Photometrische Methoden:
UV/VIS Photometer, Atomabsorptionsspektrometrie

Grundlagen 1:

- Lichtmessung
- spezifische Lichtabsorption, Monochromasie
- Zusammenhang: Konzentration und Lichtschwächung das Lambert- Beersche Gesetz

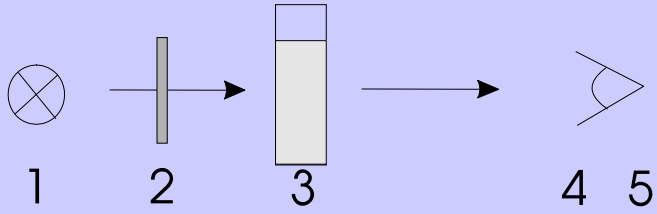
Grundlagen 2:

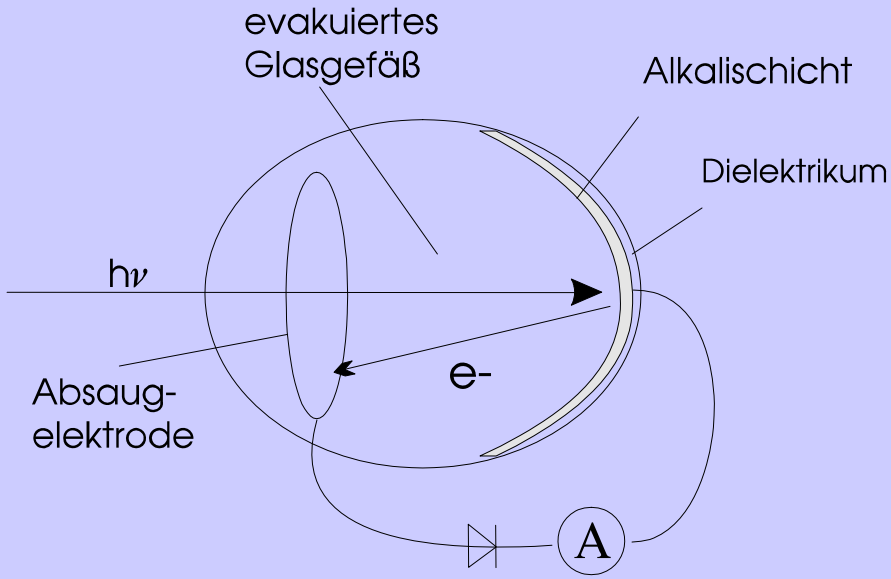
- Atomabsorption und Emission
- Thermische Dissoziation und Ionisation

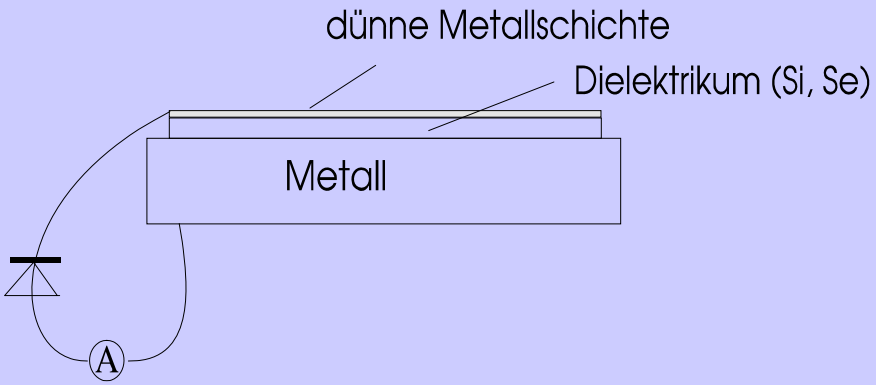
Messpraxis:

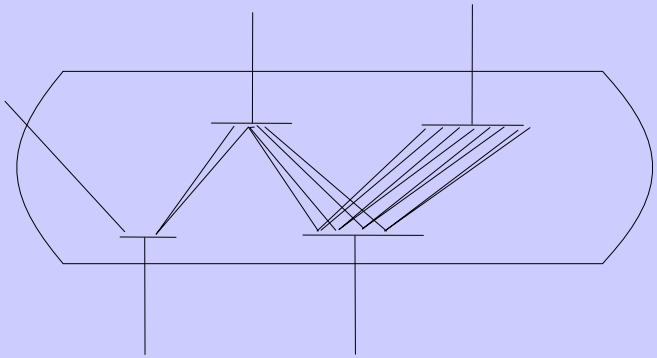
- Konzentrationsmessung (Mikroplatte), Kalibration
- Absorptionsspektrum (Diondenarray)
- Enzymkinetik (Zweistrahlphotometer)
- AAS, Interferenzen bei der AAS

