TFYA78 Molekylfysik
Föreläsning 13a – Bilder
Fotofysikaliska och fotokemiska processer

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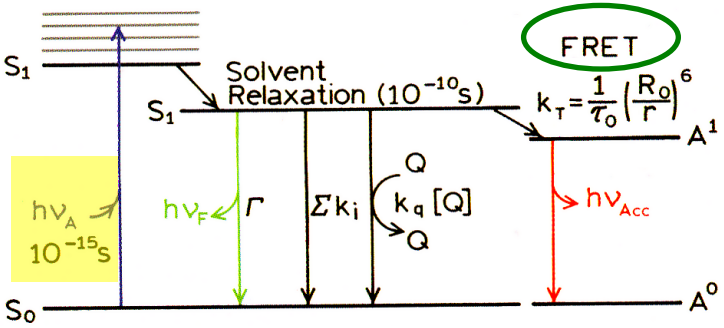
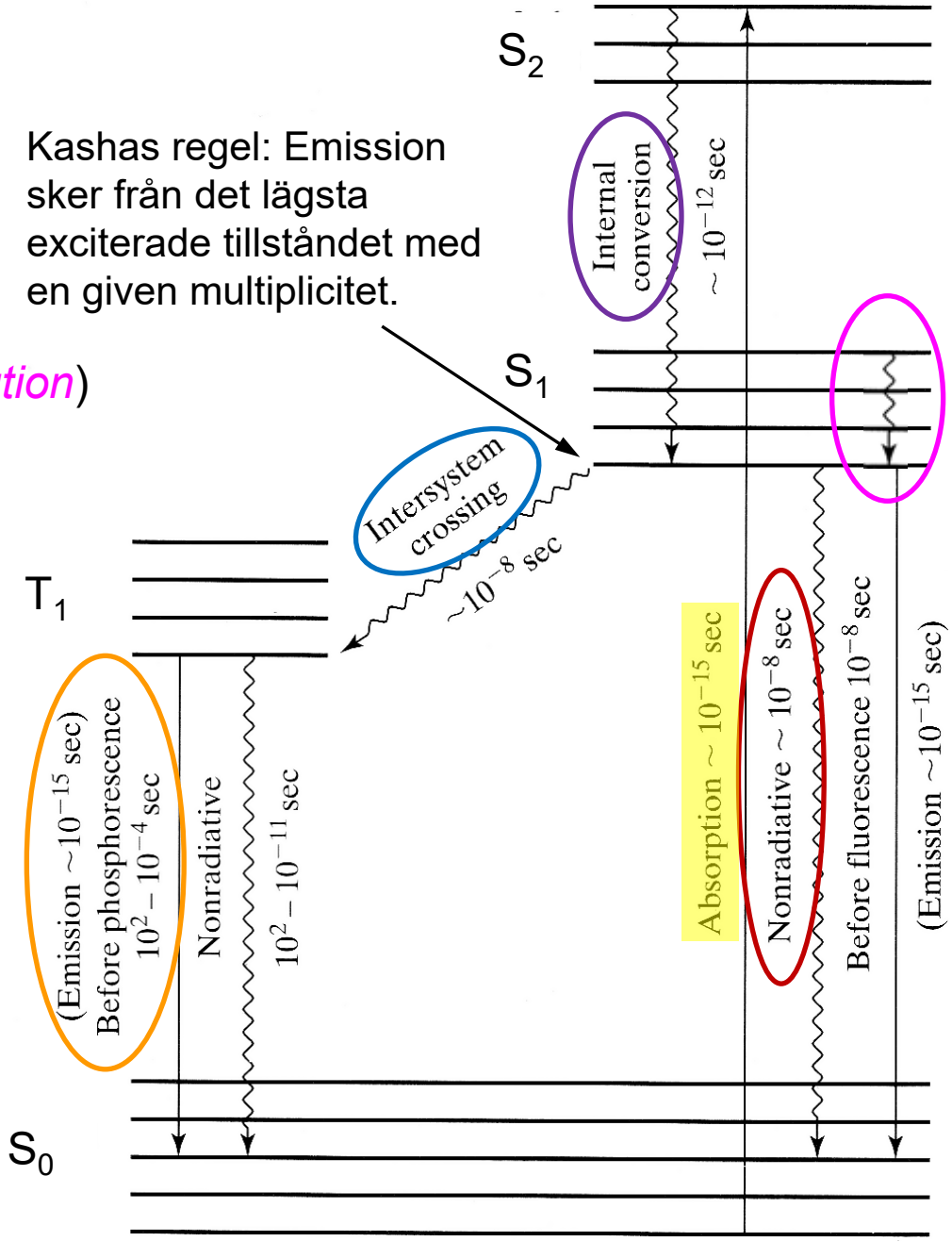
Vad händer efter en elektronexcitation?

