Methoden moderner Röntgenphysik II: Streuung und Abbildung

Lecture 1	Vorlesung zum Haupt- oder Masterstudiengang Physik, SoSe 2015 G. Grübel, M. Martins, E. Weckert	
Location	Lecture hall AP, Physics, Jungiusstraße	
Date	Tuesday	12:45 - 14:15
	Thursday	8:30 - 10:00

Methoden moderner Röntgenphysik II: Streuung und Abbildung

Lecture:	4 SWS	Tuesday and Thursday
Tutorial/Übungen:	2 SWS	Tuesday (if agreed on)

Proseminar:	*For Bachelor students*
8 credits:	For Master students

Fixed dates:	Tuesday	12:45 - 14:15
	Thursday	8:30 - 10:00
Organization meeting“Tutorial”:	Tuesday, April 7	14:30 - 16:00
Location:	Seminar room 4	

DESY

Methoden moderner Röntgenphysik II: Streuung und Abbildung

Proseminar:	For Bachelor students
Regularly date:	Tuesdays at 16:30 - 18:00
Preliminary discussion:	April 7, 2015
	Seminar room 4
Starting date:	To be discussed
Location:	To be discussed

Methoden moderner Röntgenphysik II: Streuung und Abbildung

PROSEMINAR

A. Grundlagen und Methodik

1 Quellen und hochbrillante Röntgenstrahlung
2 Fokussierung von Röntgenstrahlung: Auf dem Weg zur 1-nm-Auflösung
3 Winkelaufgelöste Photoelektronenspektroskopie
4 Röntgenabsorption
5 Fluoreszenzspektroskopie
6 Magnetische Röntgenspektroskopie und Streuung
7 Diffraction Imaging und Röntgenholographie
8 Correlation Spectroscopy
9 Near Field Speckle Spectroscopy
10 Reaktionsmikroskop

B. Anwendungen von Röntgenstrahlung

1 Ultraschnelle dynamische Prozesse
2 Untersuchung von dünnen Schichten mittels Streuung
3 Metallische Oberflächen – Vom Atom zum Kontakt
4 Reflektivität und Streuung von Grenzflächen
5 Kolloidale Suspensionen
6 Metallische Gläser
7 Röntgenspektroskopie an freien Ionen

Methoden moderner Röntgenphysik II: Streuung und Abbildung

Lecturers: Gerhard Grübel (GG), Thomas Schneider (TS), Oliver Seek (OS), Stephan Roth (SR),

Part I:	**Basics of X-ray Physics**	(GG)
Part II:	**Soft Matter**	(SR)
Part III:	**Surfaces and Interfaces**	(OS)
Part IV:	**Macromolecular Crystallography**	(TS)
	Site Visit	

Methoden moderner Röntgenphysik II: Streuung und Abbildung

Part I:

Basics of X-ray Physics

by Gerhard Grübel (GG)

Methoden moderner Röntgenphysik II: Streuung und Abbildung

Part II:

Soft Matter

by Stephan Roth (SR)

[12.5.]

[19.5.]

[21.5.]

[2.6.]

[4.6.]

Methoden moderner Röntgenphysik II: Streuung und Abbildung

Part III:
Surfaces and Interfaces
by Oliver Seek (OS)

[9.6.] **Introduction**
- Concepts of surfaces
- Scattering (Born approximation)

[11.6.] **Crystal Truncation Rods**
- The basic idea
- How to calculate
- Examples

[16.6.] **Reflectivity**
- In Born approximation
- Exact formalism (Fresnel)
- Examples

[18.6.] **Grazing Incidence Diffraction**
- The basic idea
- Penetration depth
- Example

[23.6.] **Diffuse Scattering**
- Concepts of rough surfaces
- Correlation functions
- Scattering Born-approximation
- DWBA
- Examples

Methoden moderner Röntgenphysik II: Streuung und Abbildung

Part III:
Macromolecular Crystallography
by Thomas Schneider (TS)

[25.6.] **Structural Biology & MX**
- **Proteins**
- **Protein crystals**
- **Recombinant production of proteins**
- **Radiation damage and cryogenic cooling**
- **Bragg's law**

[30.6.] **Collection and processing of Diffraction Data**
- **Real and reciprocal space**
- **The crystallographic phase problem**
- **Symmetry in real and reciprocal space**
  (+ Demonstration Practical)

[2.7.] **Phasing an Model refinement**
- **Phasing methods**
- **Ab initio**
- **Molecular Replacement – 25%. Model bias**
- **Multiple Isomorphous Replacement**
- **Multiple Anomalous Diffraction**
- **Refinement**

[7.7.] **Site Visit**

Literature

Basic concepts:

Elements of Modern X-Ray Physics
J. A. Nielsen and D. McMorrow, J. Wiley&Sons (2001)

X-Ray Diffraction
B.E. Warren, DOVER Publications Inc., New York

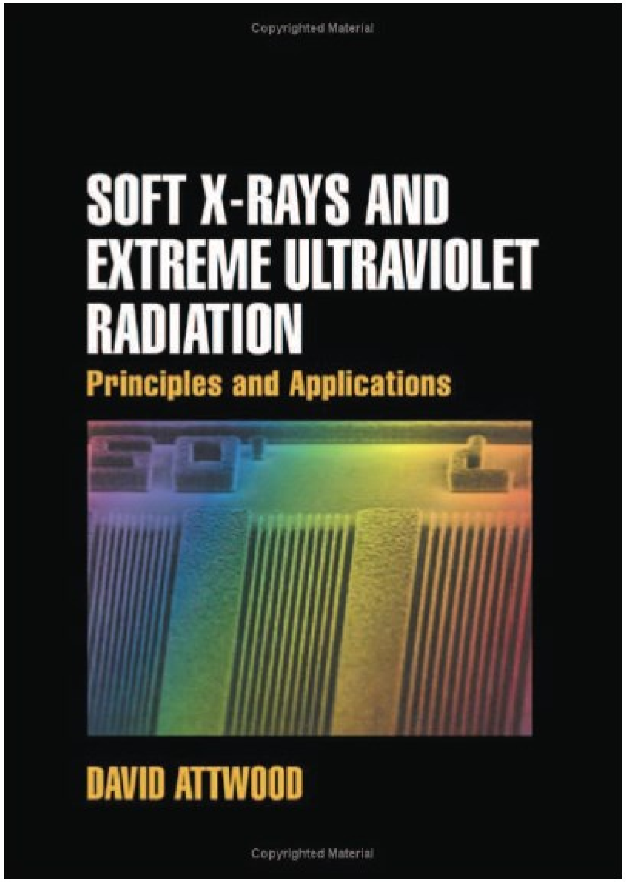
Principles of Optics
M. Born and E. Wolf, Cambridge University Press, 7th ed.

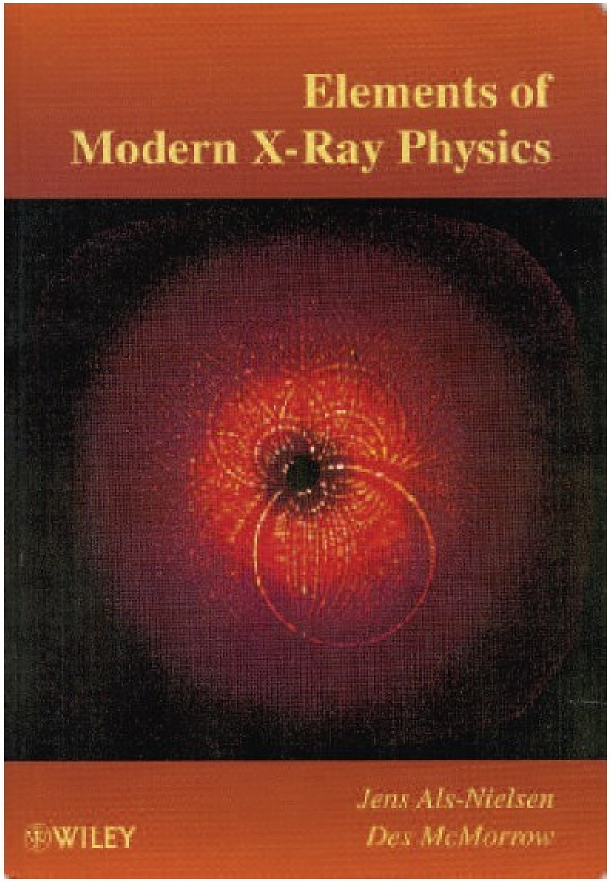
Soft X-rays and Extreme Ultraviolet Radiation
D. Attwood, Cambridge University Press (2000)
http://www.coe.berkeley.edu/AST/sxreuv/

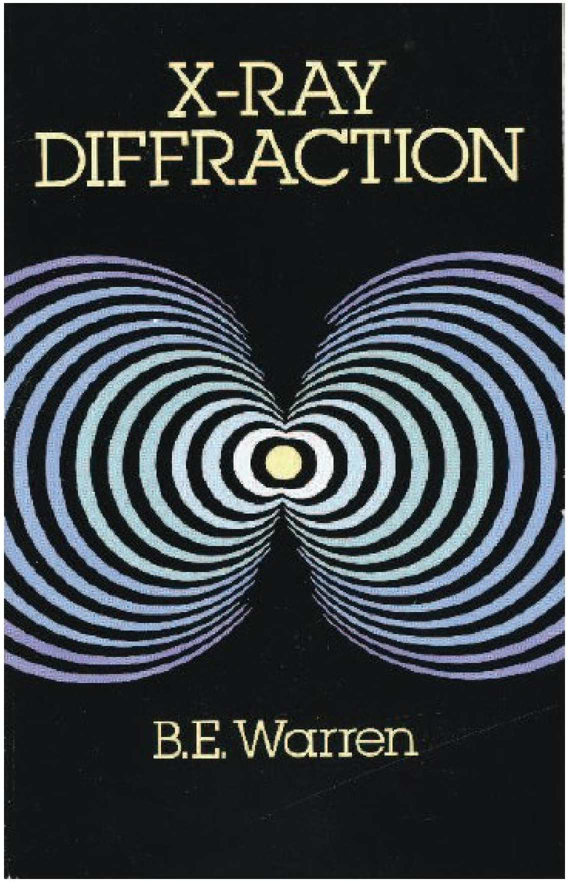
Physik der Teilchenbeschleuniger und Synchrotronstrahlungsquellen
K. Wille, Teubner Studienbücher 1996

Lecture Notes

http://photonscience.desy.de/research/studentsteaching/lectures_seminars/ss15/roentgenphysik_streuung_und_abbildung/index_eng.html







*** some of the slides are courtesy of M. Tolan, C. Gutt and A. Hermmerich**

Methoden moderner Röntgenphysik II: Streuung und Abbildung

Part I:

Basics of X-ray Physics

by Gerhard Grübel (GG)

Introduction
Overview, Introduction to X-ray Scattering

X-ray Scattering Primer
Elements of X-ray Scattering

Sources of X-rays, Synchrotron Radiation
Laboratory Sources, Accelerator Bases Sources

Reflection and Refraction from Interfaces
Snell's Law, Fresnel Equations

Kinematical Diffraction (I)
Diffraction from an Atom, a Molecule, from Liquids, Glasses, …

Kinematical Diffraction (II)
Diffraction from a Crystal, Reciprocal Lattice, Structure Factor, …

Methoden moderner Röntgenphysik II: Streuung und Abbildung

Small Angle Scattering, and Soft Matter
Introduction, Form Factor, Structure Factor, Applications, ...

Anomalous Diffraction
Introduction into Anomalous Scattering, ...

Introduction into Coherence
Concept, First Order Coherence, ...

Coherent Scattering
Spatial Coherence, Second Order Coherence, ...

Applications of Coherent Scattering
Imaging and Correlation Spectroscopy, ...