Lichtmessung



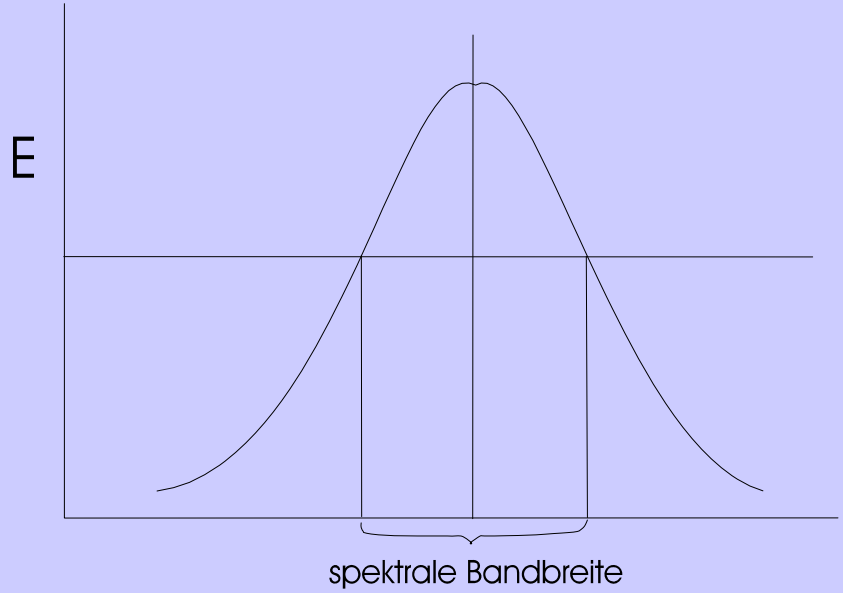


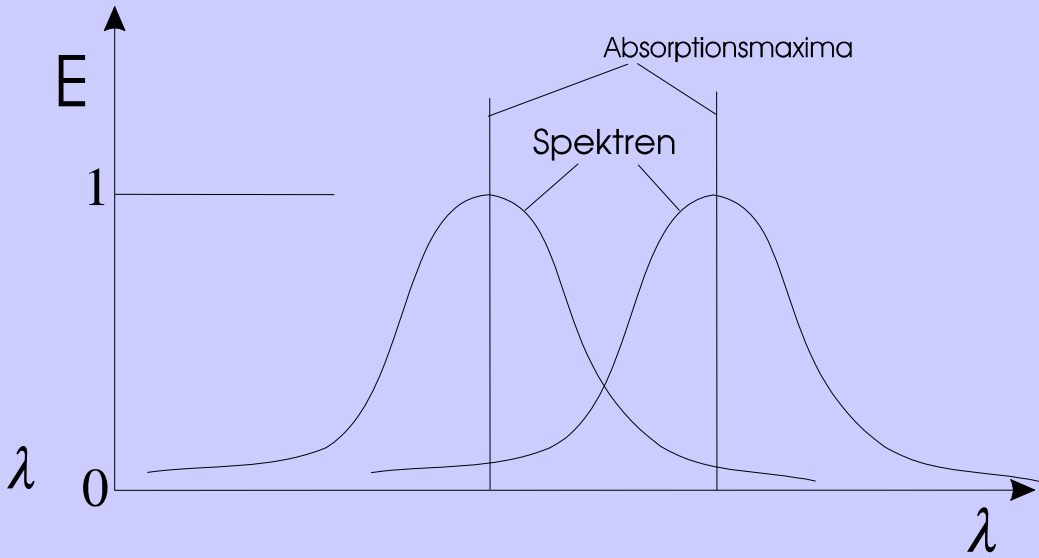


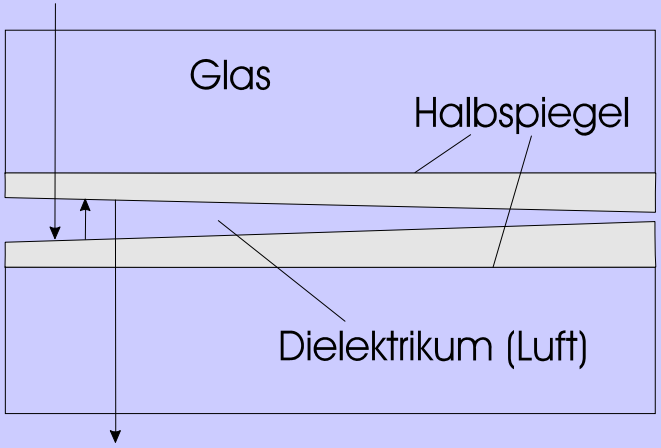


Sekundärelektronenvervielfacher - photomultiplier

Monochromatisches Licht

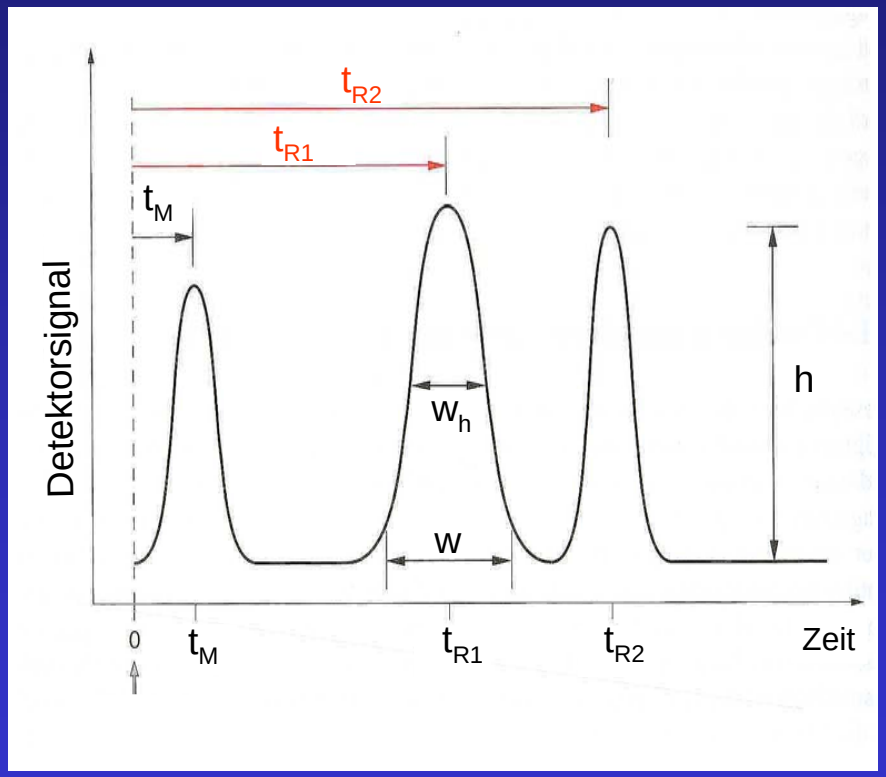






Interferenzverlauffilter

Der Peak



- t_R ... Retentionszeit
- t'_R ... Netto-Retentionszeit
- t_M ... Totzeit
- w ... Basisbreite

Kapazitätsfaktor

- $k' = \frac{t_R - t_M}{t_M} = \frac{t'_R}{t_M}$

Konzentration und Lichtschwächung

$$\frac{I}{I_0} = T \qquad \frac{I}{I_0} \cdot 100 = \%T$$

d	1	2	4	8	16
T%	50	25	12,5	6,25	3,125
T	0,5	0,25	0,125	0,063	0,031
log T	-0,30	-0,60	-0,90	-1,20	-1,51
E (A)	0,30	0,60	0,90	1,20	1,51

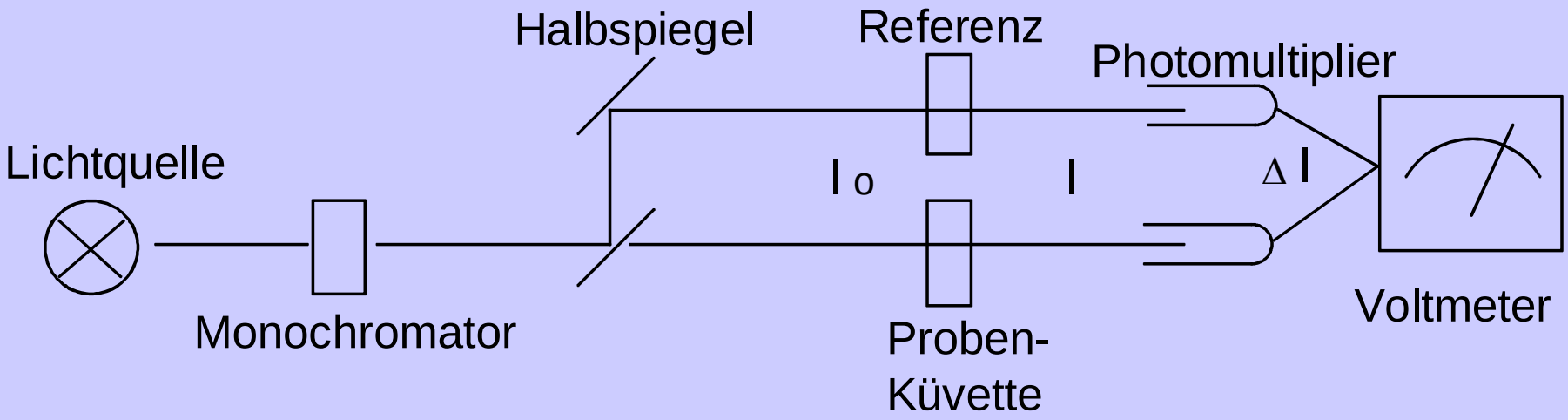
$$\log\left(\frac{I}{I_0}\right)_d = n \cdot \log\left(\frac{I}{I_0}\right)_1 \qquad E = -\log T_d = -\log\left(\frac{I}{I_0}\right)_d = n \cdot \log\left(\frac{I_0}{I}\right)_d$$

$$E = -\log\left(\frac{I}{I_0}\right)_d$$ ...Lambert 1760 $E = \varepsilon_\lambda \cdot c \cdot d$ ...Beer

$$\frac{I}{I_0} = 10^{-\varepsilon_\lambda \cdot c \cdot d}$$

$I = I_0 \cdot 10^{-\varepsilon_\lambda \cdot c \cdot d}$.... Lambert Beer` sches Gesetz

