Methoden der Strukturenuntersuchung

Elektronmikroskope

Diffraktionsmethode

CD-Spektroskopie

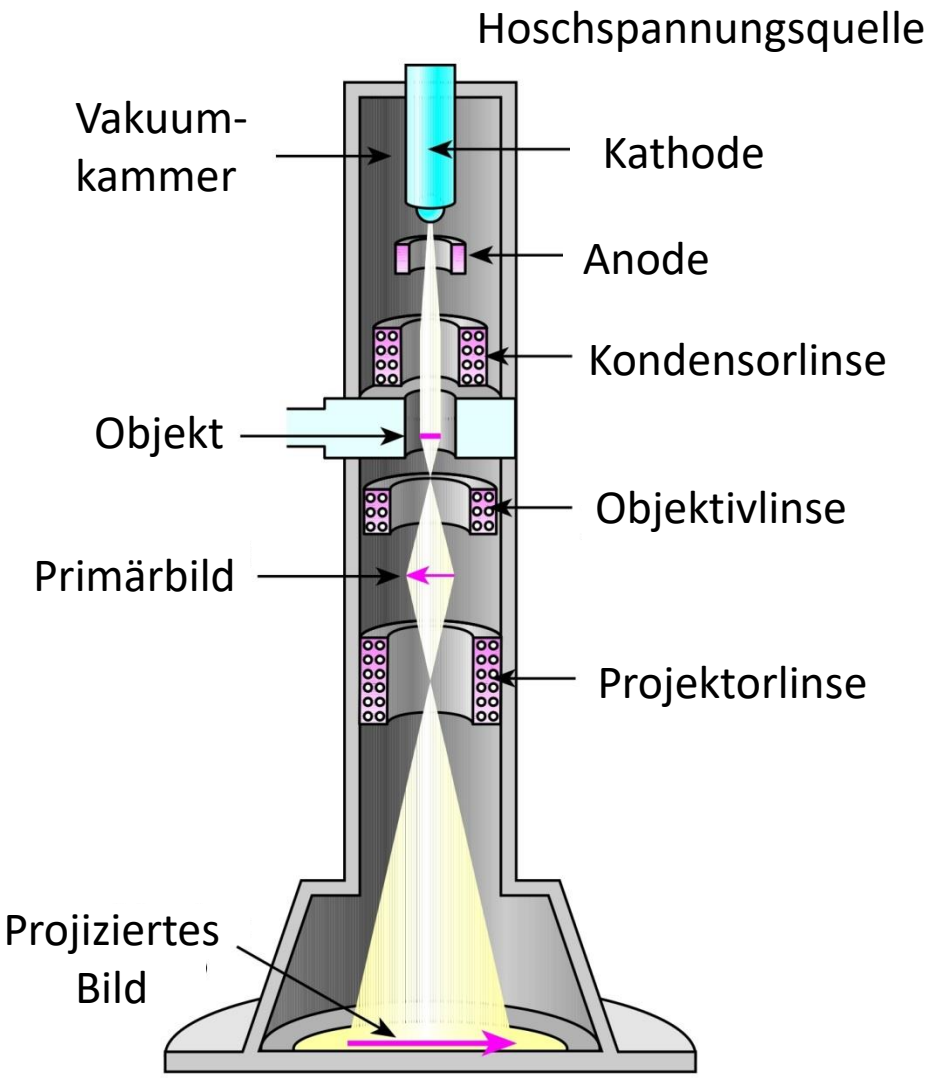
ELEKTRONENMIKROSKOPE

$$\lambda = \frac{h}{mv}$$

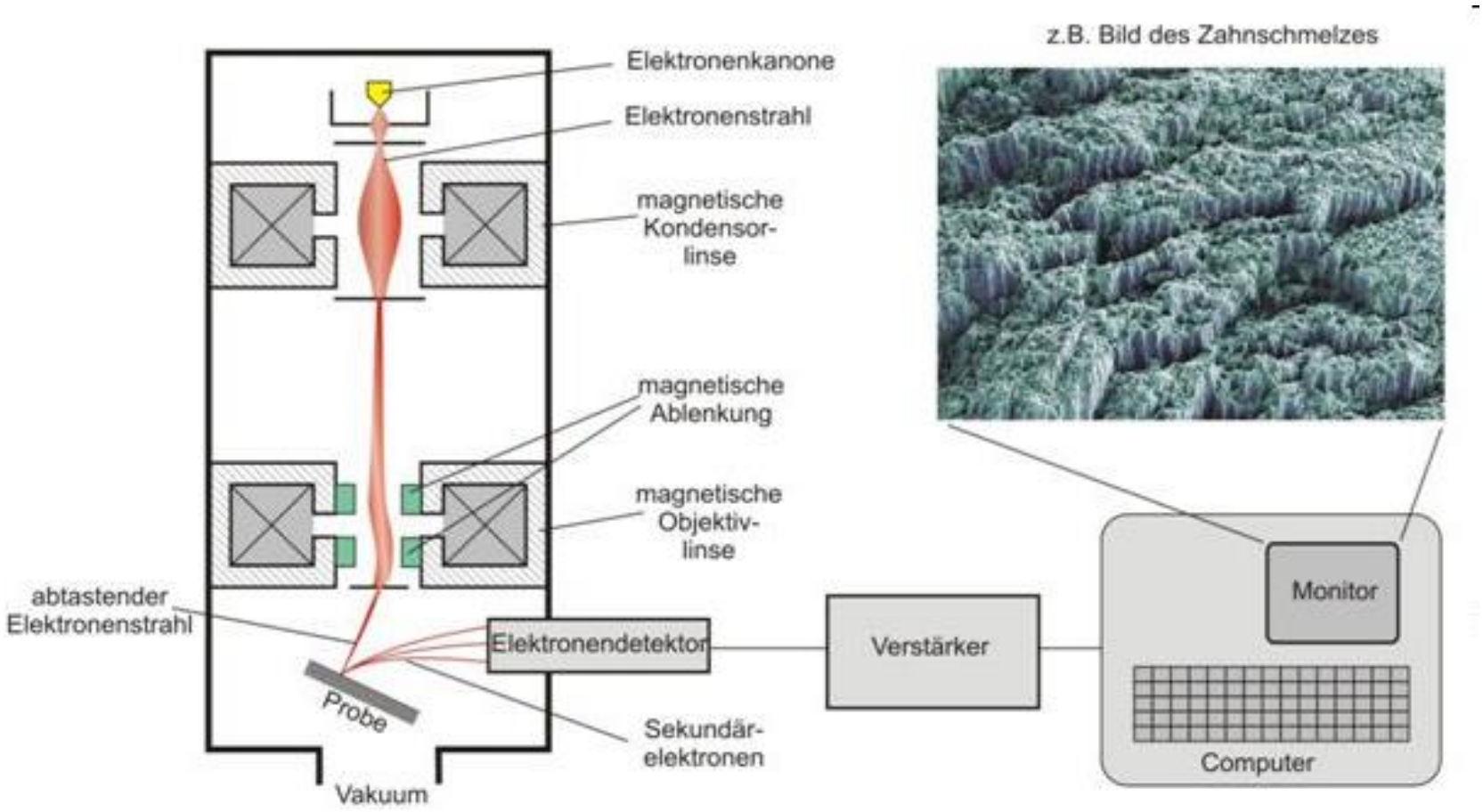
Transmissionselektronenmikroskop
Rasterelektronenmikroskop

Materienwellen!

Transmissionselektronenmikroskop



Rasterelektronenmikroskop



Auflösungsvermögen des Elektronenmikroskops
Abbe´sches Prinzip und Materialwellen

Materialwelle: Zu einem Teilchen mit m Masse und v Geschwindigkeit, kann man eine Welle (Materienwelle)

zuordnen, die eine Wellenlänge von $\lambda = \frac{h}{mv}$ hat.

Die Geschwindigkeit des Elektrons nach einer Beschleunigung mit U Spannung beträgt:

$$v = \sqrt{\frac{2eU}{m}} \quad \text{womit:} \quad \lambda = \frac{h}{\sqrt{2emU}}$$

Typisch kann λ 5 pm sein. Aber ω ist sehr klein! NA≈0,002

$\delta = 0{,}61 \cdot \lambda / (n \cdot \sin\omega) \approx$ nm

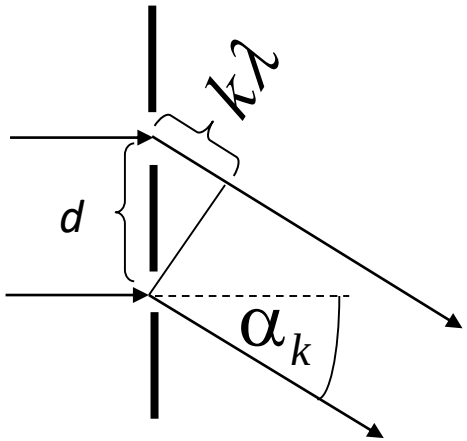
DIFFRAKTIONSMETHODE

Röntgendiffraktion

Anwendung der Röntgenstrahlung in Strukturanalyse der Materie.