Methoden moderner Röntgenphysik II: Streuung und Abbildung

Part II:

Soft Matter

by Stephan Roth (SR)

What is Soft Matter
What are the Properties and Which Properties can be Investigated by X-rays: Example Bulk Polymer and Diffuse Scattering

Soft Matter and Biological Multilayer Membranes
Investigation by X-ray Reflectivity (Born Approximation)

Soft Matter and Biological Multilayer Membranes
Investigation by X-ray Reflectivity (Parratt-, Abeles-Formalism), Examples

Structures Surfaces/Gratings and Molecular In-plane Ordering,
Off-specular Diffraction

Capillary Waves
What are Capillary Waves, Correlation Functions, Power Spectral Density

Capillary Waves, Diffuse X-ray Scattering

Methoden moderner Röntgenphysik II: Streuung und Abbildung

Part III:
Surfaces and Interfaces
by Oliver Seek (OS)

What is Soft Matter
What are the Properties and Which Properties can be Investigated by X-rays: Example Bulk Polymer and Diffuse Scattering

Soft Matter and Biological Multilayer Membranes
Investigation by X-ray Reflectivity (Born Approximation)

Soft Matter and Biological Multilayer Membranes
Investigation by X-ray Reflectivity (Parratt-, Abeles-Formalism), Examples

Structures Surfaces/Gratings and Molecular In-plane Ordering,
Off-specular Diffraction

Capillary Waves
What are Capillary Waves, Correlation Functions, Power Spectral Density

Capillary Waves, Diffuse X-ray Scattering

Methoden moderner Röntgenphysik II: Streuung und Abbildung

Part IV:

Macromolecular Crystallography

by Thomas Schneider (TS)

Methoden moderner Röntgenphysik II: Streuung und Abbildung

MX – The Method

Bragg's law
Structure Factors, Argand Diagrams
Real and Reciprocal Space
Phase Problem *What is the 'Phase Problem'
Symmetry in Real and Reciprocal Space
Anomalous Diffraction
Overview of Phasing Methods

MX – Collection and Processing of Diffraction Data

Crystals and Their Properties/Problems
Synchrotron Radiation Beams for MX
Diffractometry for MX
Detectors for MX
2D Diffraction Images. Ewald Construction?
Data Processing
- Integration
- Scaling
- Anomalous Data
- Data Quality Indicators
- Resolution
- Radiation Damage

Methoden moderner Röntgenphysik II: Streuung und Abbildung

MX – Building Models & Future

Experimental Phasing: MAD
Experimental Phasing: Derivatives
Knowledge-based Phasing: Molecular Replacement
Model bias
Model Building
Refinement
X-FEL crystallography

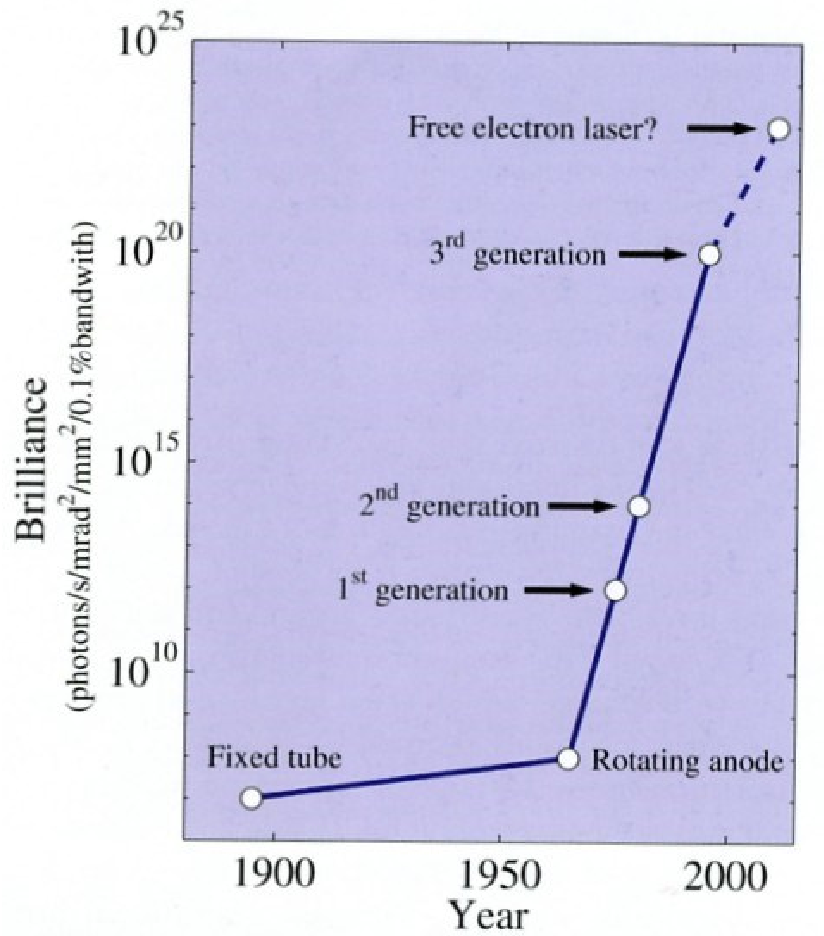
DEMO 1: Crystallographic Data Processing
DEMO 2: Crystallographic Structure Determination

Literature:

Als-Nielsen
D. Blow: Outline of Crystallography for Biologists. Oxford University press. ISBN: 978-0198510512. € 39.00 (amazon)
G. Rodes: Crystallography made crystal clear. Academic press. ISBN: 978-0-12-587073-3. € 40.95 (amazon)
http://www-structmed.cimr.cam.ac.uk/course.html
G. Taylor (2003): 'The phase problem', Acta Cryst. D59:1881.

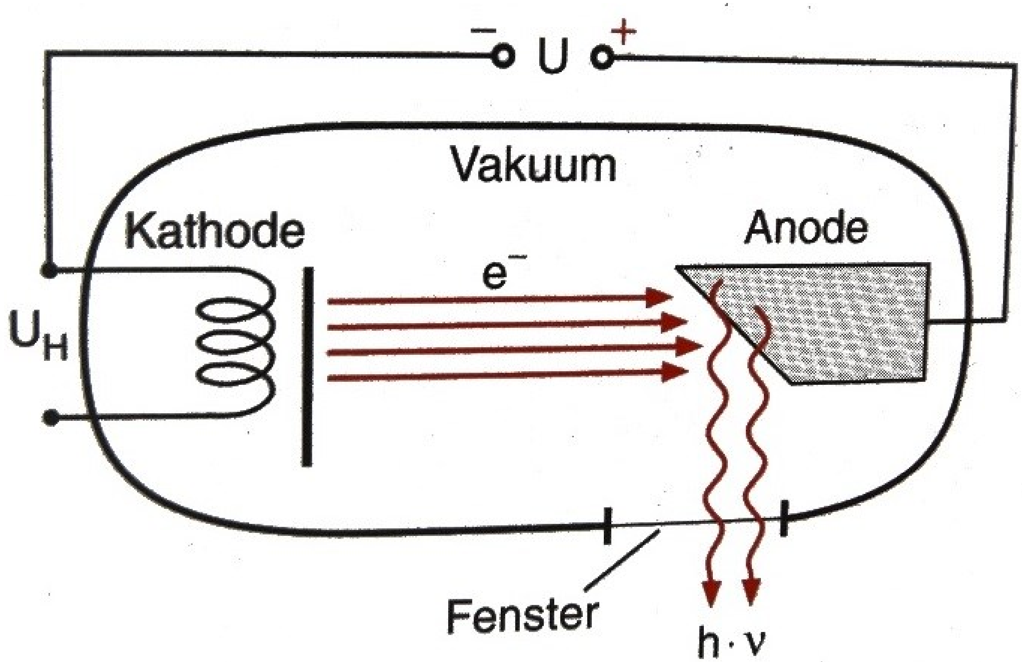
Introduction by Gerhard Grübel

1895	X-ray discovered by W.C. Röntgen
1901	Nobel Price; since then, unprecedented success in unraveling the structure of materials
1970	Synchrotron radiation revolutionizes the field
2005	Start operation FLASH (first SASE based FEL)
2009	Free Electron Lasers (XFEL)

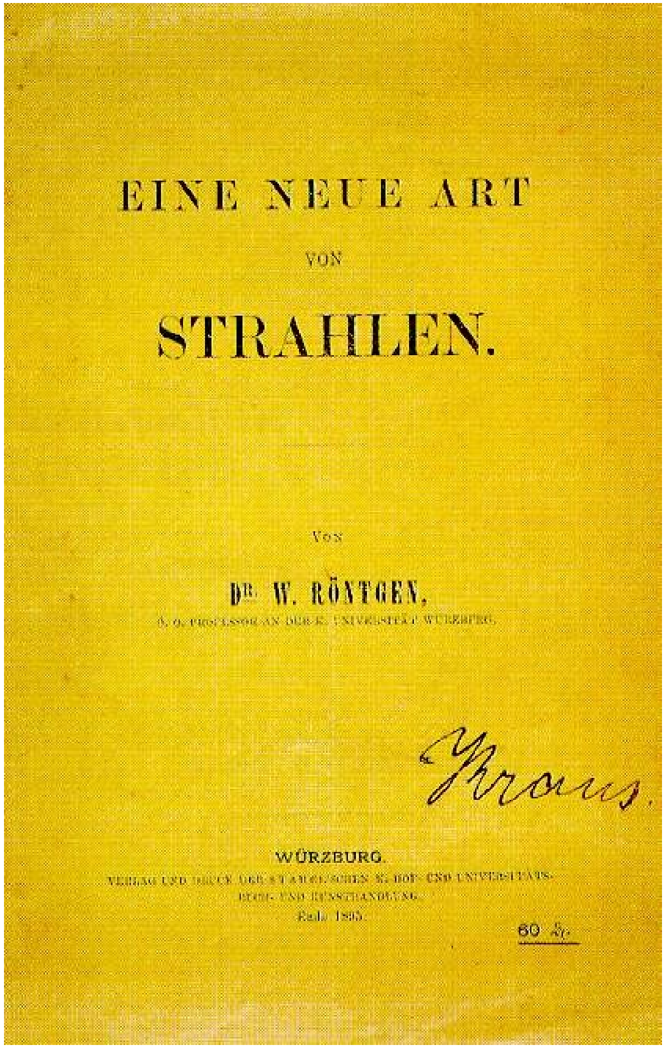


Würzburg, 8. November 1895

Wilhelm C. Röntgen (1845 - 1923)



Klassische Röntgenröhre



EINE NEUE ART

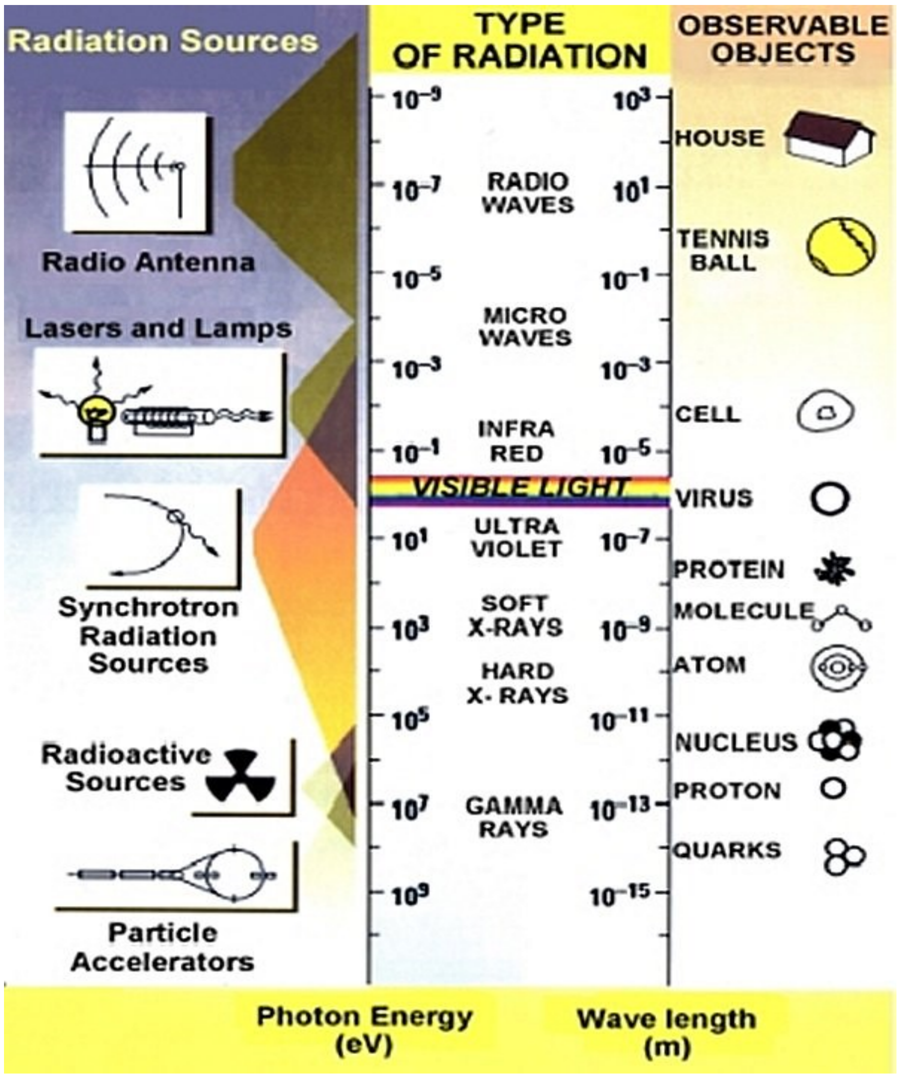
VON

STRAHLEN.

VON

DR. W. RÖNTGEN,

WÜRZBURG.