Photometrie



I_0,I ...Intensität
E ...Extinktion bzw. A ...Absorption
ε_λ ...molarer Extinktions- bzw. Absorptionskoeffizient
d ...Schichtdicke der Küvette
c ...Konzentration

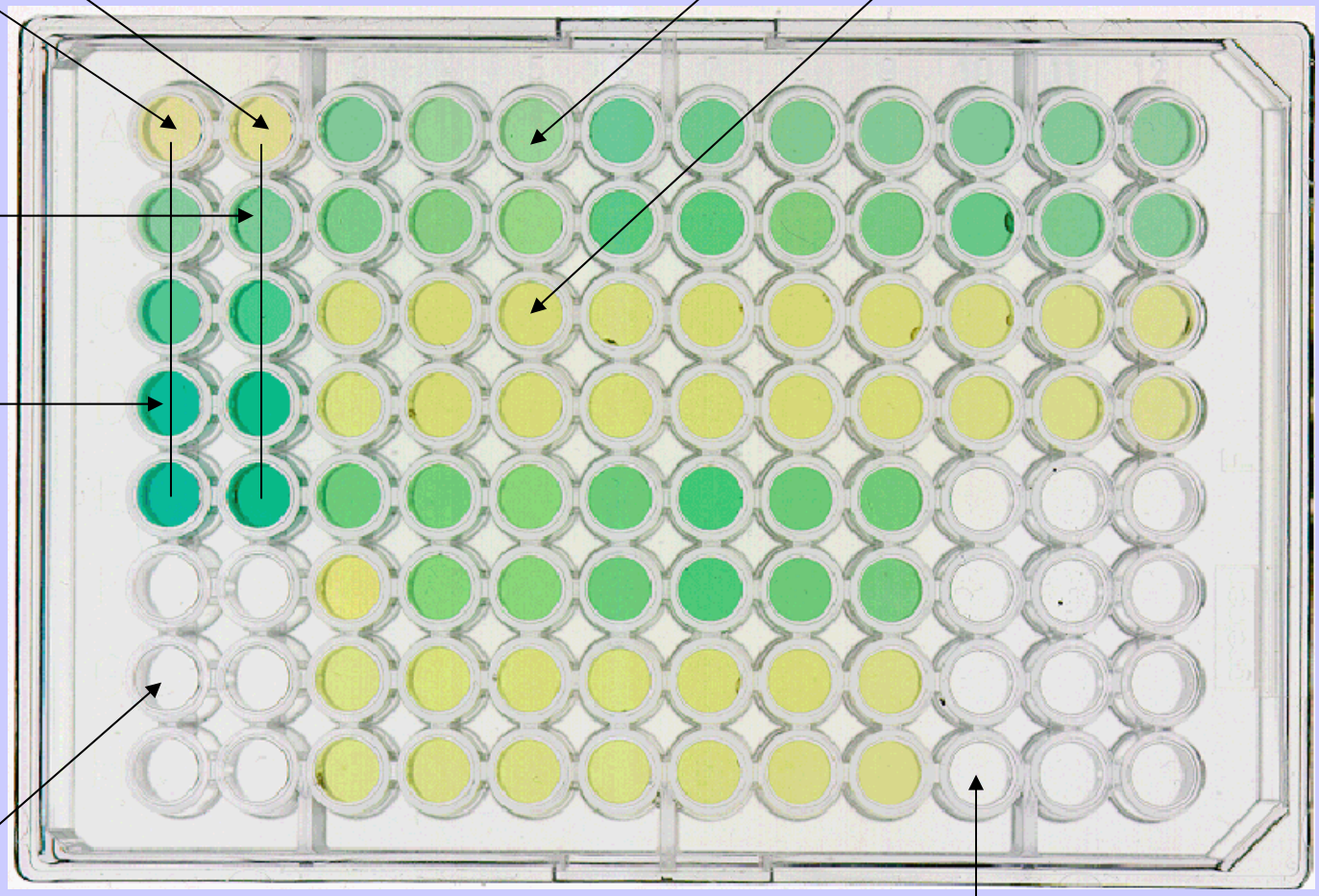
$$A = \lg \frac{I_o}{I} = \varepsilon_\lambda \, c \, d$$ ...*Lambert und Beer*

cProbe / aProbe = cRef / a Ref

Mikroplattenphotometer

Vollproben
Blindproben

Reagenzienleerwerte



Kalibrationsreihen

Reines Lösungsmittel (A.demin.)

V = konstant!

Auswertung (Rechnung) über Kalibrationskurve

Position	Sample ID	Abs	Conc
A 1	Kalibrationspunkt 1	1	2
B 1	Kalibrationspunkt 2	2	3
C 1	Kalibrationspunkt 3	3	5
D 1	Kalibrationspunkt 4	4	7
E 1	Kalibrationspunkt 5	5	9
F 1			
G 1			
H 1			

