87.8	
			Xst. TAxis		25.9	28.1		25.9	25.8	
			Xst. TAngle		16.4	23.3		16.4	28.0	
			-							

• •	•	MloK1-film   2dx (4.0.0)	
*		🏛 📪 📜	¢°
2D	Merge Once	Refine PhaseOrigins Once	
	Refine PhaseOrigins Once		
3D \$	<ul> <li>Refine PhaseOrigins Once</li> <li>Refine &amp; Merge (Iterative)</li> <li>Final Merge</li> <li>Generate Merged Map</li> <li>Refine Merged Map (3D only)</li> <li>Sharpen Refined Map</li> <li>Quality Evaluation</li> <li>Generate Image Maps</li> </ul>	Setup       Level: Simplified <ul> <li>Level: Simplified</li> <li>Phaseorigin Refinement HK Max</li> <li>20</li> <li>Phaseorigin Refinement HK Max</li> <li>20</li> <li>Yes</li> <li>No</li> </ul> 3D Merging       3D: zstarwin (Window in z* for consideration in phase origin search) <ul> <li>0.0025</li> <li>Merging Refinement</li> </ul> Refine tilt geometry (only in 3D mode)? <ul> <li>Yes</li> <li>No</li> </ul> Refine Beamtilt (only in 3D mode, only if better than 5 Angstroems)? <ul> <li>Yes</li> <li>No</li> </ul>	Parameter     Value       initialization_reset     y       initialization_reset     y
	2dx more materin 2dx more materin 2dx select APP om normalize TAN 2D ecise 2dx origili refine com initialize can calc	Output (Double click for logbrowser)       Verbosity Level: Low         ####################################	CSH: First (01) refinement script LOG: First (01) refinement postprocessing script CSH: First (01) refinement postprocessing script LOG: Last (04) refinement script LOG: Last (04) refinement postprocessing script CSH: Last (04) refinement postprocessing script """ CYStal),
		15	6-2dx-2016-Tilt-Geometry.key - 22 Aug 201

				N	MloK1-fi	lm   2dx (4.0.0	)								
语					1	1 🔁									
(3) Mioł	K1-ł	K2-2014-11-17_MloK1_cAMP/2014-11	-17_12-05-02 🛛 🖓 🛋 MioK100/MioK10	099001	200										
		Init Program	Generate Map												
-	*	Init Files and Parameters													
Ċ.	R	Calculate FFTs	Setup				Level:	Simplified							
	4	Get Defocus & Tilt	Keep Large Temporary Files ? 🔒	ZStar	range			0.083333							
	R	Get Lattice & Tilt													
	*	Refine Lattice	Symmetry												
	-	Calculate Crystal Tilt from Carb	Symmetry						-						
		CTF Correction (Part 1)	Symmetry		212	0									
	R	Get SpotList	Phase Origin	<b>a</b> -85			3.500								
	Ξ.		Phase Origin after Fourier Filtration	_			3.500								
		Unbend I	Phase Origin for ML result	0.0			0.0								
		Get SpotList (or refine)													
	10	Unbend II	Output (Double click for logbrowser)		0	Verbos	ity Level: Low	0	Images						
	*	Unbend MovieA1	The location of Anaconus Py			ted in Fieles	ences.		Unb.I		symmet	rized Map			
	R	Unbend MovieA2	==== Calculating zstarrange as 0	.08333	33 (wit	h 0.5 = Nyqu	ist resoluti	on). ======	PS: Unb.11 Non-symmetrized Map						
		Unboard Mayle P	==== Sourcing generateMap script	cript for Unbending reconstruction in pl symmetry =======							Projected Reference				
	8	Unbend MovieB	2dx lencalc: To calculate s ORIGTILT - in pl symmetry t	ome val	lues form t	or the tilt g	eometry dete	rmination. ==							
	×	CTF correction (Part 2)	==== Using APH file APH/image_ct ==== avrgamphs - to transform me												
		Generate Map 													
			==== laserplot - to create PS/mP	loK100	9900120	00MAP-pl.ps =									
	-	DEPENDENT SCRIPTS	Done.												
	4	2dx_makedirs	==== 2dx generateMAP - normal en	d. ====											
	92	2dx_generateMap_sub.com 2dx_checklattice_sub.com	Note: The following floating-poi	nerate	eptions	are signall	ing: IEEE DI	VIDE BY ZERO							
		initialize	Status									review			
	-		Calc. Mag=	22007.	-	103	201		QVAL=		460.4 OVAL	1000	CONTRACT OF		
			Unbend IQ Unb.I 3		102	103	104	105	1Q6 219		QVAL 415.3	10110	1000		
			Unb.II 3		83	65	113		228		460.4	COLUMN.	1		
			MovieA	-	-	-	-	-	-			4960.7	0.04667		
			MovieB	-	-	-	-	-	-			10000	100 100 100		
			PowerBins (noise=7)			[A]	15 818	12 9 77 5		5	3	34 0	2019		
			Tilt Info			Defoc.	Latt.		SpSplit	,	Merge	Sec. 20	Sales and the		
			Grid TAxis			89.7	86.3		89.7		87.8	10 Mar	1.000		
			Grid TAngle			-16.4	-23.3		-16.4		-28.0	COLUMN.	1 100 M		
			Xst. TAxis			25.9	28.1		25.9		25.8	4500 3	BALL OF		
			Xst. TAngle			16.4	23.3		16.4		28.0	and the second	100 B		







How does the CTF look like, if the sample is tilted?

This is called "Tilted Transfer Function" (TTF)