X-rays

≡

Electromagnetic Radiation

≡

Wavelength

$$(\lambda[\text{Å}] = \frac{12.398}{E\ [keV]})$$

≡

Object Size

≡

Angstroms for Condensed Matter Research

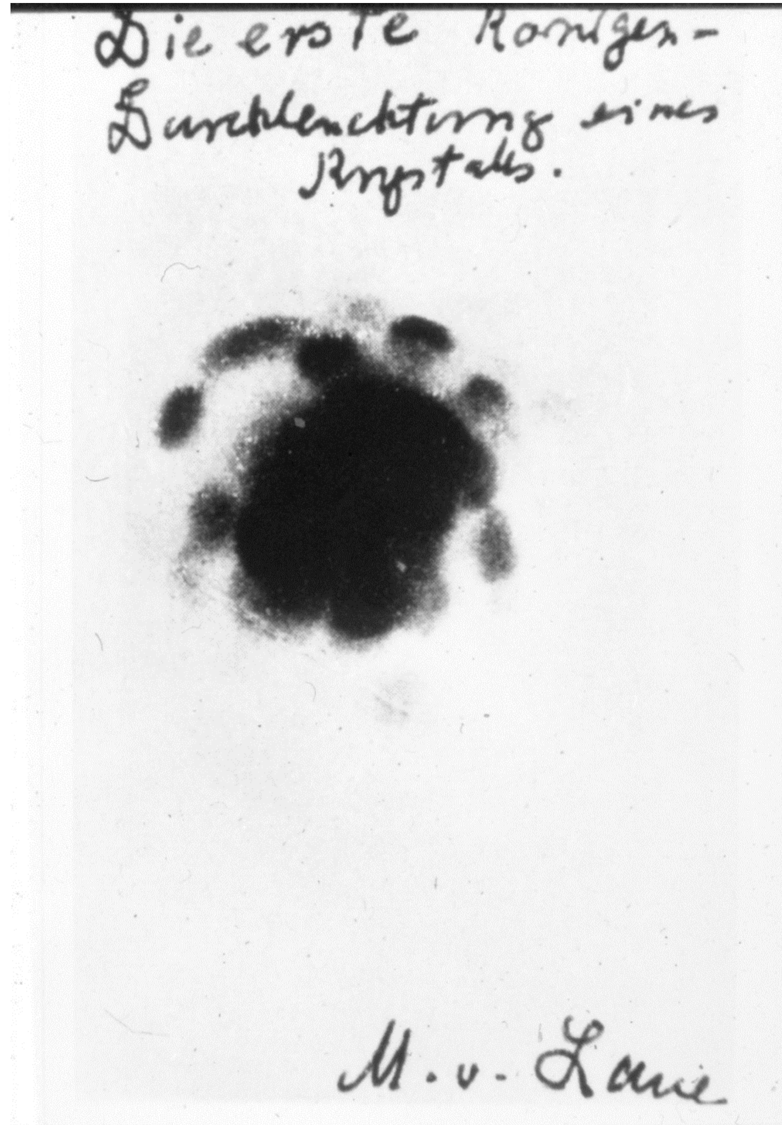
Nobel Prices

1901 W.C. Röntgen in **Physik** für die **Entdeckung der Röntgenstrahlen**
1914 M. von Laue in **Physik** für **Röntgenbeugung an Kristallen**
1915 W.H. Bragg und W.L. Bragg in **Physik** für Bestimmung der **Kristallstruktur mit Röntgenbeugung**
1917 C.G. Barkla in **Physik** für die **charakteristische Strahlung der Elemente**
1924 K.M.G. Siegbahn in **Physik** für **Röntgenspektroskopie**
1927 A.H. Compton in **Physik** für **Streuung von Röntgenstrahlen durch Elektronen**
1936 P. Debye in **Chemie** für **Beugung von Röntgenstrahlen und Elektronen in Gasen**
1946 H.J. Muller in **Medizin** für die Entdeckung von **Mutationen durch Röntgenstrahlung**
1954 L. Pauling in **Chemie** für Entwicklungen in der **Strukturchemie**
1956 A.F. Cournand, W. Forssmann und D.W. Richards in **Medizin** für die **Entwicklung des Herzkatheters unter Röntgenkontrolle**
1962 J. Watson, M. Wilkins und F. Crick in **Medizin** für die **Strukturaufklärung des DNA-Moleküls**
1962 M. Perutz und J. Kendrew in **Chemie** für die **Strukturaufklärung von Hämoglobin**
1964 D.C. Hodgkin in **Chemie** für die **Röntgenstrukturanalyse von Penicillin** und wichtigen biochemischen Substanzen
1976 W.N. Lipscomb in **Chemie** für **Röntgenstrukturuntersuchungen an Boranen**
1979 A.M. Cormack und G.N. Hounsfield in **Medizin** für **Computertomographie**
1981 K.M. Siegbahn in **Physik** für **hochaufgelöste Elektronenspektroskopie**
1985 H.A. Hauptman und J. Karle in **Chemie** für die Entwicklung direkter Methoden zur **Bestimmung von Röntgenstrukturen**
1988 J. Deisenhofer, R. Huber und H. Michel in **Chemie** für die **Bestimmung der dreidimensionalen Struktur von Proteinen für die Photosynthese**
1997 P.D. Boyer, J.E. Walker und J.C. Skou in **Chemie** für **Aufklärung der Funktion des Enzyms ATP**
2002 R. Giacconi in **Physik** für die **Entwicklung der Röntgenastronomie**
2003 R. MacKinnon in **Chemie** für **Röntgenstrukturbestimmung von Ionenkanälen in Zellmembranen**
2009 V. Ramakrishnan, T. A. Steitz, A. E. Yonath in **Chemie** für **Studies of the Structure and Function of the Ribosome**

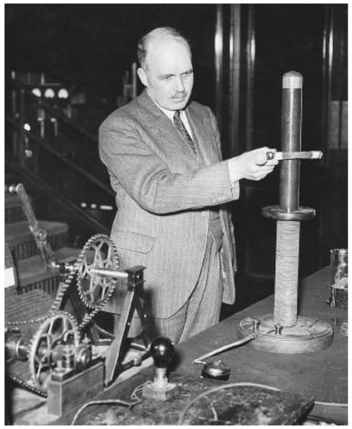
v. Laue et. al.

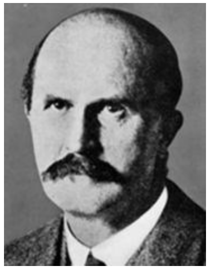
Interferenzen am Kristallgitter

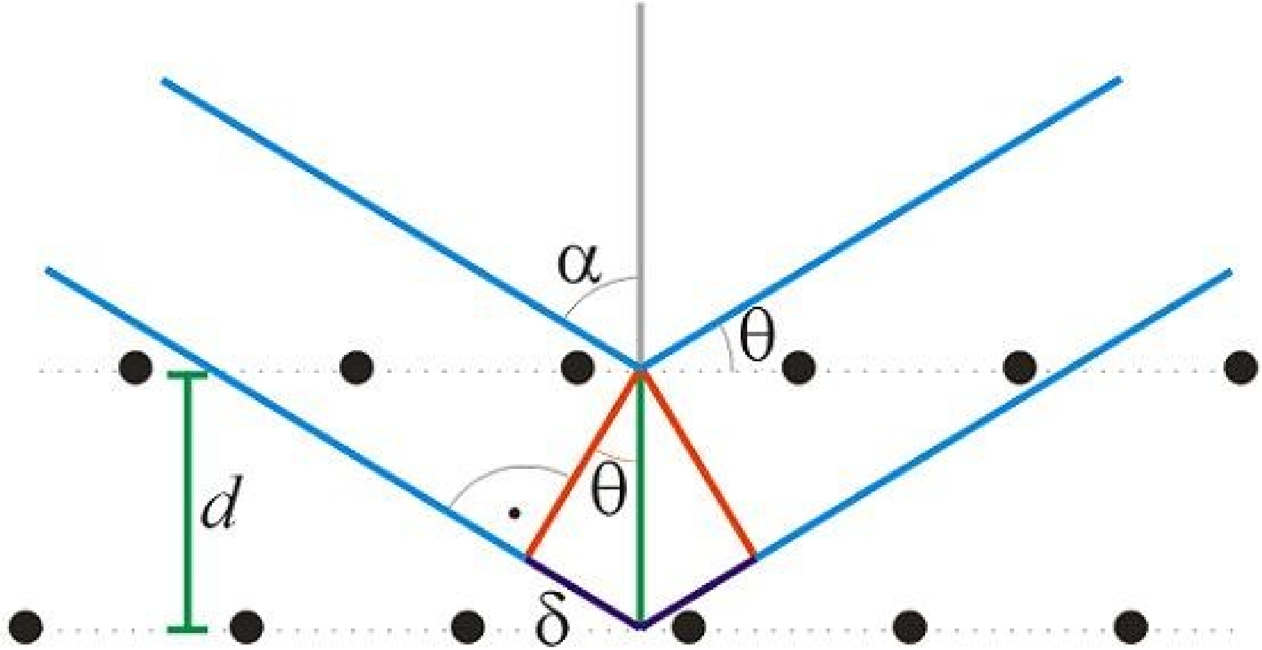
Laue, Friedrich und Knipping 1912



Bragg's Law

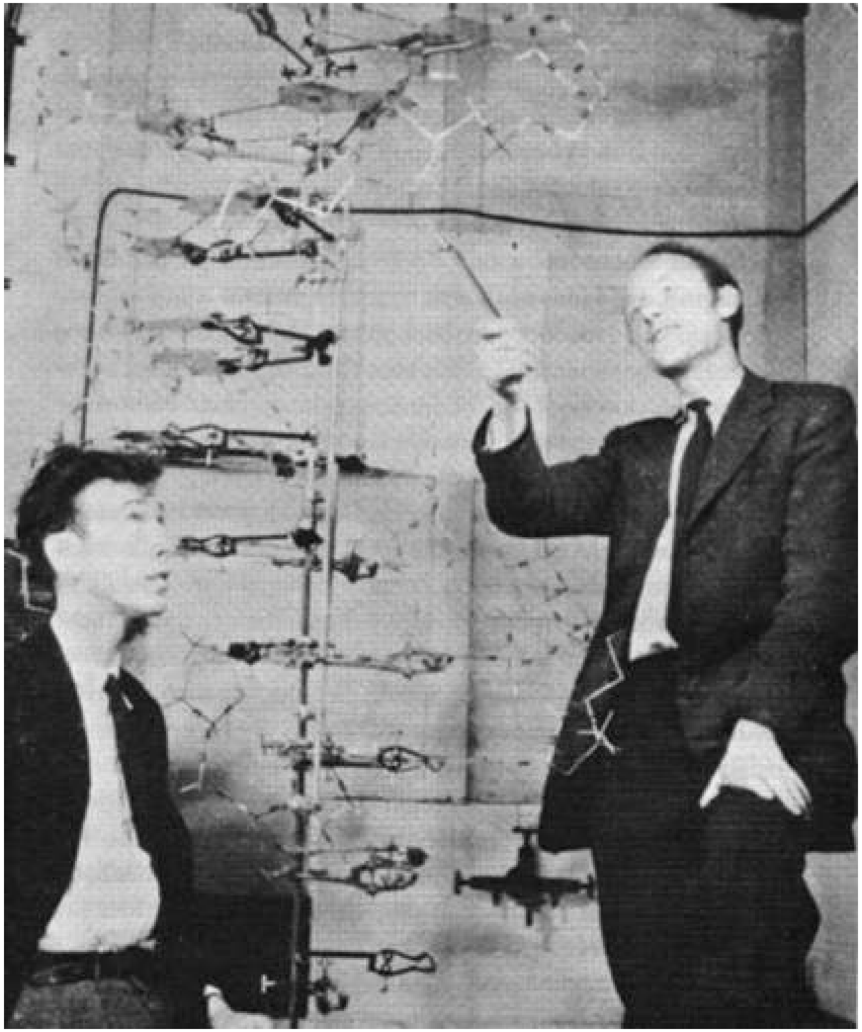






Scattering intensity only if: $n\lambda = 2d\sin(\Theta)$

Watson & Crick 1953



A Structure for DNA

James D. Watson and
Francis H.C. Crick (1953)

It has not escaped our notice that the specific pairing we have postulated immediately suggests a possible copying mechanism for the genetic material.
Full details of the structure, including the conditions assumed in building it, together with a set of co-ordinates for the atoms, will be published elsewhere.
We are much indebted to Dr. Jerry Donohue for constant advice and criticism, especially on inter-atomic distances. We have also been stimulated by a knowledge of the general nature of the unpublished experimental results and ideas of Dr. M. H. F. Wilkins, Dr. R. E. Franklin and their co-workers at

James D. Watson and Francis H. C. Crick
"Molecular Structure of Nucleic Acids: A Structure for Deoxyribose Nucleic Acid" (1953)

No. 4356 April 25, 1953 NATURE 737

equipment, and to Dr. G. E. R. Deacon and the captain and officers of R.R.S. *Discovery II* for their part in making the observations.