y´ = kx + d

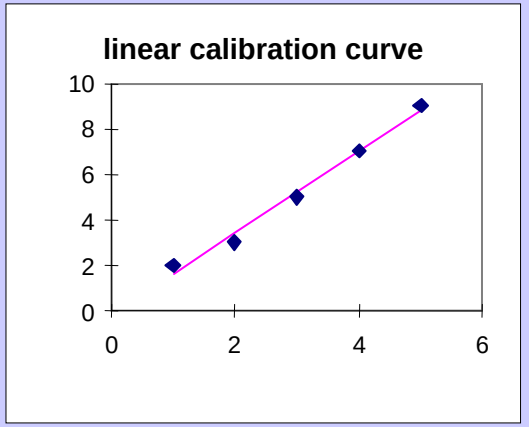
k = 1,80

d = -0,2

r = 0,9938837

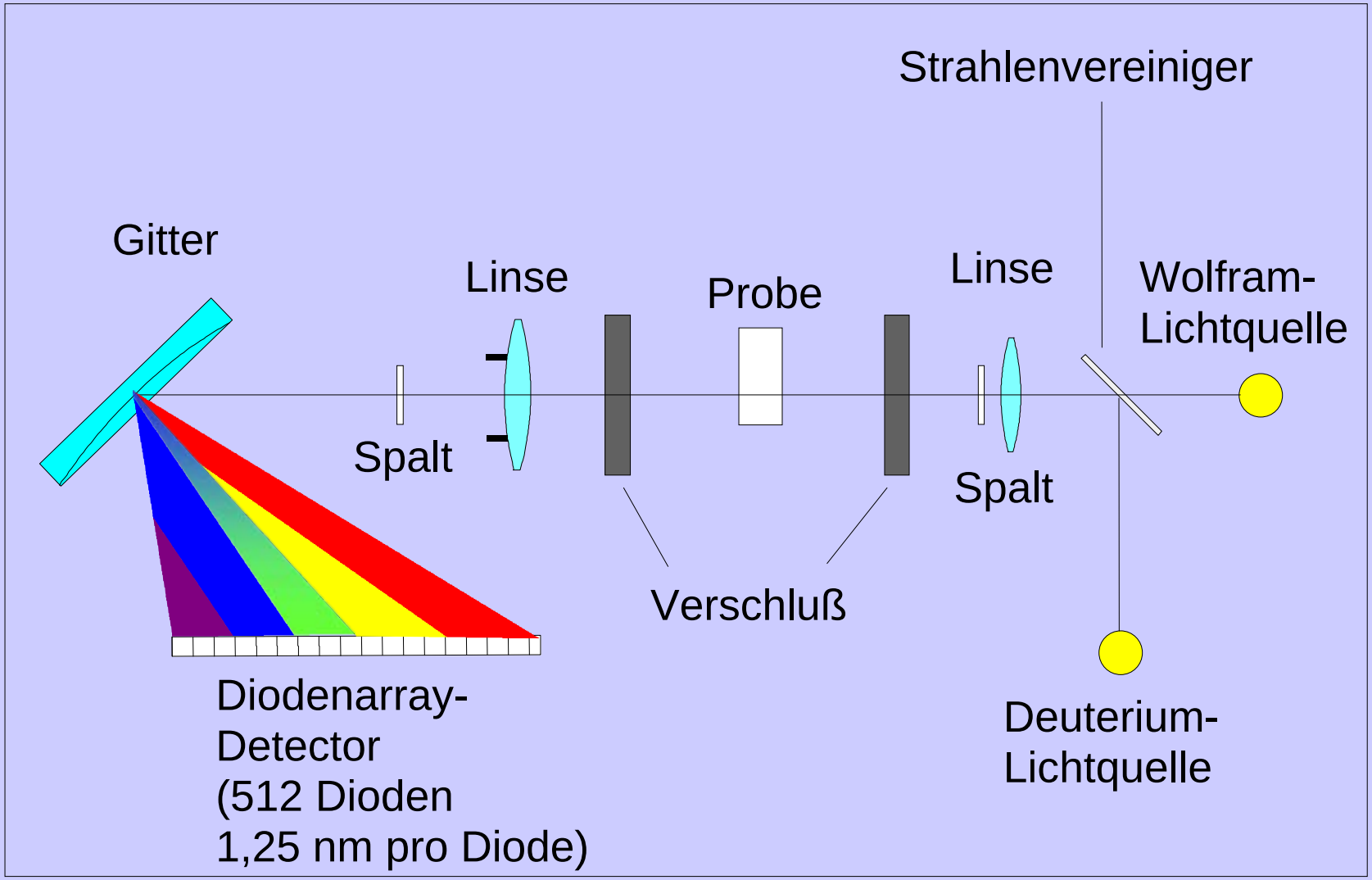
Factor	Abs	Conc
1	1	**1,6**

Copy this formula to column D to calculate the y´ values!