- Intern omvandling $S_n \rightarrow S_1$ (*internal conversion*)
- Vibrationsdeaktivering (*intramolecular vibrational redistribution*)
- Resonant energiöverföring (*resonance energy transfer*, FRET)
- Livstidsförkortning, utan emission (*quenching*)
- Övergång (tunnling) till triplett-tillstånd (*intersystem crossing*) → fosforescens.



Fluorescens } Exempel på
Fosforescens } luminiscens

Kashas regel: Emission sker från det lägsta exciterade tillståndet med en given multiplicitet.



Fotofysikaliska processer

Primary absorption	$S + h\nu \rightarrow S^*$
Excited-state absorption	$S^* + h\nu \rightarrow S^{**}$
	$T^* + h\nu \rightarrow T^{**}$
Fluorescence	$S^* \rightarrow S + h\nu$
Stimulated emission	$S^* + h\nu \rightarrow S + 2h\nu$
Intersystem crossing (ISC)	$S^* \rightarrow T^*$
Phosphorescence	$T^* \rightarrow S + h\nu$
Internal conversion (IC)	$S^* \rightarrow S$
Collision-induced emission	$S^* + M \rightarrow S + M + h\nu$
Collisional deactivation	$S^* + M \rightarrow S + M$
	$T^* + M \rightarrow S + M$
Electronic energy transfer:	
Singlet–singlet	$S^* + S \rightarrow S + S^*$
Triplet–triplet	$T^* + T \rightarrow T + T^*$
Excimer formation	$S^* + S \rightarrow (SS)^*$
Energy pooling	
Singlet–singlet	$S^* + S^* \rightarrow S^{**} + S$
Triplet–triplet	$T^* + T^* \rightarrow S^* + S$

Redan behandlat!

→ *Exciplex* om två olika molekyler ingår

† An asterisk (*) denotes an excited state, ** a more excited state, S a singlet state, T a triplet state, and M a third body.

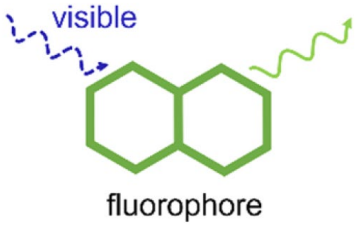
Kvantutbyte:
Antal gånger en händelse inträffar, per absorberad foton.

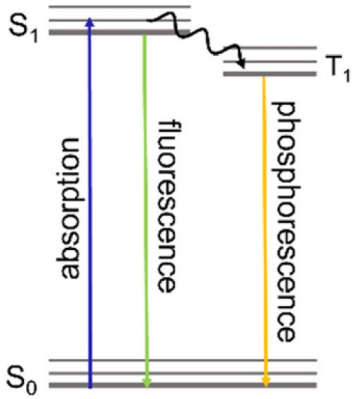
T.ex. för fluorescens:

$$\Phi = \frac{\text{Emitterade fotoner}}{\text{Absorberade fotoner}}$$

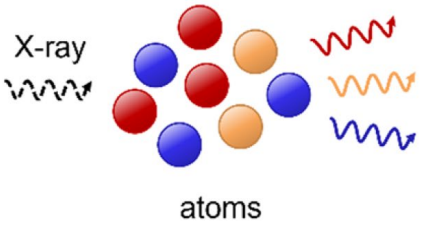
”Fluorescens” är inte begränsat till synligt ljus