Zur Erinnerung:
Diffraktion des Lichtes

$$\sin \alpha_k = \frac{k\lambda}{d}$$

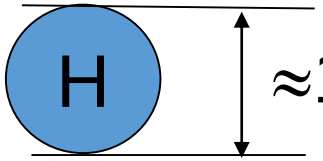


Röntgendiffraktion

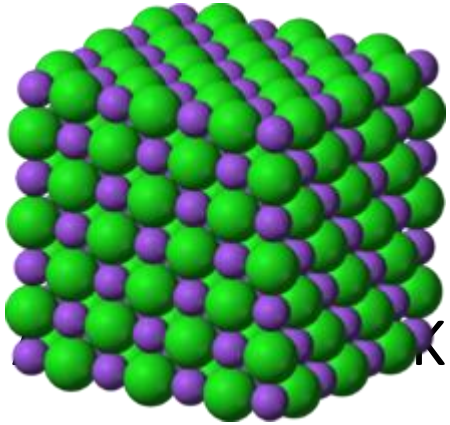
Was für ein Gitter passt zur Röntgenstrahlung?

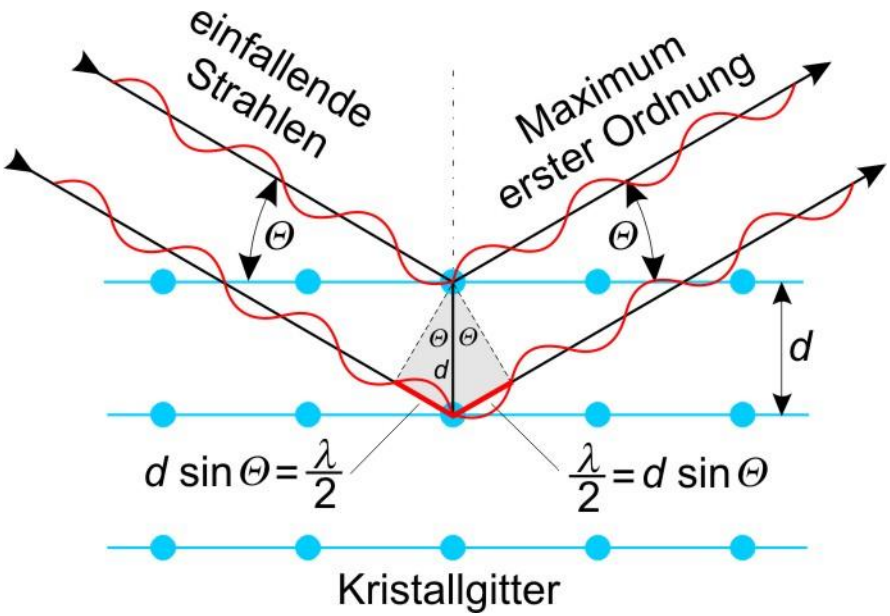
$\lambda < d$

λ_{Rtg} 10-100 pm



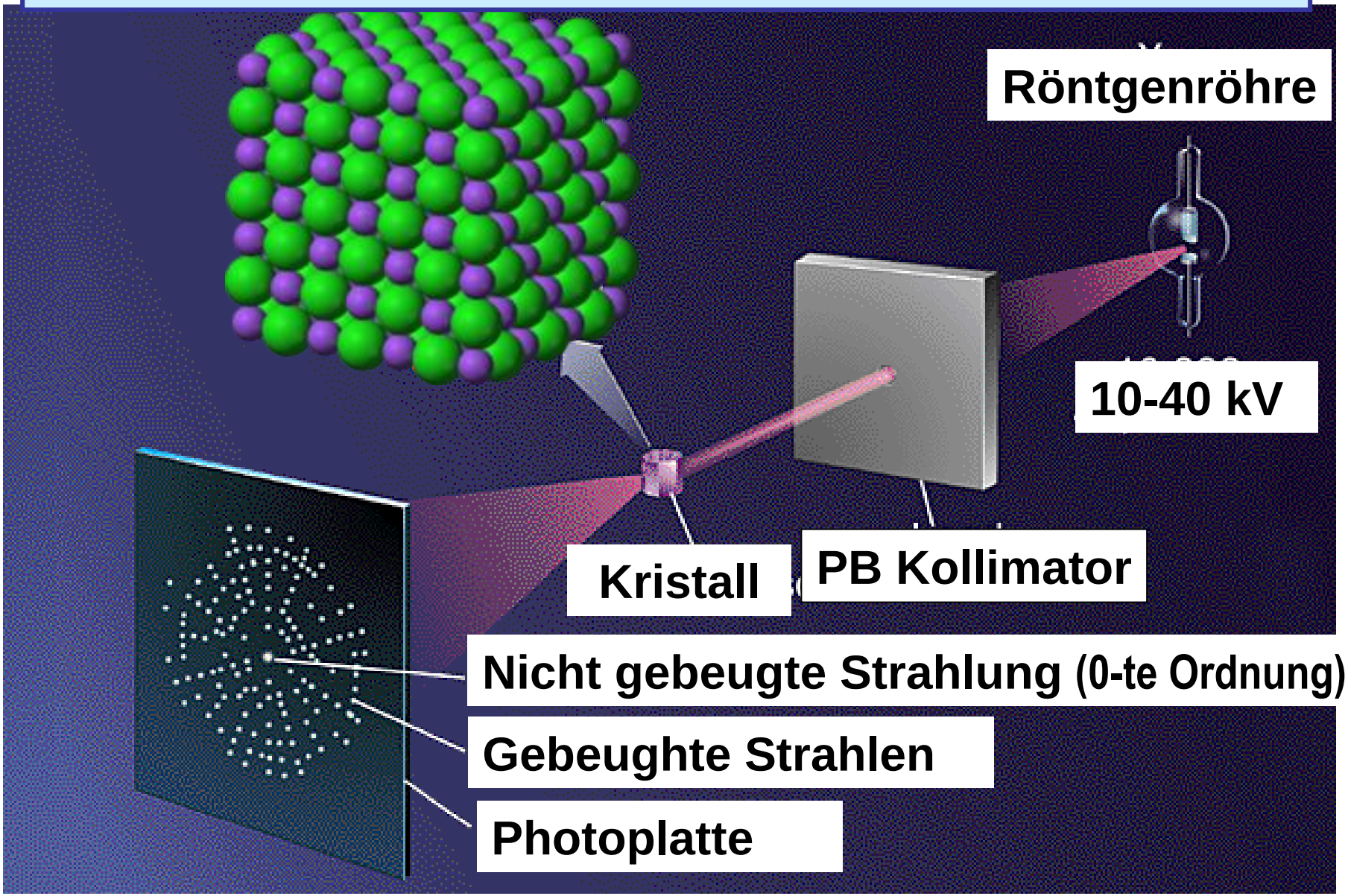
≈100 pm

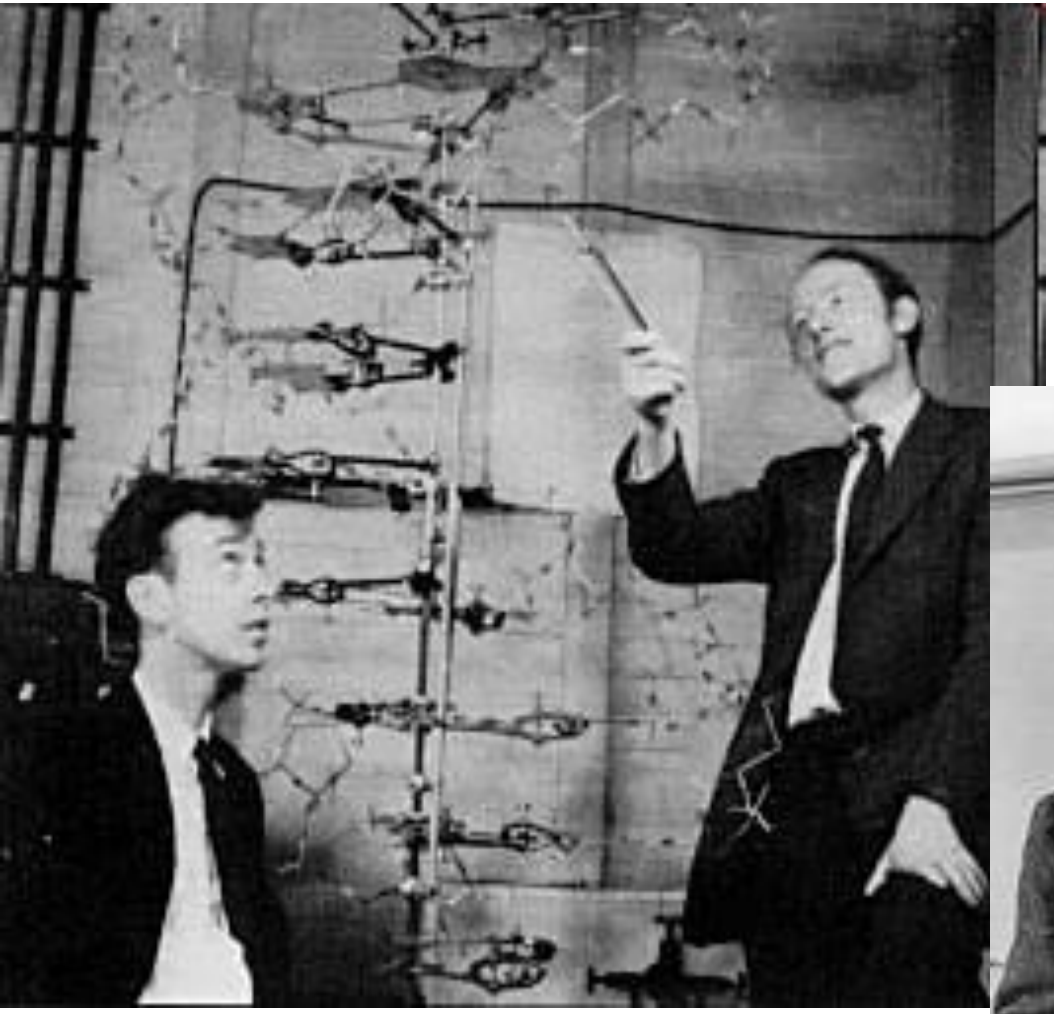




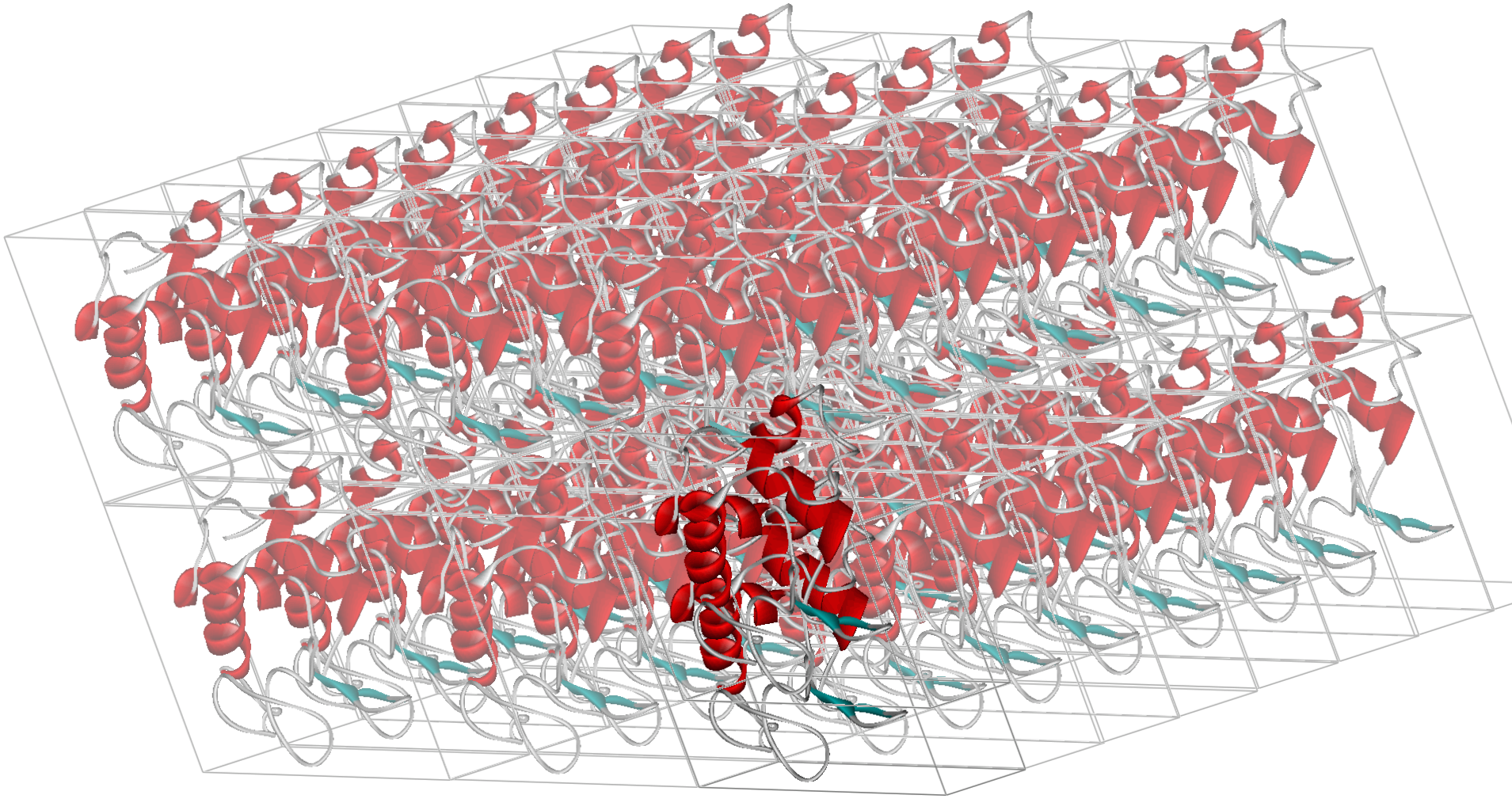
Kristall → auch DNS $n\lambda = 2d \sin\Theta$ all!

Aufbau des Röntgendiffraktionsgerätes



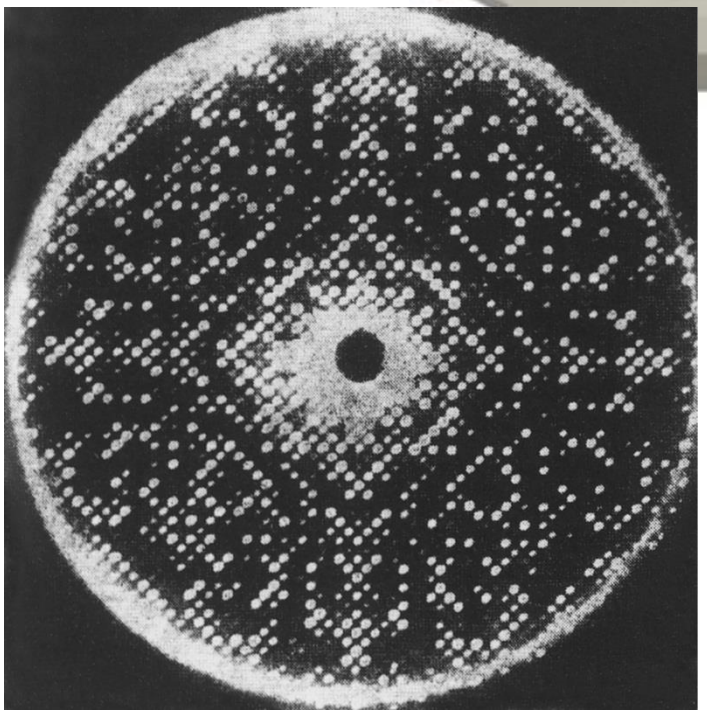