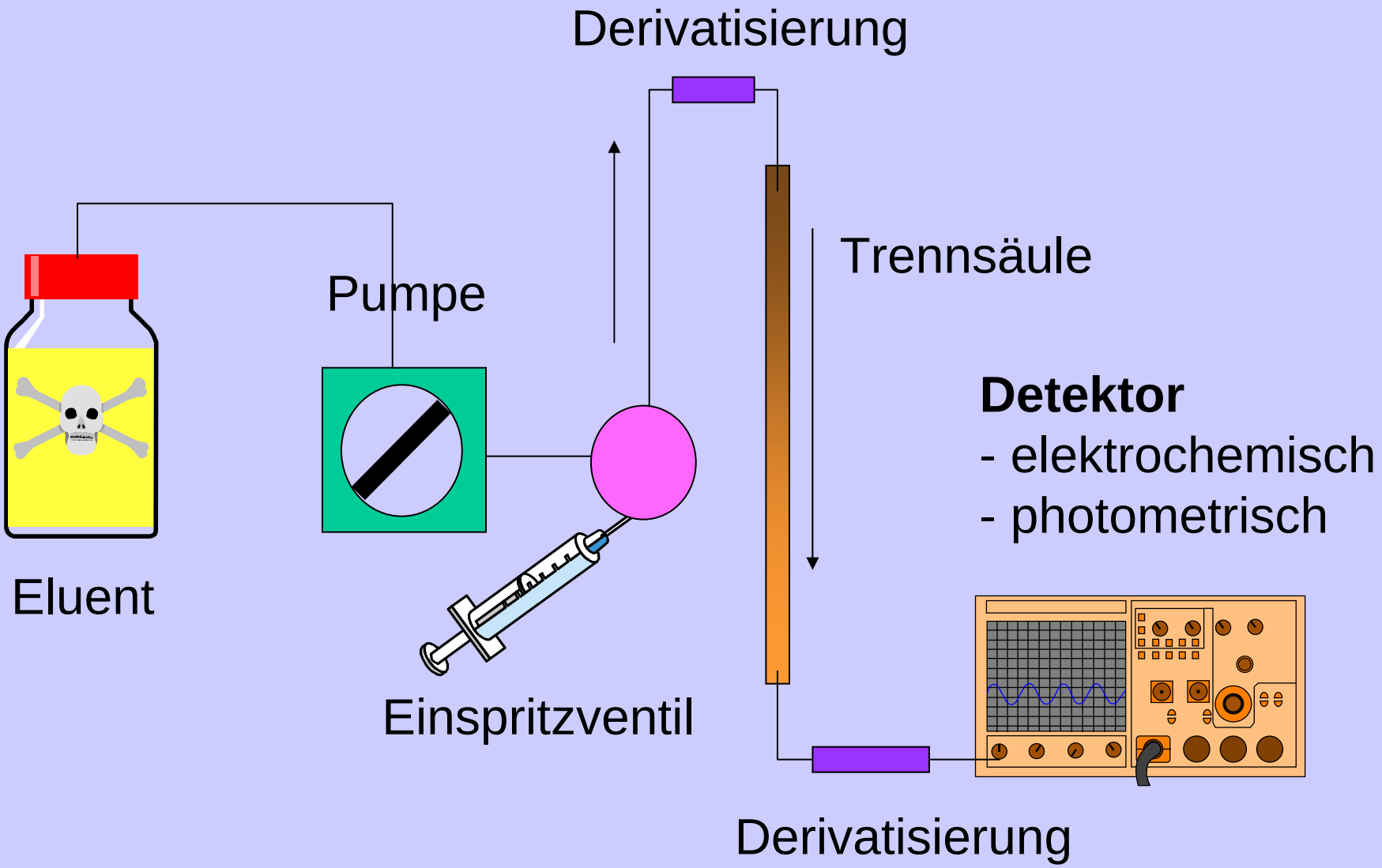


y'
1,6
3,4
5,2
7
8,8
-0,2
-0,2
-0,2

DIODENARRAY- SPEKTRALPHOTOMETER

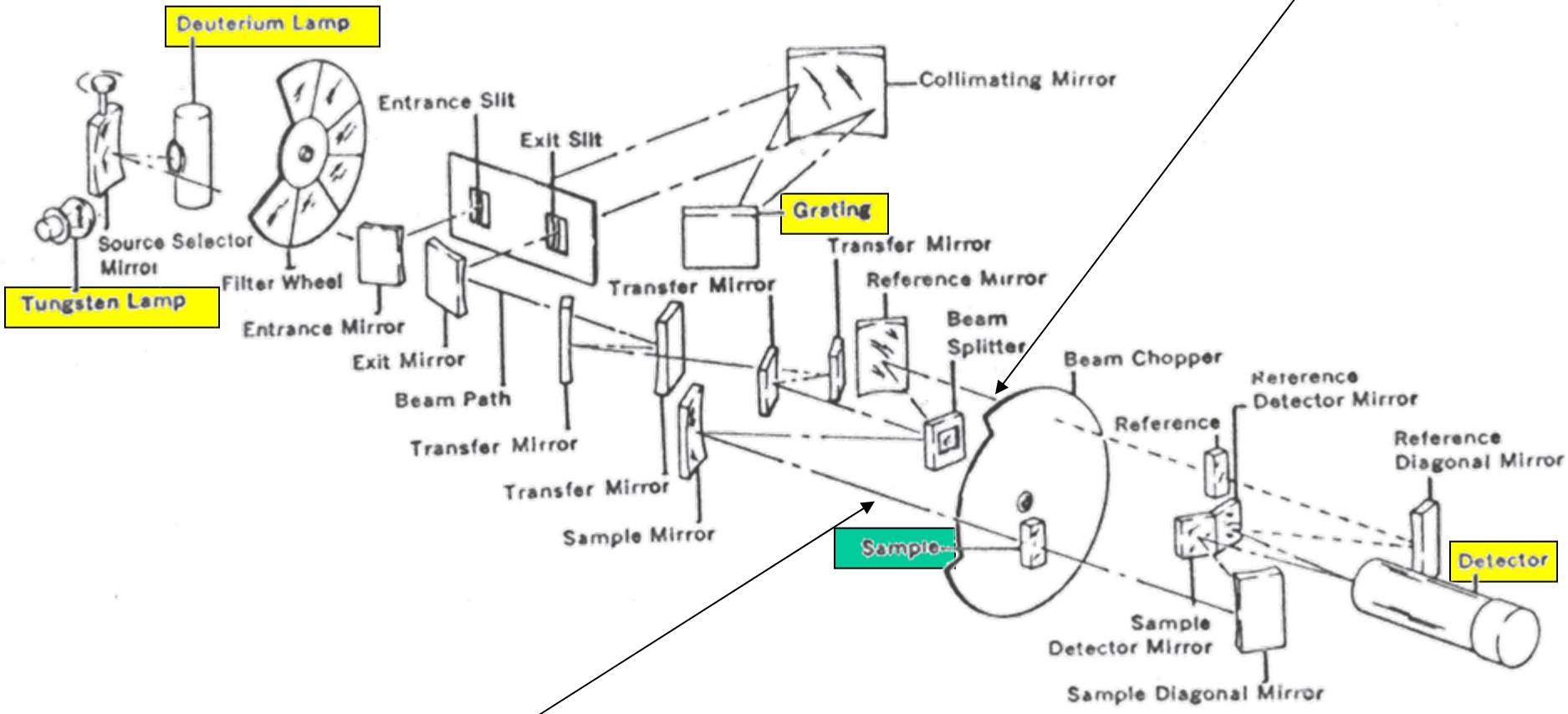


Flüssigkeitschromatographie

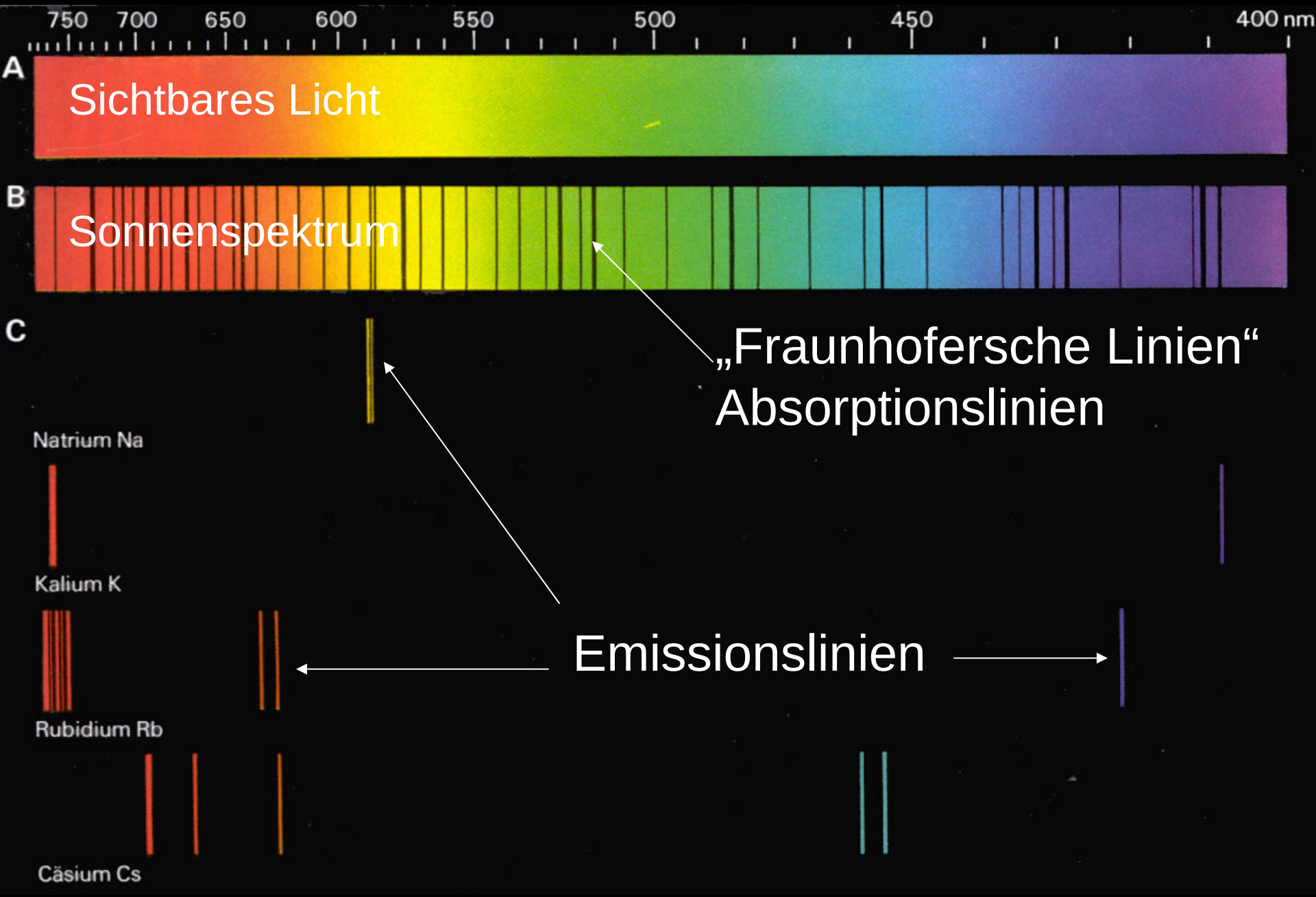


Zweistrahlphotometer:

Referenzstrahlengang



Probenstrahlengang


750 700 650 600 550 500 450 400 nm
A
Sichtbares Licht
B
Sonnenspektrum
C
„Fraunhofersche Linien“
Absorptionslinien
Natrium Na
Kalium K
Emissionslinien
Rubidium Rb
Cäsium Cs

Temperatur und Licht

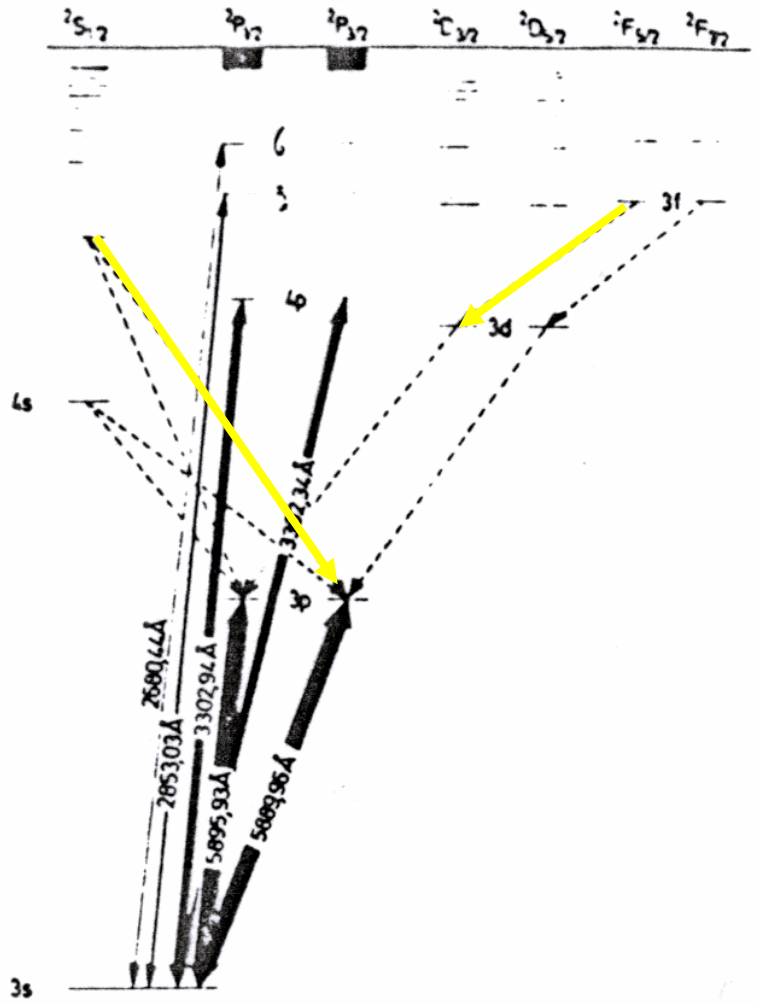
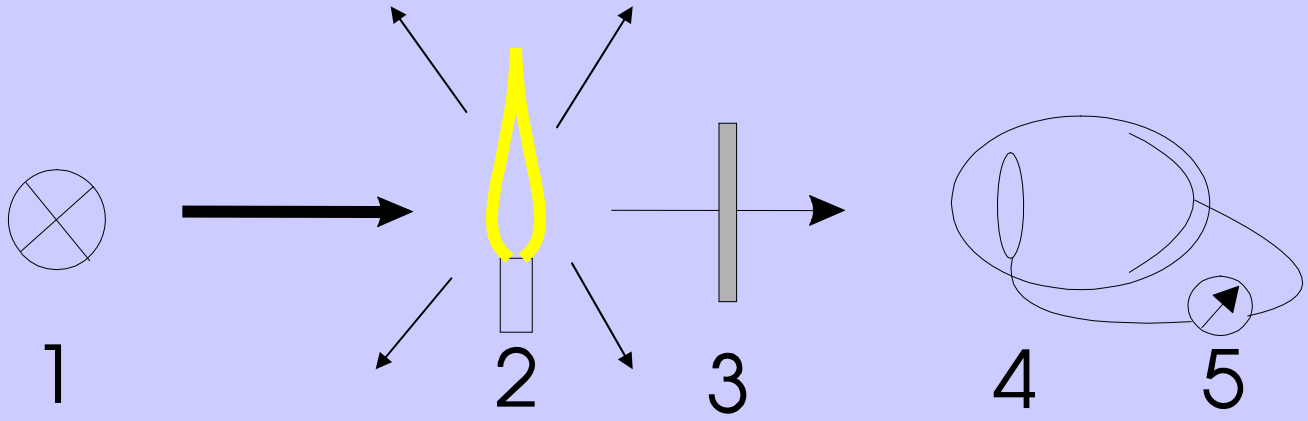


Abb. 3. Termschema des Natriums – Die mit Doppelpfeilen versehenen, durchgezogenen Linien sind die ersten Übergänge der Hauptserie und treten in Absorption und Emission auf. Die gestrichelten Linien gehören zu den verschiedenen Nebenserien und treten unter den hier interessierenden Temperaturverhältnissen nur in Emission auf.