[1] Young, F. B., Gerrard, H., and Jevons, W., *Phil. Mag.*, **40**, 149 (1920).
[2] Longuet-Higgins, M. S., *Mon. Not. Roy. Astro. Soc., Geophys. Supp.*, **5**, 285 (1949).
[3] Von Arx, W. S., Woods Hole Papers in Phys. Oceanog. Meteor., **11** (3) (1950).
[4] Ekman, V. W., *Arkiv. Mat. Astron. Fysik. (Stockholm)*, **2** (11) (1905).

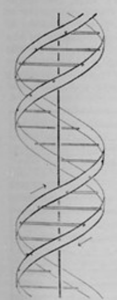
MOLECULAR STRUCTURE OF NUCLEIC ACIDS

A Structure for Deoxyribose Nucleic Acid

WE wish to suggest a structure for the salt of deoxyribose nucleic acid (D.N.A.). This structure has novel features which are of considerable biological interest.

A structure for nucleic acid has already been proposed by Pauling and Corey[1]. They kindly made their manuscript available to us in advance of publication. Their model consists of three inter-twined chains, with the phosphates near the fibre axis, and the bases on the outside. In our opinion, this structure is unsatisfactory for two reasons: (1) We believe that the material which gives the X-ray diagrams is the salt, not the free acid. Without the acidic hydrogen atoms it is not clear what forces would hold the structure together, especially as the negatively charged phosphates near the axis will repel each other. (2) Some of the van der Waals distances appear to be too small.

Another three-chain structure has also been suggested by Fraser (in the press). In his model the phosphates are on the outside and the bases on the inside, linked together by hydrogen bonds. This structure as described is rather ill-defined, and for this reason we shall not comment on it.



This figure is purely diagrammatic. The two ribbons symbolize the two phosphate—sugar chains, and the horizontal rods the pairs of bases holding the chains together. The vertical line marks the fibre axis

We wish to put forward a radically different structure for the salt of deoxyribose nucleic acid. This structure has two helical chains each coiled round the same axis (see diagram). We have made the usual chemical assumptions, namely, that each chain consists of phosphate di-ester groups joining β-D-deoxy-ribofuranose residues with 3′,5′ linkages. The two chains (but not their bases) are related by a dyad perpendicular to the fibre axis. Both chains follow right-handed helices, but owing to the dyad the sequences of the atoms in the two chains run in opposite directions. Each chain loosely resembles Furberg's[2] model No. 1; that is, the bases are on the inside of the helix and the phosphates on the outside. The configuration of the sugar and the atoms near it is close to Furberg's 'standard configuration', the sugar being roughly perpendicular to the attached base. There is a residue on each chain every 3·4 A. in the z-direction. We have assumed an angle of 36° between adjacent residues in the same chain, so that the structure repeats after 10 residues on each chain, that is, after 34 A. The distance of a phosphorus atom from the fibre axis is 10 A. As the phosphates are on the outside, cations have easy access to them.

The structure is an open one, and its water content is rather high. At lower water contents we would expect the bases to tilt so that the structure could become more compact.

The novel feature of the structure is the manner in which the two chains are held together by the purine and pyrimidine bases. The planes of the bases are perpendicular to the fibre axis. They are joined together in pairs, a single base from one chain being hydrogen-bonded to a single base from the other chain, so that the two lie side by side with identical z-co-ordinates. One of the pair must be a purine and the other a pyrimidine for bonding to occur. The hydrogen bonds are made as follows: purine position 1 to pyrimidine position 1; purine position 6 to pyrimidine position 6.

If it is assumed that the bases only occur in the structure in the most plausible tautomeric forms (that is, with the keto rather than the enol configurations) it is found that only specific pairs of bases can bond together. These pairs are: adenine (purine) with thymine (pyrimidine), and guanine (purine) with cytosine (pyrimidine).

In other words, if an adenine forms one member of a pair, on either chain, then on these assumptions the other member must be thymine; similarly for guanine and cytosine. The sequence of bases on a single chain does not appear to be restricted in any way. However, if only specific pairs of bases can be formed, it follows that if the sequence of bases on one chain is given, then the sequence on the other chain is automatically determined.

It has been found experimentally[3,4] that the ratio of the amounts of adenine to thymine, and the ratio of guanine to cytosine, are always very close to unity for deoxyribose nucleic acid.

It is probably impossible to build this structure with a ribose sugar in place of the deoxyribose, as the extra oxygen atom would make too close a van der Waals contact.

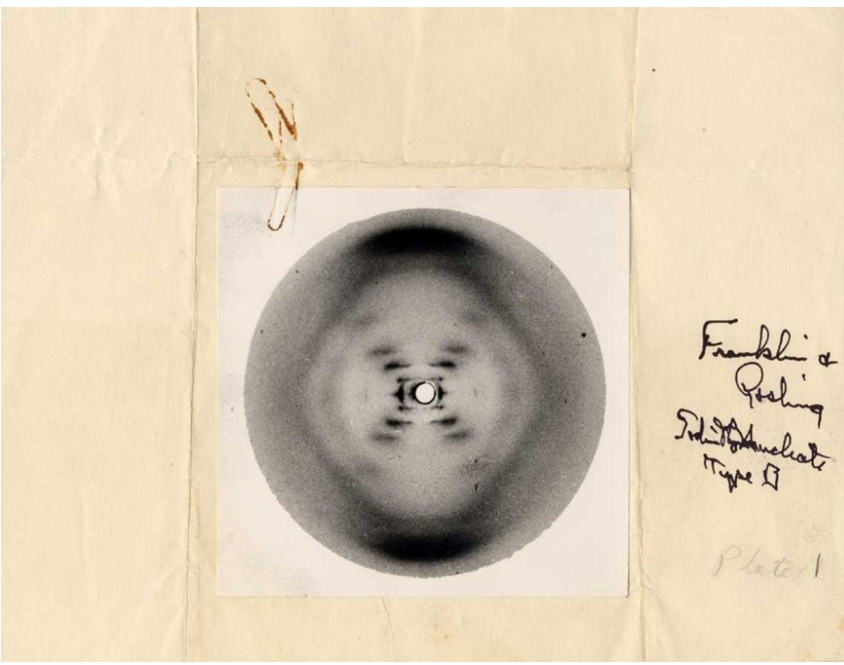
The previously published X-ray data[5,6] on deoxyribose nucleic acid are insufficient for a rigorous test of our structure. So far as we can tell, it is roughly compatible with the experimental data, but it must be regarded as unproved until it has been checked against more exact results. Some of these are given in the following communications. We were not aware of the details of the results presented there when we devised our structure, which rests mainly though not entirely on published experimental data and stereo-chemical arguments.

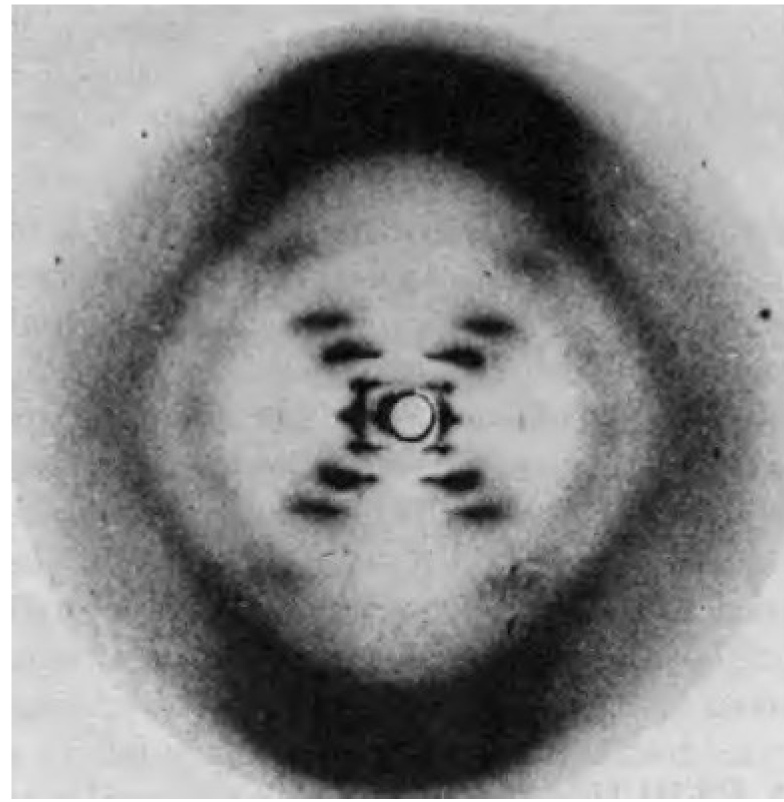
It has not escaped our notice that the specific pairing we have postulated immediately suggests a possible copying mechanism for the genetic material.

Full details of the structure, including the conditions assumed in building it, together with a set of co-ordinates for the atoms, will be published elsewhere.

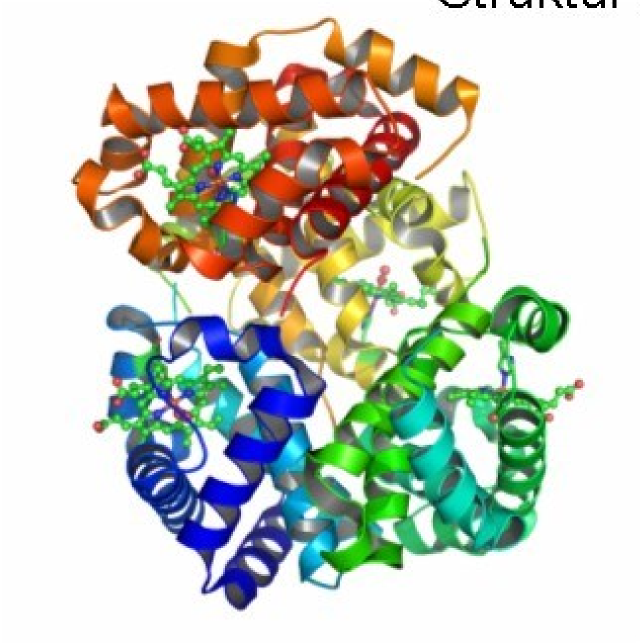
We are much indebted to Dr. Jerry Donohue for constant advice and criticism, especially on inter-atomic distances. We have also been stimulated by a knowledge of the general nature of the unpublished experimental results and ideas of Dr. M. H. F. Wilkins, Dr. R. E. Franklin and their co-workers at

Rosalind Franklin's Famous X-ray that provided enough evidence to establish that DNA was a helix.

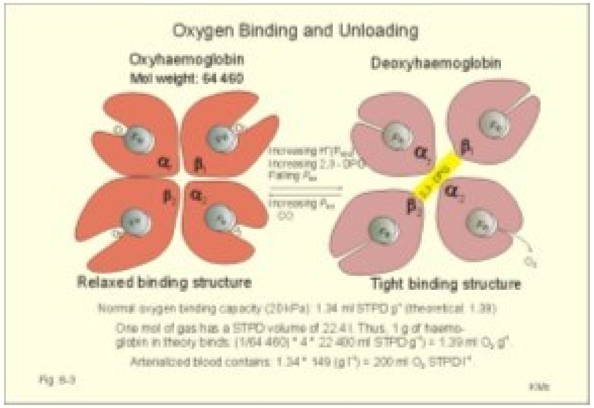




Hämoglobin und Myoglobin
Struktur mit Röntgenstrahlen

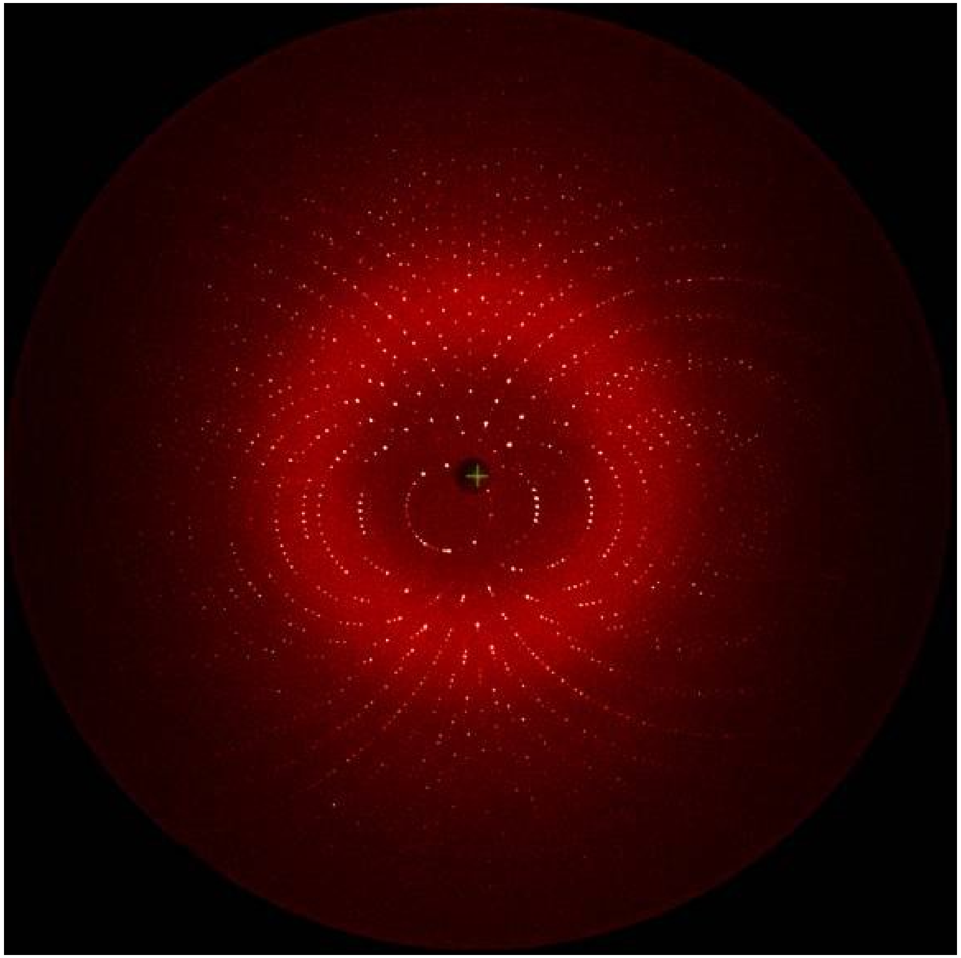


John Cowdery Kendrew
Max Perutz
Nobelpreis 1962



X-ray Scattering Research Today

Modern Protein Crystallography



BioCARS 14-ID-B station of APS using an undulator with a gap of 25 mm from a crystal of the M37V mutant of CO-bound dimeric clam hemoglobin.