Eiweißkristalle



Bestimmung der Raumstruktur der Eiweiße

Lysozyme



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A Structural View of Biology

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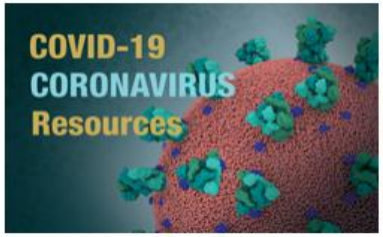
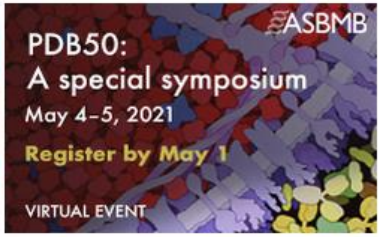
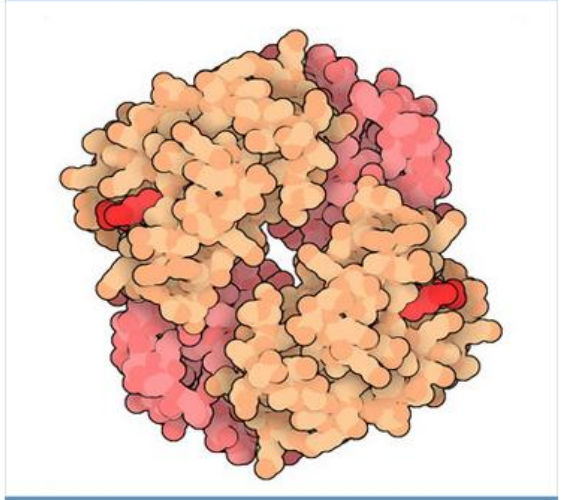
As a member of the wwPDB, the RCSB PDB curates and annotates PDB data.

The RCSB PDB builds upon the data by creating tools and resources for research and education in molecular biology, structural biology, computational biology, and beyond.





