

Electron Crystallography Workshop

at UC Davis, August 7-11, 2006

Scientific Organization:

Ben Hankamer (B.Hankamer@imb.uq.edu.au)
Henning Stahlberg (HStahlberg@ucdavis.edu)

Local Organization:

Rena Hill (RMHill@ucdavis.edu)

Web Site:

<http://2dx.org/workshop>

Emergency Phone Numbers:

Henning Stahlberg: +1 (530) 574 0835 (mobile)
Rena Hill: +1 (530) 400 0339 (mobile)

Lodging:

UCD Conference Housing, Tercero Area

Seminars:

1022 Life Sciences, University of California, Davis, CA 95616

Practicals:

WetLab: 15 Briggs Hall, University of California, CA 95616 (Phone: (530) 754 8285)
TEM: 29 Briggs Hall, University of California, CA 95616 (Phone: (530) 754 8338)
Computing: Science Lecture Building Computer Room

Sponsors:



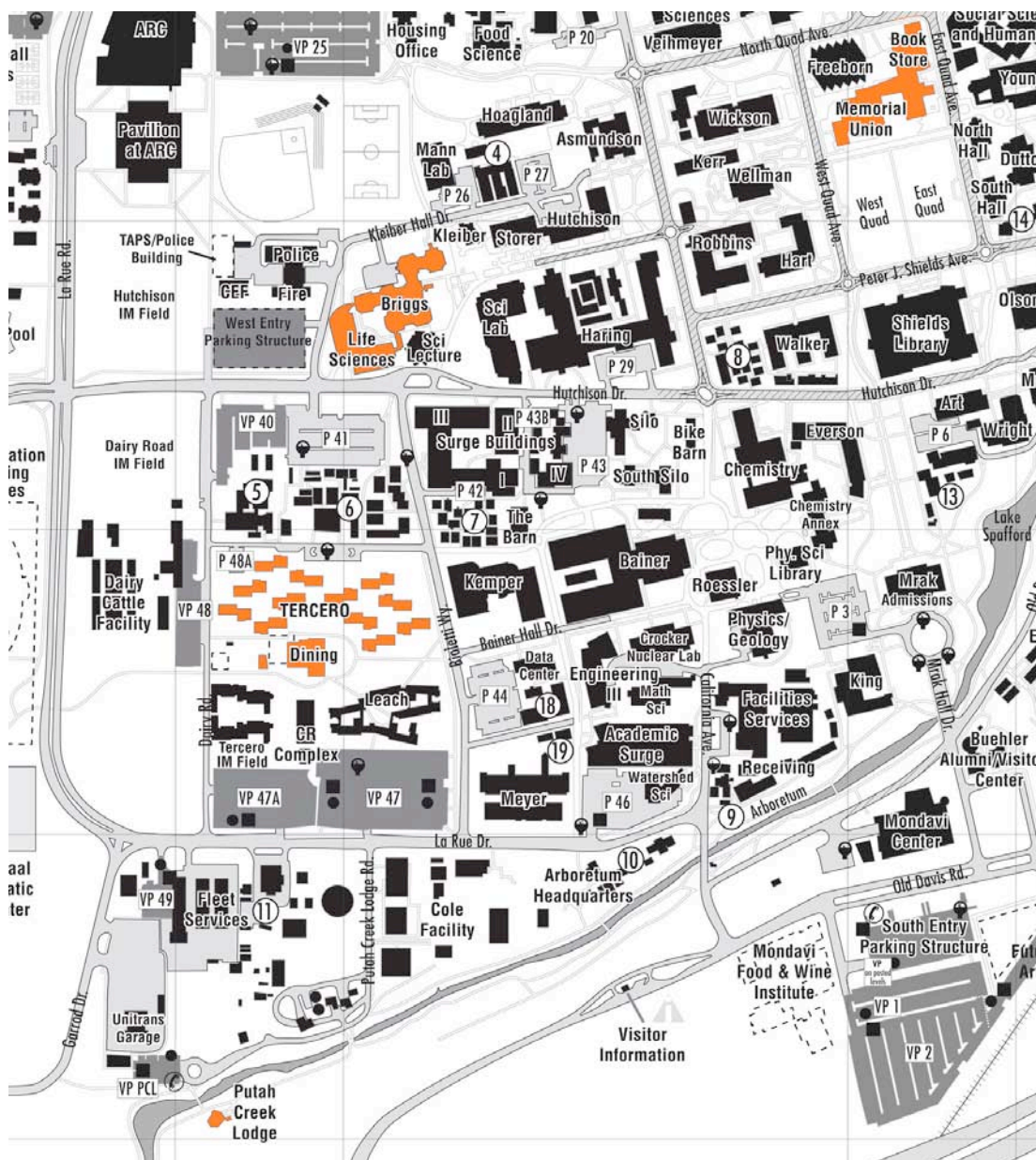
Program

Sunday, August 6, 2006

Afternoon Arrival
Arrival Desk: 1022 LSA
Pickup badge, walk to lodging area

Rena Hill
Ben Hankamer
Henning Stahlberg
Hui-Ting Chou
Po-Lin Chiu
Ludovic Renault

7:00 pm Dinner Buffet. Putah Creek Lodge (at the bottom of the map below)



Monday, August 7, 2006

8:00 am	Breakfast.		
9:00 am	Welcome	Ben Hankamer Rena Hill Henning Stahlberg	
9:30 am	Detergents and their properties <ul style="list-style-type: none">- General overview of 2D crystallization process: Key components = Detergents, lipids and proteins- Definition of a detergent - briefly describe amphipathic nature- Main detergent types: Ionic, non-ionic- Common classes of non-ionic detergents (malt, gluc. etc)- CMC, Micelle size, hydrophobic lipophobic balance.- Important properties of detergents for membrane protein stabilization.- Detergent exchange- Important properties of detergents for membrane protein crystallization- Interesting new detergents coming onto the market or under development- Specialized properties: temperature dependent phase separation and hydrophobicity- Statistics: Best detergents- Detergent suppliers - and purity (eg. alpha /beta story)- Synthetic amphipathic peptides	Ben Hankamer	
10:20 am	Coffee Break		
