The SSRL Structural Biology Program & SSRL and the Early SPEAR3 Era

SSRL 50th Anniversary Celebration

Keith Hodgson

April 20, 2023

U.S. DEPARTMENT OF ENERGY | Stanford University
SLAC NATIONAL ACCELERATOR LABORATORY

Outline of Presentation

- The SSRL Structural Molecular Biology Program
  - Early science – building the foundations
  - Innovations in instrumentation and beam lines
  - Scientific discovery

- SSRL and the Early SPEAR3 Era
  - SPEAR3 – realizing funding and delivering the project
  - And in parallel – building the foundations for LCLS

SSRL SMB Program – a Brief Chronology and Key Milestones

- Macromolecular crystallography and EXAFS applications over 1974-1980
- First NIH funding from NIH NCRR began with a Research Resource Grant (P41) in 1980
- Program scope (added SAXS) and user base expanded, and DOE OHER (now BER) funding began in 1993
- Since 1993, managed and operated as an integrated center for structural biology with primary funding from NIH and BER augmented by private and corporate investments
- In 2023, NIH (from NIGMS) and BER support continues with planning ongoing for the next 5 years of the program

Foundations of the SSRL SMB Program – Early Science – Macromolecular Crystallography

- Earliest applications of synchrotron radiation to study biological structure were fiber diffraction studies by Holmes and Rosenbaum at DESY (first images in the summer of 1970)
- One of SSRL's first 5 experimental stations, BL1-4, was built by Caltech and SSRL for fiber diffraction. My group modified the BL1-4 instrument for crystal diffraction studies during 1974-75
- Subsequently, there were experiments involving anomalous scattering – including by my group, David and Lilo Templeton, and Wayne Hendrickson and collaborators

Foundations of the SSRL SMB Program – Early Science – Macromolecular Crystallography on BL1-4

Reprinted from
Proc. Nat. Acad. Sci. USA
Vol. 73, No. 1, pp. 128–132, January 1976
Biophysics

1974-75

Applications of synchrotron radiation to protein crystallography: Preliminary results

(x-ray diffraction/anomalous dispersion/rubredoxin/azurin/nerve growth factor/glutaminase-asparaginase)

JAMES C. PHILLIPS, ALEXANDER WLODAWER, MARGUERITE M. YEVITZ, AND KEITH O. HODGSON*
Department of Chemistry and Stanford Synchrotron Radiation Project, Stanford University, Stanford, California 94305

Communicated by Richard H. Holm, October 23, 1975

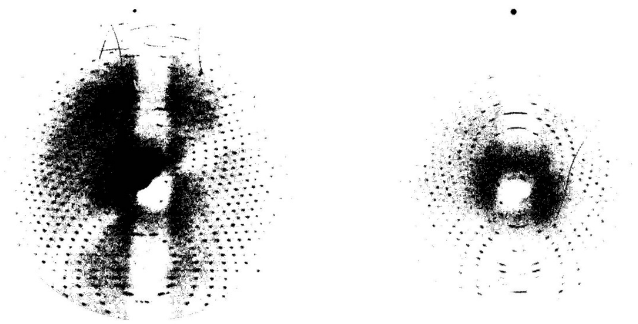

FIG. 3. Cone-axis oscillation photographs of the same azurin crystal. Precession angle 6.5°, oscillation angle 20°. (*left*) Synchrotron source, E = 3.7 GeV, I = 40 mA, only electrons present, λ = 1.740 Å, exposure time 10 min. (*right*) Philips fine-focus sealed Cu anode tube, operated at 40 kV, 30 mA, exposure time 6 hr, Ni filter.

- Measured with much shorter data collection times - 60x < fine focus x-ray tube
- Demonstrated successful use of smaller crystals
- Observed anomalous scattering effects at Fe (rubredoxin) and measured effects of good collimation
- Reported striking observation that higher intensity of SR offered significant advantages with regard to recording higher quality data to higher resolution

Foundations of the SSRL SMB Program – Early Science – Macro

Reprinted from
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Biophysics

Applications of syn
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JAMES C. PHILLIPS, ALEXAND
Department of Chemistry and Stanford Sync

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FIG. 3. Cone-axis oscillation photograph
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Foundations of the SSRL SMB Program – Early Science – Macromolecular Crystallography on BL1-4

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Proc. Nat. Acad. Sci. U
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JAMES C. PHILLI

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Foundations of the SSRL SMB Program – Early Science – Macromolecular Crystallography on BL1-5

1985-87

Reprint Series
12 August 1988, Volume 241, pp. 806–811

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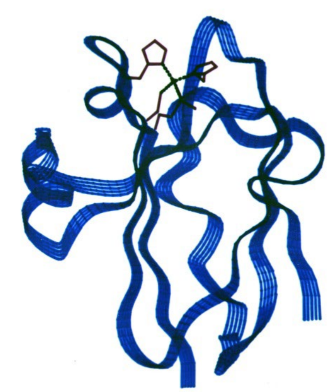