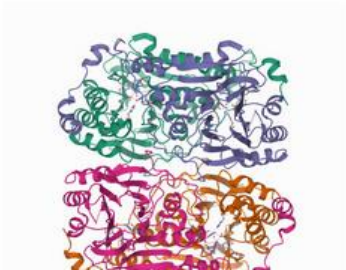
May Molecule of the Month



Fetal Hemoglobin

Latest Entries

As of Tue Apr 27 2021



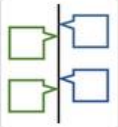
Features & Highlights

EXTENDED: Enter the 2021 CellPAINT Contest by May 6

Create images inspired by vaccines and by 50 years of the PDB with CellPAINT and win prizes

Future Planning: Entries with extended PDB and CCD ID codes will be distributed in PDBx/mmCIF format

News | Publications



Explore the History of the PDB

Explore PDB's historical highlights alongside milestone advances in structural biology » *04/27/2021*

Annual Report Published

Download the 2020 Annual Report for an overview of recent RCSB PDB activities, PDB structures and the

Elektronen und Neutronendiffraktion

λ: Materialwellen

Elektronen: Kleine Eindringstiefe: Oberflächen

Elektronen und Neutronen werden an den Atomkernen gestreut.

(Rtg wird durch Elektronenwolken gestreut.)

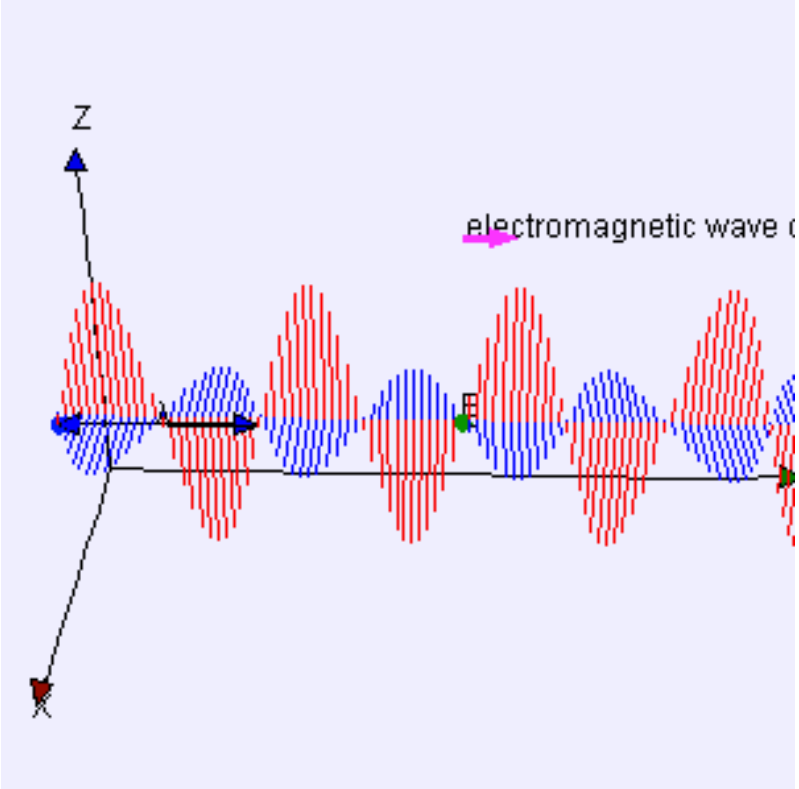
Elektronen werden an den schwereren Kernen gestreut

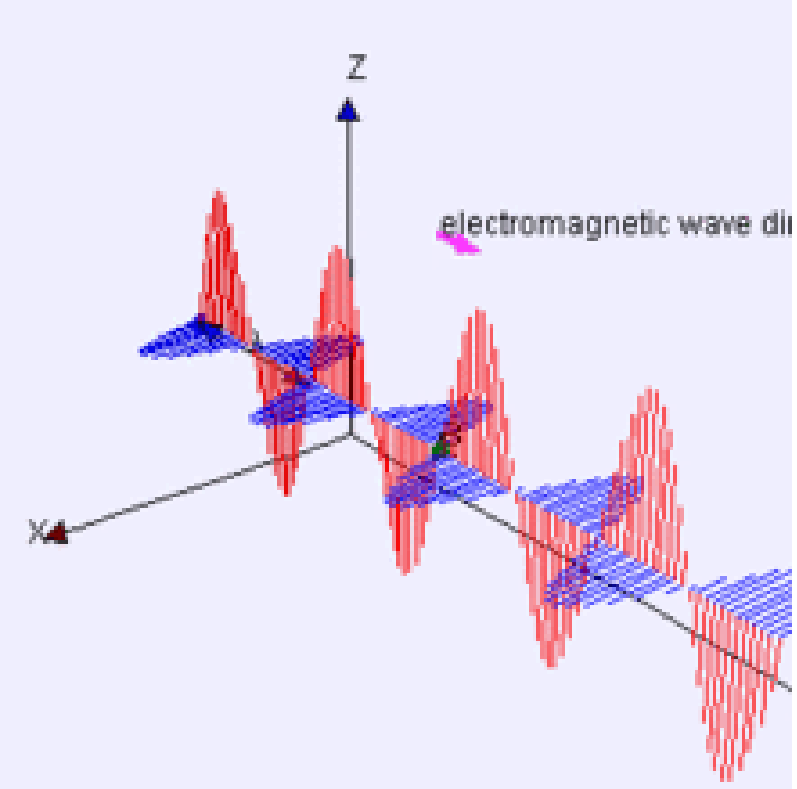
Neutronen auch an den Protonen, => Neutronendiffraktion gut zur Strukturuntersuchung von wasserstoffhaltigem Material.

CD

Circular dichroism spectroscopy

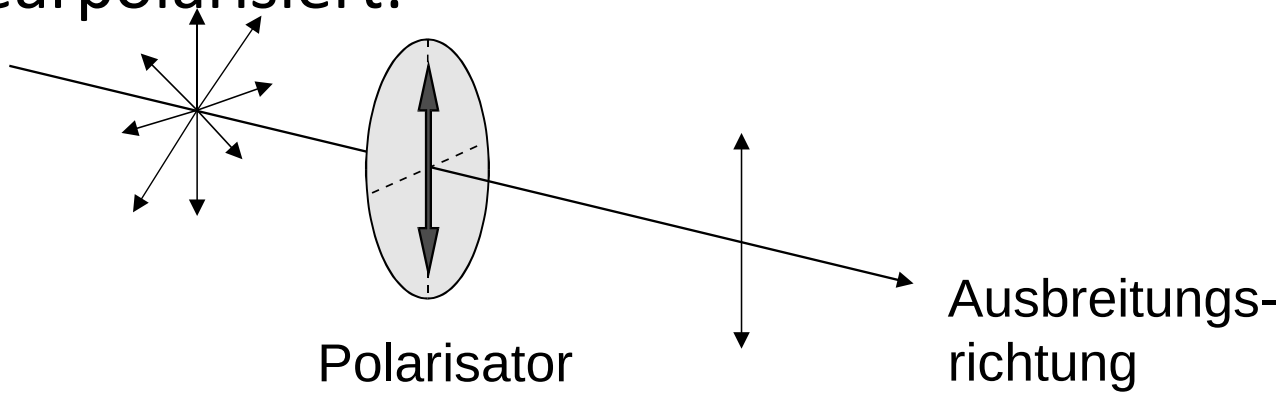
Elektro (**E**) magnetiche (**B**) Welle = Licht



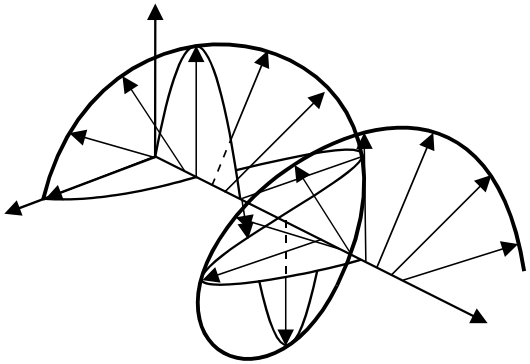


Polarisiertes Licht

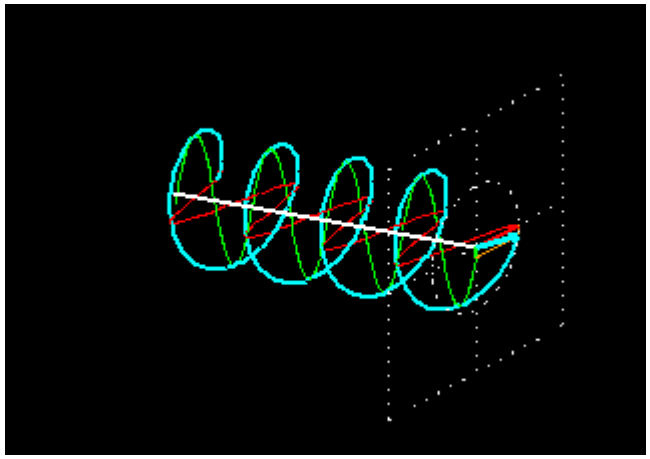
Linearpolarisiert:



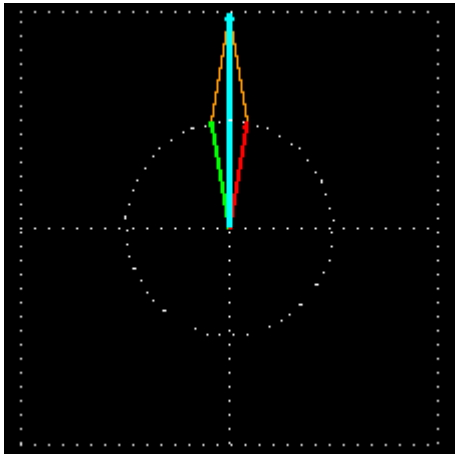
Cirkulär polarisiert



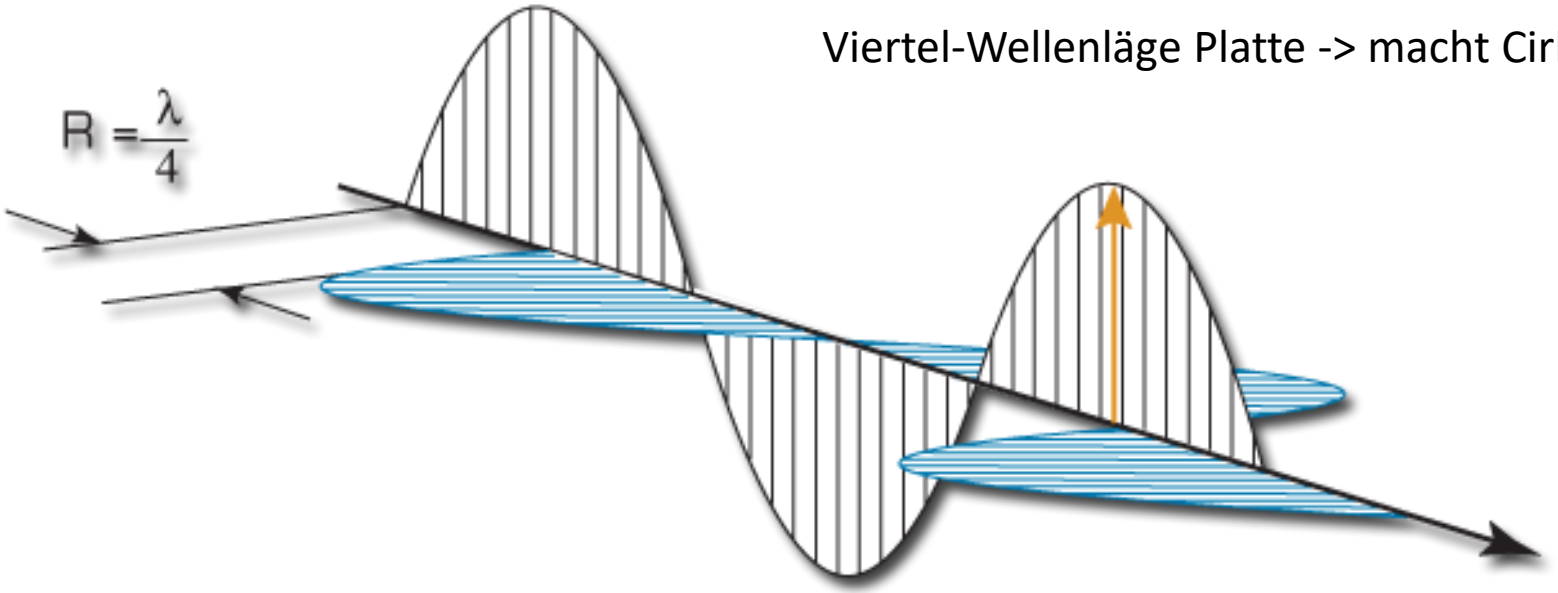
Cirkulärpolarisiertes Licht



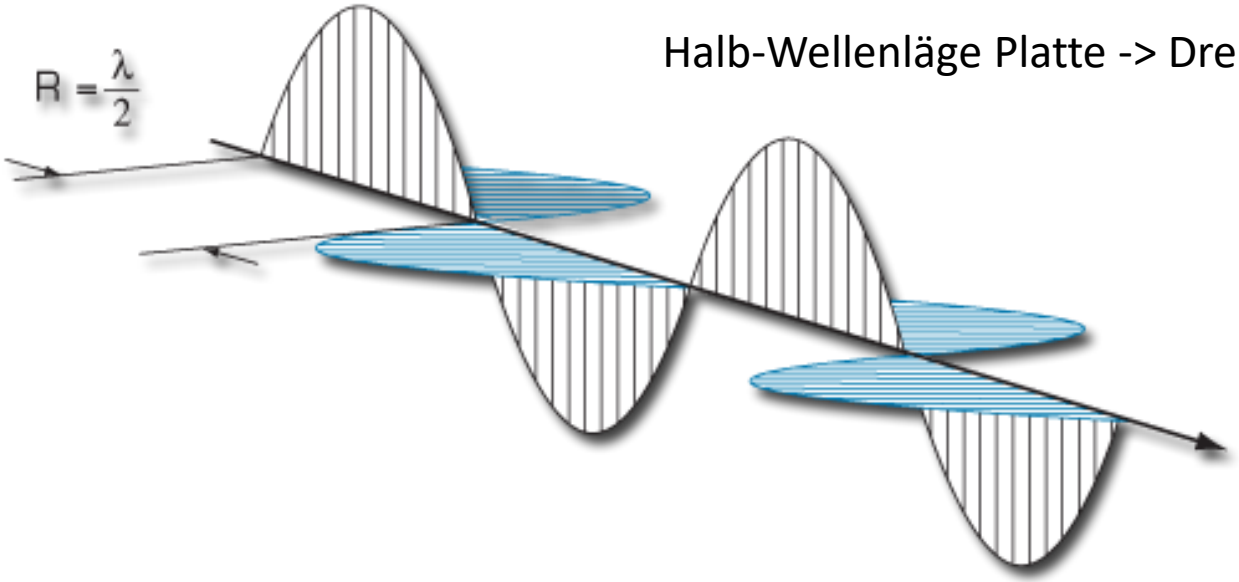
Linear = Links + Rechts



Viertel-Wellenläge Platte -> macht Cirk.Pol.

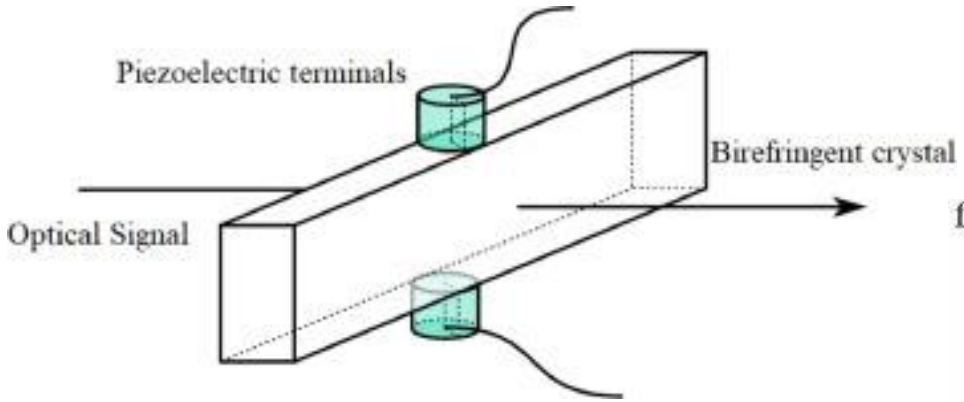


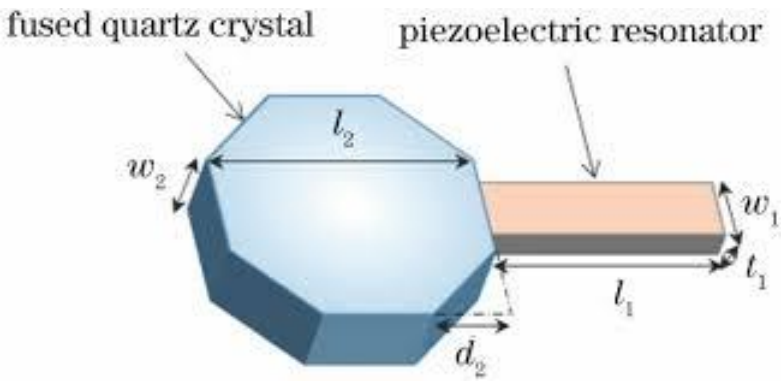
Halb-Wellenläge Platte -> Dreht Pol.Richtung um 90 Grad.



PEM: Photo Elastic Modulator

Dies ist ein Kristall, welche unter Stauchungsstress Doppelbrechend wird.





