Our History

The WMEN conference has been held for the past 17 years during the month of May in Los Cabos, Mexico. The meetings originated from a grant from the Rockefeller Foundation supporting research collaborations between scientists at UCSF and Scripps Research Institute, now called TSRI. Drs. Daniel Santi and Ian Wilson started the meetings and created the unique scientific ambience. The meeting style has remained unchanged, but nine years ago the venue moved from Cabo San Lucas to the all-inclusive El Presidente Hotel in San Jose del Cabo. The meeting includes approximately 70 selected participants, 40 of which are laboratory heads. The spirit of scientific research is enhanced and refreshed in this stunning setting.

FINAL SCHEDULE
World Molecular Engineering Network Seventeenth Annual Meeting on Structural Biology
April 30-May 4, 2006

Sunday Evening, 30 April **San Jose Conference Room**

17:30 – 17:45 Dan Santi and Ian Wilson **Introduction and Welcome**

17:45 – 20:00 ***Self-Introductions by Non-Presenting Sponsors and Others***

Darcy, Maureen	Fluidigm
Barbas, Carlos	TSRI

Short Presentations (5 min.) by TSRI and UCSF Graduate Students, Postdoctoral Fellows, and Others

Apriletti, James	UCSF	Structural Determinants for Selective Thyroid Hormone Receptor Antagonists
Chandonia, John-Marc	UCBerkeley	The Impact of Structural Genomics
Chen, Yu	UCSF	Sub-Angstrom Resolution Protein Crystal Structure: WYSIWYG
Deu Sandoval, Edgar	UCBerkeley	Engineering of an obligate homodimeric enzyme into a heterodimeric hybrid with dual specificity
Egea, Pascal	UCSF	Targeting Proteins to and across Membranes
Friedland, Gregory	UCSF	Using computational models to interpret NMR experiments of protein dynamics
Ghosh, Kingshuk	UCSF	Small Numbers Fluctuations in Biology
Howard, Rebecca	UCSF	A C-Terminal Coiled-Coil Domain Drives Assembly and Specificity in KCNQ Channels
Ivanetich, Kathryn	UCSF	Application of DNA sequencing & quantitative PCR microbial source tracking assays to identify host sources of fecal pollution in watersheds

	Lang, Paula Theresa	UCSF	Computational Method Development for Targeting RNA: Applications to HIV TAR
	Scheer, Justin	UCSF	A conserved allosteric site and mechanism in caspases identified by disulfide-trapping
	Topf, Maya	UCSF	Combining Comparative Modeling and CryoEM Density Fitting
	Barglow, Katy	TSRI	Discovering disease associated enzymes by proteome reactivity profiling
	Chappie, Joshua	TSRI	Towards the structural understanding of dynamin GTPase
	Debler, Erik	TSRI	Crystal structure of the IL-2 signaling complex
	Evans, Michael	TSRI	Preparation and reactivity of a natural product-like library of activity-based probes
	Grbic, Jovana	TSRI	Investigating the regulatory role of BMI-1 in hematopoietic stem cell regulation and cancer
	Hills, Ron	TSRI	Cooperative self-association in amyloidogenic peptides
	Kerkow, Don	TSRI	Characterization of a potentially novel RNA binding motif found within the *C. elegans* protein NSF-2
	Lander, Gabriel	TSRI	CryoEM reconstruction reveals key insights into viral DNA packaging machinery
	Manuell, Andrea	TSRI	Structure of the chloroplast ribosome and implications for light-regulated translation
	Ward, Andrew	TSRI	Structural and functional analysis of the ABC transporter MsbA
	Yoshioka, Craig	TSRI	Automating random conical data collection
20:15 – 21:00	**Reception**		**In the Main Garden**

FINAL SCHEDULE
World Molecular Engineering Network Seventeenth Annual Meeting on Structural Biology

April 30-May 4, 2006, San Jose del Cabo, Baja, Mexico

Monday Morning, 1 May

			Advances in Structural Biology & Proteomics (Chair: Brian Shoichet)
09:00	Brenner, Steven	UCBerkeley	Phylogenomics and Metagenomics
09:20	Sjolander, Kimmen	UCBerkeley	New algorithms for phylogenomic inference of protein function
09:40	Sali, Andrej	UCSF	Modeling the structures of proteins and macromolecular assemblies
10:00	Yeates, Todd	UCLA	Do thermophiles stabilize their protein complexes by entangling them in links and knots?
10:20	**Break**		
10:50	Nollert, Peter	deCODE	Going the extra mile: Rational surface mutagenesis
11:10	Williamson, Jamie	TSRI	Postproteomic approach to ribosome assembly
11:30	Byram, Susan	Bruker AXS	Latest equipment and crystallographic results from Bruker
11:50	Ruth, Ronald	Lyncean Tech	Recent tests of the compact light source

Monday Afternoon

			Protein Engineering and Drug Discovery (Chair: Jack Kirsch)
16:30	Wells, James	UCSF	Site-directed ligand discovery
16:50	Shoichet, Brian	UCSF	Relating proteins by their ligands
17:10	Kortemme, Tanja	UCSF	Reengineering specificity in signaling domains
17:30	Ruben Abgayan	TSRI	Advances in virtual lead discovery and structure annotation
17:50	**Break**		
			Immune System and Membrane Proteins (Chair: Todd Yeates)
18:05	Paulson, James	TSRI	Sweet spots in B cell signaling
18:25	Stroud, Robert	UCSF	Membranes 4 billion years What's new‘
18:45	Minor, Dan	UCSF	Ion channels in pieces: Structural dissection of channel action and regulation
19:05	Stout, Dave	TSRI	Experiments toward crystallization of transhydrogenase

FINAL SCHEDULE
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Tuesday Morning, 2 May

09:00	Santi, Dan	UCSF	Synthetic biology approaches to polyketide bio-synthesis
09:20	Vieth, Michel	Eli Lilly	Advancing drug discovery with computational chemistry - from docking to chemogenomics
09:40	Muskal, Steven	Eidogen-Sertanty	Broader visibility after proliferating the structural proteome – Are promiscuous drugs inevitable?
10:00	McRee, Duncan	ActiveSight	Fishing for fragments with the ACTOR Robot
10:20	**Break**		
10:50	Syed, Rashid	Amgen	P38 MAP kinase inhibitors: Discovery and selection of drug candidates
11:10	He, Molly-Min	Sunesis	A novel mechanism for small molecule inhibition of TNF-alpha
11:30	May, Andy	Fluidigm	Aspects of crystallization using free interface diffusion

Tuesday Afternoon

SPONSORS
(Chair: James Wells)

16:30	Gottesfeld, Joel	TSRI	DNA and chromatin therapeutics for triplet repeat diseases
16:50	Millar, David	TSRI	Nucleic acid-protein complexes as therapeutic drug targets
17:10	James, Thomas	UCSF	Interactions of Poly(C) binding protein with telomeric DNA and telomerase RNA
17:30	Baxter, John	UCSF	Selective modulation of the thyroid hormone receptor
17:50	**Break**		
18:05	Yang, Xiang-Lei	TSRI	Two conformations of a crystalline human tRNA synthetase tRNA complex: Implications for protein synthesis
18:25	Johnson, Jack	TSRI	An asymmetric CryoEM reconstruction of infectious P22 virus shows that a component of the DNA packaging motor is also a sensor for chromosome condensation density
18:45	Yeager, Mark	TSRI	Native hepatitis B virions and capsids visualized by electron cryomicroscopy
19:05	Finn, M.G.	TSRI	Viruses and click chemistry: polyvalent platforms and connectivity tools of use in structural and functional biology

Nucleic Acids & Viruses
(Chair: Jamie Williamson)

FINAL SCHEDULE
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April 30-May 4, 2006, San Jose del Cabo, Baja, Mexico

Wednesday Morning, 3 May

Assemblies and Computation (Chair: Andrej Sali)

08:30	Dill, Kenneth	UCSF	Using folding physics for protein structure prediction
08:50	Kirsch, Jack	UCBerkeley	Protein/protein interactions in the gas phase
09:10	Sanner, Michel	TSRI	Computational representation of protein flexibility: Applications to automated docking
09:30	Finer-Moore, Janet	UCSF	The structure of the ER-luminal domain of Ire1, a sensor for unfolded protein
09:50			
	Break		
	Deniz, Ashok	TSRI	Shedding light on protein aggregation using fluorescence
10:20	Milligan, Ronald	TSRI	Structural studies on microtubule binding proteins
10:40	Balch, William	TSRI	Structural and functional design of cargo nanocages
11:00	Olson, Arthur	TSRI	Computational biology from atoms to cells
11:20			

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The following pages are summaries of presentations and comments on the meting and venue.

WMEN Conference
San Jose del Cabo
El Presidente Hotel

For more information, contact:

Daniel Santi
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Ian Wilson
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Karin Asensio
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Investigator: Ruben Abagyan
Dept./Institution: Scripps-Laboratory of Computational Biology and Bioinformatics, TSRI
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Overview: My research focuses on problems in computational biology and chemistry. We have developed methods for predicting the functional map of a protein with a known 3D structure, accurate docking of compounds to a binding site and virtual ligand screening of large chemical databases, and, structure prediction by global energy optimization, e.g. characterizing mutants and SNPs, homology modeling, protein-protein or peptide docking, and accurate loop prediction.

Presentation: Two stories in which novel compounds were discovered through "ligand-guided" receptor pocket modeling followed by virtual screening of large compound libraries, were presented. First, we developed models of the androgen receptor in an antagonist-bound conformation. These models were used to discover computationally the secondary activity of antipsychotic drugs. These drugs were then chemically altered and "re-purposed" to loose their binding to the serotonin and dopamin receptors, and improve their anti-androgen properties. The experimental side of this project was performed by the labs of Xiaokun Zhang and James Dalton. Second, in collaboration with the David Lomas lab at Cambridge, we identified the first small molecules to inhibit pathological polymerization of an alpha1-antitrypsin mutant which is the most common genetic cause of a lethal liver disease in childhood. Computationally this project was particularly difficult because the target of a small molecule was a dynamic protein-protein interface.

Impressions of the meeting: All the aspects of the meeting were just right for me. In particular, I liked the high pace of

the meeting, the 5 minute post-doc slots, and strict enforcement of the timing including short discussions.

Name: Jim Apriletti
Supervisor: John Baxter
Dept./Institution: Diabetes Center / Metabolic Research Unit, UCSF
Topic: Structural Determinants for Selective Thyroid Hormone Receptor Antagonists
Mailing Address: UCSF, Box 0540, San Francisco, CA 94143-0540
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Phone: (415) 476-9292 **FAX:** (415) 564-5813

Summary of Research: My research focuses on understanding the molecular mechanisms of action of the nuclear thyroid hormone receptor, primarily through the use of X-ray crystallography and hormone binding studies. We have developed several thyroid hormone receptor-beta selective agonists and antagonists, and have been examining the basis of their selectivity. We found that changing thyroid hormone's amino group to an oxoacetate group was responsible for the beta selectivity in the analog GC-1. Para-substitution with strong electron withdrawing groups on a 5'-phenylethynyl GC-1 derivative gave several thyroid hormone antagonists. The antagonists promote corepressor release but don't recruit coactivators, preventing transactivation and relieving transrepression. Antagonist potency increased as the electron withdrawing character of the 5'-para-substituted aryl extension increased.