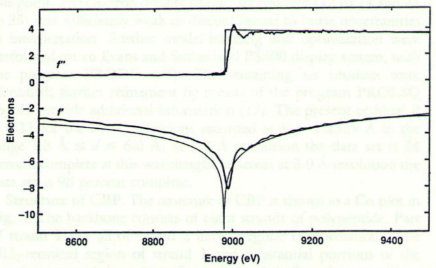
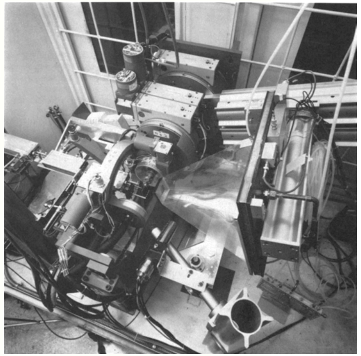


Fig. 1. Energy dependence of the anomalous dispersion terms *f''* and *f'* in the region of the CuK absorption edge. Values of *f''* and *f'* are in electrons. Experimental values for *f''* (heavy line) were obtained from x-ray fluorescence from a single crystal of CBP; ideal *f''* values (thin line) for atomic Cu are from (58). Experimental values for *f'* are derived by numerical integration from the *f''* spectrum with the Kramers-Kroenig relation; ideal *f'* values (thin line) are from Hönl theory (59). Derivation of the experimental *f''* and *f'* values was performed with an in-house program DISCO (60).



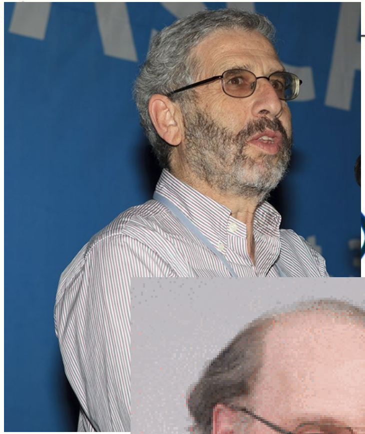
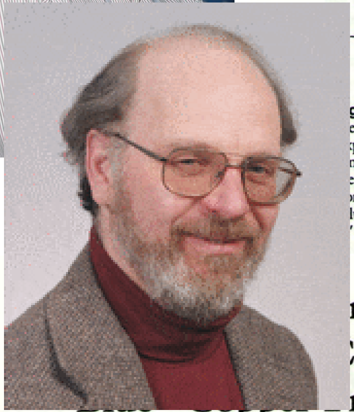
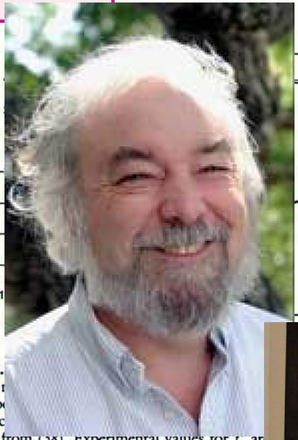
Phase Determination by Multiple-Wavelength X-ray Diffraction: Crystal Structure of a Basic "Blue" Copper Protein from Cucumbers

J. Mitchell Guss, Ethan A. Merritt,* R. Paul Phizackerley, Britt Hedman, Mitsuo Murata,† Keith O. Hodgson, and Hans C. Freeman

- Earliest success in high resolution "*de novo*" MAD phasing for structure solution using synchrotron radiation
- MAD data collection at SSRL BL1-5 with MWPC – 4 wavelengths – required 8 days
- Phasing of the 10-kD protein using native Cu atom gave excellent electron density map at ~2.5 Å resolution

Foundations of the SSRL SMB Program – Early Science – Macromolecular Crystallography on BL1-5

1985-87

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J. MITCHELL GUSS, ETHAN A. MERRITT,* R. PAUL PHIZACKERLEY
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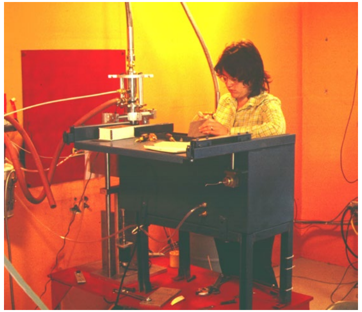
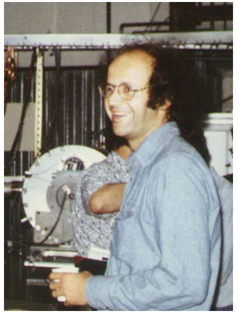
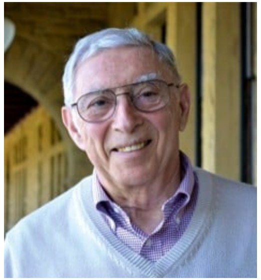
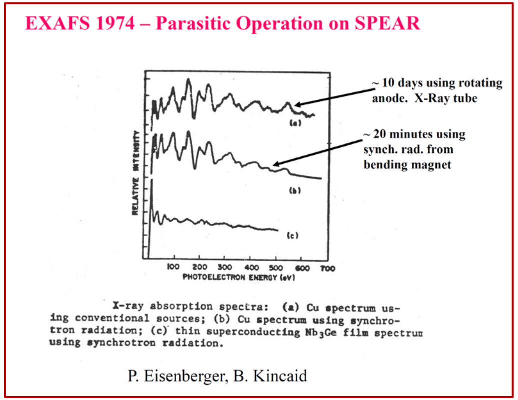
Foundations of the SSRL SMB Program – Early XAS and Science

- SSRL's BL1-5 designed in cooperation with Bell Labs and Seattle Groups for XAS studies
- Many key players, including Bienenstock and Hunter, Doniach, Ashley and Kincaid, Bell Labs group (Eisenberger and collaborators) and Sayers, Lytle and Stern
- Earliest bio applications at BL1-5 were on Fe in hemoglobin and rubredoxin