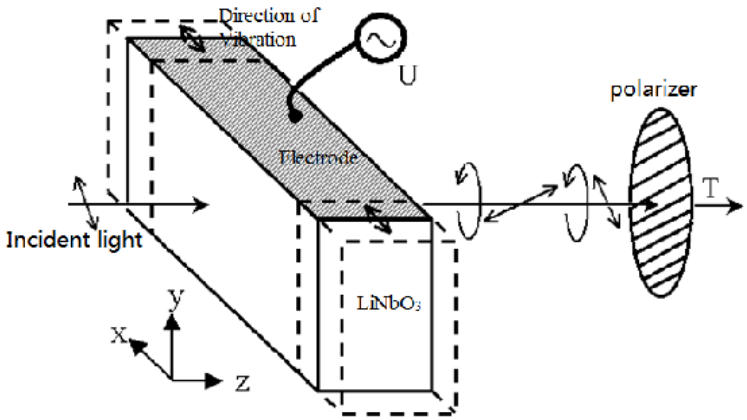
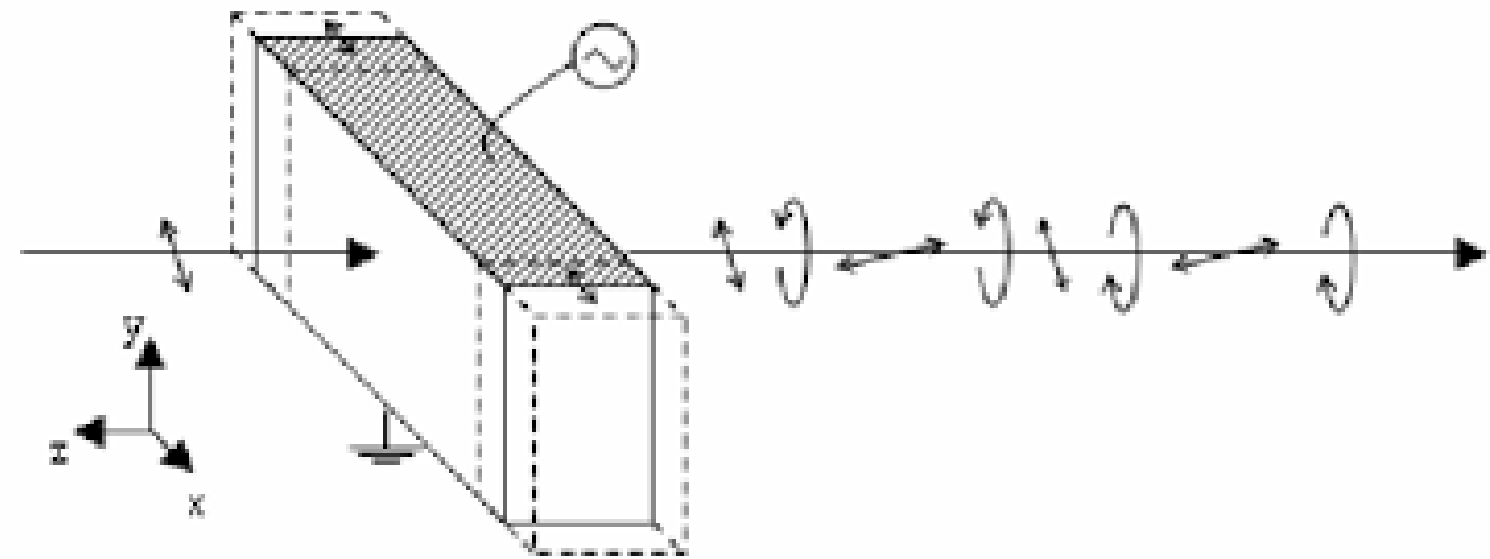


Figure 3. Sketch of basic structure of single crystal photo-elastic modulator (SCPEM)

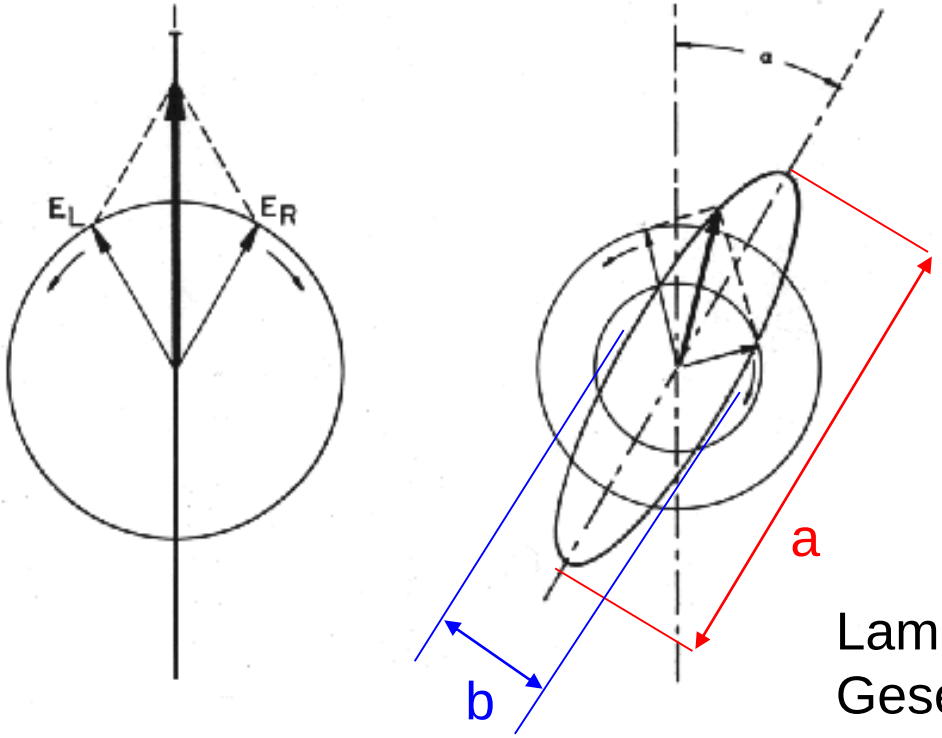


Am einfachsten benutzt man Resonanz, also treibt den Kristall mit dem Eigenfrequenz.

Dann bekommen wir periodische Lichtdrehung: Manchmal Linear, dann Rechts, dann wieder linear, dann Links, etc..

Chirale Stoffe: Abrobanz und Brechzahl sind Drehrichtungsabhängig:

$A_{Links} \neq A_{Rechts}$ und $n_{Links} \neq n_{Rechts}$



$$\Delta A = A_L - A_R = \Delta\varepsilon \; c \; x$$

$$\Delta\varepsilon = \varepsilon_L - \varepsilon_R$$

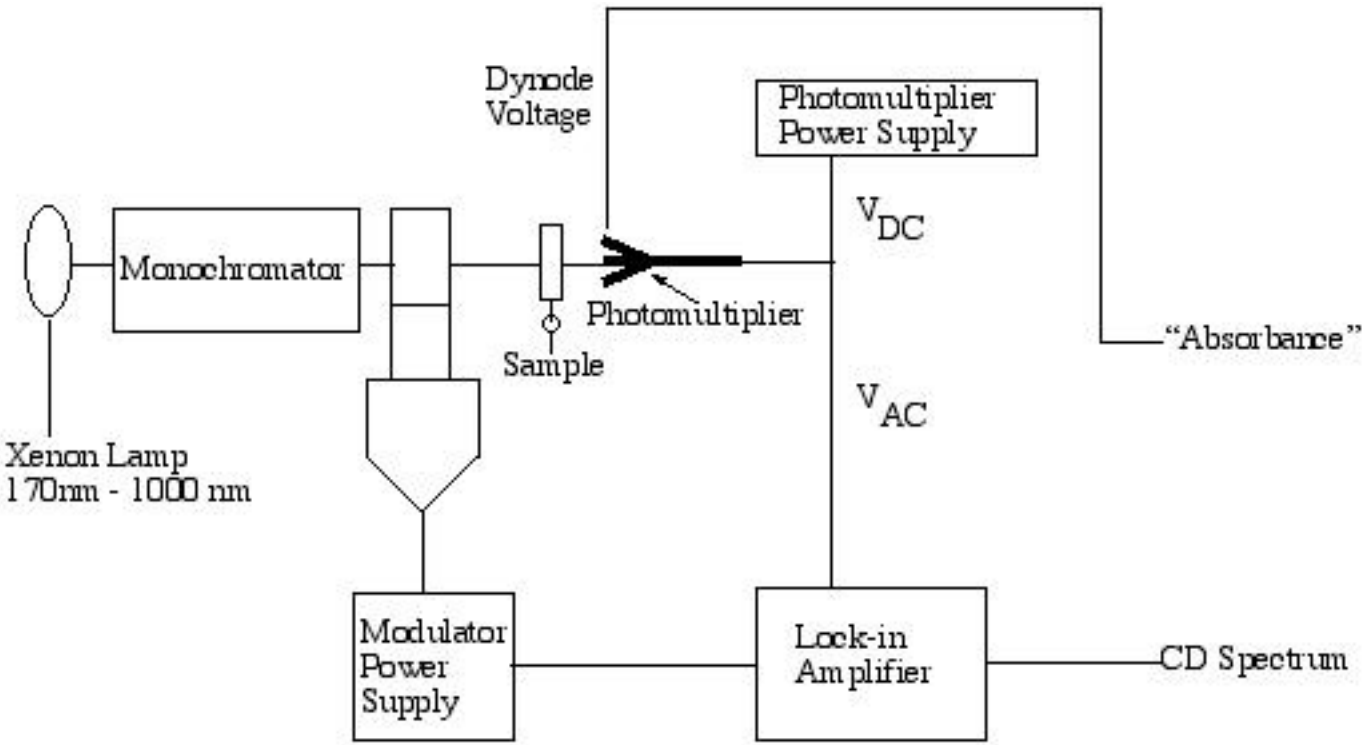
Elliptizität: θ $\quad$ tg θ = b/a

$$\theta = \frac{\ln 10}{4}(A_L - A_R) \cdot \frac{180}{\pi} \quad \text{[Grad]}$$