Impressions of the Meeting: The meeting was excellent. I enjoyed the beautiful location, and it was a great opportunity to meet many scientists I would not normally interact with. The research presentations were interesting and thought provoking. The size and length of the meeting was about right, and I can't think of any changes that I would make in the format of the meeting.

Name: Katy Barglow
Supervisor: Benjamin F. Cravatt
Dept./Institution: Department of Chemistry and Cell Biology, TSRI
Topic: Profiling the proteome with structurally diverse libraries of activity-based probes
Mailing Address: 10550 N. Torrey Pines Road, BCC 159, The Scripps Research Institute, La Jolla, CA 92037
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Summary of Research: My work focuses on using structurally diverse small molecule libraries to probe proteomes towards two main goals. First, we look for enzymes activities altered in disease states. Second, we use the reactivity patterns of these small molecules to gain insight into endogenous biochemical functions of enzymes. One enzyme from mouse liver, ureidopropionase, showed a very restricted probe reactivity profile. Labeling was sensitive to both the functionalities and the chirality of the probe binding group. All probes that labeled the enzyme were close mimics of the known natural substrate, N-carbamoyl-beta-alanine. All these probes had a negative charge beta to the site of reactivity, indicating that the enzyme selectively accepts substrates with this functionality.

Impressions of the Meeting: This meeting was great! The small size allowed for a lot of interaction between participants. I thought the structure of short postdoc and graduate student talks on the first night, followed by longer PI presentations and sponsor presentations on the following days worked very well. The meetings content, location, and participants were all fantastic.

Investigator: Steven E. Brenner
Dept./Institution: Department of Plant & Microbial Biology, University of California, Berkeley
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Overview: My research is in the area of computational genomics using evolutionary principles. There are several related projects underway in my group. We have developed and are enhancing an automated statistical method for reliably predicting protein function from sequence (SIFTER project). We provide resources based on automated analysis of protein structures associated with the manually-curated SCOP database (ASTRAL project), and we are developing methods for automatically recognizing ancient and subtle protein homology from structure—even when too distant to be seen in sequence (CAPER project). Similarly, we have created a comprehensive classification of RNA structures and are interpreting the evolutionary and functional roles of RNA motifs (SCOR project). We are initiating studies of protein complexes characterized with electron microscopy and tomography (PCAP project).
We discovered that a large number of natural human mRNAs are targets of the RNA surveillance system, nonsense-mediated mRNA decay (RUST project). We are currently demonstrating the significance of this coupling as a critical and well-conserved mode of gene regulation. *Drosophila* uses a novel mode of detecting premature stop codons in fly, and we have profiled its alternative mRNA isoforms to understand how these systems operate. We are also exploring the *cis* sites that are responsible for triggering alternative splicing.
We recently analyzed domain demographics as part of the Global Ocean Survey metagenomics project, and we are initiating medical metagenomics projects with an initial focus on profiling the intestinal microbes associated with Crohn's disease.

Presentation: Current BLAST-based methods of annotating genomes are error-prone, and they have polluting databases for a decade. Protein functions can be reliably predicted by intuitive phylogenetic analysis, but this process can be exquisitely labor intensive. We have developed an

automated method for protein function prediction using a phylogenomic approach. We have formalized protein function using the GO hierarchy and taken explicit account of reliability of proteins' functions using GOA database evidence codes. At its core, our method is based on a simple model of protein function evolution, parameterized from existing data. When applied to a reconciled phylogenetic tree, this method provides posterior probabilities of the functions for each protein in a family. On our test set, our current early prototype of SIFTER accurately predicts more proteins' functions than any other method.

Impressions of the meeting:

Location: Excellent

Number of participants: Excellent

Length of meeting: Just right

The meeting location was excellent. Cabo is an easy flight from San Francisco, but offers a remoteness and seclusion conducive to an interactive conference environment. The all-inclusive resort encouraged participants to stay together and mingle in overlapping groups rather than dissipate in predefined cliques. Especially since many attendees brought family, having the all-inclusive resort as a "home" helped facilitate scientific interactions throughout the meeting (rather than having each family go its own way).

I thought the size of the meeting was great. I knew of nearly all of the participants in advance of the meeting, but had only seriously interacted with a handful. The quality of the presentations was almost uniformly excellent, and so it was extremely educational. Discussions at the meeting have helped nucleate one new effort in my lab and provided key direction to another.

The length of the meeting was great. The free afternoons encouraged interactions for most participants, and they also allowed those with ineluctable pressing deadlines to complete critical tasks without having to miss conference sessions. Were the return flights deliberately scheduled for a full day after the meeting ended? I probably would not have recommended this, but it did actually seem useful to ensure that attendees went to all of the sessions knowing that they

had designated time to explore and interact more freely at the end of the meeting.

Investigator: Sue Byram
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Overview: The Bruker is a $700 million per year scientific instrument company which develops and manufactures research equipment, using methods in X-ray, NMR, MS, FT-IR and FT-Raman.

Presentation: New products developed for protein crystallography since the last Cabo meeting were described. The well-accepted 400 plate Crystal Farm is now complemented by a benchtop version and a large 5,000 plate system to incubate and image protein crystal growth plates automatically. A prototype flexible robotic system for harvesting crystals directly from the growth plates has been developed by Square One System Design and Bernhard Rupp of UC Irvine, with Bruker as the commercial partner. A new line of detectors for x-ray screening and full data collection has been developed using the novel 486 scientific grade 4K CCD. This new chip offers lowest noise, fastest readout and much higher dynamic range. The APEXII detector has no taper, offering maximum gain for fast screening in a compact, low maintenance X8 PROSPECTOR system. The bigger Platinum 135 CCD is the premium choice coupled to our high brightness rotating anode in the X8 PROTEUM system, for superb quality full data sets in addition to fast screening. Three atomic resolution data sets in the home lab demonstrate its capability. The biggest Platinum 200 CCD employs the biggest possible fiberoptic taper but at 17 electrons gain is still three to four times higher gain than any other beamline CCD.

A new technology AXIOM200 microgap detector was presented, offering ultimate quantum efficiency (QE) of 80%, compared to ~50% for the best CCD detectors and ~20% for typical image plates. This unbeatable QE coupled with zero noise makes it a new choice for x-ray screening of looped and frozen crystals, as well as novel applications for screening crystals in situ in plates or capillaries.

Impressions of the meeting:
Location: Excellent
Number of participants: Good size
Length of meeting: Fine
This is the fourth Cabo meeting I have attended, and I am still impressed with the content and the participants. It is a unique blend where all the participants from grad students to post docs to PI's in academia and industry present. The interaction is excellent, and this meeting was the largest and most energetic to date. I really appreciated the opportunity.

Name: John-Marc Chandonia
Supervisor: Steven E. Brenner
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Summary of Research: My research focus is the study of protein structure, function, and evolution from a computational biology and bioinformatics perspective. I am a member of several large-scale structural genomics (Berkeley SG Center, http://strgen.org) and proteomics (Protein Complex Analysis Project, http://pcap.lbl.gov) projects, and also work on databases that provide a foundation for studies of protein structure and evolution (SCOP, http://scop.berkeley.edu, and ASTRAL, http://astral.berkeley.edu). At the meeting, I presentedcurrent results of a study of the impact of

Structural Genomics projects. We quantitatively reviewed the novelty, cost, and scientific impact of structures solved by structural genomics centers, and contrast these results with traditional structural biology.

Impressions of the Meeting:
Location: Excellent
Number of Participants: could invite a few more UCB/LBNL people
Length: suggest extending talks through Wednesday evening.
The effort that participants put into crafting talks specifically for this audience was evident, resulting in excellent scientific quality. I thought it was at least four times better than a traditional structural biology meeting!

Name: Yu Chen
Supervisor: Brian K. Shoichet
Dept./Institution: Department of Pharmaceutical Chemistry, UCSF
Topic: Sub-Ångstrom resolution protein crystal structure: WYSIWYG
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Summary of Research: I am interested in using ultrahigh resolution X-ray crystallography in studying enzyme catalysis and molecular docking. The structure of CTX-M-9 β-lactamase has been determined to 0.88 Å, revealing many hydrogen atom positions. In the active site, the catalytic serine, Ser70, accepts a proton from Lys73 and donates its own proton to the catalytic water, which in turn acts as a donor in hydrogen bonding with Glu166. This suggests that Glu166 is the general base to deprotonate Ser70 in the first step of the reaction that hydrolyzes β-lactam antibiotics such as penicillin.

Impressions of the Meeting: A great meeting! Both the short presentations by the students/postdocs and the longer ones by the PIs were very insightful and inspirational. The sponsor's talks were very informative as well. The casual environment encouraged interactions among the scientists. I would recommend no changes in the meeting.

Name: Erik Debler
Supervisor: Ian A. Wilson
Dept./Institution: Department of Molecular Biology, TSRI
Topic: Crystal structure of the IL-2 signaling complex
Mailing Address: 10550 North Torrey Pines Road, BCC206, La Jolla, CA 92037
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Summary of Research: My research focuses on the understanding of the relationship between the structure and function of the interleukin-2 receptor. The crystal structure of this heterotrimeric cytokine receptor in complex with its ligand interleukin-2 has provided important insights into receptor assembly, interleukin-2 signaling, and receptor disassembly, as well as into the structural basis loss-of-function mutations found in patients with X-linked severe combined immunodeficiency diseases (X-SCID).

Impressions of the Meeting: This meeting was great! I am very pleased to have had the opportunity to listen to and talk with many outstanding scientists. The relatively small size of the meeting offers many opportunities to talk about projects and science in detail, as well as to get to know colleagues better and to meet new people in a unique environment. The sponsors' presentations were interesting and relevant to my research.

Investigator: Ashok Deniz
Dept./Institution: Molecular Biology Department, The Scripps Research Institute.
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Overview: My research primarily focuses on developing and utilizing high-sensitivity single molecule and ensemble fluorescence methods to study a range of biophysical problems. In particular, single molecule methods provide a means to access detailed mechanistic information in biology, which is lost to averaging in standard ensemble experiments. We utilize these state-of-the-art methods along with a highly interdisciplinary approach to gain detailed mechanistic insights into several important areas including protein/RNA folding, protein aggregation, assembly of molecular machines, and RNAi mechanisms.