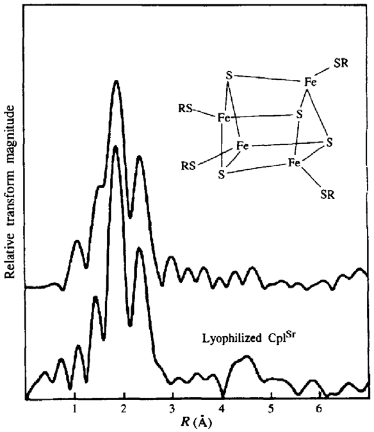


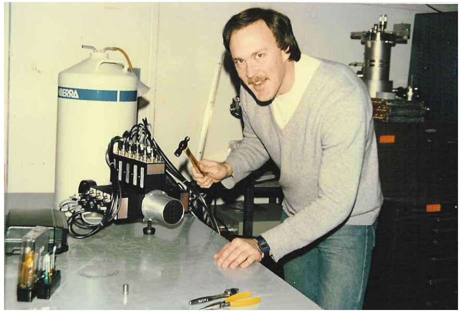
Foundations of the SSRL SMB Program – Early XAS Bioscience

"The promise of EXAFS as an important new tool to investigate the structure and function in biological and chemical systems was quickly realized" (Doniach, Hodgson, Lindau, Pianetta and Winick, *J. Synch. Rad*. (1997) 380-395)

Studies by students and postdocs in my group with collaborators in 1976-79 period included:

- First *de-novo* determination of metal cluster active site – nitrogenase
- Active site of cytochrome P-450 and details of key axial sulfur ligand to Fe
- Finding of unusually short Cu-S bond in the "blue copper protein" azurin

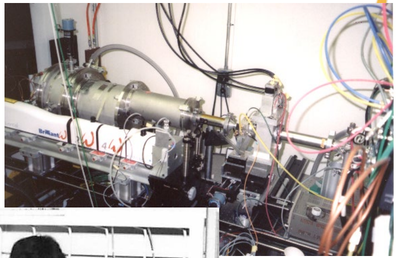




Steve Cramer, Tom Eccles, Tom Tullius and collaborators

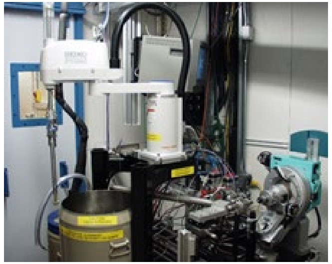
Foundations of the SSRL SMB Program – Early SAXS Science

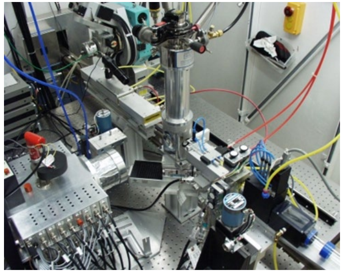
- Biological SAXS/diffraction started in 1974 at SSRL – BL1-4 – Caltech – John Baldeschwieler, Nick Webb – frog muscle
- Early 80ties, a SAXS camera was built and used part time on BL2-1 as element of SSRL SMB's NIH NCRR-funded resource
- Early focus on anomalous scattering (Doniach, Fairclough, Miake-Lye, Stroud, Hubbard)
- In 1991, Hiro Tsuruta arrived to lead building the dedicated SAXS program in its current "home" BL4-2

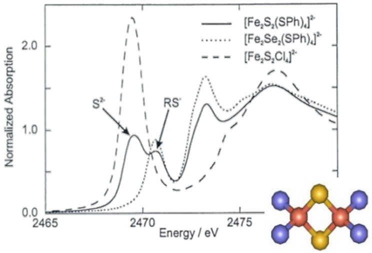


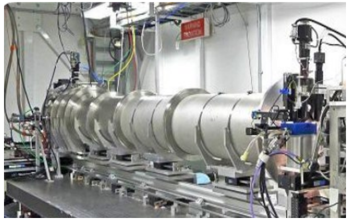
Selected Innovations from the SSRL SMB Program

- Macromolecular Crystallography - Robotics and Remote Access
  - Instruments fully automated with robotic sample handling at cryogenic temperatures (>1M samples handled)
  - Users perform experiments remotely (~95%)
- Dilute metalloprotein EXAFS with extension to tender energies
  - Stable beams, LHe sample environment, high-performing detector arrays for high-resolution structures
  - Ligand (S, Cl) as reporter for enzyme active sites
- Time-resolved solution x-ray scattering
  - Automation and robotics with millisec time resolution
  - Integrated microfluidics and sample purification









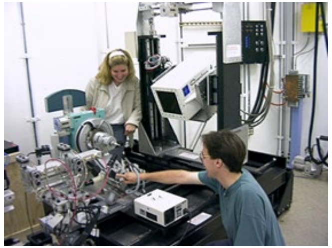
Selected Innovations from the SSRL SMB Program

- Macromolecular Crystallography - Robotics and

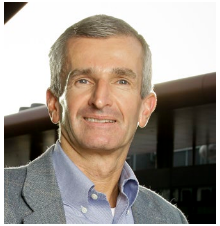
  - Automation and robotics with millisec speed
  - Integrated microfluidics and sample purification