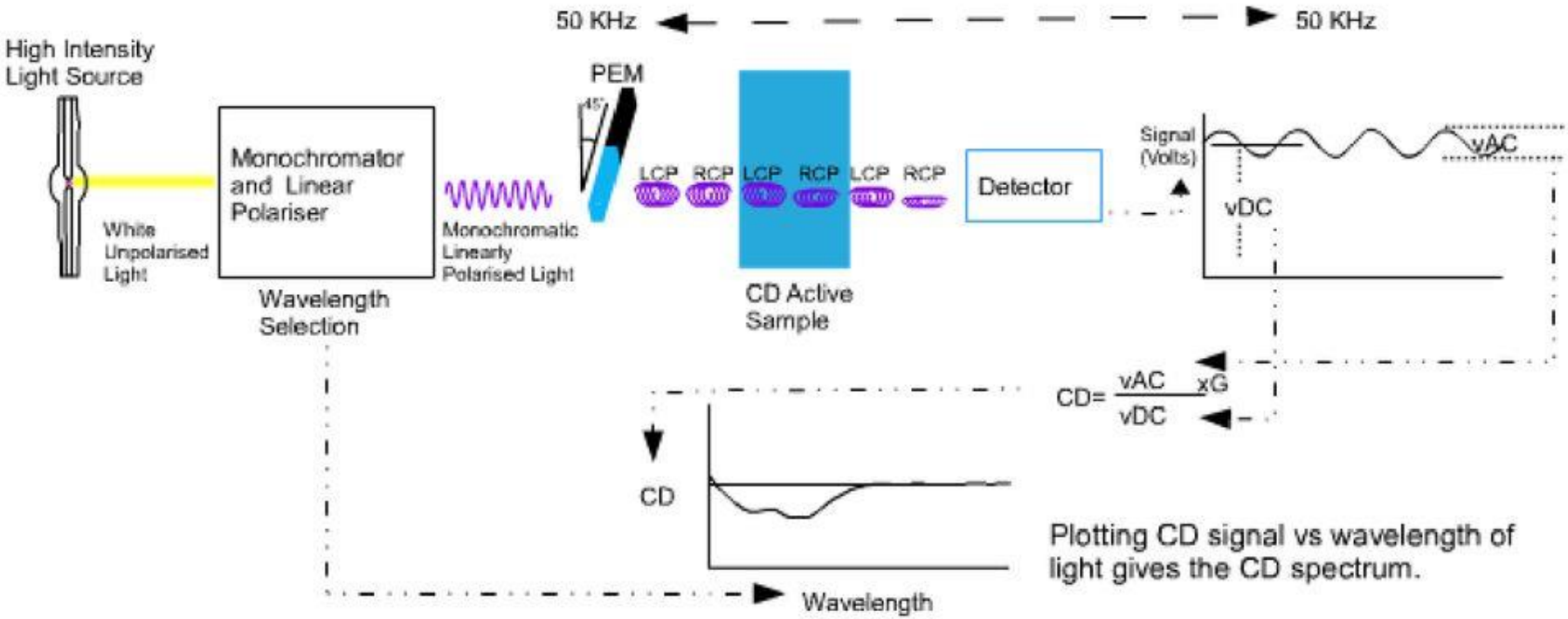
Lambert-Beer ähnliches Gesetz: $\theta = c \cdot l \cdot \theta_m$

(θ_m: molare Elliptizität)

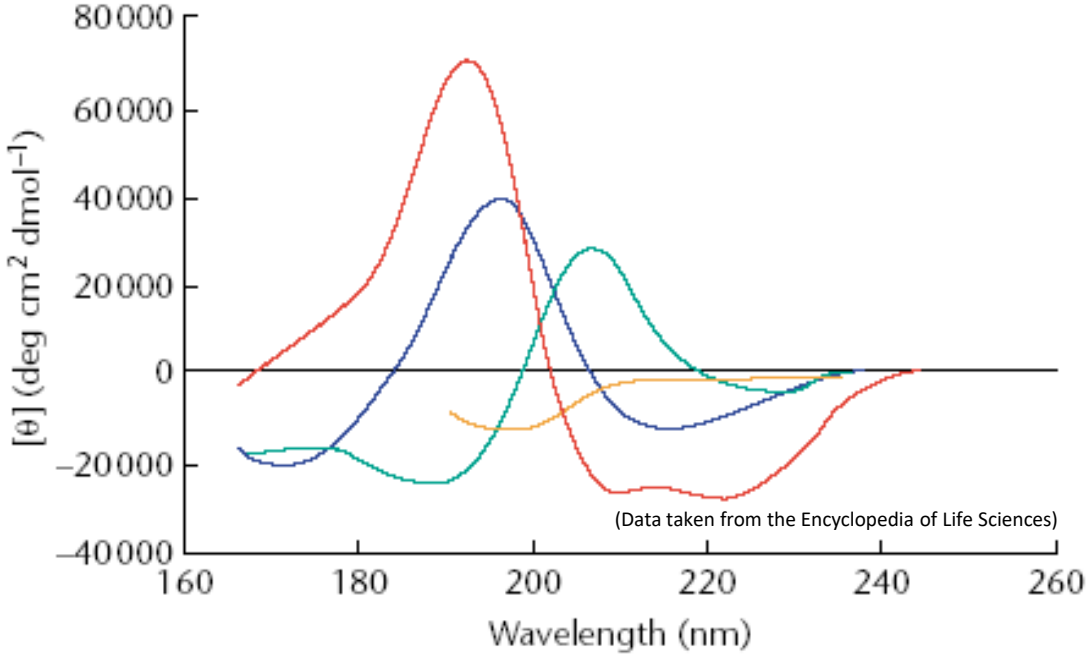
CD-spektrometer



CD spectrophotometer



im tief-UV ist θ empfindlich für die sekundäre Struktur der Eiweißstoffe.



α-helix

Typ I. β-turn

antiparallel β

unstructuriert

BeSTSel

Single spectrum analysis Results

Choose wavelength range:
- (●) 175-250 nm
- ( ) 180-250 nm
- ( ) 185-250 nm
- ( ) 190-250 nm
- ( ) 195-250 nm
- ( ) 200-250 nm

Scale factor 1

Recalculate

Recalculation for the spectrum multiplied by the scale factor in the chosen wavelength range.

Best factor

Dependence of the secondary structure estimation and NRMSD of fitting upon the spectral amplitude (use of NRMSD as hint for correction is adviced only for the 175-250 and 180-250 nm ranges).

Fold recognition

Prediction of fold class, architecture and topology (CATH classification) based on the results of the secondary structure determination. This function is available when the secondary structure is estimated in the Single spectrum analysis.

Back

Back to the starting page. Data will be lost.

References: Micsonai et al. Nucleic Acids Res. 46:W315-22 (2018), Micsonai et al. PNAS 11:E3095-103 (2015)

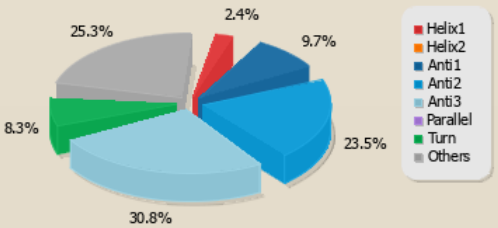
Header

Title		Wavelength range	**175-250 nm**
Date/time	**5-05-2023 19:16 (GMT+1:00)**	Scale factor	**1**

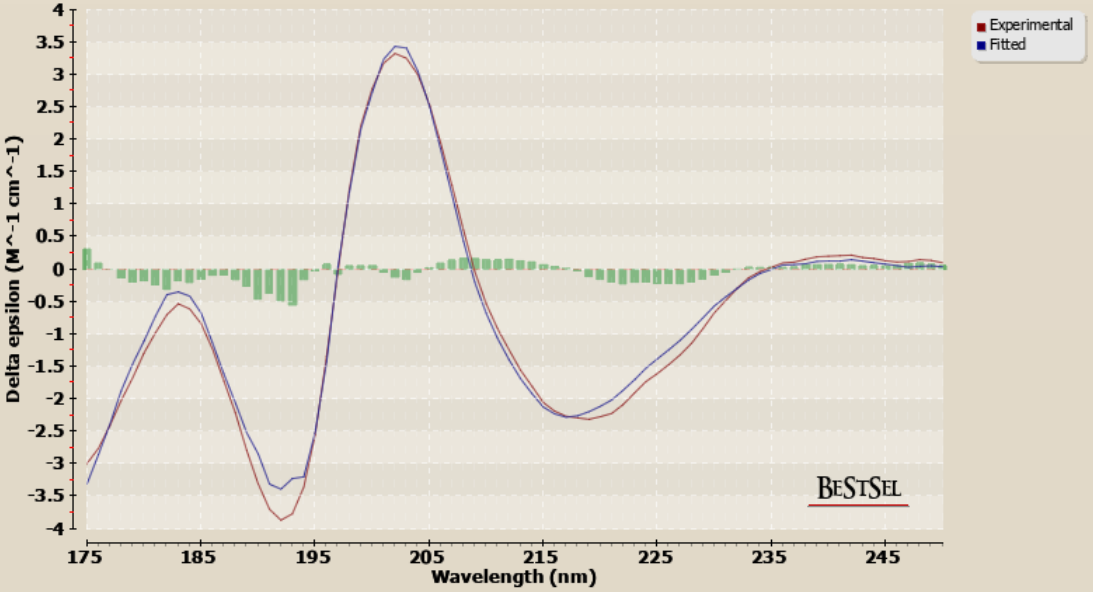
Secondary structure

Estimated secondary structure content (%)