Presentation: My presentation mainly focused on high-resolution fluorescence approaches to structural studies of proteins that are involved in aggregation and amyloid formation, in particular for the yeast prion protein, Sup35, which has been found to play a role as a protein based genetic element in yeast. Key aspects discussed were using single molecule FRET to study protein conformations, as well as using a fluorescence quenching technique to study intermolecular distances in soluble aggregates and fibrils. Initial data were presented, and more detailed studies of the coupling between conformational changes and the protein aggregation process will be discussed at the next Cabo meeting.

Impressions of the meeting:
Location: Excellent
Number of participants: Optimal
Length of meeting: Optimal
The number of participants in this year's meeting was optimal. The small number and excellent setting provided an outstanding opportunity for close interactions and discussion between participants. The quality of the presentations was also uniformly very high, and a wide range of interesting topics were discussed. The combination of small size, high quality, and possibilities for close and extended discussions

at the meeting provides an excellent opportunity for encouraging novel ideas and collaborations.

Investigator: Ken A. Dill
Dept./Institution: Department of Pharmaceutical Chemistry, University of California, San Francisco
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Overview: My research is in three main areas, at the interface between statistical physics and biology. First, we are interested in protein folding, physical principles and structure prediction. Second, we are interested in developing a better understanding of water and solvation models, principally through better models of the statistical physics, but ultimately for better practical algorithms for computational biology. Third, we are interested in foundations of dynamics, and fluctuations in the small-numbers limit.

Presentation: We have combined all-atom forcefields, with implicit solvation models, and replica-exchange molecular dynamics sampling, with a new "mechanism-based conformational search method we call Zipping & Assembly, to do protein structure prediction. So far, the method appears promising for the 9 small single-domain soluble proteins we have tested to date.

Impressions:
Location: Outstanding
Number of participants: Perfect
Length of meeting: No complaints
I agree with Jack Kirsch: this is one of the best meetings of the year. Very high caliber talks, from academic PI's, from the grad students and postdocs, and from the people from industry: all uniformly high. My only real complaint is that the travel agency was difficult to deal with: I needed to change flights, and found they were inaccessible and unresponsive,

and a normally trivial change became a half-day's hassle. Otherwise, superb meeting!

Name: Pascal EGEA
Supervisor: Robert STROUD
Dept./Institution: Department of Biochemistry and Biophysics, UCSF
Topic: Targeting Proteins to Membranes
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Email Address: pascal@msg.ucsf.edu
Phone: (415) 476-3937 **FAX:** (415) 476-1902

Summary of Research:
I am interested in the mechanism responsible of proper protein targeting to biological membranes. The correct functioning of every living cell requires that secreted soluble proteins be translocated across, and membrane proteins be integrated into, a cellular membrane.. The translocation of most proteins across the endoplasmic reticulum or bacterial inner membrane occurs through an aqueous pore that spans the membrane, the translocon. In all three kingdoms of life, the co-translational targeting of secreted and membrane proteins is mediated by a ribonucleoprotein complex, the signal recognition particle (SRP), and its membrane-associated receptor (SR). The principal components of this pathway (SRP, SR and the translocon) are conserved throughout all evolution. Proteins translocated co-translationally across the membrane are directed to the translocation pore via an interaction between the cytosolic SRP and its membrane-bound receptor. I seek to study the structure-function relationships between the machineries involved in membrane protein co-translational targeting and translocation. My primary goals are to determine the three dimensional structures of essential macromolecular complexes involved in the co-translational targeting and the translocation of proteins to biological membranes.

Impressions of the Meeting: This meeting was very interesting! The short 5 minute presentations by graduate

and post doc gave me the opportunity to see the variety of subjects, systems and techniques used in the different research centers represented at the meeting. I am very pleased to have the opportunity to listen to and talk with many outstanding scientists. The PI's presentations were inspirational and the sponsor's presentations were very informative about the most recent technical developments and improvements in my field. I would recommend no changes in the meeting.

Name: Michael J. Evans
Advisor: Benjamin F. Cravatt III, PhD
Dept/Institution: Department of Chemistry, TSRI
Topic: Target discovery in small-molecule cell-based screens by in situ proteome reactivity profiling
Mailing Address: TSRI, Rm BCC159, 10550 N. Torrey Pines Road, La Jolla, CA 92037
Email: mjevans@scripps.edu
Phone: (858) 784-8201

Summary of Research: Chemical genomics aims to discover small molecules that impact biological processes through the perturbation of protein function[1,2]. However, determination of the protein targets of bioactive compounds remains a formidable challenge[3]. Here, we address this problem through the creation of natural products-inspired small molecule libraries bearing protein-reactive elements. Cell-based screening identified a compound MJE3 that inhibited breast cancer cell proliferation. *In situ* proteome reactivity profiling revealed that MJE3, but not other library members, covalently labeled the glycolytic enzyme phosphoglycerate mutase 1 (PGAM1), resulting in enzyme inhibition. Interestingly, MJE3 labeling and inhibition of PGAM1 were observed exclusively in intact cells. These results support the hypothesis that cancer cells depend on glycolysis for viability and promote PGAM1 as a potential therapeutic target. More generally, the incorporation of protein-reactive compounds into chemical genomic screens offers a powerful way to discover targets of bioactive small

molecules in living systems, thereby propelling downstream mechanistic investigations.

Impressions of the Meeting: In general, I thought the meeting was an enriching experience. The intimate and relaxing location, the scholarly content of the presentations, and the spectrum of topics discussed distinguished this conference from most that I've previously attended. Moreover, I profited greatly from the opportunity to interact closely with representatives from many of the several communities contributing to chemical and structural biology. I sincerely hope that future generations of young scientists will be afforded the chance to participate in a similarly formatted conference.

Investigator: Janet Finer-Moore
Dept./Institution: Department of Biochemistry and Biophysics, UCSF
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Overview: I have been collaborating with Robert Stroud in research that combines protein crystallography, site directed mutagenesis, and kinetics or other function assays to understand how proteins work. We have primarily focused on thymidylate synthase, which is a target for anticancer drugs. More recently, we have been working on two classes of RNA-modifying enzymes whose mechanisms are related in some respects to that of thymidylate synthase. We have also collaborated on determining structures of membrane proteins involved in signaling (Ire1, erythropoietin receptor/erythropoietin complex).

Presentation: Ire1 is an endoplasmic reticulum (ER) transmembrane sensor of unfolded proteins that triggers the unfolded protein response (UPR), a signaling pathway that adjusts the protein folding capacity of the ER. We have determined the crystal structure of the luminal domain of Ire1

to 3Å resolution. While we have found that Ire1 is a monomer in solution, the protein crystallizes with a dimer in the asymmetric unit. The dimer interface is extensive and creates a continuous b-sheet that spans the dimer. a-helices from both monomers pack against this b-sheet to form a shared central groove. The crystallographic interface between neighboring dimers is also extensive, Mutagenesis of residues at either interface seen in the crystal structure reduces the UPR, suggesting that Ire1 may form higher-order oligomers during signaling. Mutagenesis of residues in the central groove also reduce the UPR, suggesting a mechanism in which Ire1 initiates signaling by binding unfolded proteins directly to its central groove.

Impressions: The meeting was in a very pleasant location, and the format of the meeting (leaving afternoons free) made it especially enjoyable. The number of attendees and the length of the talks were just right. The quality of the science was very high. I enjoyed hearing talks from the sponsors.

Investigator: M.G. Finn
Dept./Institution: Department of Chemistry and The Skaggs Institute for Chemical Biology, TSRI
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Overview: Our group develops virus platforms for the polyvalent display of biologically active structures, and the directed evolution of enzymes. These efforts in molecular biology and protein design are complemented by the synthesis of organic compounds and the development of new catalysts and bioconjugation methods. In the past several years we have developed and applied “click chemistry” methods to the synthesis of active small molecules, protein conjugates, derivatized surfaces, and polymers. Of particular relevance to this conference are our fledging efforts to devise new heavy atom labeling reagents

and attachment strategies for use in cryo-electron microscopy.

Presentation: An overview of our capabilities and targets in the areas of viral platforms and chemical attachment methodologies was presented. We are now able to introduce unnatural amino acids with excellent positional control into two virus-like particles, one of which (bacteriophage Qb) maintains excellent stability toward the subsequent steps used to address these altered positions with excellent selectivity. The reaction most often used is the copper-catalyzed cycloaddition process between terminal alkynes and organic azides. These two functional groups exhibit no side reactions with protein or polynucleotide molecules, and so precise control of the spacing and orientation of attached units can be achieved.

Impressions of the meeting:
Location: Excellent
Number of participants: Excellent
Length of meeting: Just right
The meeting bears a happy resemblance to Gordon conferences in its length, degree of interaction among the participants, and format. Especially valuable was the strong representation from three top-quality institutions (Scripps, UCSF, UC Berkeley). Overall, a first-rate enterprise.

Name: Greg Friedland
Supervisor: Tanja Kortemme
Dept./Institution: Graduate group in Biophysics, UCSF
Topic: Understanding mechanisms of protein flexibility by modeling NMR dynamics experiments
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Summary of Research: My research focuses on understanding the mechanisms of protein flexibility. I'm approaching this by using computational modeling to

compare different plausible types of protein motion of varying amplitude and locality. The different models of motion are evaluated against NMR relaxation and residual dipolar coupling experiments which measure amplitudes of motion of numerous sites in a protein over timescales of nanoseconds and milliseconds, respectively. Some of the key questions are: Do core residues of some proteins undergo correlated rotameric transitions? How do motions accessible within picoseconds and nanoseconds differ? Are motions local on the energy landscape or significantly non-local? A future goal is to apply the resulting model of flexibility to understand the specificity of different PDZ domains for their peptide ligands.

Impressions of the Meeting: I'm very grateful for the opportunity to take part in this meeting. The small size and distant/exotic location contributed to create an intimate atmosphere in which to meet researchers from diverse fields and have stimulating discussions. I don't think I would change anything.

Name: KINGSHUK GHOSH
Supervisor: DILL. KEN
Dept./Institution: Department of Pharmaceutical Chemistry, UCSF
Topic: Small number fluctuations in Biology
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Summary of Research:
Biology deals with non-equilibrium problems in the small number limit unlike branches in
Physical sciences where we have large number of molecules. It is thus, important to
have dynamical laws which govern quantities beyond averages. Following the principle of maximum caliber, we developed a method to study fluctuations in non-equilibrium systems.

We test our theoretical predictions against the experiments done in the system of nano-diffusion. We find Fick's law to be valid in the small numbers limit, also we predict the fluctuations as observed in experiments. The distribution of the flux distribution is Gaussian consistent with the theory of maximum caliber. We are in the process of using this principle for other non-equilibrium systems of biological relevance.

Impressions of the Meeting:
This meeting was very useful and interactive. Presentations from leading scientists were encouraging and stimulating. I also enjoyed the presentations from PostDocs and students. Location of the meeting was great. I highly recommend the continuation of this meeting in the future.