Innovations from the SSRL SMB Program – SMB Beam Lines and Partnerships

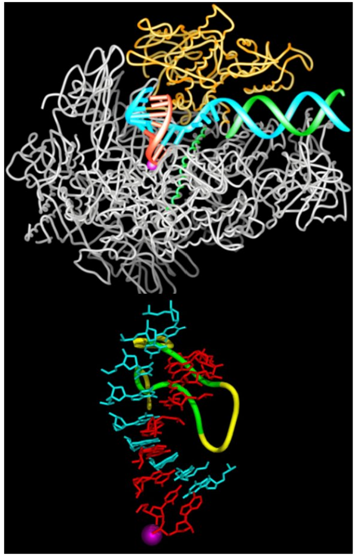
- Beam Line 9 (1994)
  - Funded by DOE-BER, 1st to take x-rays from SPEAR3
  - Dedicated stations for MC and bio-XAS
- Beam Line 12-2, a “Molecular Observatory” (2008)
  - Partnership with CalTech and funding from the Gordon and Betty Moore Foundation
  - SSRL’s first high brightness undulator beam line for MC
- Beam Line 12-1 (2020)
  - Partnership with Scripps Research Institute and Stanford, funding from NIGMS donors and foundations
  - 2nd high brightness mircofocus MC BL with most advanced x-ray area detector

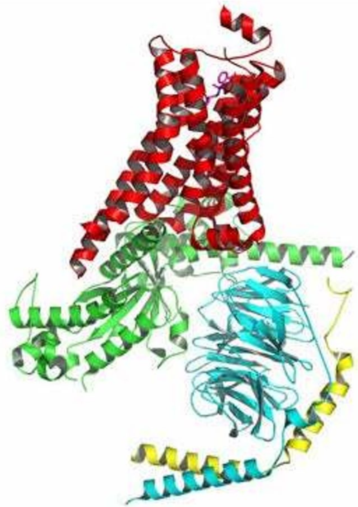


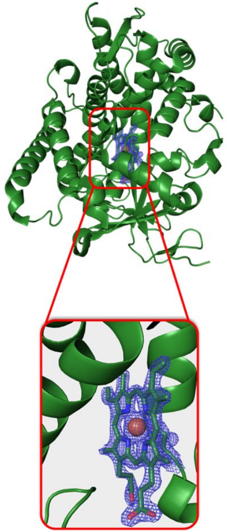
Scientific Discovery from the SSRL SMB Program – Nobel Prizes



Roger Kornberg – 2006 - RNA Polymerase II



Brian Kobilka and Robert Lefkowitz– 2012 – G-Coupled Protein Receptors



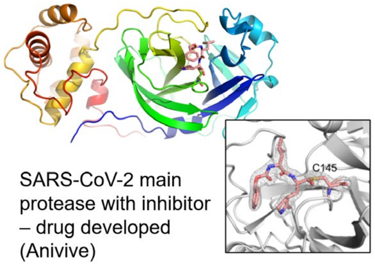
Frances Arnold – 2018 – Directed Evolution

- Synchrotron-based macromolecular crystallography has contributed directly to 7 Nobel Prizes
- Two of these made extensive use of data from SSRL beam lines, and 2 others more limited use

The SSRL SMB Program – A Few Reflections

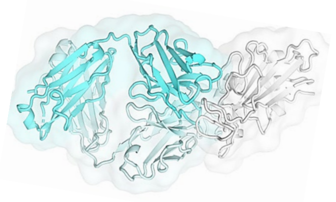
- SSRL SMB and its user community responded to the challenge during the COVID-19 pandemic to gain molecular insight into the SARS-CoV-2 virus, since 2020:
  - About 50 SARS-CoV-2 related publications
  - Over 1500 fragments/inhibitors screened
  - More than 160 structures deposited in the Protein Data Bank
  - Contributed to 4 drugs in clinical trials
  - Partner in the DOE NVBL

- Looking back over the SMB program’s 43 years:
  - More than 3500 beam time proposals
  - More than 5800 pubs
  - Contributed to more than 1000 Ph.D. and M.Sc. theses



SARS-CoV-2 main protease with inhibitor – drug developed (Anivive)

MJ Lemieux et al. Nat. Commun. (2020) doi: 10.1038/s41467-020-18096-2

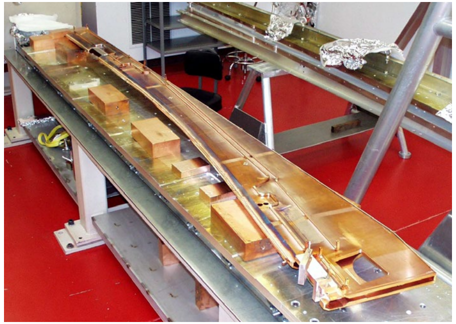


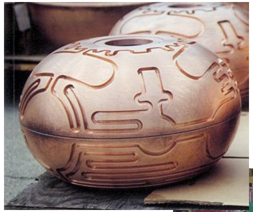
SARS-CoV-2 – antibody complex with surface antigen spike protein – detailing receptor-binding domain