Flammenphotometrie, Atomabsorption

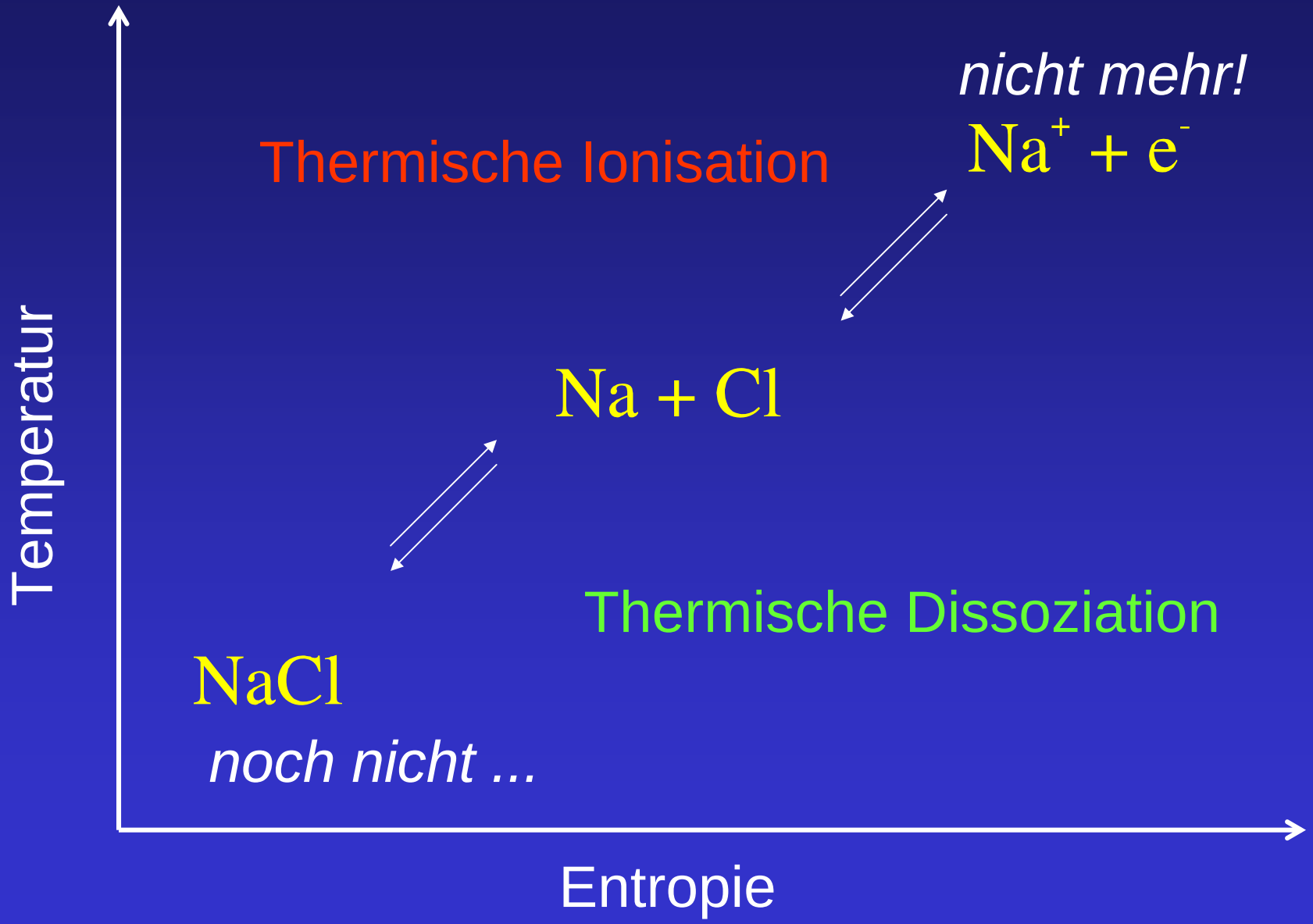


As	Cu	Sr	Li	Cs
197,7	324,7	460,7	670,8	852,1

UV	PHAR	IR

Fe	Ti	Na	K
248,3	365,3	589,6	766,5

AAS - Meßbedingungen



Flammen, Gasgemische

Flamme	Temperatur	geeignet für
Propan/Luft	1930	A (Na,K) es sollen möglichst viele Atome im Grund-zustand bleiben
Acetylen/Luft	2300	A (E)
Acetylen/N_2O	2700	A
H_2/O_2	2660	E
Acetylen/O_2	3100	E
$(CN)_2/O_2$	4500	(E) schon zu starke thermische Ionisation

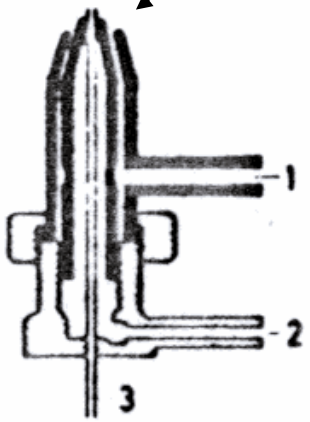
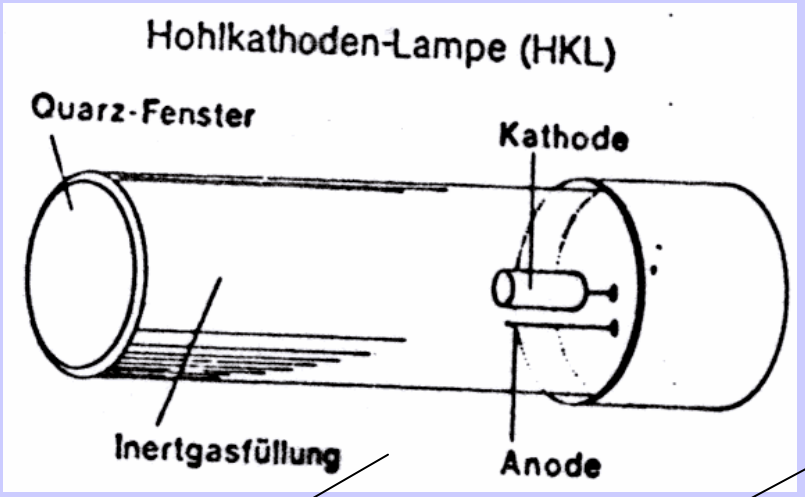
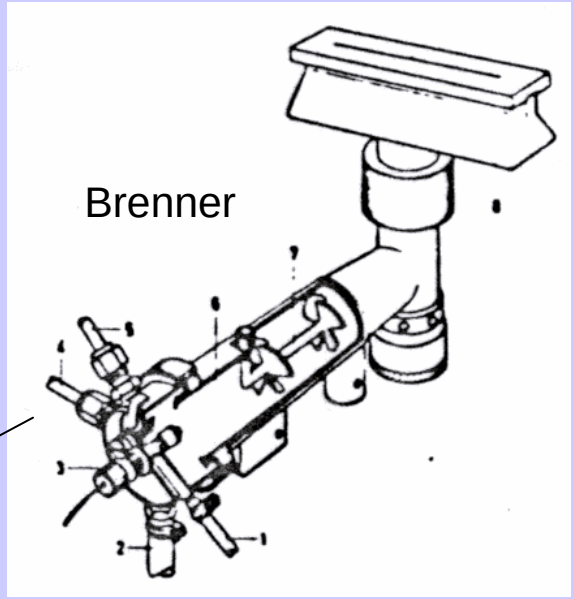
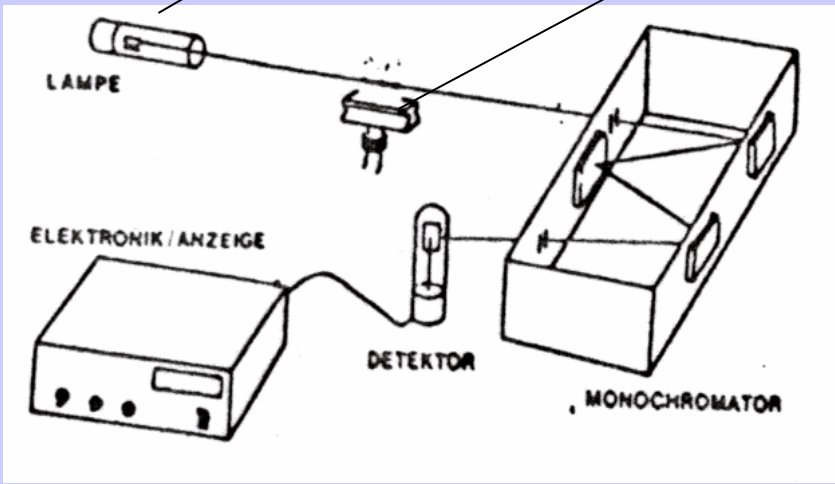