Dynamics of Biomolecules

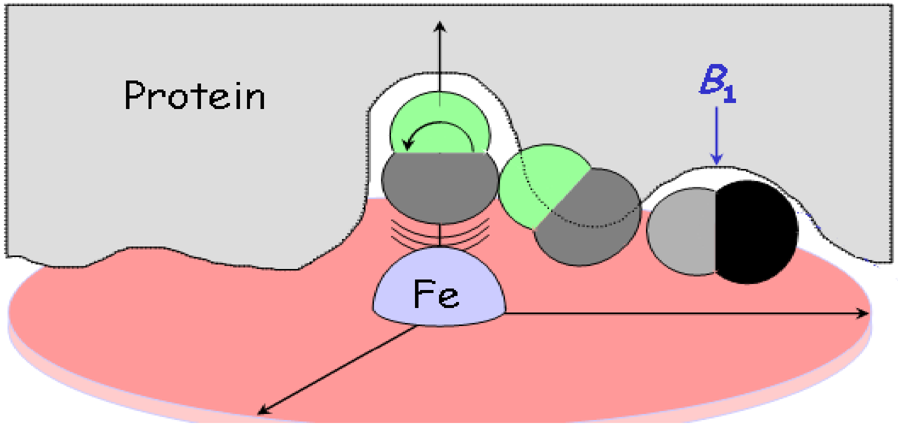
<u>Example:</u> Myoglobin protein found in muscle, stores oxygen for conversion into energy. Structure solved in 1960 (Kendrew).

<u>Puzzle:</u> How does the oxygen get into and out of the myoglobin molecule?

The protein is not static but dynamic with channels opening and closing?

➔ Time resolved Laue Diffraction

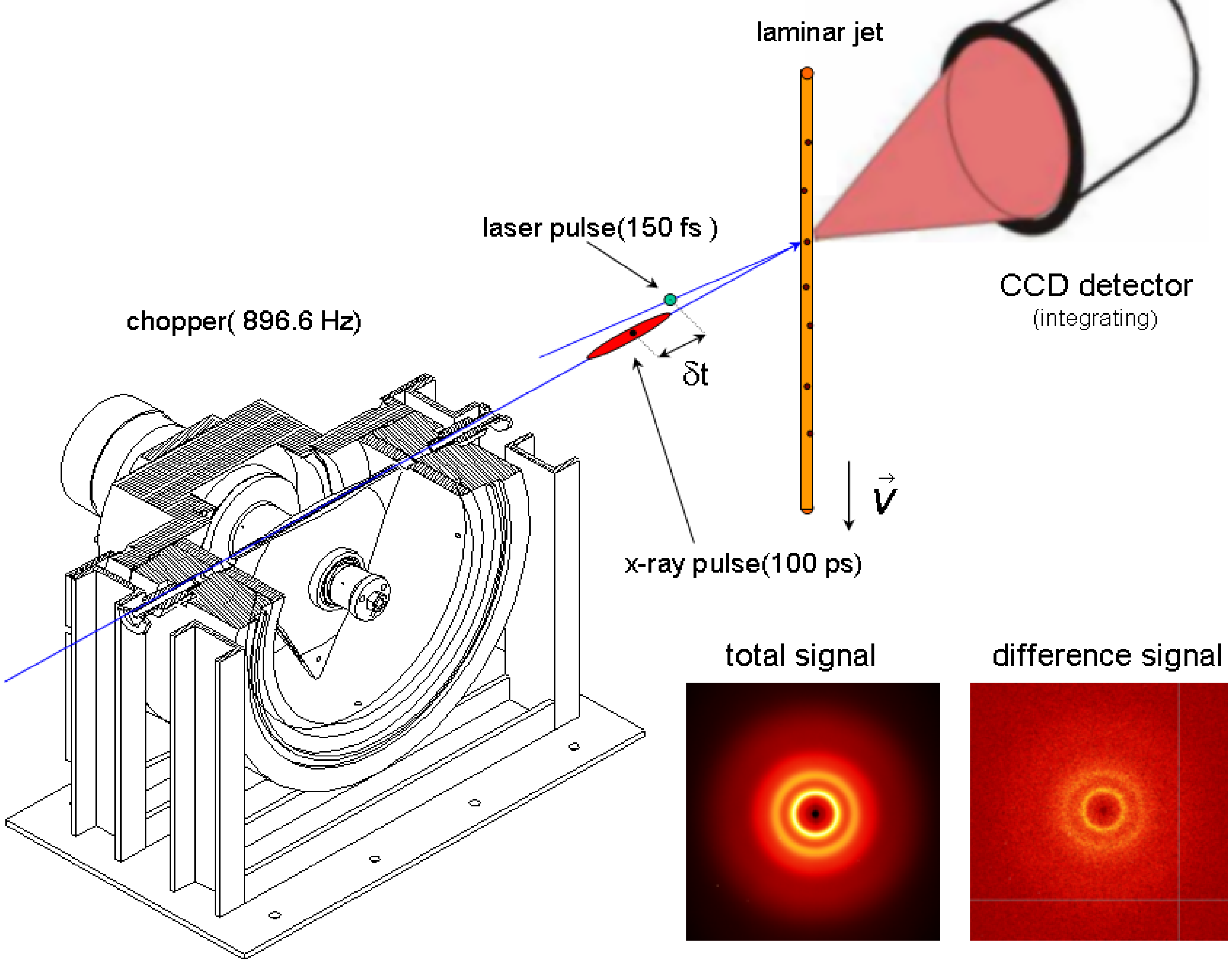
Use CO instead of O2. Use 10 ns optical pulse to CO from the Fe docking site. Probe dynamics with a 100 ps x-ray pulse.



Courtesy M. Wulff

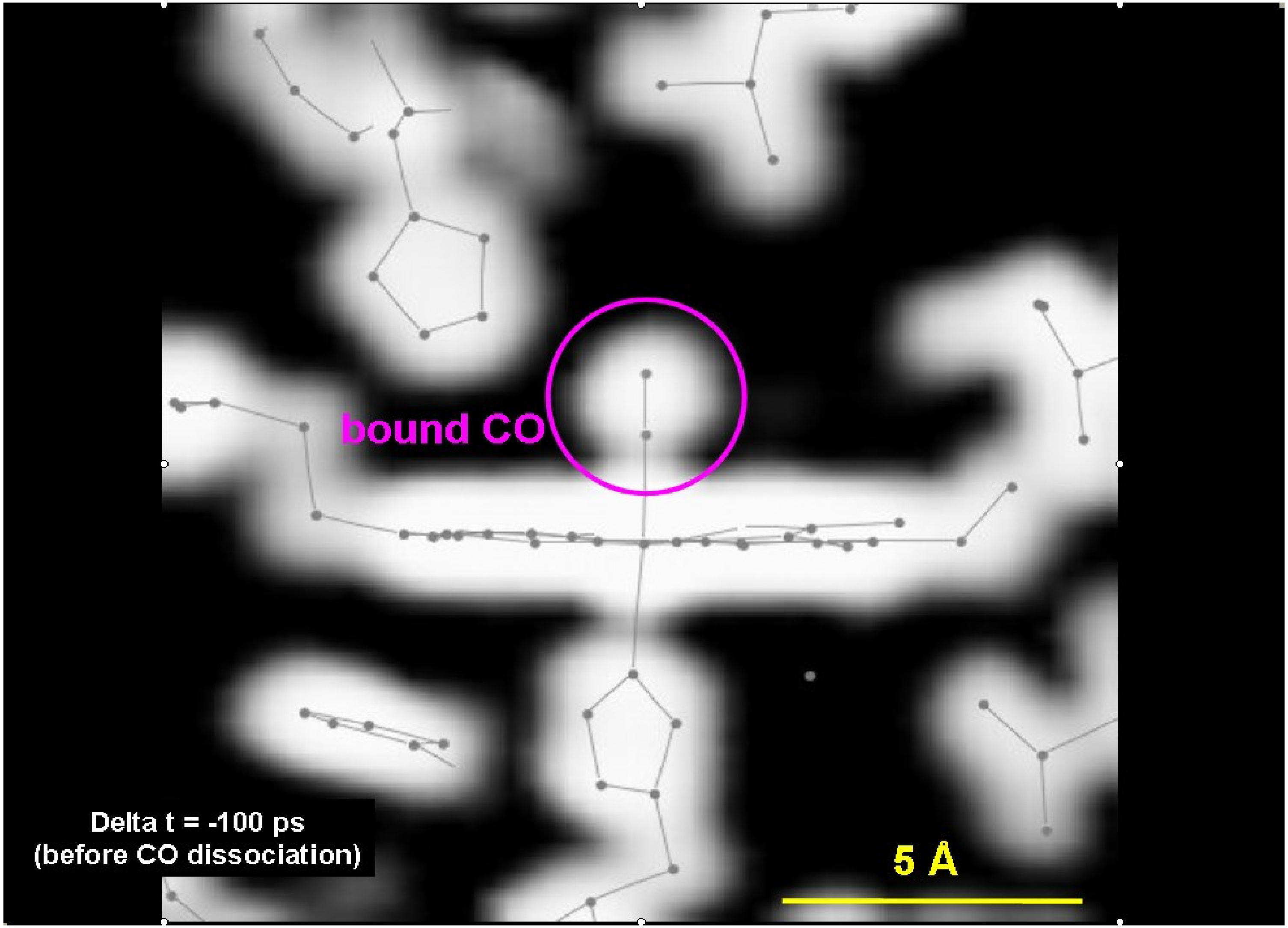
Time Resolved Protein Dynamics

Optical pump & x-ray probe(ID09B)