IA Wilson *et al. Science* 2020 doi: 10.1126/science.abd2321

SPEAR3 - a 3rd Generation Light Source Dedicated to the SSRL Users from 2004 →

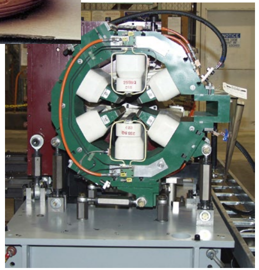


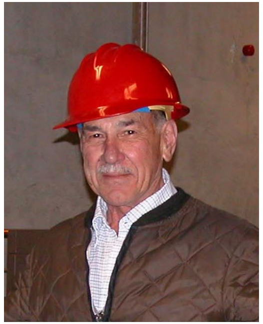







The SPEAR3 Era – Funding and the SPEAR3 Project



Tom Elioff,
Director

Bob Hettel,
Deputy Director

- Concept dates back to mid-1990ties, discussions with BES about a new SSRL light source, planning workshops, etc.
- Within an interagency working group framework, NIH proposed to support upgrades at SSRL and NSLS and an agreement was reached with DOE. News of project approval arrived on May 25, 1999
- Project was jointly funded ($58M), starting June 1, 1999, and completed (CD4) November 24, 2003 (ahead of schedule and within budget)
- Superb organization - management of more than 400 people at SLAC contributing to the project
- Built upon SLAC and SSRL core competencies – especially PEP-enabled RF and vacuum system
- Removal of SPEAR2 and installation of SPEAR3 completed in only 7 months. No reportable accidents over life of the project

SPEAR3 – Installation

SPEAR3 - Transition from 2nd to 3rd Generation

SPEAR3 Installation Time Line

- Begin: Mar 31, 2003
- Remove SPEAR2: Mar 31 – May 8
- B118 renovation: Mar 31 – Oct 6
- Pour concrete floor: May 8 – June 20
- Install new monuments: June 20 – July 25
- Install mounting plates & holes: July 1 – Aug 11
- Install girders & straights: Aug 11 – Aug 29
- Install cable plant: Aug 25 – Oct 1
- Install vacuum hardware: Aug 18 – Oct 16
- Leak-check, pump-down: Sept 8 – Oct 20
- Final survey: Oct 20 – Oct 30
- Lock ring-Installation complete: Oct 30

Richard Boyce
Project
Engineer

- Complete removal of all SPEAR2 components (~1 million lbs of magnets and equipment)
- Preparation of tunnel with new concrete foundation and new survey monuments
- Installation of all SPEAR3 systems and components (about 1.25 million lbs)
- Completed on schedule in an amazingly short ~7 months

SPEAR3 – Installation

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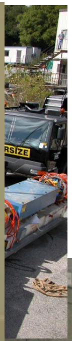

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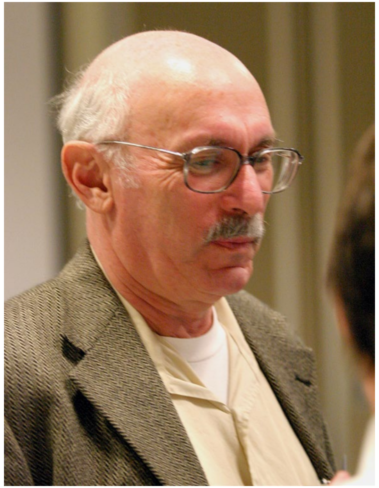
The SPEAR3 Era – Funding and the SPEAR3 Project



Bob Hettel,
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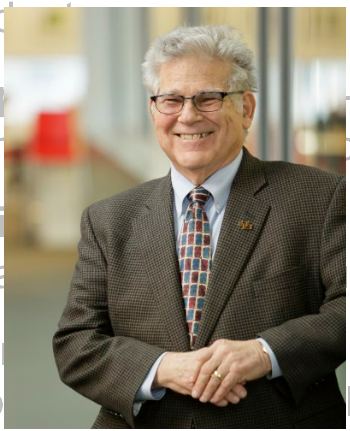

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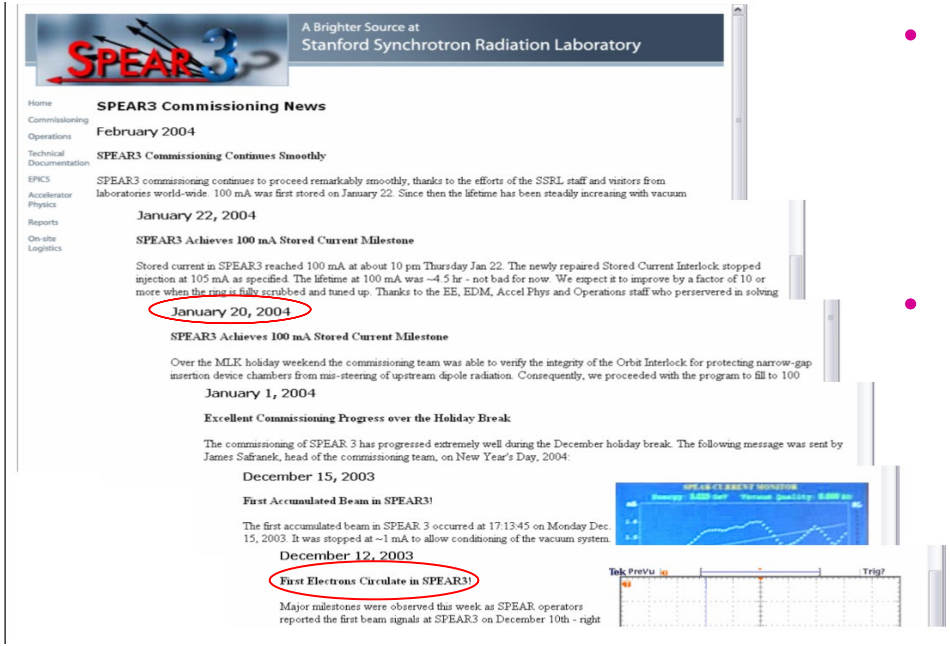
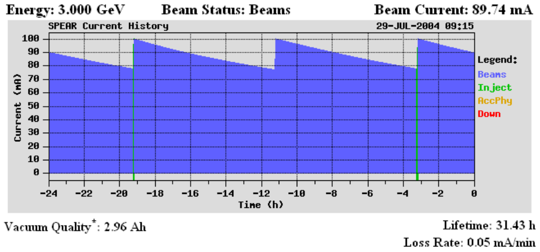
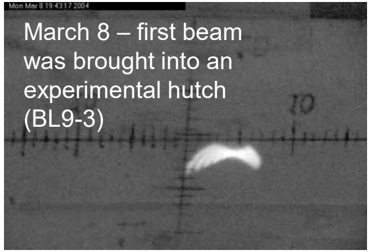
The SPEAR3 Era – Commissioning – A Spectacular Start!