10:30 am	Lipids and their properties <ul style="list-style-type: none">- Definition of a lipid - briefly describe biophysical properties- How do lipids differ from detergents?- Main types of lipids- Important properties of lipids for 2D crystallization. (Saturation, liquid crystal:crystalline phase transition).- Statistics: Best lipids for crystallization.- Choosing lipids for crystallization	Daniel Levy	
12:00 pm	Lunch.		
1:00 pm	Detergents: Purifying, solubilizing and crystallizing membrane proteins <ul style="list-style-type: none">- Naturally occurring 2D arrays (bR, PSII etc)- Membrane protein expression systems - What's out there, what works, what are the problems.- Tags: What tags are out there, what are their properties and limitations?- Towards streamlining crystal production: Coupling protein purification and crystallization (just briefly as it will be covered later. Eg. Ni affinity chromatography and Ni monolayer lipids).- Organizing membrane proteins be packed into crystals: Type 1, type 2 3D crystals, cubic phase, 2D bilayer stacked 2D (e.g Gonen paper) which has links with the early stages of cubic phase crystallization.- Advantages of 2D crystallization- Template assisted monolayer crystallization- Non-fluorinated and Fluorinated lipid monolayers- Examples of the successes of each technique (Biobeads, dialysis, dilution, monolayer).- Incomplete factorial designs or 3 parameter searches.....and other methods of screening crystallization conditions.- Crystallization intermediates	Ben Hankamer	
2:00 pm	Practical: 2D crystallization <ul style="list-style-type: none">- Monolayer Trial (Daniel Levy, Amy Anderson)- Dialysis Trial (Tom Walz, Hui-Ting Chou, Po-Lin Chiu, Ludovic Renault)- Biobead Trial (Ben Hankamer, Rena Hill)	Practical: Image Processing <ul style="list-style-type: none">- Installation of MRC, 2dx, possibly IPLT on the private portable computers- Introduction to the image processing in small sub-groups	Ben Hankamer Daniel Levy Stahlberg Lab Tom Walz
5:30 pm	Dinner, Tercero Dining Area.		
6:30 pm	Student Posters and Coffee, in 1022 LSA		
7:30 pm	2D crystallization: Examples and Interpretation. <ul style="list-style-type: none">- Crystal Quiz (Ben Hankamer)	Tom Walz	
8:00 pm	Round table discussion: Improved approaches to 2D crystal production	Tom Walz	
9:00 pm	Open hour.		