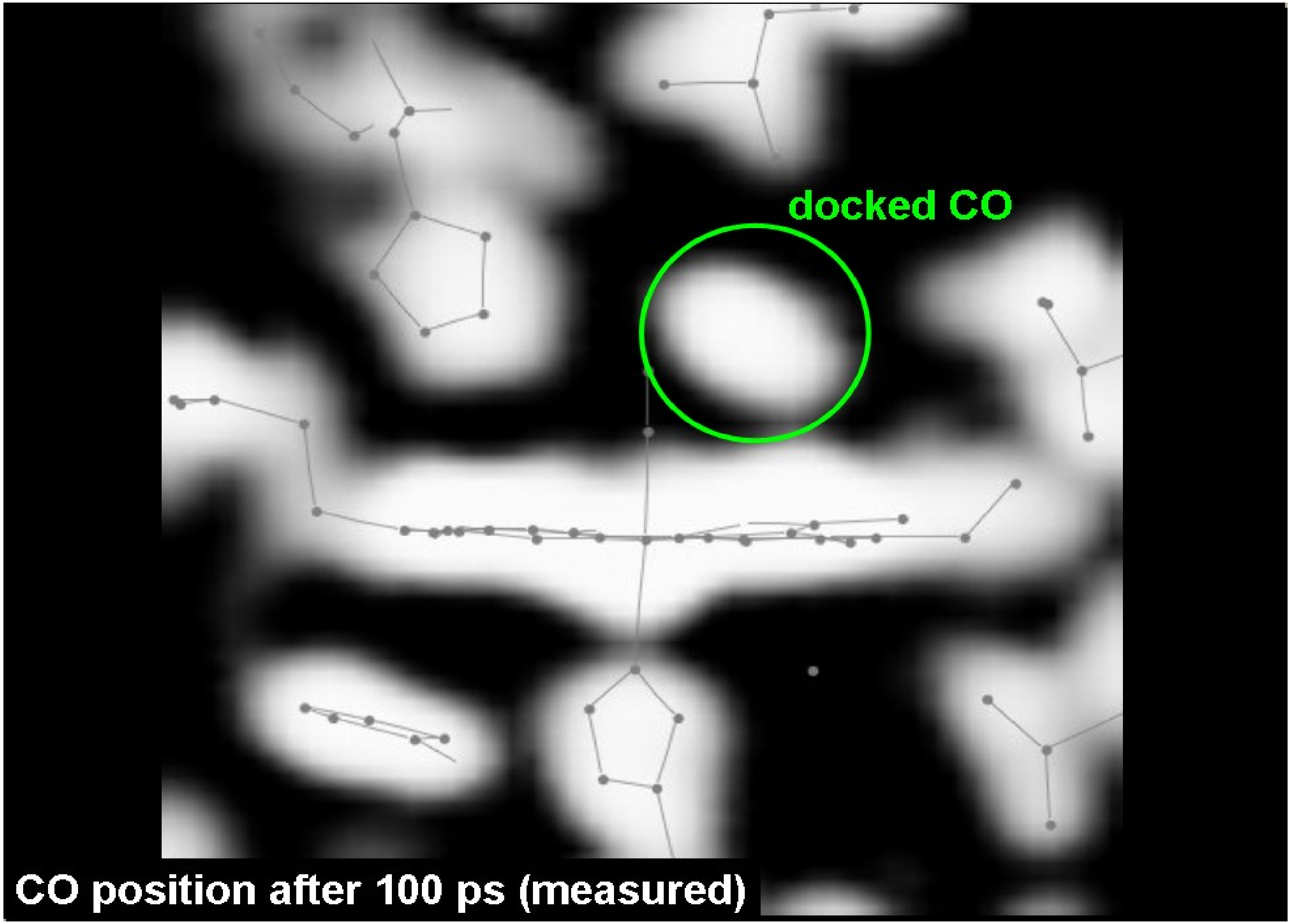
Courtesy M. Wulff

Time Resolved Protein Dynamics



Courtesy M. Wulff

Time Resolved Protein Dynamics



Courtesy M. Wulff

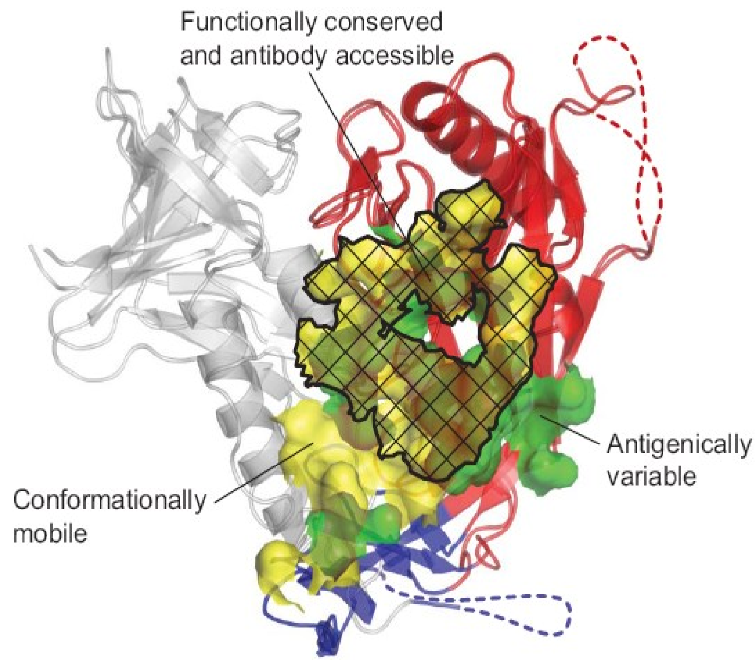
nature Vol 445|15 February 2007|**doi:10.1038/nature05580**

ARTICLES

Structural definition of a conserved neutralization epitope on HIV-1 gp120

Tongqing Zhou[1], Ling Xu[1], Barna Dey[1], Ann J. Hessell[3], Donald Van Ryk[2], Shi-Hua Xiang[4], Xinzhen Yang[4], Mei-Yun Zhang[5], Michael B. Zwick[3], James Arthos[2], Dennis R. Burton[3], Dimiter S. Dimitrov[5], Joseph Sodroski[4], Richard Wyatt[1], Gary J. Nabel[1] & Peter D. Kwong[1]

The remarkable diversity, glycosylation and conformational flexibility of the human immunodeficiency virus type 1 (HIV-1) envelope (Env), including substantial rearrangement of the gp120 glycoprotein upon binding the CD4 receptor, allow it to evade antibody-mediated neutralization. Despite this complexity, the HIV-1 Env must retain conserved determinants that mediate CD4 binding. To evaluate how these determinants might provide opportunities for antibody recognition, we created variants of gp120 stabilized in the CD4-bound state, assessed binding of CD4 and of receptor-binding-site antibodies, and determined the structure at 2.3 Å resolution of the broadly neutralizing antibody b12 in complex with gp120. b12 binds to a conformationally invariant surface that overlaps a distinct subset of the CD4-binding site. This surface is involved in the metastable attachment of CD4, before the gp120 rearrangement required for stable engagement. A site of vulnerability, related to a functional requirement for efficient association with CD4, can therefore be targeted by antibody to neutralize HIV-1.



DESY

BESCHLEUNIGER | FORSCHUNG MIT PHOTONEN | TEILCHENPHYSIK
| FORSCHUNG | AKTUELLES | ÜBER DESY | INFOS & SERVICES | KARRIERE | KONTAKT

Deutsches Elektronen-Synchrotron
Ein Forschungszentrum der Helmholtz-Gemeinschaft

FORSCHERWELT.

Wir machen Erkenntnis möglich

Home

INFORMATIONEN FÜR

- » Journalisten
- » Wirtschaftspartner
- » Schüler und Lehrer

DESY beglückwünscht Ada Yonath zum Chemie-Nobelpreis

Die israelische Forscherin Prof. Ada E. Yonath hat zusammen mit zwei Amerikanern den Nobelpreis für Chemie verliehen bekommen. Ihre Forschungen zur Struktur und Funktion der Ribosomen, denjenigen Molekülkomplexen, die aus der DNA-Erbinformation die für das Leben notwendigen Eiweißmoleküle herstellen, führte sie hauptsächlich an DESYs DORIS-Beschleuniger durch.
» mehr

50 Jahre DESY

» Alle Infos zum Jubiläumsjahr

Veranstaltungen

» VERANSTALTUNGSKALENDER FÜR DESY IN HAMBURG UND ZEUTHEN

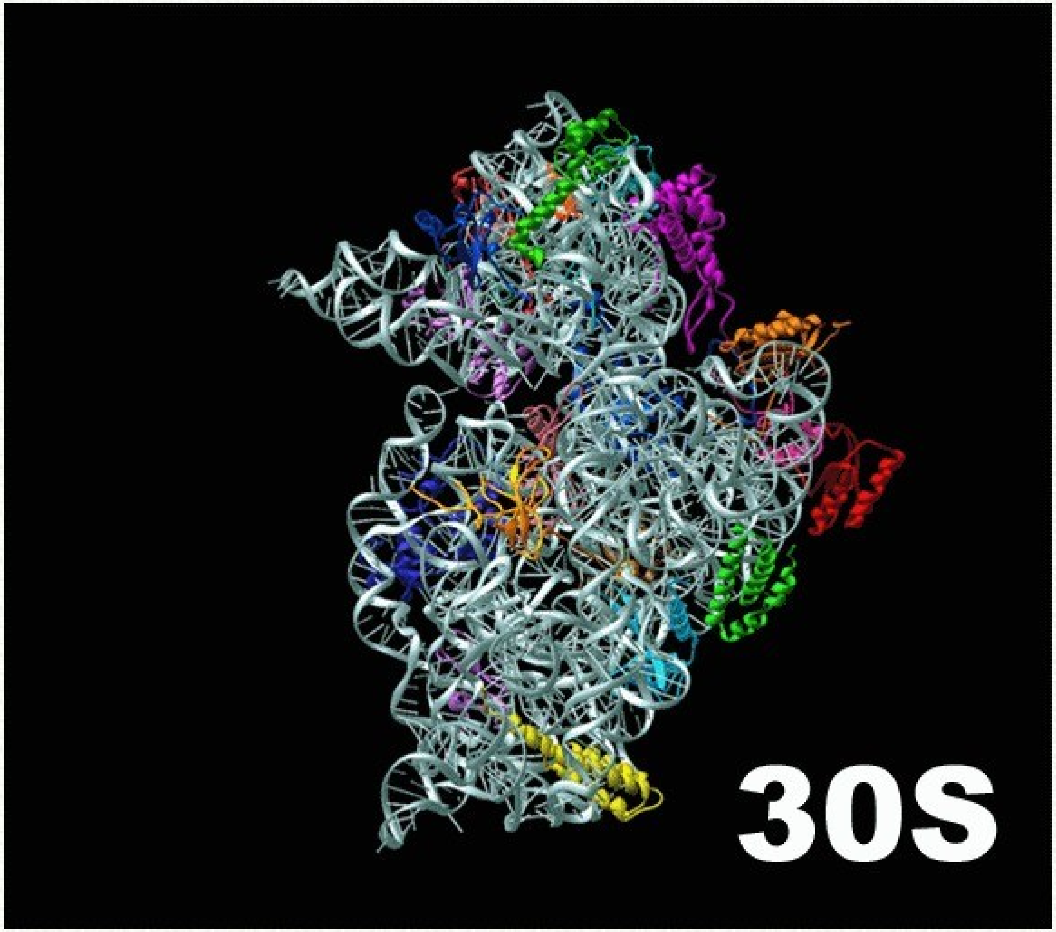


Figure A:

The Small Ribosomal Subunit

Schluenzen et al., Cell, 102, 615-23 (2000)

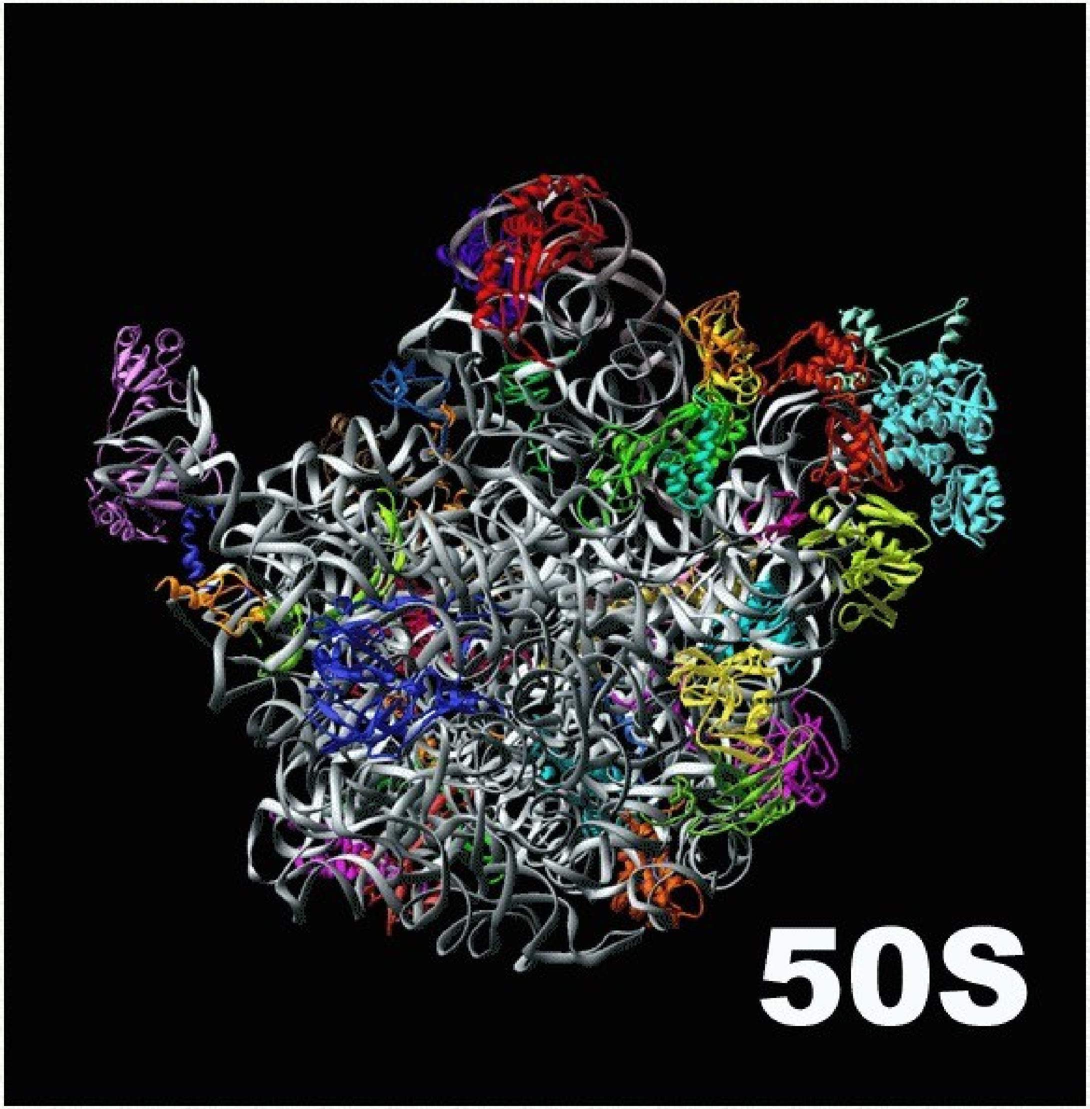


Figure B:

The Large Ribosomal Subunit

Harms et al., Cell, 107, 679-88 (2001)

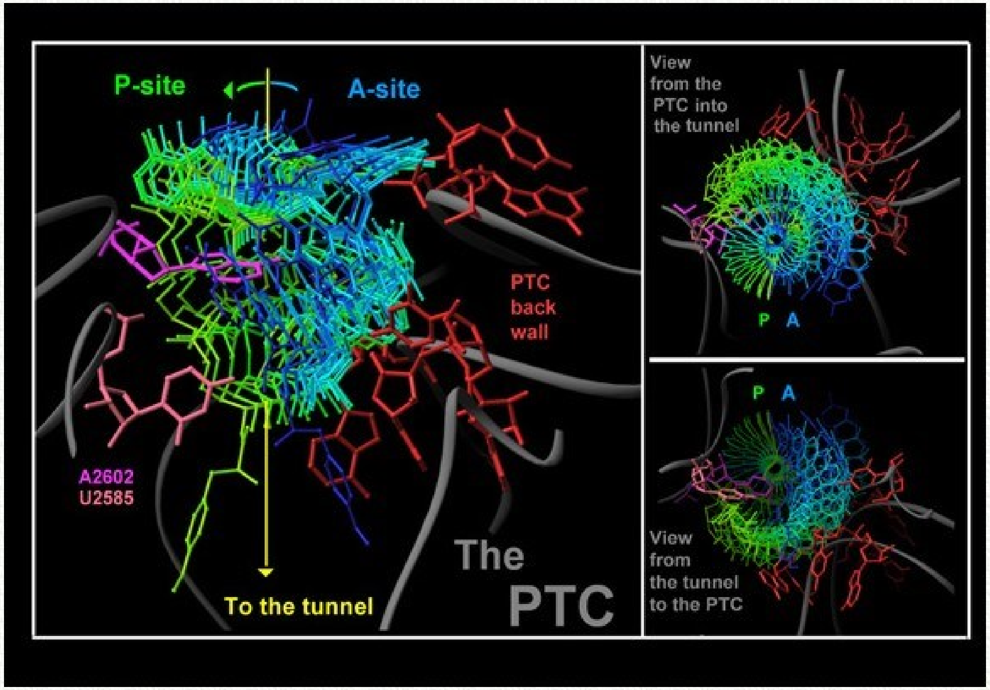


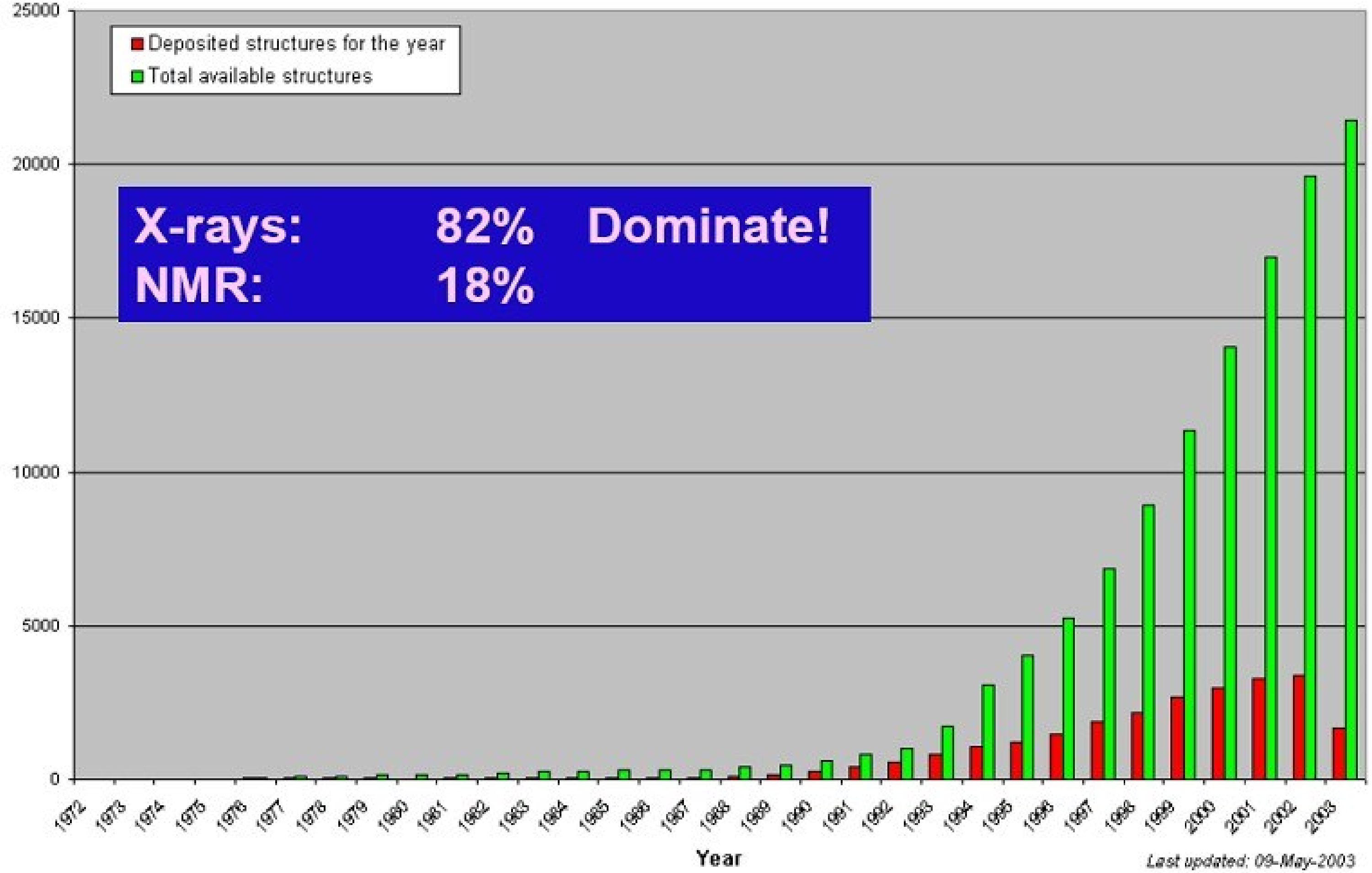
*Figure C:

tRNA A-site --->P site

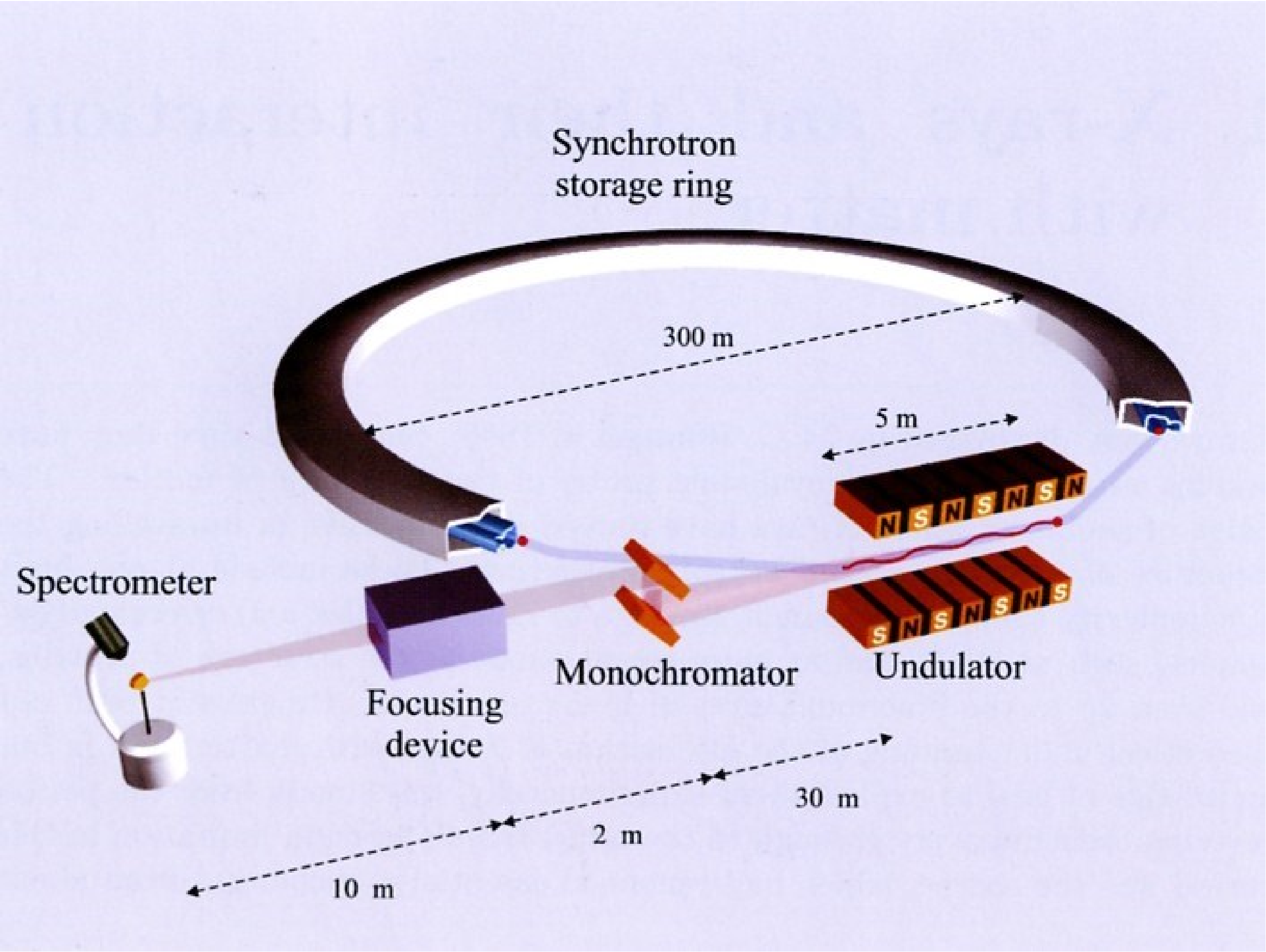
Agmon et al., 2003; Bashan et al., 2003

*For details go to "Scientific Activities" (Figure 5)