- As of ***mid July, 2004*** – 10 of 11 beam lines had been certified by Radiation Physics and opened. On these beam lines, 20 experimental stations were operational/scheduled

- On ***July 31, 2004*** – first user run ended – integrated delivery since first users in mid-March was 97.1%





- On ***January 29, 2004*** – SLAC held a gala celebration to dedicate the new SPEAR3 accelerator

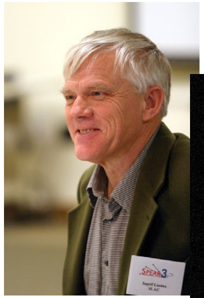
- On ***August 13, 2004*** – the SPEAR3 project team was recognized by a Secretary of Energy's Project Management Award

The SPEAR3 Dedication – January 29, 2004 – a New Era for SSRL

SPEAR3 provided SSRL and SLAC new opportunities for delivery of discovery science and capacity for growth and a very bright future for synchrotron science!

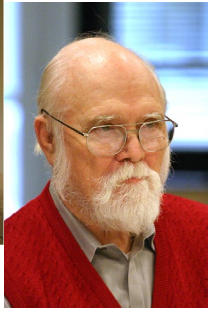




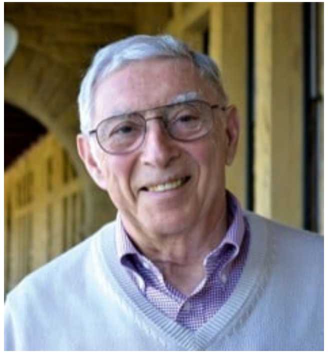


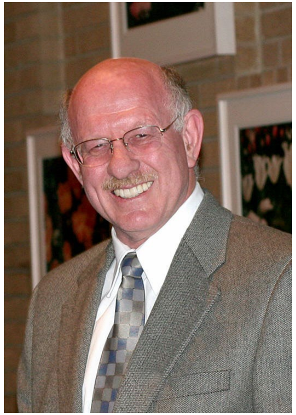




SPEAR3 - SLAC and Stanford Leadership – Vision and Commitment

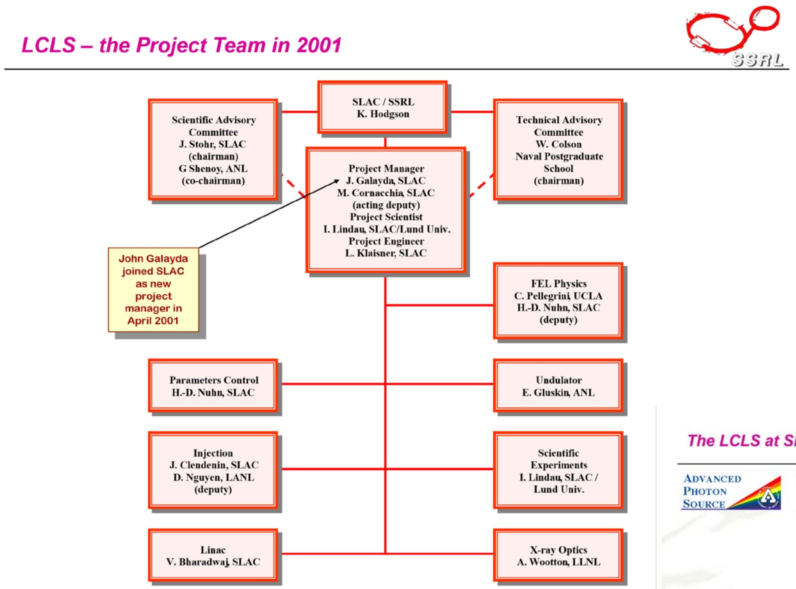
- Stanford
  - John Hennessy and John Etchemendy and Artie Bienenstock
- SLAC
  - Directors Burt Richter and Jonathan Dorfan