Tuesday, August 8, 2006

8:00 am	Breakfast.				
9:00 am	TEM: Instrumentation - Vacuum, Gun (FEG/Thermal, Acc. Voltage), Stage (LN2, Helium), Low-Dose, Spot Scanning, Detection (Film/CCD)	Ken Downing			
9:45 am	Sample Preparation - Methods for enriching crystals from a mixture of crystals and vesicles. - Harvesting crystals: preventing damage to crystals during transfer (monolayer, bilayers). - Benefits of glucose, trehalose etc. - Back injection, Carbon sandwich and other techniques	Nobuhiko Gyobo			
10:30 am	Coffee Break				
10:45 am	Data collection: Imaging - Alignment (Coherence, Beam tilt, stability) - Data collection (Defocus, Astigmatism) - Dose limitation and dose measurement	Dieter Typke			
11:20 pm	Data collection: Diffraction - Principle, Alignment, Operation mode (Exp. time, etc.), Film/CCD	Janet Vonck			
12:00 pm	Lunch.				
1:00 pm	TEM development at JEOL	Barbara Armbruster			
1:30 pm	CCD camera development at TVIPS	Hans Tietz			
2:00 pm	<table border="0" style="width: 100%;"> <tr> <td style="vertical-align: top;"> Practical: Sample Preparation / CryoEM data collection - RT & Cryo: Neg. stain (Hui-Ting Chou) - Cryo: Back injection (Nobuhiko Gyobo) - Cryo: Sandwich (Nobuhiko Gyobo) - Cryo-EM imaging (Dieter Typke, Nobuhiko Gyobo, James Evans) - e-Diffraction (Janet Vonck, Hui-Ting Chou) </td> <td style="vertical-align: top;"> Practical: Image Processing - Installation of MRC, 2dx, possibly IPLT on the private portable computers - Introduction to the image processing in small sub-groups </td> <td style="vertical-align: top;"> Janet Vonck Nobuhiko Gyobo Dieter Typke Bryant Gipson Ansgar Philippsen Andreas Schenk </td> </tr> </table>	Practical: Sample Preparation / CryoEM data collection - RT & Cryo: Neg. stain (Hui-Ting Chou) - Cryo: Back injection (Nobuhiko Gyobo) - Cryo: Sandwich (Nobuhiko Gyobo) - Cryo-EM imaging (Dieter Typke, Nobuhiko Gyobo, James Evans) - e-Diffraction (Janet Vonck, Hui-Ting Chou)	Practical: Image Processing - Installation of MRC, 2dx, possibly IPLT on the private portable computers - Introduction to the image processing in small sub-groups	Janet Vonck Nobuhiko Gyobo Dieter Typke Bryant Gipson Ansgar Philippsen Andreas Schenk	
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5:30 pm	Dinner, Tercero Dining Area				
6:30 pm	Student Posters and Coffee, in 1022 LSA				
7:30 pm	Deceleration CCD	Ken Downing			
8:00 pm	Phase Plate Technology	Bob Glaeser			
8:30 pm	Experiences with a Cs corrected and energy filtered TEM	James Evans			
9:00 pm	Dynamic Transmission Electron Microscopy (DTEM)	Nigel Browning			
9:30 pm	Open Hour				