Spectacular growth of structural biology

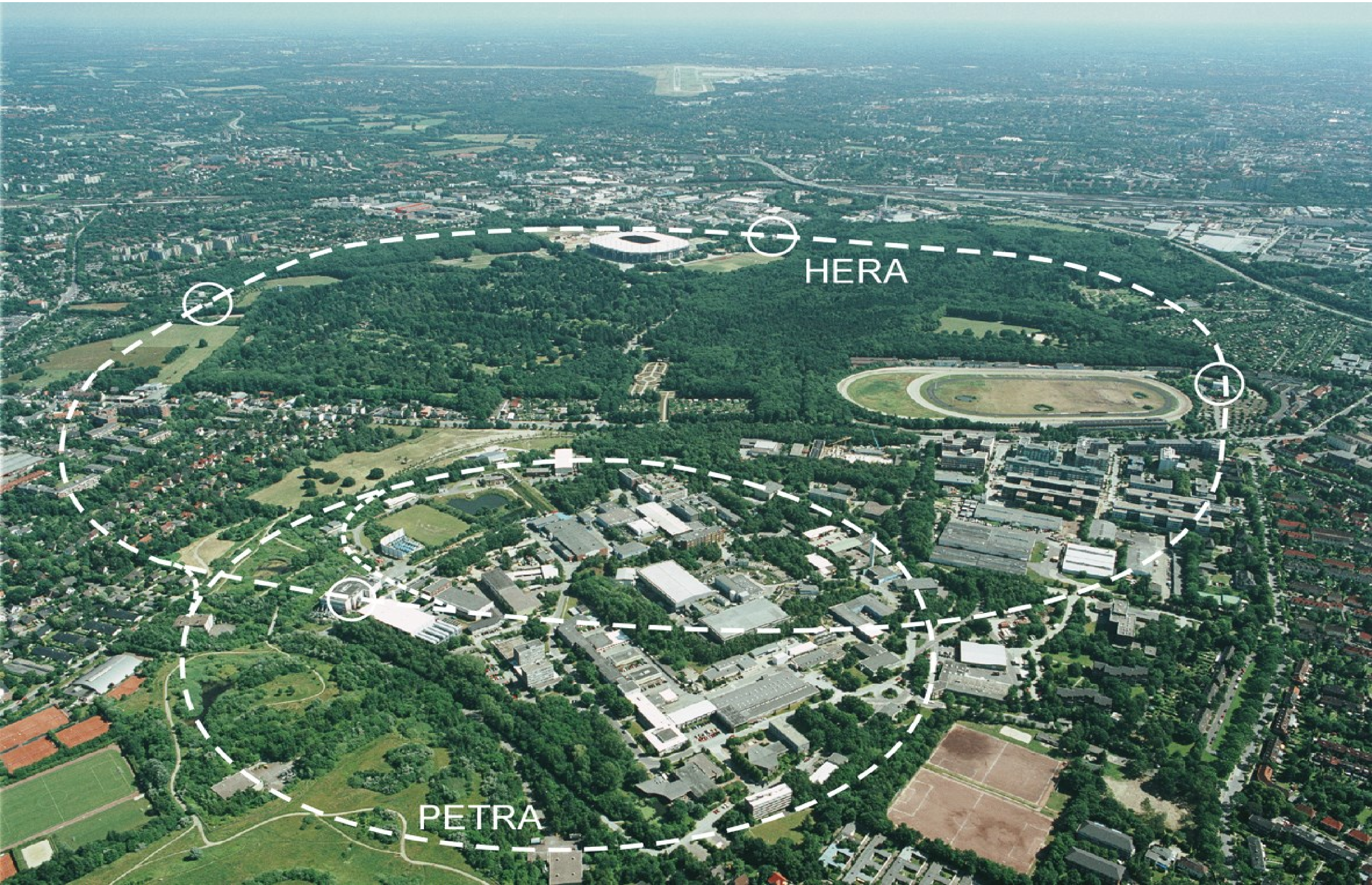


Made Possible by Storage-Rings

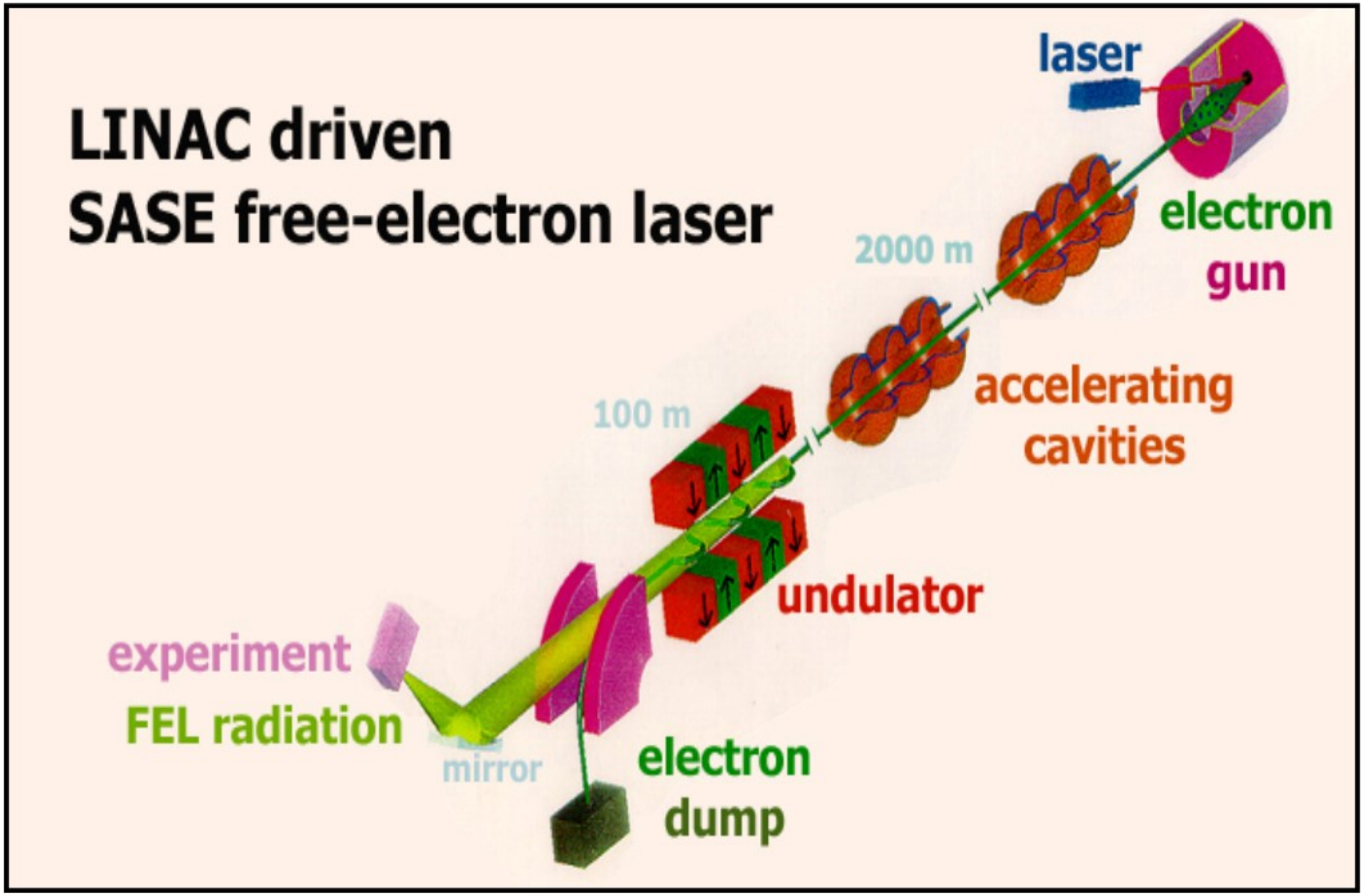


European Synchrotron Radiation Facility (ESRF)

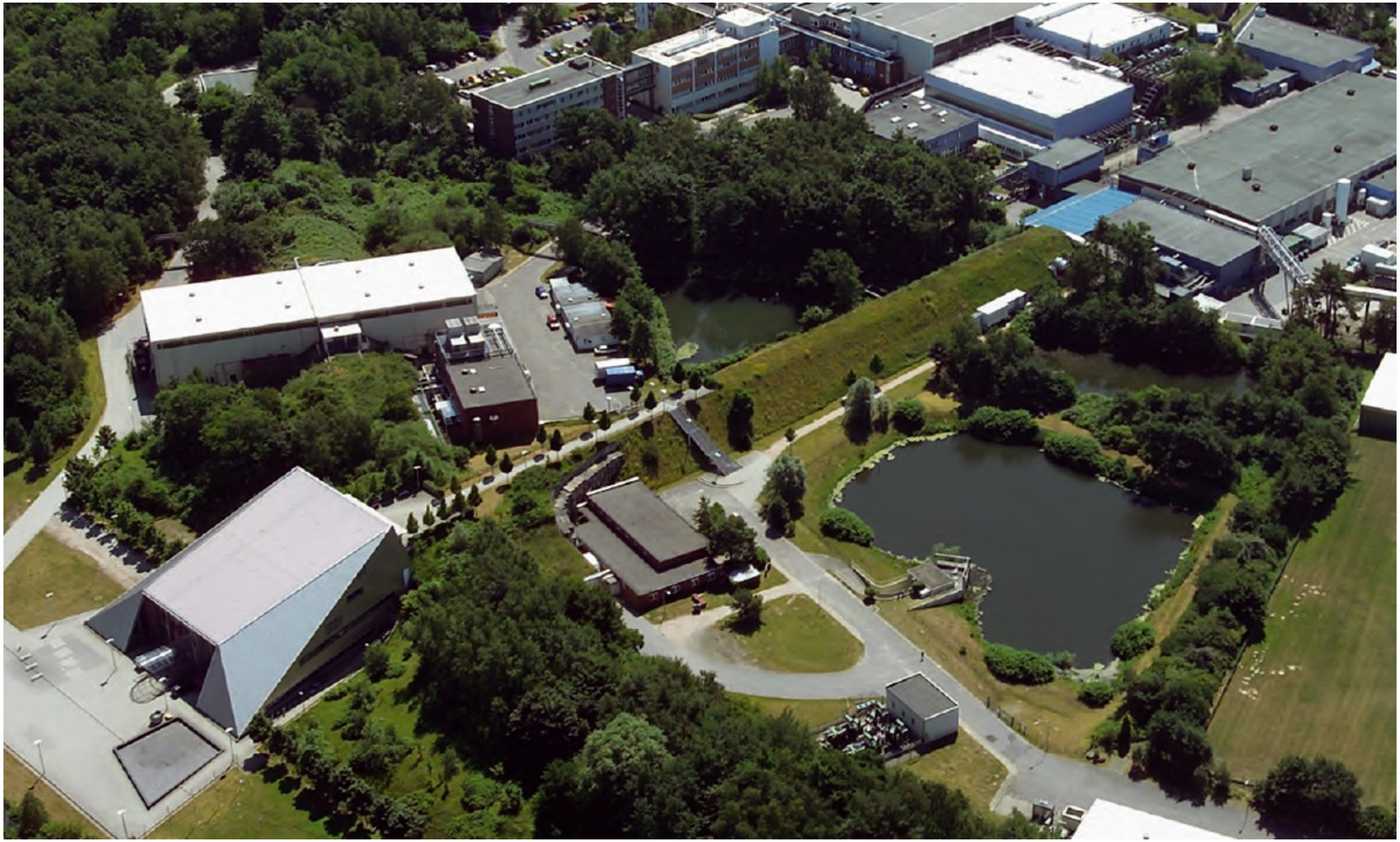
Deutsches Elektronen Synchrotron (DESY)



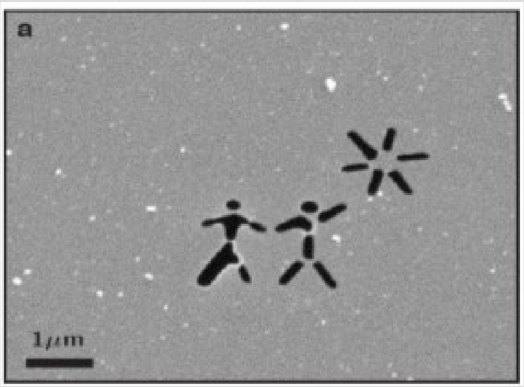
Free Electron Lasers (FELs)

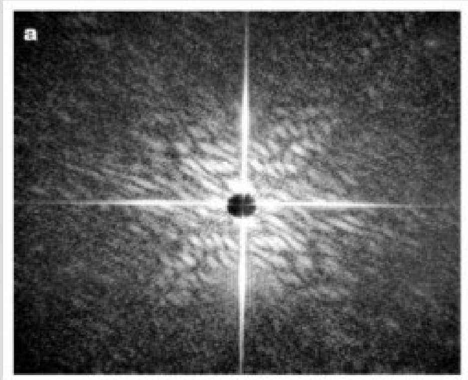


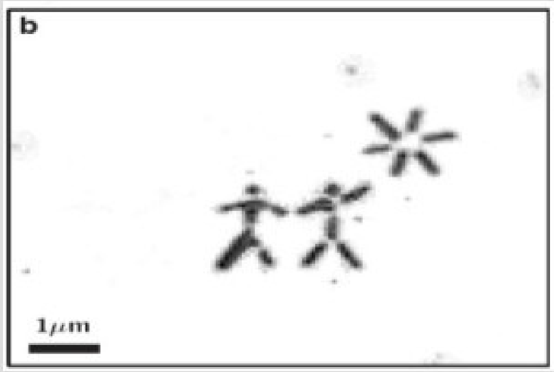
The FLASH Facility in Hamburg



The Femtosecond World



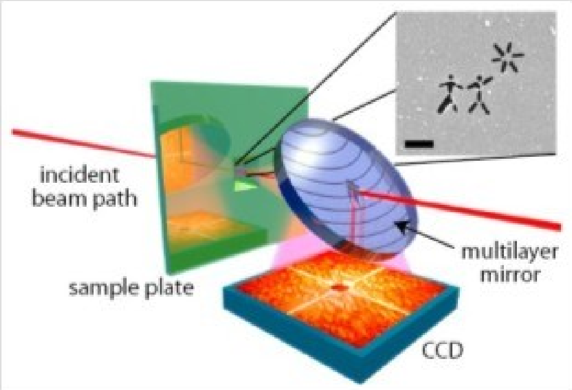




Model structure in 20 nm SiN membrane

Speckle pattern recorded with a single (25 fs) pulse

Reconstructed image



Incident FEL pulse:
25 fs, 32 nm,
4 x 10^{14} W cm^{-2}
(10^{12} ph/pulse)

H. Chapman et al., Nature Physics, 2,839 (2006)