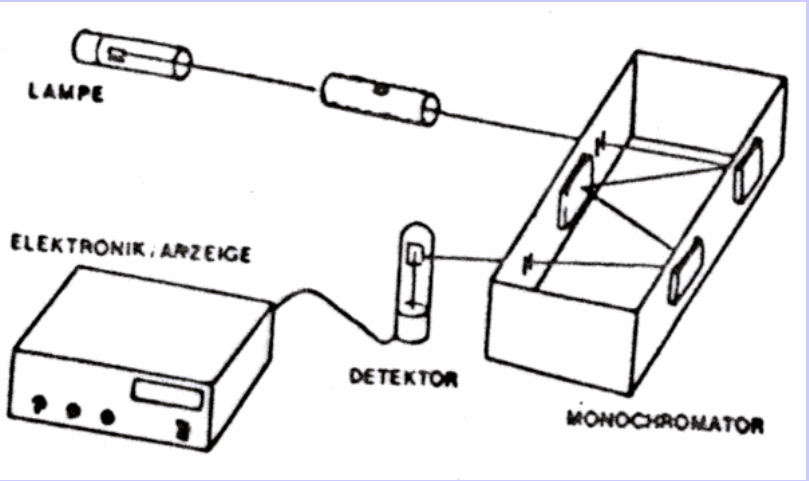
Abb. 15. Direktzerstäubender Turbulenzbrenner. – 1 Brenngaszufuhr, 2 Oxidans-Zufuhr, 3 Ansaugkapillare für die Probenlösung.

AAS - Atomabsorptionsspektroskopie









DCP und ICP/ICPMS

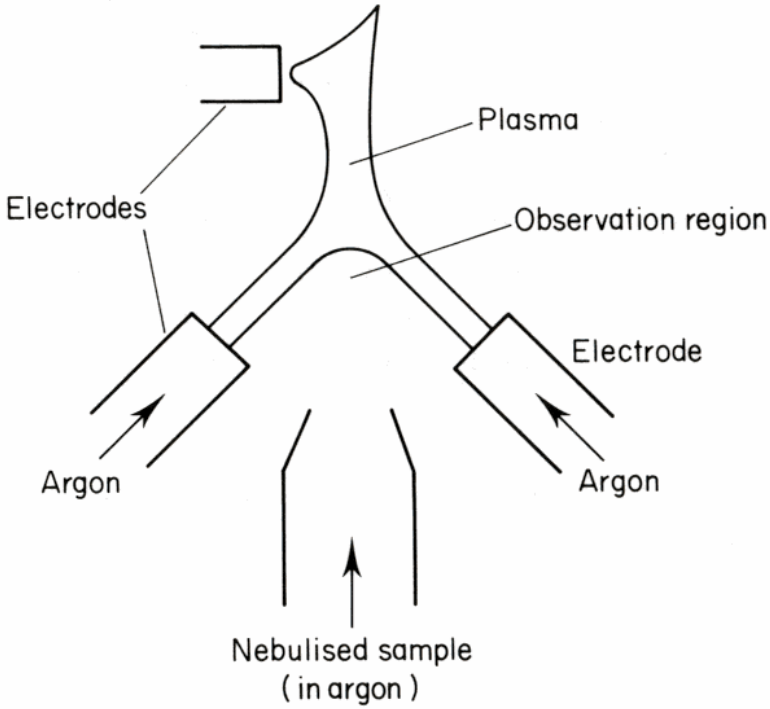


Fig. 7.4a. *Three electrode direct current plasma*

The plasma has a characteristic doughnut shape with the sample being introduced into a relatively cool (6000–8000 K) hole in the centre of the doughnut (see Frontispiece). The light emission from the white hot fireball of the plasma is very intense, so analytical measurements are made in the cooler tail plume of the plasma. The spectral background in the plume, particularly in the useful 190–300 nm region, is relatively simple, consisting mainly of argon emission lines.

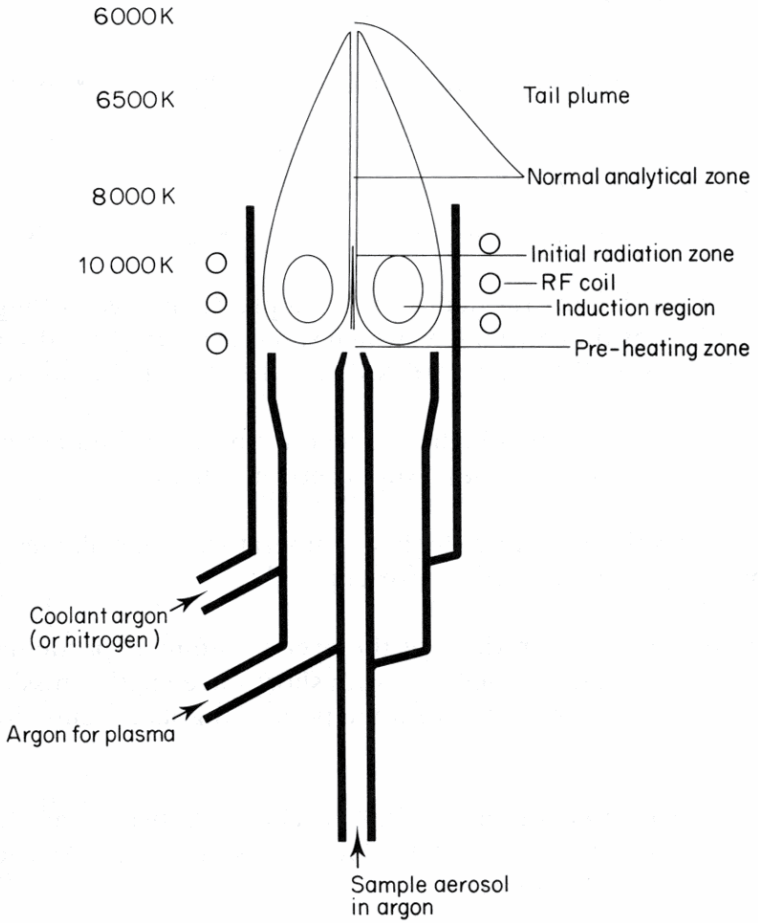


Fig. 7.6b. *An ICP torch*

Atomabsorption: Interferenzen

Art	**Abhilfe**
Chemische I.	Flammenwahl, Fällungen
Ionisations-I.	Strahlungspuffer (CsCl)
Spektrale I.	Andere Wellenlänge
Physikalische I.	Nullpunktabgleich, **Matrix-** abgleich (Viskosität !) Additionsmethode - Spiken