Helix	**2.4**	Helix1 (regular)	**2.4**
		Helix2 (distorted)	**0.0**
Antiparallel	**64.0**	Anti1 (left-twisted)	**9.7**
		Anti2 (relaxed)	**23.5**
		Anti3 (right-twisted)	**30.8**
Parallel	**0.0**		
Turn	**8.3**		
Others	**25.3**		



Fitting results



RMSD	**0.1736**	NRMSD	**0.02414**

Comments

- Input in text window.
- No project title provided. Title will be generated automatically.
- Maximum usable wavelength range: 175-250 nm

Secondary structure

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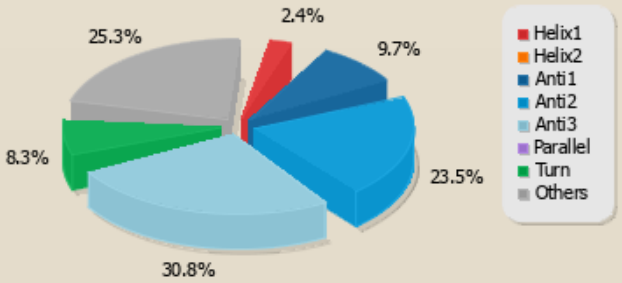
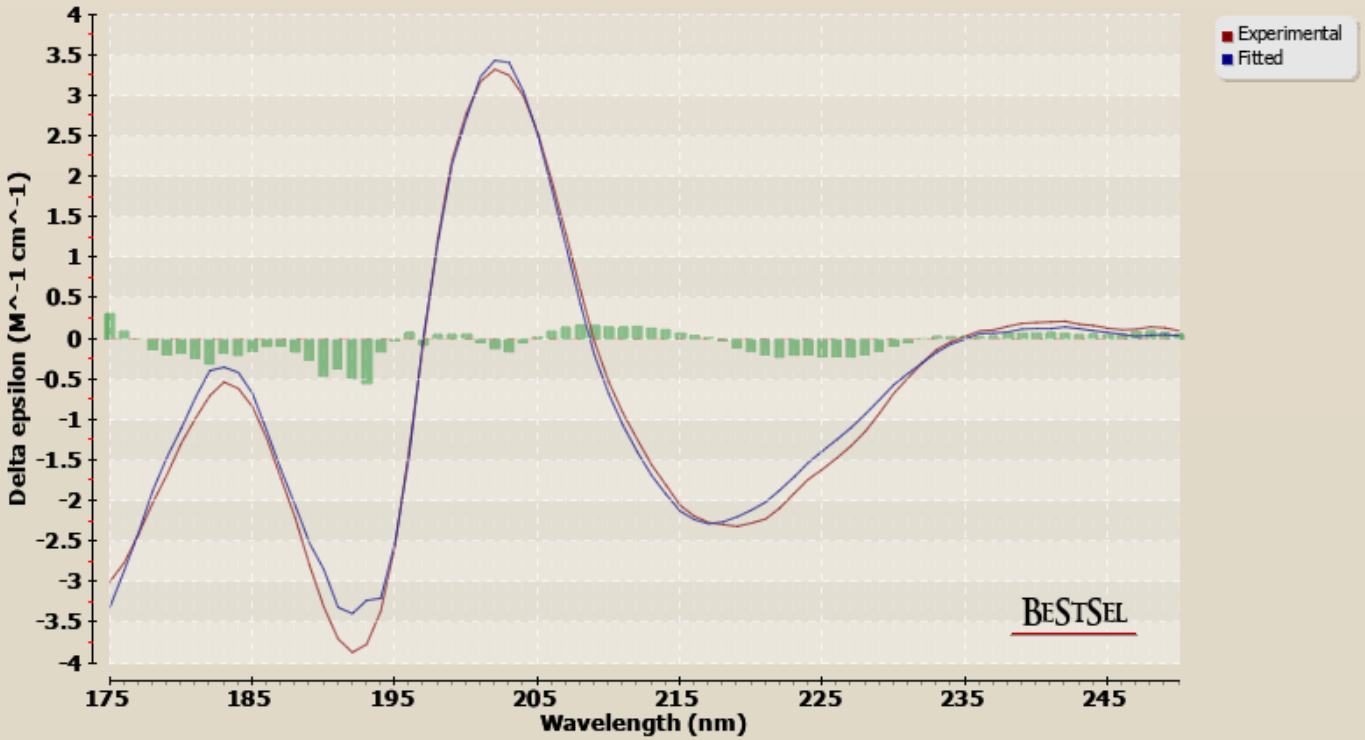


Fitting results



RMSD	0.1736	NRMSD	0.02414

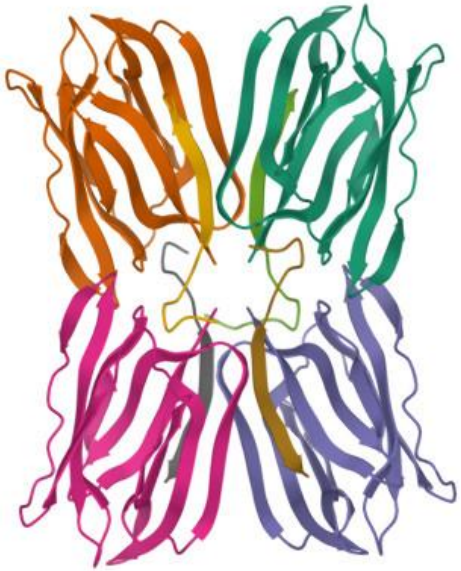
Comments

- Input in text window.
- No project title provided. Title will be generated automatically.
- Maximum usable wavelength range: 175-250 nm

1KU8

jakalin

Zukerbindendes Eiweißstoff.



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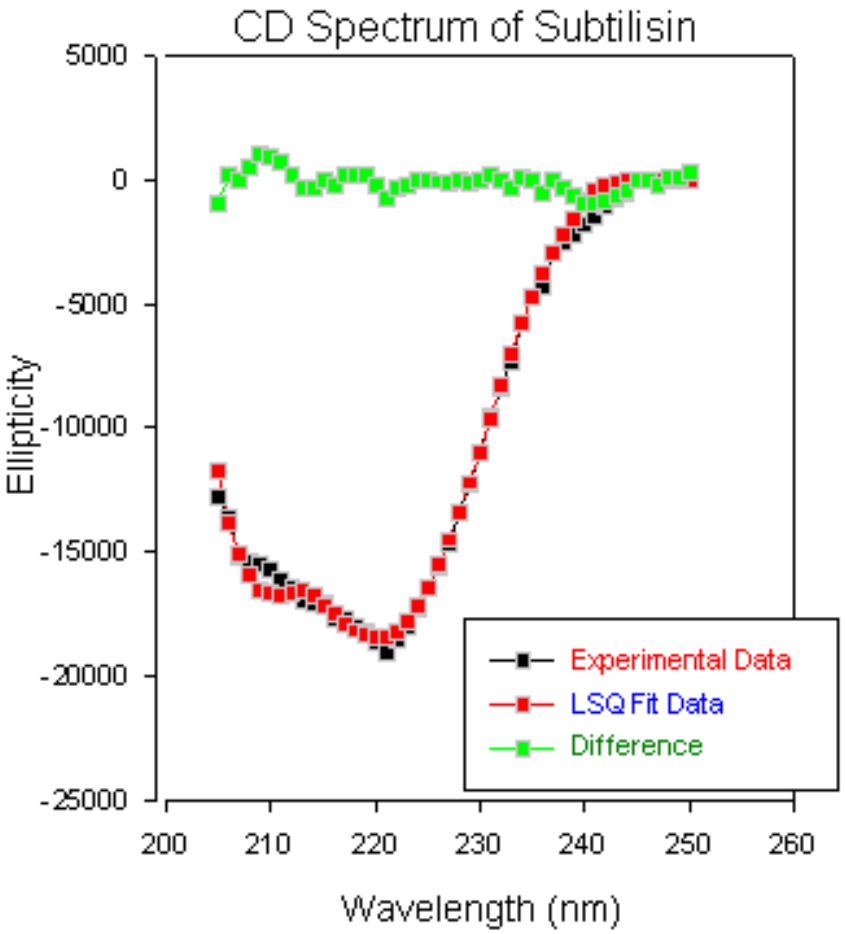
The Structure and CD spectrum of Subtilisin



helix	sheet	coil
57.92	26.22	15.85

A) triosephosphate isomerase
B) hen egg lysozyme
C) myoglobin
D) chymotrypsin