Wednesday, August 9, 2006

8:00 am	Breakfast.		
9:00 am	Fourier Theory - Real space / Fourier space - FFT, Correlation, Convolution, Autocorrelation - Lattice lines		Bob Glaeser
9:30 am	Symmetry - Plane groups, definitions, appearances in 2D/3D		Michael Landsberg
10:00 am	MTF - Determination of the MTF of CCD - Scanner vs. Film		Dieter Typke
10:30 am	Coffee Break		
10:45 am	Overview of algorithms for 2D crystal image processing - Fourier-space solutions (filtering, unbending) - Real-space solutions		Henning Stahlberg
11:15 am	Image Processing in 2D: MRC software for real-space images - Format - Documentation - Work-flow - Programs		Anchi Cheng
12:00 pm	Lunch.		
1:00 pm	Practical: Sample Preparation / CryoEM data collection - RT & Cryo: Neg. stain (Hui-Ting Chou) - Cryo: Back injection (Nobuhiko Gyobo) - Cryo: Sandwich (Nobuhiko Gyobo) - Cryo-EM imaging (Dieter Typke, Nobuhiko Gyobo, James Evans) - e-Diffraction (Janet Vonck, Hui-Ting Chou)	Practical: Image Processing - Introduction to the image processing in small sub-groups	Janet Vonck Nobuhiko Gyobo Dieter Typke Bryant Gipson Ansgar Philippsen Andreas Schenk
4:30 pm	Beam-induced movement of samples - SpotScanning - Beam-induced drum-head movement vs. charging		Bob Glaeser
5:30 pm	Dinner, Tercero Dining Area		
6:45 pm	Scanning Transmission Electron Microscopy - Comparison of STEM vs. TEM - How is STEM implemented - Aberration corrected STEM - What are the applications and implications to biological samples - High-resolution results.		Nigel Browning
7:30 pm	High resolution membrane protein structure		Tom Walz
8:15 pm	Tubulin		Ken Downing
9:00 pm	Open hour		

Thursday, August 10, 2006

8:00 am	Breakfast.		
9:00 am	Tilt geometry		Henning Stahlberg
9:30 am	2dx: Image processing of 2D crystal images using 2dx_image		Bryant Gipson
10:15 am	Coffee Break		
10:30 am	Image Processing: MRC software for e-diffraction evaluation - Algorithm - Programs		Janet Vonck
11:15 am	IPLT architecture		Ansgar Philippsen
12:00 pm	Lunch.		
1:00 pm	IPLT: Image processing of e-diff pattern		Ansgar Philippsen
1:45 pm	Practical: Image processing of non-tilted 2D crystal data - MRC: Processing of 2D crystal images (Janet Vonck, Anchi Cheng) - 2dx: Processing of non-tilted and tilted images (Bryant Gipson, Henning Stahlberg) - IPLT: Processing of e-diff pattern (Ansgar Philippsen, Andreas Schenk)	Practical: Image processing trouble shooting session - Targeted problem sessions in small groups with requested teachers	Bryant Gipson Ansgar Philippsen Andreas Schenk Henning Stahlberg Janet Vonck
5:30 pm	Dinner, Tercero Dining Area		
6:45 pm	Resolution: Definition, Determination		Anchi Cheng
7:30 pm	A Maximum Likelihood approach to 2D crystals		Xiangyan Zeng
8:15 pm	Advantages of higher (icosahedral) symmetries		Holland Cheng
9:00 pm	Open hour		

Friday, August 11, 2006

8:00 am	Breakfast.	
9:00 am	Tilted Transfer Function (TTF): SpotSplitting and Fourier spot deconvolution	Henning Stahlberg
9:40 am	A new method for Tilted Transfer Function correction	Ansgar Philippsen
10:15 am	Coffee Break	
10:30 am	Merging with the MRC software (2D) - Phase origin, Defocus, Beam-tilt - ORIGINILT - MAKETRAN	Henning Stahlberg
11:00 am	Merging with the MRC software (3D) - Tilt geometry refinement - ORIGINILT - Lattice lines	Anchi Cheng
11:30 am	3D reconstruction - Lattice line interpolation, 3D volume generation, NCS - 3D Visualization with O / Dino	Andreas Schenk
12:00 pm	Lunch.	
1:15 pm	Calculation of the variance of a 3D map by the bootstrap method	Anchi Cheng
2:00 pm	Practical: Image processing of tilted 2D crystal data - MRC: Processing of 2D crystal images (Janet Vonck, Anchi Cheng) - 2dx: Processing of non-tilted and tilted images (Bryant Gipson, Henning Stahlberg) - IPLT: Processing of e-diff pattern (Ansgar Philippsen, Andreas Schenk)	Practical: Image processing trouble shooting session - Targeted problem sessions in small groups with requested teachers Bryant Gipson Ansgar Philippsen Andreas Schenk Henning Stahlberg Janet Vonck
5:00 pm	Data Deposition: PDB, EBI	Ben Hankamer
5:30 pm	Closing Remarks	Ben Hankamer Henning Stahlberg
6:00 pm	Free Time	
7:00 pm	Banquet	

Saturday, August 12, 2006

8:00 am	Breakfast.
9:00 am	Departure

Instructors

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