I**nvestigator**: Joel M. Gottesfeld, PhD
Dept./Institution: Department of Molecular Biology, The Scripps Research Institute
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Overview: Research in our laboratory concerns the development of small molecules to regulate gene expression. Recently, small interfering RNAs (siRNAs) have received considerable attention as potential therapeutics for human disease; however, siRNAs elicit off-target effects and like all nucleic acids, these molecules suffer from poor cellular uptake and poor stability in live animals. In contrast, synthetic pyrrole-imidazole polyamides can be programmed by chemical synthesis to recognize a wide range of DNA sequences with affinities and specificities that are comparable to eukaryotic transcriptional regulatory proteins. In previous studies, we have shown that polyamides bind their cognate DNA sequences in the context of cellular chromatin, both in simple model systems and in the nucleus of cultured cells. We have used these molecules as either activators or repressors of gene expression.

Presentation: My presentation this year focused on the development of polyamides and other small molecules as therapeutics for the neurodegenerative disease Friedreich's ataxia (FRDA). FRDA is caused by hyper-expansion of GAA•TTC repeats in the first intron of a nuclear gene that encodes the mitochondrial protein frataxin. Expanded triplet repeats cause gene silencing either by altering DNA or chromatin structure. We find that polyamides targeting GAA•TTC repeats partially alleviate transcription repression of the frataxin gene in a cell line derived from white blood cells from a FRDA patient. These molecules also increase frataxin protein levels in these cells, and microarray studies show that a limited number of genes in the human genome are affected by polyamides targeting GAA•TTC repeat DNA.

We also examined the chromatin structure of the frataxin gene in cell lines derived from a FRDA patient and a normal sibling of this patient using antibodies to the various modification states of the core histones and chromatin immunoprecipitation methods. We find that gene silencing at expanded *frataxin* alleles is accompanied by hypoacetylation of histones H3 and H4, and methylation of histone H3 at lysine 9, consistent with a heterochromatin-mediated repression mechanism. We screened commercially available histone deacetylase inhibitors for their effects on *frataxin* transcription in the FRDA cell line, and identified one compound, BML-210, that partially reverses silencing in the FRDA cell line. Based on the structure of this compound, we synthesized and assayed a series of derivatives of BML-210 and identified histone deacetylase inhibitors that reverse *frataxin* silencing in primary lymphocytes from Friedreich's patients. We showed that these molecules act directly on the histones associated with the *frataxin* gene, increasing acetylation at particular lysine residues on histones H3 and H4. Unlike many triplet-repeat diseases (for example, the polyglutamine expansion diseases such as Huntington's disease and the spinocerebellar ataxias), expanded GAA·TTC triplets do not alter the coding potential of the *frataxin* gene; thus, gene activation would be of therapeutic

benefit. Animal studies are currently underway to explore the bioavailability and efficacy of these molecules.

Impressions of the Meeting:
Location: excellent
Number of participants: optimal for discussions
Length of meeting: exactly right
For a non-structural molecular biologist, with interests in structure-function relationships in DNA-binding proteins, this meeting is very valuable in learning the nuances of biophysical approaches to this field. As in previous years, I am continually impressed by the quality of structural data that comes from the institutions represented at this meeting. I think the mix of senior PI's, more junior PI's and postdocs and students is excellent. There are far too few meetings where such a mix is possible. I would favor longer presentations by some of the more advanced postdocs and graduate students. These could be in place of some of the talks given by the PI's who have spoken at previous meetings. Perhaps PI's should only speak every other year, and I would be willing to speak on such a schedule. I think the location and length/scheduling of the meeting is outstanding and I hope to be included in future years.

Small Molecule Regulation of Gene Expression

The ability to control gene expression at will has been a longstanding goal in molecular biology and human medicine. Recently, small interfering RNAs (siRNAs) have received considerable attention as potential therapeutics for human disease; however, siRNAs elicit off-target effects and like all nucleic acids, these molecules suffer from poor cellular uptake and poor stability in live animals. In contrast, our collaborators at the California Institute of Technology have developed a series of small molecules, called pyrrole-imidazole polyamides, that can be programmed by chemical synthesis to recognize a wide range of DNA sequences with affinities and specificities that are comparable to eukaryotic transcriptional regulatory proteins. In previous studies, we have shown that polyamides bind their cognate DNA

sequences in the context of cellular chromatin, both in simple model systems and in the nucleus of cultured cells. Polyamides can be used as either activators or repressors of gene expression, depending upon the chosen target site in particular genes. Below we summarize recent efforts toward the development of polyamides as therapeutics for human disease, and the identification of another class of small molecules that offer promise in the treatment of neurodegenerative diseases.

Investigator: Molly He
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Overview: Sunesis has developed a proprietary fragment-based drug discovery approach called Tethering®. We combine Tethering® with other drug discovery tools, such as structure-based design and medicinal chemistry, to discover and develop novel product candidates. Tethering® allows us to screen drug fragments based on binding properties rather than function, which allows us to potentially identify compounds that may not be discovered through conventional methods of drug discovery. We believe that this capability allows us to efficiently design drug candidates that bind to sites or regions on a specific protein not readily accessed by other discovery methods.

Presentation: We have been interested in discovery of small molecule antagonists of protein-protein interactions, in particular, tumor necrosis factor (TNFa) and TNFa receptor interaction that is involved in autoimmune diseases such as rheumatoid arthritis and Crohn's disease. Here, a novel mechanism of inhibition was presented where a small molecule inhibitor disrupted the TNFa trimer and disabled the TNFa molecules from binding to its receptor. Structural, biophysical, and biochemical assay results demonstrated

that the design of appropriate assays may allow for the identification of potent small-molecule inhibitors that inactivate multimeric proteins via a rapid predissociation-independent subunit dissociation process.

Impressions of the meeting:
Location: Excellent
Number of participants: Excellent
Length of meeting: Excellent
I really enjoyed the meeting. This is one of the best structural biology meetings that I have been to. The presentations were of very high quality. The scientists were outstanding. The atmosphere of the meeting was very friendly and scientifically stimulating. The meeting provides an excellent forum for discussion and fostering of new ideas and collaborations. I do not recommend any changes to the meeting.

Name: Ron Hills
Supervisor: Charles L. Brooks III
Dept./Institution: Department of Molecular Biology, TSRI
Topic: Cooperative self-association in amyloidogenic peptides
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Summary of Research: The kinetics of amyloid fibril formation are in most cases consistent with classical nucleation theory. Here molecular dynamics simulations were employed to measure the cooperativity in the early association events of the amyloidogenic peptide STVIYE(-). Sufficient cooperativity was found in the hydrophobic interactions gained upon association to allow for nucleation-dependent polymerization with a pentamer critical nucleus. Simulations performed with the non-amyloidogenic peptide STVIYE suggest an interesting role for electrostatics in determining amyloidogenicity. Novel considerations of the electrostatic solvation energy using the Born-Onsager

equation were put forth to rationalize the aggregation of charged peptides and provide new insight into the energetic differences between parallel and antiparallel beta-sheets.

Impressions of the Meeting: The meeting was very good. The number of attendees was just right. I particularly thought the balance between computation and experiment was excellent. The length of the meeting was good, and the amount of free time during the day was good. It would be nice to have the meeting in Cabo San Lucas instead, though.

Name: Rebecca J. Howard
Supervisor: Daniel L. Minor, Jr., Ph.D.
Dept./Institution: Chemistry and Chemical Biology Graduate Program, UCSF
Topic: Structural Insight Into KCNQ Channel Assembly and Channelopathy
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Summary of Research: My research focuses on the assembly properties of KCNQ voltage-gated potassium channels, which regulate cell excitability and have prominent roles in human diseases. The five KCNQ subtypes have distinct assembly preferences that are encoded by a C-terminal cytoplasmic assembly domain. By biochemical experiments and crystal structure determination, I have shown that the channel assembly specificity domain is a self-assembling parallel left-handed four-stranded coiled-coil that is conserved in all five KCNQ isotypes. The determinants of assembly specificity are encoded by a limited set of hydrophobic and electrostatic interactions that bridge the subunits. These interactions vary with assembly properties among channel subtypes. A cluster of disease mutants on the intersubunit surface indicates a likely binding site for proteins that organize intracellular signaling components. Together, my data show how a naturally occurring coiled-coil

provides a modular unit that can direct channel assembly and specificity.

Impressions of the Meeting:

(a) the optimal size of the group: I was inspired to see such a small and diverse group of scientists interacting with such familiarity. The meeting was large enough to incorporate scientists from well outside my usual community, but small enough for the uninitiated to gain a reasonable understanding of everyone's major contributions, and—moreover—to find connections between seemingly disparate research areas.

(b) location of the meeting: The location, of course, was beautiful, and I appreciated not having to worry about getting around or finding basic necessities in an unfamiliar country. The facility was a little extravagant, but very comfortable, and all the personnel were friendly.

(c) attendees and presenters: The meeting's intimate scale left me the unique impression that highly reputable professionals were, for the most part, genuinely attending to the contributions of students and postdocs, as well as more junior faculty. It was also instructional to see industry representatives presenting new research alongside academics. I appreciated meeting people outside my research community, and would have enjoyed even more opportunities for that sort of formal interaction. Most of the presentations were quite effective (although I was frustrated that so many of the short talks went over their allotted time, diminishing the impact of those of us who spoke later and more briefly).

(d) length of the meeting: The length of the meeting was pleasant. Although it was nice to have several hours free each afternoon, I would have sacrificed some of that to allow more time in the shorter talks—or even better, a poster session to continue the science dialog.

(e) other comments: Thanks very much for inviting me!

Name: Kathryn Ivanetich
Dept./Institution: Biomolecular Resource Center and Department of Pharmaceutical Chemistry, UCSF
Topic: Application of DNA sequencing & quantitative PCR microbial source tracking assays to identify host sources of fecal pollution in watersheds
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Summary of Research: My research centers on developing new methods for microbial source tracking, viz. identifying the host sources of fecal pollution in watersheds, with a long term goal of making a significant contribution toward improving water quality in California and nationwide. Specifically, we have applied bioinformatics and biotechnologies that directly sample gene sequence, such as automated DNA sequencing and Taqman Quantitative PCR, toward development of novel microbial source tracking assays. The DNA sequencing assay for the mdh gene target can distinguish the horse, dog/deer pair, gull and in some cases human host sources. The second approach utilizes host specific microorganism strains and species to selectively assay individual host sources. The positive and negative predictivities and false positive rates for these hosts specificity were comparable to or better than most published microbial source tracking methods. Taqman QPCR assays for the human specific gene Esp gene target and the 16srRNA gene target indicative of total fecal pollution were developed and validated. The assays were further validated and applied to environmental samples to determine host sources of fecal pollution. The application of these assays to the San Francisco Bay and San Pedro Creek (Pacifica, CA) was outlined.