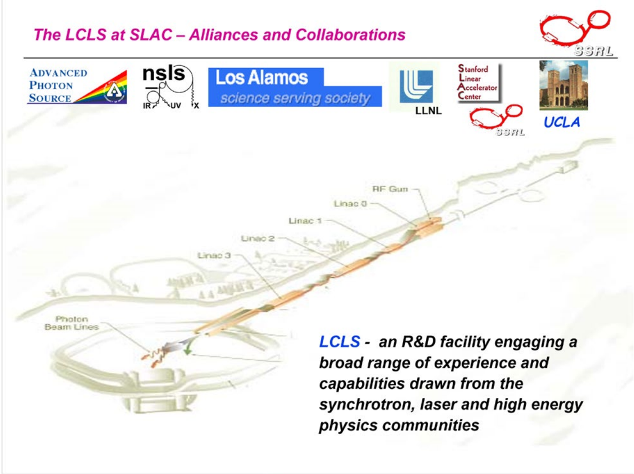


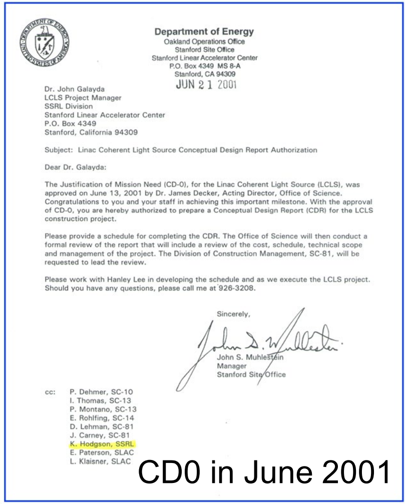


SSRL - Building the Case and Organization for the LCLS

- SSRL scientists and collaborators began to develop the case for an XFEL based on the SLAC Linac in the early-to-mid 1990ies (Artie’s talk)
- SSRL provided the organizational infrastructure within SLAC to move from a concept to a project – including interactions with BES/BESAC and overseeing an LCLC SAC







Department of Energy
Oakland Operations Office
Stanford Site Office
Stanford Linear Accelerator Center
P.O. Box 4349 MS 8-A
Stanford, CA 94309

JUN 21 2001

Dr. John Galayda
LCLS Project Manager
SSRL Division
Stanford Linear Accelerator Center
P.O. Box 4349
Stanford, California 94309

Subject: Linac Coherent Light Source Conceptual Design Report Authorization

Dear Dr. Galayda:

The Justification of Mission Need (CD-0), for the Linac Coherent Light Source (LCLS), was approved on June 13, 2001 by Dr. James Decker, Acting Director, Office of Science. Congratulations to you and your staff in achieving this important milestone. With the approval of CD-0, you are hereby authorized to prepare a Conceptual Design Report (CDR) for the LCLS construction project.

Please provide a schedule for completing the CDR. The Office of Science will then conduct a formal review of the report that will include a review of the cost, schedule, technical scope and management of the project. The Division of Construction Management, SC-81, will be requested to lead the review.

Please work with Hanley Lee in developing the schedule and as we execute the LCLS project. Should you have any questions, please call me at 926-3208.

Sincerely,

John S. Muhlestein
Manager
Stanford Site Office

cc: P. Dehmer, SC-10
I. Thomas, SC-13
P. Montano, SC-13
E. Rohlfing, SC-14
D. Lehman, SC-81
J. Carney, SC-81
K. Hodgson, SSRL
E. Paterson, SLAC
L. Klaisner, SLAC

CD0 in June 2001

Building the Foundation for the LCLS – The Scientific Case

Joachim Stöhr: First Experiments Document (Sept. 2000)

Members of the LCLS Scientific Advisory Committee (SAC)

Phil Bucksbaum	Univ. of Michigan
Roger Falcone	Univ. of California, Berkeley
Rick Freeman	Univ. of California, Davis
Andreas Freund	European Synchrotron Radiation Facility
Janos Hajdu	Uppsala Univ.
Jerry Hastings	National Synchrotron Light Source
Richard Lee	Lawrence Livermore National Laboratory
Ingolf Lindau	Stanford Synchrotron Radiation Laboratory
Gerd Materlik	HASYLAB
Simon Mochrie	Univ. of Chicago
Keith Nelson	Massachusetts Institute of Technology
Francesco Sette	European Synchrotron Radiation Facility
Sunil Sinha	Advanced Photon Source
Brian Stephenson	Argonne National Laboratory
Z.X. Shen	Stanford Univ.
Gopal Shenoy	**Advanced Photon Source (Co-Chairman)**
Joachim Stöhr	**Stanford Synchrotron Radiation Laboratory (Chairman)**

- Initiated by SSRL Director Keith Hodgson
- Developed by international team of ~ 50 scientists
- Approved by LCLS SAC

Team leaders and BESAC talks:

Femtochemistry	Dan Imre, BNL	*Plasma and Warm Dense Matter*	Richard Lee, LLNL
Nanoscale Dynamics in Condensed Matter	Brian Stephenson, APS	*Structural Studies on Single Particles and Biomolecules*	Janos Hajdu, Uppsala Univ.
Atomic Physics	Phil Bucksbaum, Univ. of Michigan	*X-ray Laser Physics*	Jerry Hastings, BNL

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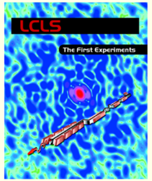
LCLS