Impressions of the Meeting: Scientifically, the meeting was excellent, and the quality of the presentations was

outstanding. I felt that UCSF representation was significantly improved, and that the number of UCSF PIs should be maintained for next year.
I felt that the quality of hotel's food had significantly declined since last year, and that overall the food was abominable. For example, the orange juice was often too watery and the snack shack guacamole was runny and the chicken too stringy to eat. Even the gourmet restaurant had uneven quality, with one meal basically inedible (cold food plus warm drinks). As always, Jorge and his staff ensured that the meeting room was run extremely well. As last year, I feel that the El Presidente's dinner reservation system which requires a reservation to be made with the concierge each day for that night's dinner is inefficient and irritating, and should be scrapped. Perhaps it is time to look at other resorts in San Jose del Cabo, with either all-inclusive or room only offerings.

Investigator: Thomas L. James
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Overview: Major goals of our research are (a) to develop the means of ascertaining conformational flexibility in protein and nucleic acid structures in solution primarily using multidimensional NMR data, (b) investigate small molecule-macromolecule interactions as well as biomolecular structure and dynamics using NMR, and (c) use three-dimensional nucleic acid structures with computational search algorithms and subsequent NMR screening to discover novel ligands. The subjects for study are often chosen to be targets for subsequent drug design.

Presentation: *Interactions of Poly(C) Binding Protein (PCBP) with Telomeric DNA and Telomerase RNA*. I reported some of our recent results on poly (C) binding proteins (PCBPs), which occur in both cytoplasm and nucleus of human cells. They participate in regulation of many cellular processes, and they are co-opted by some invading viruses. They appear to play a role in tumor growth and in viral growth. PCBPs exert their influence via interactions with RNA, DNA and protein. Each PCBP contains three KH domains as the only nucleic-acid recognizing motif. We have used both NMR and XRD to gain structural insights into the specificity of interactions of PCBP2 KH domains with RNA and DNA, with an emphasis on the human telomere cytosine-rich strand and the RNA template from human telomerase. Specifically, we have found: (a) The KH domains recognize both RNA and DNA with similar binding modes; (b) specific recognition of the poly(C) sequence is achieved by a network of strong hydrogen bonds; (c) specific recognition of the poly(C) sequence is achieved by a network of strong hydrogen bonds, (d) the KH domain has two non-overlapping surfaces for interaction with nucleic acids and proteins, respectively, so RNA/DNA binding and protein-protein interaction can occur simultaneously; and (e) RNA/DNA binding may affect protein-protein interactions, thus effecting function.

Impressions of the meeting:
Location: Excellent
Number of participants: Just right
Length: Just right
The presentations and quality of science presented were first-rate. The size of the meeting, focus of interests, location in an isolated setting conducive to strong interactions among participants, and quality of the science presented made this one of the best meetings I have attended. Although I had some notion of the research areas of my colleagues, it was good to have those updated as well as learn much more about the

exciting research at Scripps, at UC Berkeley, and at the industrial labs represented.

Investigator: John E. (Jack) Johnson
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Overview: Our laboratory is interested in the structure and function of viruses. We use crystallography, electron cryo microscopy (cryoEM), molecular biology and a variety of other methods to study the entry, assembly and maturation of RNA and DNA viruses. Four different systems have been extensively investigated by structural and genetic methods. (1) Nodaviruses are simple positive strand RNA viruses that encode a polymerase and a capsid protein in two different segments of its bi partite genome. The 320Å diameter capsids have T=3 symmetry and utilize the RNA genome as part of the molecular switch that controls particle formation. Many aspects of its cell entry appear to be comparable to picornaviruses. We have solved structures of four different members of this group and have developed detailed and, in some cases, well tested hypotheses for the molecular mechanisms associated with virus entry, translocation of the genome across membranes and steps in the particle maturation. (2) Tetraviruses are positive strand RNA viruses with bi partitite genomes and a T=4 capsid, 410Å in diameter. The x-ray structure of one tetravirus and cryoEM studies of a procapsid (450Å diameter) have recently been published. Employing recombinant expression-assembly systems, we have been able to demonstrate, for the first time, the existence of a procapsid in the assembly of a simple RNA virus. (3) Picorna-like plant and insect viruses have been studied by crystallography and molecular biology. These positive strand RNA viruses are similar to the mammalian picorna viruses and are likely to be part of their evolutionary development. Plant viruses are now used as bio materials by placing peptides or reactive groups on the

virus with an infectious clone. (4) dsDNA phage capsids were recently crystallized for the first time in our laboratory. The structure of the 650Å diameter mature particle of the l-like phage, HK97, was recently completed at 3.5Å resolution and procapsid (450Å diameter) was solved at 5.5Å resolution. Time resolved cryoEM has allowed us to model intermediates in the maturation and to generate a molecular movie of a large-scale protein reorganization.

Presentation: "An asymmetric cryoEM reconstruction of infectious P22 virus shows that a component of the DNA packaging motor is also a sensor for chomosome condensation density"

Bacteriophages, herpesviruses and other large dsDNA viruses contain molecular machines that pump DNA into preassembled procapsids, generating internal capsid pressures exceeding, by 10-fold, that of bottled champagne. A 17Å resolution asymmetric reconstruction of the infectious P22 virion reveals that tightly spooled DNA about the portal dodecamer forces a conformation that is significantly different from that observed in isolated portals assembled from ectopically expressed protein. We propose that the tight dsDNA spooling activates the switch that signals the headful chromosome packing density to the particle exterior. We have also shown that "cementing" proteins that stabilize the capsid in the presence of the intense pressures are interchangeable between different bacteriophage and that they can discriminate between closely similar quasi-equivalent sites.

References

Lander, G. C., Tang, L., Casjens, S. R., Gilcrease, E. B., Prevelige, P., Poliakov, A., Potter, C. S., Carragher, B., and Johnson, J. E. 2006. The structure of an infectious P22 virion shows the signal for headful DNA packaging. *Science* 312:1791-5.

Tang, L., Gilcrease, E. B., Casjens, S. R., and Johnson, J. E. 2006. Highly discriminatory binding of capsid-cementing proteins in bacteriophage L. *Structure* 14:837-45.

Impressions of the meeting
Location: Excellent
Number of participants: About right
Length of meeting: appropriate
This was my fifth Cabo meeting and I enjoyed it at least as much as the previous four(1998, 2000, 2002, 2004). The scientific presentations were of uniform high quality and the exposure to new developments outside of my own field was excellent. The meeting had the advantages of a large gathering because of the diversity of science, but also the advantages of a small meeting because there were no parallel sessions and plenty of time to discuss the science and presentations with colleagues. It is a remarkable meeting that I hope will continue. I found it very worthwhile and plan to attend again if possible.

Name: Don Kerkow
Supervisor: James R Williamson
Dept./Institution: Department of Molecular Biology, TSRI
Topic: [New Title] Characterization of a potentially novel RNA binding motif in the *C. elegans* protein NXF-2
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Summary of Research: My research focuses on the biochemical and structural characterization of a unique *C. elegans* orthologue of the Human nuclear mRNA exporter NXF-1/TAP. Unlike NXF-1/TAP, *C. elegans* NXF-2 is not implicated in general mRNA nuclear export and, therefore, may possess a more unique and specific set of functions. One example of NXF-2's unique behavior within the nuclear export family (nxf) comes from its interaction with *C. elegans tra-2* mRNA. NXF-2 bound *tra-2* appears to prevent normal NXF-1 mediated export of that transcript and promotes export through a CRM-1 dependent mechanism. I have identified that NXF-2 contains a potentially novel RNA binding domain that is used to interact with tra-2 mRNA. Structural work is being pursued to better understand this

potentially novel interaction. This work may reveal a new family of RNA binding domains.
Impressions of the Meeting: I would not recommend any changes to the meeting. It was a great location and a good number of people. Professor and Sponsor talks were interesting and informative. It was also nice to see what students at my institution and others are researching. I look forward to next year!

Investigator: Tanja Kortemme
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Overview: Our research focuses on prediction and design of structures and dynamics of proteins, protein-protein interactions and networks. We develop computational models at the atomic level of detail, and apply these methods to engineer proteins and their interactions. Using a combination of approaches from computational biology, experimental biophysics and cell biology, we aim to characterize the properties of sets of protein-protein interactions in regulating biological processes at the cellular level.

Presentation: We have developed computational models to analyze determinants of specificity and promiscuity in protein-protein interfaces. We experimentally tested our predictions through structural and functional characterization of new protein pairs with altered interaction specificity, and will use these engineered protein interactions to investigate the quantitative requirements for signal transfer in biological assemblies.

Impressions of the meeting: Location: Excellent
Number of participants: Excellent

Length of meeting: Just right
The meeting provides a great environment for scientific exchange, with high-quality presentations throughout and ample time for informal discussions of new ideas. The size was perfect. I particularly liked the fact that most of the presented research was unpublished, which makes this meeting very unique. I would also like to stress that the science presented by the post-docs and graduate students was very exciting, and a real highlight of the meeting.

Name: Gabriel C. Lander
Supervisors: Jack E. Johnson & Bridget Carragher
Dept./Institution: Department of Molecular Biology/ Department of Cell Biology
Topic: Characterization of virus assembly and maturation by CryoEM
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Summary of Research: I have been working towards building a better understanding of the role that each gene product plays in the process of bacteriophage p22 assembly and maturation. A reconstruction of the intact virion with fully packaged DNA has been determined to 17 Angstroms resolution, and each gene product has been identified. The portal complex within this reconstruction, when compared to the reconstructed density of the isolated portal complex, displays a significant conformational change. We hypothesize that this dodecameric portal structure functions as a pressure-sensitive switch that signals the packaging complex to shut off once the capsid has reached an optimum DNA density.

Impressions of the Meeting: If anything, I believe that this meeting was too short, I would have liked to have heard slightly longer talks from the PI's present. Many of the presentations I felt were slightly rushed due to the limited time they had to speak, such that their ideas were not fully

explained. The relaxed atmosphere and small size of the meeting provided a perfect stage for interesting discussions with people from a wide range of backgrounds, an aspect which I found most enjoyable. The schedule was well planned, and at no point did I feel worn out from long days of talks and discussions, as is the case at many other conferences I have attended.

Investigator: Duncan McRee
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Overview: ActiveSight is a structure-based drug design contract services organization. We provide a portfolio of drug targets with which we do co-crystal structures of client's lead compounds. Basically our clients send us compound and we send them back the coordinates with a rapid turnaround since we have the proteins targets ready-to-go. We also provide crystallization, structure-determination services for custom projects, fragment-based screening services. See www.active-sight.com for more information.

Presentation: I described our recent work in automating fragment-based screening in the home-lab environment. In fragment-based screening crystals are soaked with mixtures of small "drug-like" fragments and binding is determined by doing the X-ray structure. While these fragments do not have high potency, they have a high ligand efficiency, the ratio of binding energy per atom, which makes then excellent starting cores for new and novel lead compounds. Thus the technique is complimentary to HTS. We have built an automated pipeline for screening including a crystal-mounting application Mt. GUI, the ACTOR robot for sample mounting, and new automated software for structure-solution. I presented the results of a fragment screen on Hsp90 which produced 11 hits from our library of 420 compounds.

Impressions of the meeting:
Location: Excellent
Number of participants: Excellent
Length of meeting: Just right

Investigator: Andrew May
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Overview: Fluidigm Corporation develops and manufactures Integrated Fluidic Circuits (IFCs) –microfluidic chips that can be used for a broad variety of applications in the life sciences. The TOPAZ® system for protein crystallization is centered around IFCs that allow up to 8 samples to be screened by free-interface diffusion against 96 reagents using approximately 1 ml of sample. IFCs are being developed for other applications, including gene expression and immunoassays.

Presentation: TOPAZ screening chips have been developed that can screen up to 8 samples against 96 reagents. The talk described an approach whereby chips can be used for analytical crystallization experiments. Rather than relying on orthogonal biophysical methods for characterisation, samples can be tested directly for crystallizability in TOPAZ chips. This early crystallization data can be obtained from small scale samples before committing resources to scale-up of the sample prep and moving to standard crystallization experiments. The parallel architecture in 8.96 chips allows this approach to be applied to many variants in the sample under study as early as possible. Data were shown from successful application of this technique by RX3 pharmaceuticals, a biotech company located in San Diego.

The final section of the talk showed examples of IFCs that have been developed at Fluidigm for other applications, including Dynamic Arrays for gene expression and immunoassays, and Digital Arrays for high sensitivity detection of rare genetic mutations.

Impressions of the meeting:
Location: Good
Number of participants: Excellent
Length of meeting: Excellent

The quality of the talks was excellent and the size of meeting provided great opportunities for interesting discussions. This was the first meeting that I have been to for a while where people were presenting new, unpublished results from their laboratories as well as more complete, well developed stories. It would be great to see a few more grad students/postdocs given the opportunity to give longer talks during the main sessions, given the quality of work being presented by their PIs. The sponsors' dinner provided a good chance to meet with people and some lively discussion.

Name: Jonathan Melnick
Supervisor: Peter G. Schultz
Dept./Institution: Chemistry, TSRI
Topic: Genome-wide screening for modulators of Integrin-alpha4 dependent migration
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Summary of Research: My research focuses on the identifying novel genes associated with alpha4-dependent migration. Using cDNA overexpression and shRNA-based knock-down, I screened genetic libraries in the scratch-wound healing assay using the adherent cell line HEK293. The resulting hits were further validated in Jurkat cells by monitoring for inhibition of transwell migration. Current

efforts focus on following up on the novel genes idetified during this process.

Impressions of the Meeting: The conference ran very smoothly and was very fun and informative. All of the attendees were very enthusiastic and friendly. The presentations were informative and well presented. All conferences should be as efficacious as this one.

Investigator: David P. Millar
Dept./Institution: Department of Molecular Biology, The Scripps Research Institute
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Overview: My laboratory applies modern fluorescence spectroscopic methods, including single-molecule and time-resolved techniques, to studies of nucleic acid folding processes and protein-nucleic acid interactions. The major projects in our laboratory include: (a) Molecular biophysics of DNA replication. Single-molecule fluorescence methods are being used to visualize the dynamic interactions of DNA polymerases with their DNA and nucleotide substrates. (b) RNA folding and ribonucleoprotein assembly. We are studying the assembly pathway of the signal recognition particle and the oligomerization of HIV-1 Rev on the Rev Response Element RNA.

Presentation: My presentation focused on the Rev/RRE system from HIV-1. Rev is a key regulatory protein from HIV-1 that controls the transition from the early to late phases of viral gene expression. Rev binds to the Rev Response Element within the viral RNA, where it assembles as an oligomeric RNP. The formation of the oligomeric complex is essential for the subsequent export of unspliced viral mRNA from the nucleus to the cytoplasm. Hence, the binding of Rev to the RRE and the subsequent oligomerization are potential targets for new therapeutic

agents for the treatment of AIDS. We are applying a variety of fluorescence methods to study the basic mechanism of RNP assembly and to develop novel screening assays to identify small molecule inhibitors of the Rev/RRE interaction.

I also described a novel nucleic acid-protein interaction that provides a potential therapeutic target for treatment of Leishmaniasis. The DNA from kinetoplastids contain an unusual modified base, β-D-glucosyl(hydroxymethyl)uracil, called J. Kinetoplastids also contain a J-binding protein (JBP) that specifically recognizes base J in the context of double helical DNA. Since the interaction of JBP with J base is essential in *Leishmania*, but does not occur in eukaryotes, this interaction provides a potential therapeutic target. We have developed a fluorescence polarization assay to monitor this DNA-protein interaction *in vitro* and to screen chemical libraries for potential inhibitors. To identify the molecular basis for recognition of J base, we have quantified the binding of JBP to a series of DNA duplexes containing analogs of the J base (in collaboration with Paul Wentworth at TSRI).

Impressions of the Meeting:
Location: Excellent.
Number of participants: Good size and mix of faculty, postdocs and graduate students.
Length of meeting: Just right.
This was another successful and highly enjoyable meeting. The Cabo meeting is one of the most informative meetings that I typically attend, and has become a highlight of the scientific year for me. The breadth of topics and the quality of the science presented are always outstanding. This meeting provides a unique opportunity to learn about exciting new areas of research at both Scripps and UCSF. I also enjoy the chance to interact with Scripps and UCSF colleagues in a friendly, relaxed atmosphere. The El Presidente Hotel continues to provide a convenient venue for the meeting.

Investigator: Ron Milligan
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Overview: We use cryo-electron microscopy and image analysis to study the structure and mechanism of action of several macromolecular assemblies. Current interests include actin-myosin and microtubule-kinesin complexes, proteins that affect microtubule dynamics, AAA proteins and membrane proteins. We use the three-dimensional maps calculated from electron images of the machines together with biochemical data and high-resolution x-ray structures of the individual components to provide insight into molecular mechanisms.

Presentation: Studies on microtubule binding proteins. Time resolved cryo-EM has revealed that assembling microtubules have blunt ends with straight protofilaments, and that disassembling microtubules have flared ends with outwardly curling protofilaments. These data have contributed to understanding the mechanisms underlying dynamic instability. I presented data on doublecortin – a microtubule stabilizing protein – showing that it binds on the outside of the microtubule at the junction of 4 tubulin monomers. This is an ideal location for stabilization as it allows doublecortin to potentially strengthen both longitudinal and lateral interactions of tubulin monomers. I also presented data on a kinesin 13 microtubule depolymerizing motor. The data reveal that the motor bends microtubule protofilaments outwards, mimicking the protofilament conformation in disassembling microtubules. These two proteins appear to have evolved by co-opting basic properties of tubulin to bring about regulatable changes in the cellular microtubule cytoskeleton.

Impressions of the meeting:

size of the group-	excellent
location of the meeting	excellent
attendees and presenters	excellent
length of the meeting,	excellent

All the presentations were of very high quality. I found the student presentations on the first evening particularly useful, although a short 15 minute break in the middle of that session would have been very welcome.

Investigator: Daniel L. Minor, Jr.
Dept./Institution: Cardiovascular Research Institute, UCSF
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Overview: My lab studies the structure and function of ion channel proteins and the molecules that regulate their function.
Our work in ion channels covers two broad areas: structural investigation of ion channel architecture and function, and the development of new methods to identify and map the sites of action of ion channel regulatory molecules. Ion channel proteins are central to the function of the brain, heart, and nervous system. Understanding how ion channels work at the molecular level should facilitate the development of a range of potential therapeutics for treatment of cognitive disorders, cardiac diseases, and pain.

Presentation: My presentation revealed the first structures from a voltage-gated calcium channel (Ca_V) subunit. We showed that the voltage-gated calcium channel b-subunit (Ca_Vb) is a two domain protein that is related to the MAGUK family of protein scaffolds. Our co-crystal structure of the Ca_Vb with the a-subunit interaction domain (AID) defines the molecular basis for how Ca_Vb interacts with the pore forming subunit. The co-crystal structure suggests that Ca_Vb may regulate channel function via direct control of the movement

of a continuous a-helix formed by the AID and the pore-lining segment IS6 of the a-subunit. Thermodynamic experiments demonstrate that the $Ca_V\tilde{b}$ AID interaction is particularly high-affinity (Kd ~ nM) and that the largest share of this high affinity interaction arises from a limited set of residues on the AID.

I also presented our recent on dissecting the structural basis of calcium-dependent autoregulation of Ca_Vs. The ability of Ca_Vs to sense calcium entry through their own pore forms the basis for two opposing processes that are mediated by calmodulin (CaM): calcium-dependent inactivation (CDI) and calcium-dependent facilitation (CDF). We determined the structure of Ca^{2+}/CaM bound to the IQ domain of $Ca_V1.2$. IQ domains are widespread CaM binding motifs and our structure is the first high-resolution structure of a Ca^{2+}/CaM-IQ complex. By combining titration calorimetry studies of the Ca^{2+}/CaM-IQ interaction with electrophysiological measurements on structure-based mutants of the entire channel, we were able to define a previously unknown role for Ca^{2+}/CaM N-lobe interactions in CDF of L-type channels.

Impressions of the meeting:

Location: Excellent

Number of participants: The meeting is a good size for facilitating interactions

Length of meeting: Fine

Investigator: Arthur J. Olson

Address: Department of Molecular Biology The Scripps Research Institute

Overview: My laboratory is engaged in the development and application of computational techniques to analyze, visualize and predict biomolecular interactions. Projects currently underway in the group include 1) development of computer graphics approaches for visualizing macromolecular structure and properties using surface and volumetric techniques, 2) development of automated docking codes for predictiong the interaction of substrates with protein active sites 3) prediction of the evolution of drug

resistance by co-evolutionary algorithms 4) application of drug design techniques to the problem of HIV anti-viral therapeutics, 5) Approximation and representation of protein surfaces using analytical functions 6) development of techniques for predicting protein-protein interactions 7) application of protein-protein docking techniques to study the molecular initiation of thrombosis and 8) development of methods to construct large molecular assemblies from individual molecular structures.

Presentation: This year I presented new results in the laboratory on our project "Tangible
Interfaces for Molecular Biology." We are investigating the application of computer-driven autofabrication and augmented-reality to develop novel tangible interfaces for the study of biomolecular structure and interaction. I discussed the nature of self-assembly, and showed a model of the pentameric subunits of the poliovirus that demonstrated the process of self assembly of the physical model into a complete dodecahedral shell. I explained the essence of the model and how it can be applied to other systems.

Impressions the meeting:
Location: very good, though it may be time for a change
Number of participants: ideal
Presentations: Of very high quality. I was glad to see more participation from the UCSF group this year.

Investigator: James C. Paulson
Dept./Institution: Departments of Molecular Biology and Molecular and Experimental Medicine, TSRI
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Overview: Our group investigates the roles of carbohydrate binding proteins that mediate cellular processes central to

immune regulation and human disease. In particular we are interested in the siglec family of glycan binding proteins that are expressed on most white blood cells and mediate cell-signaling events.