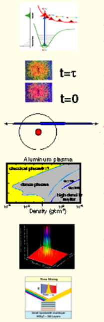
Stanford Linear Accelerator Center

Stanford Synchrotron Radiation Laboratory

LCLS Science Program – New Opportunities for Discovery



Program recommended by the LCLS SAC and being developed by SSRL/LCLS working with an international team of scientists working with accelerator and laser physics communities



***Femtochemistry and Biology** – watching motions of atoms and molecules*

***Nanostructured Materials** – structure dynamics and function at sub nm scales*

***Atomic Physics** – exploring how electrons move*

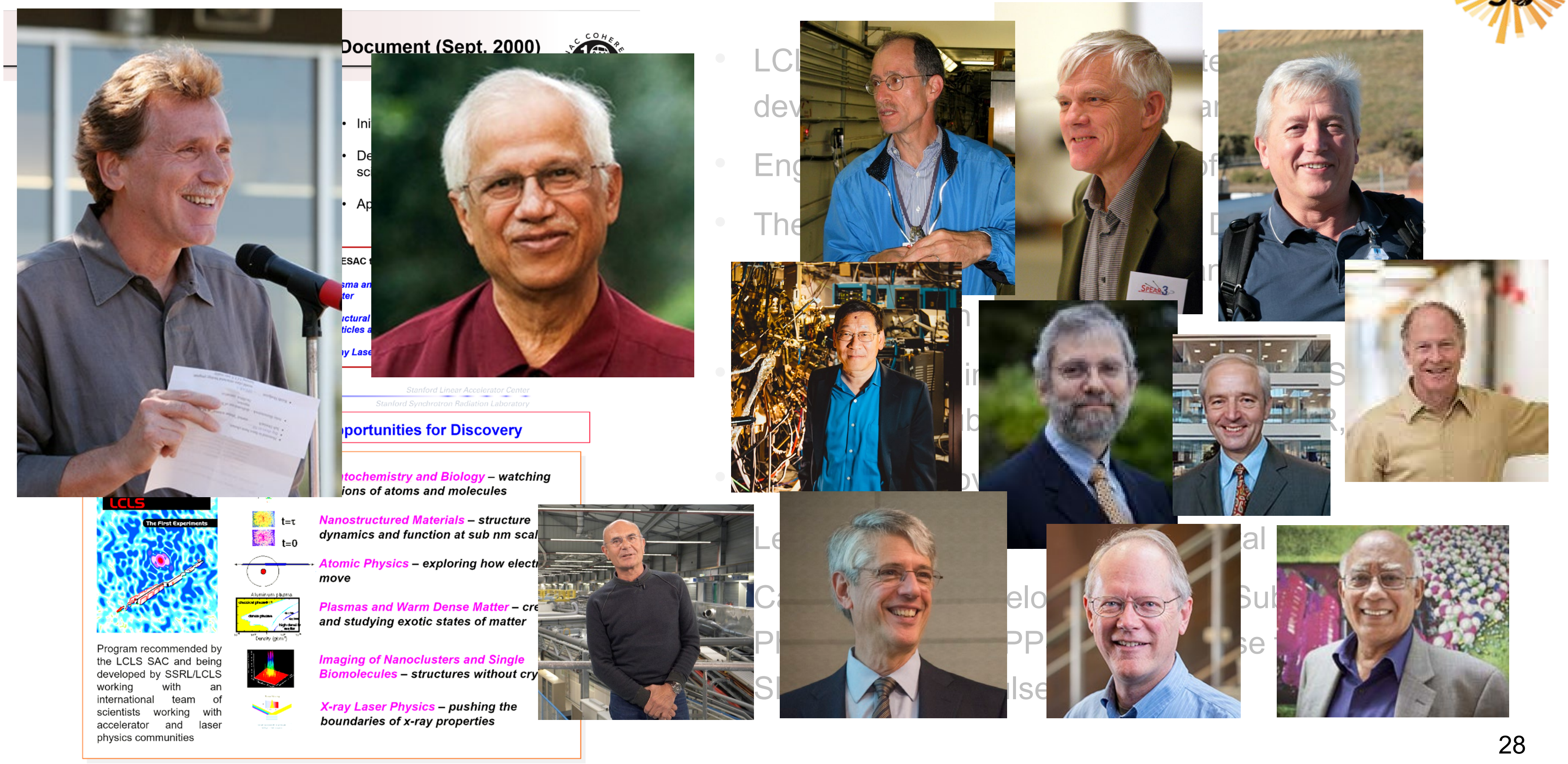
***Plasmas and Warm Dense Matter** – creating and studying exotic states of matter*

***Imaging of Nanoclusters and Single Biomolecules** – structures without crystals*

***X-ray Laser Physics** – pushing the boundaries of x-ray properties*

- LCLS Science Advisory Committee charged to develop the initial science program
- Engaged an international team of experts
- The resulting "First Experiments Document" was completed in September, 2000 and presented to DOE BESAC in October, 2000
- Received unanimous endorsement of BESAC to prepare and submit the formal LCLS CDR
- CD0 was approved in June, 2001
- Led to near and far hall experimental halls concept
- Catalyzed the development of the Sub-picosecond Photon Source (SPPS) and the case for the SLAC/Stanford PULSE Institute
- Also organized a Technical Advisory Committee (TAC)

Building the Foundation for the LCLS – The Scientific Case



And 50 Years! Thanks to the Vision, Commitment and Effort of so Many Dedicated SSRL Staff