Presentation: *Sweet Spots in B cell Signaling.* CD22 is a regulator of B cell receptor signaling mediated by its ability to recruit phosphatases to the receptor complex via its cytoplasmic domain. CD22 is also a member of the Siglec family with an extracellular domain that recognizes sialic acid containing carbohydrates (glycans) of glycoproteins as a ligand. Our current focus is to understand how the glycan-binding domain of CD22 modulates its function as a modulator of B cell signaling. At the last CABO meeting we reported that cell surface CD22 is predominately localized to clathrin coated pits, which is the likely site of its regulation of the B cell receptor following activation and endocytosis of the activation complex. We have now developed high affinity synthetic ligand probes of CD22 that compete with endogenous cell surface ligands, and are endocytosed by the B cell in a CD22 dependent manner. By attachment of a toxin to this glycan ligand we created a Trojan horse that is endocytosed and kills B cell lymphomas, but is inert to cells that do not carry CD22. The results suggest a novel approach to development of therapeutics for B cell leukemias by targeting the ligand binding domain of CD22.

Impressions of the meeting:
Location: Excellent
Number of participants: Just right
Length of meeting: Just right
As in past years the format of the meeting is unique and highly effective. Hearing each participant speak or introduce themselves reduces barriers for stimulating discussions with new and old participants alike. It is amazing how effective the short 5-7 minute student and post-doc presentations are. This year the quality and diversity of the presentations by academic and industry presenters was outstanding.

Investigator: Andrej Sali
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Overview: We are using computation grounded in the laws of physics and evolution to study the structure and function of proteins. We aim to improve and apply methods for: (i) predicting the structures of proteins; (ii) determining the structures of macromolecular assemblies; (iii) annotating the functions of proteins using their structures. This research contributes to structure-based functional annotation of proteins and thus enhances the impact of genome sequencing, structural genomics, and functional genomics on biology and medicine.

Presentation: We developed a method for structure characterization of assembly components by iterative comparative protein structure modeling and fitting into cryo-electron microscopy (cryoEM) density maps. Specifically, we calculate a comparative model of a given component by considering many alternative alignments between the target sequence and a related template structure while optimizing the fit of a model into the corresponding density map. The method is being implemented in our program MODELLER for protein structure modeling by satisfaction of spatial restraints and will be applicable to the rapidly increasing number of cryoEM density maps of macromolecular assemblies.

Impressions of the meeting:
Location: Good
Number of participants: Perfect
Length of meeting: Just right
There was plenty of time to be engaged in free format discussions with other participants. A large fraction of presentations were inspiring and informative.

I**nvestigator**: Michel F. SANNER
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Overview: My research interests are in the computational representation and simulation of molecular interactions. I am the author of the program MSMS to compute such surfaces, which has been distributed to over 6000 laboratories worldwide. I have also pioneered a software development strategy centered on the high-level, object-oriented, interpretive language Python. This approach promotes the development of components that are re-usable and that can be combined at a high level to rapidly prototype novel combinations of known computational methods. It also facilitates the rapid integration of newly developed computational methods. We have developed several basic components to take care of specific computational aspects of our research. We have built several applications from these software components and have distributed them to over 13000 users. We are currently working on adding flexibility to the computational representation of biological macromolecules during simulations such as docking.

Presentation:
Computational Representation of Protein Flexibility: Application to Automated Docking
We have developed a new hierarchical, multi-resolution, representation of protein flexibility called the Flexibility Tree. This structure allows the combination of a wide variety of motions within a protein in a way that is computationally tractable. We have used this data structure in the context of automated docking and demonstrated significantly higher levels of success in cross docking multiple ligands into receptors.

Impressions of the meeting: Location: Excellent.

Number of participants: Good size
Length of meeting: Just right
This was the forth time I attended this meeting and I was very impressed both by the quality of the science presented and the number of people I interacted with. I think the size of the meeting is perfect because it give you a chance to get to know everyone and it really favors interactions. I learned a lot about research conducted by colleagues from both my own institution (TSRI) as well as UCSF. I truly appreciated the opportunity to "advertise" my own work and I am working with several new collaborators as a direct consequence of attending this meeting.

Investigator: Daniel V. Santi
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Overview: Some of my past research, and that which I described, focused on synthetic biology approaches to combinatorial biosynthesis of polyketides.

Presentation: Type I polyketide synthase (PKS) genes consist of modules ~3-6 kb long that encode the structures of 2-carbon units in a polyketide product. Alteration or replacement of PKS modules can lead to "unnatural" natural products, but technology development to do so has been slow. We have describe a generic design of synthetic PKS genes where modules and domains are flanked by a repeated set of restriction sites, allowing facile cassette assembly and interchange. To test feasibility, we synthesized 17 modules from nine PKS clusters and associated them in 204 bimodular combinations spanning over 2 million bp of novel PKS gene sequences. Remarkably, nearly half the combinations made a polyketide

in *Escherichia coli*. We also resuscitated inactive bimodule pairs by exchanging ketosynthases in the second, ketide-accepting modules to those that are naturally cognate to the first. Finally, we were almost 100% successful at producing trimodules by coupling overlapping bimodules that were individually active.

Impressions of the meeting:
Location: Excellent
Number of participants: Excellent
Length of meeting: Perfect
The size and quality of the meeting this year was perfect. The size provided a basis for interaction amongst all participants. The presentations were of extremely high quality throughout. As always, the meeting provided an excellent forum for discussion and fostering of new ideas and collaborations.

Name: Justin Scheer
Supervisor: Jim Wells
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Topic: A common allosteric site in caspases.
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Summary of Research: Using a high-throughput screening approach called disulfide-trapping, I identified several compounds that allosterically inhibit caspase-1. The mechanism of inhibition is to stabilize an inactive conformation of the enzyme. This conformation is remarkably similar to naturally occurring inactive forms of the enzyme and is shared among very divergent caspases.

Impressions of the Meeting: I liked the meeting, although there were a slightly disproportionate number of computational talks. The industry talks were definitely a plus.

Investigator: Kimmen Sjolander
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Overview: Research in the Berkeley Phylogenomics Group focuses on three areas:

(a) Algorithm development for protein functional and structural classification, including homolog detection, multiple sequence alignment, phylogenetic tree construction, protein structure prediction, active site prediction, subfamily identification, hidden Markov model (HMM) construction, and improving the accuracy of comparative model construction.

(b) Web server development, as a service to the scientific community. We provide servers for tools developed in our lab, e.g., active site detection, protein structure prediction, phylogenetic tree construction, etc. Our flagship resource is the PhyloFacts phylogenomic library, containing protein family "books" for almost 10K protein families and domains, and over 700K HMMs for classification of novel sequences to families and subfamilies.

(c) Research in innate immunity. We collaborate with scientists around the world in investigation of the complex evolutionary history of the molecules involved in eukaryotic innate immunity.

Presentation: Protein families evolve a multiplicity of functions and structures through gene duplication, domain shuffling, speciation and other processes. As numerous studies have shown, function prediction by homology is associated with systematic errors on these data. Phylogenomic analysis, combining phylogenetic tree construction, integration of experimental data, and differentiation of orthologs and paralogs, has been shown to address these errors and improve the accuracy of functional

classification. The explicit integration of structure prediction and analysis in this framework, which we call *structural phylogenomics*, provides additional insights into protein superfamily evolution, and improves function prediction accuracy.

Impressions of the meeting:
Location: Excellent. Encouraged interaction among participants. Hotel room was very comfortable. I don't normally stay at all-inclusive resorts, but this was very enjoyable.
Number of participants: Just right. It should be kept to this size or slightly smaller.
Length of meeting: Just right
Other comments: Very well organized. I'm very glad I was able to attend, and look forward to future meetings.

Investigator: C. David Stout, Ph.D.
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Overview: Our research is focused on the structure and function of membrane bound enzymes, and the development of methods for membrane protein crystallization. We study the mechanism of transhydrogenase (TH), a mitochondrial respiratory enzyme complex that couples proton translocation with hydride transfer. We employ x-ray crystallography, biochemical and spectroscopic methods, electron microscopy in collaboration with M. Yeager, and NMR in collaboration with J. Dyson. Crystal structures of TH soluble domains, alone and in complex have been determined. We also work on cytochrome P450s, cytochrome oxidase, and nanodiscs, which are particles composed of apo-lipoprotein and phospholipids, for solubilizing integral membrane proteins.

Presentation: At this year's meeting I reviewed efforts toward crystallization of intact *E. coli* TH in its membrane bound form, including the development of His-tagged constructs, protein expression and purification methods, the use of specific mutants, and the efficacy of various detergents and additives in stabilizing a monodisperse form of the enzyme. These experiments illustrate the challenges in crystallizing large integral membrane proteins, and are a necessary prelude to structure determination. Biochemical characterization indicates that TH exists in membranes as a larger assembly of subunits than previously expected.

Impressions of the meeting:
Location: Outstanding
Number of participants: Ideal
Length of meeting: Just right
This year's Cabo meeting was truly outstanding with very strong representation from both TSRI and UCSF. As in previous years, the breadth of scientific problems and diversity of disciplines covered in the presentations made it highly stimulating and enriching. In particular, a strong theme of the role of symmetry in macromolecular assembly and structure emerged. The unique juxtaposition of speakers also highlighted themes in drug design, and protein structure and folding, in a manner that just does not occur at other scientific gatherings. The Cabo meeting is unique in anticipating upcoming research trends, and at the same time in presenting new applications in biotechnology. The meeting provides a very favorable format and venue for extensive discussions with colleagues and graduate students. The setting, facilities and program are excellent.

Investigator: Robert M. Stroud
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Overview: The main project in my laboratory discussed concerned the structural determinations of the ammonia channel.

Presentation: The mechanism was defined in structures that show how the channel works to conduct ammonia in the gas form. Protein crystal structure has a key role in the understanding of an essential process in cell biology.

Impressions of the Meeting:
Location: Excellent.
Number of participants: Good size
Length of meeting: Just right

Convenient for access from California, and sufficiently remote to concentrate people's time and attention. Cabo San Lucas, is excellent after refinement of location over the years.
Number of participants: A comfortable size for the meeting is about 40 people, with 20 speakers. Attendees and presenters were excellently chosen from the superb groups in structural biology at Scripps and at UCSF.
Length of meeting: The meeting of 3 days length is quite adequate and more would probably be too much.
The science presented was absolutely first rate with many important new breakthroughs in the fields of immunology, drug design, chemical basis for inhibition, chemical basis for understanding enzyme mechanisms and cell surface receptor interactions.

Investigator: Rashid Syed
Dept./Institution: Molecular Structure Group, Amgen Inc.
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Overview: My research as a structural biologist in the Drug Discovery group at Amgen focuses on therapeutic targets of

interest based on medical need and its amenability to protein crystallography methods. My recent work has involved a number of different target classes e.g. kinases and prolyl hydroxylases.

Presentation: "**p38 MAP Kinase Inhibitors: Discovery and Selection of Drug Candidates"**
My presentation was on p38 MAP kinase target describing various aspects of its drug-discovery process from preclinical research to clinical trial stages. I presented challenges involved and showed how crystallography plays a critical part in it.

Impressions of the meeting:
Location: Great
Number of participants: Perfect
Length of meeting: Perfect
I enjoyed the meeting as the location provides a perfect environment for discussion, meeting old friends and making new ones.

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Supervisor: Andrej Sali
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Summary of Research: My research focuses on improving the structural characterization of macromolecular assemblies by combining comparative modeling and electron cryo-microscopy density maps at intermediate resolutions (5-15 Å). I developed a density fitting program (Mod-EM) which is implemented in the comparative modeling program MODELLER. My protocol for refining comparative models by simultaneously optimizing their fit into the density map and the alignment between their target sequence and a given template structure is currently being applied to model components of macromolecular assemblies such as the

RyR1 calcium release channel, TRiC chaperonin, Acrosomal bundle, as well as ribosomes from two different species.

Impressions of the Meeting: This meeting was great! The location is beautiful, and the presentations were very interesting and informative. Presentations by post-docs were too short and too close to one another.

Investigator: Jim Wells
Dept./Institution: Pharmaceutical Chemistry & Cellular and Molecular Pharmacology
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Overview: My research focuses on site-directed ligand discovery to probe and identify allosteric sites in proteins. In particular we have found a common allosteric site in the family of caspases which are broadly participate in innate immunity and apoptosis.

Presentation: We identified highly selective compounds using disulfide-trapping that react with cysteine residues in the dimer interface of caspases. The compounds drive the enzyme into a natural “off-state” that mimics the zymogen or apo-forms of the protease. These compounds disrupt a hydrogen-bonding network that radiates some 15 angstroms away to each of the two active sites. These studies reveal and unique allosteric mechanism for these proteases and identify an alternate site for drug discovery in this important class of proteases.

Impressions of the meeting:
Location: Excellent
Number of participants: Excellent
Length of meeting: Just right
The size of the meeting good. Big enough for diversity in talks and small enough to encourage interactions.

Investigator: Ian A. Wilson
Dept./Institution: Department of Molecular Biology, TSRI
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Overview: My laboratory focuses on the structural biology, particularly on interaction of microbial pathogens with the immune system. We investigate antigen recognition in the innate and adaptive immune system with emphasis on antibodies, T-cell receptors, Toll-like receptors, CD1, and other pattern recognition peptides. I also direct one of the four NIH-funded structural genomics production centers as part of the NIGMS Protein Structure Initiative (PSI). The Joint Center for Structural Genomics has now determined over 300 structures in the last five years, and our goal is to increase structure coverage of large protein families with no structure representatives and on our biomedical theme on "The Central Machinery of Life."

Presentation: I did not for the first time in 17 years give a presentation due to the great turnout this year and, hence, the need to let new people speak.

Impressions of the meeting: Fantastic – one of our best ever. The turnout from UCSF and TSRI was terrific, which when combined with our sponsors, led to a great meeting.
Location: As usual, excellent
Number of participants: About right
Length of meeting: Perfect

I**nvestigator**: Xiang-Lei Yang
Dept./Institution: Department of Molecular Biology, TSRI
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Overview: Our research is focused on a group of enzyme called Aminoacyl-tRNA synthetases, which catalyze the ligation of amino acids onto the 3' of the cognate tRNAs. This is the first reaction in the protein biosynthesis pathway. Along with their role in the establishment of the genetic code, over a long period of evolution, tRNA synthetases from mammalian cells pick up new domains, new motifs, new interactions results in new functions, which may or may not related to translation.

Presentation: We report the co-crystal structure of human tryptophanyl-tRNA synthetase and $tRNA^{Trp}$. This enzyme is reported to interact directly with elongation factor 1a, which carries charged tRNA to the ribosome. Crystals were generated from a 50%/50% mixture of charged and uncharged $tRNA^{Trp}$. These crystals captured two conformations of the complex, which are nearly identical with respect to the protein and a bound tryptophan. They are distinguished by the way tRNA is bound. In one, uncharged tRNA is bound across the dimer, with anticodon and acceptor stem interacting with separate subunits. In the other conformation, presumptive aminoacylated tRNA is bound only by the anticodon, the acceptor stem being free and having space to interact precisely with EF-1a, suggesting that the product of aminoacylation can be directly handed off to EF-1a for the next step of protein synthesis.

Impressions of the meeting:
Location: Excellent
Number of participants: Excellent
Length of meeting: Just right
The size of the meeting this year was perfect. The small size provided a perfect basis for interaction amongst all participants. The presentations were of very high quality throughout and in particular those who departed from the standard conference presentation. The meeting provides an excellent forum for discussion and fostering of new ideas and collaborations.

Investigator: Mark Yeager, M.D., Ph.D.
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Summary of research:

Macromolecular assemblies visualized by electron cryo-microscopy and image processing

In our laboratory a major theme is the use of electron cryo-microscopy (cryo-EM) and image analysis to examine the structure of large, multicomponent, supramolecular complexes. In electron cryomicroscopy, biological specimens are quick frozen to preserve their native structure and functional properties. A special advantage of this method is that we can capture dynamic states of functioning macromolecular assemblies, such as open and closed conformations of membrane channels and viruses actively transcribing RNA. Threedimensional density maps are obtained by digital image processing of the highresolution electron micrographs. The rich detail revealed in the density maps demonstrates the power of this approach to reveal the structural organization of complex biological systems that can be related to the functional properties of such assemblies. For our work in structural virology, maps at low resolution (~20 Å) reveal the overall shape and symmetry of the protein capsid layers, as well as the location, shape, dimensions, quaternary arrangement and stoichiometry of the component proteins. For the structure analysis of membrane proteins, electron crystallography of highly ordered, two-dimensional crystals enables a higher resolution analysis (~6 Å resolution) from which the secondary structure of the protein within the lipid bilayer can be discerned.

Research projects under way include the structure analysis of (1) membrane proteins involved in cell-to-cell communication (gap junctions), water transport (aquaporins), ionic transport (potassium channels), transmembrane

signaling (integrins), and viral recognition (rotavirus NSP4); (2) viruses responsible for significant human diseases (retroviruses, hepatitis B, rotavirus, astrovirus); and (3) viruses used as model systems to understand mechanisms of pathogenesis (arenaviruses, reoviruses, nodaviruses, tetraviruses and sobemoviruses).

Presentation:

Native Hepatitis B Virions and Capsids Visualized by Electron Cryo-microscopy

Hepatitis B virus (HBV) infects more than 350 million people, of which one million will die every year. The infectious virion is an enveloped capsid containing the viral polymerase and double-stranded DNA genome. The structure of the capsid assembled *in vitro* from expressed core protein has been studied intensively. However, little is known about the structure and assembly of native capsids present in infected cells, and even less is known about the structure of mature virions. We used electron cryomicroscopy and image analysis to examine HBV virions (also called Dane particles) isolated from patient sera and capsids positive and negative for HBV DNA isolated from the livers of transgenic mice. Both types of capsids assembled as icosahedral particles indistinguishable from previous image reconstructions of capsids. Likewise, the virions contained capsids with either T=3 or T=4 icosahedral symmetry. Projections extending from the lipid envelope were attributed to surface glycoproteins. Their packing was unexpectedly nonicosahedral but conformed to an ordered lattice. These structural features distinguish HBV from other enveloped viruses.

Impressions of the Meeting: This continues to be one of my favorite meetings! The size and informal workshop format of this meeting is similar to a Gordon conference with the advantages of a broad scientific scope and resort location. Frankly, I think the quality of the science presented at this meeting is outstanding. I also appreciate the participation of scientists in industry in order to gain insight into translational research and the potential for commercial

applications of basic research. For instance, it was very informative, but also sobering, to hear the Amgen stories about the development of p38 inhibitors that failed in Phase I trials.

Investigator: Todd Yeates
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Overview: Research in my group is in the broad areas of computational and structural biology and genomics. In our structural work we are increasingly interested in large self-assembling complexes. Along that line we are working to obtain high resolution crystal structures of actin oligomers to allow an atomic level construction of the F-actin filament. We are also elucidating the structures of giant protein shells called bacterial microcompartments. Although they are not well studied, these virus-like compartments are present in many bacteria where they serve as primitive organelles for sequestering certain metabolic reactions. We are also interested in thermophilic proteins and have discovered that certain organisms use widespread disulfide bonding, as well as topological linking to stabilize their proteins under extreme conditions; that was the topic presented this year. We also continue our work in computational and comparative genomics, with the goal of inferring the functions of uncharacterized proteins on the basis of 'genomic context', of which 'protein phylogenetic profiles' and our recent extension to triplet relationships using logic analysis are examples.

Presentation: We have been studying the proteins from certain hyperthermophilic, archaeal microbes. We have found – contrary to the general idea that the reducing environment of the cytosol eliminates the utility of disulfide bonding as a stabilizing mechanism – that certain

thermophiles have apparently evolved mechanisms that allow them to take advantage of widespread disulfide bonding to stabilize their cytosolic proteins. Furthermore, at least in *P. aerophilum* which we have studied the most thoroughly, disulfide bonding is used in an apparently intermolecular fashion to hold protein-protein complexes together. After identifying several such complexes using 2D gels and mass spectrometry, we determined the structure of one of them, citrate synthase, which we had shown experimentally was a homodimer held together by disulfide bonding. However, the crystal structure revealed an unexpected surprise. The disulfide bonds that hold the dimer together are in fact intramolecular. Each of the chains is cyclized by an intramolecular disulfide bond, but in such a way that the two chains of the dimer are topologically linked, or concatenated. Although there are no direct bonds between the two distinct subunits, they are held together irreversibly in a topological sense. Investigations are underway to see how widespread the phenomenon might be, and whether other variations on topological linking or knotting might be used by thermophiles for stability.

Impressions of the meeting: The meeting was wonderful as usual, and was the source of lots of new ideas. The meeting site was enjoyable as always. The duration and format were fine.

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Presentation: Automating Random Conical Tilt Data Collection.
Abstract: We have adopted cutting edge techniques from the field of computer vision and applied them to the problem

of collecting Random Conical Tilt Data on transmission electron microscopes. This ability will hopefully enable a new high throughput approach to finding initial models for EM reconstructions.

Impressions of the meeting: The meeting location is phenomenal, good weather and relaxed atmosphere are conducive to both conversation between peers and productive talks. The meeting schedule also provides ample opportunity to enjoy the best part of the day in leisure and arrive refreshed for evening talks. The quality of the talks are all excellent, and the small group size makes interaction from all the attendees common. The short talk time given to students and post-docs is an excellent example of forcing speakers to be concise and direct with their information and the impact of their work. All in all one of the best meetings within the structural biology community.