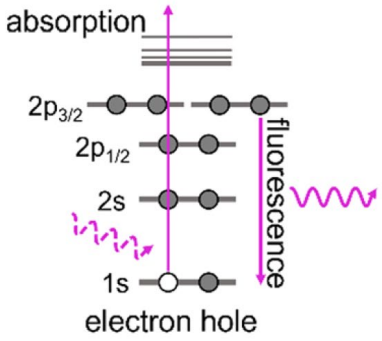
Optisk fluorescens



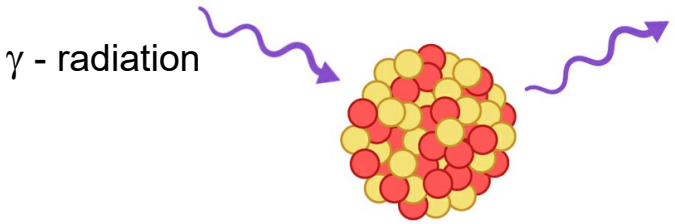


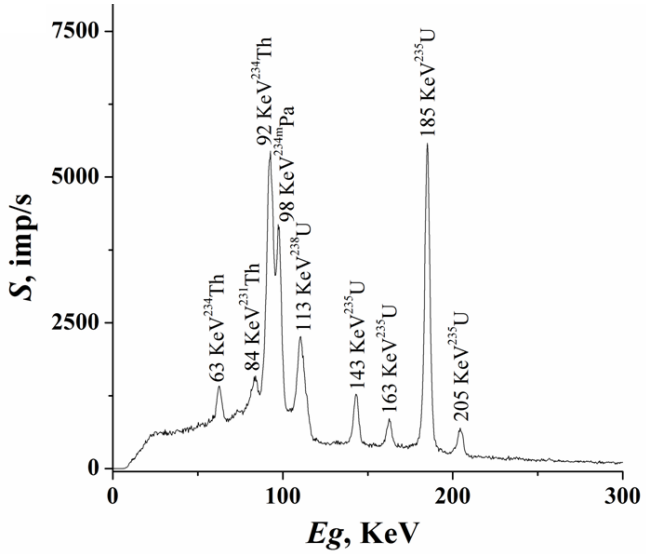
Röntgenfluorescens



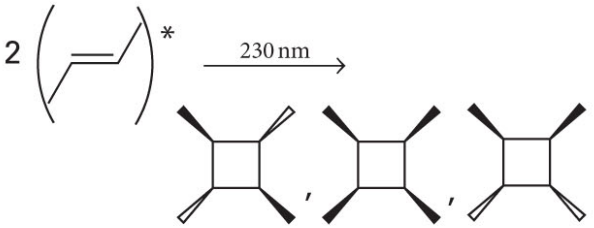
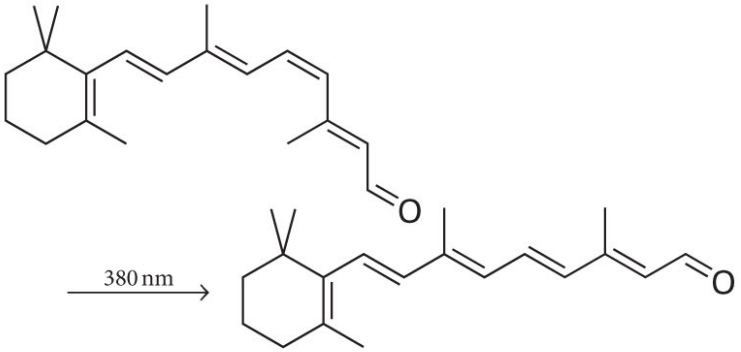


Nukleär fluorescens
(*Nuclear Resonance Fluorescence,* NRF)





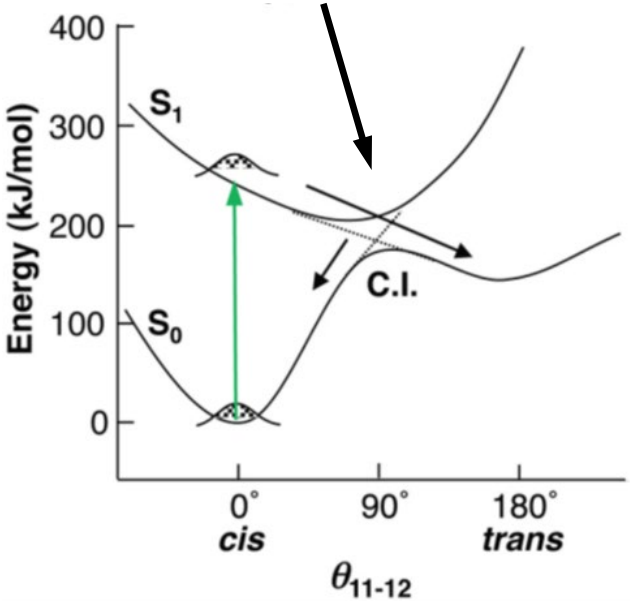
Fotokemiska processer

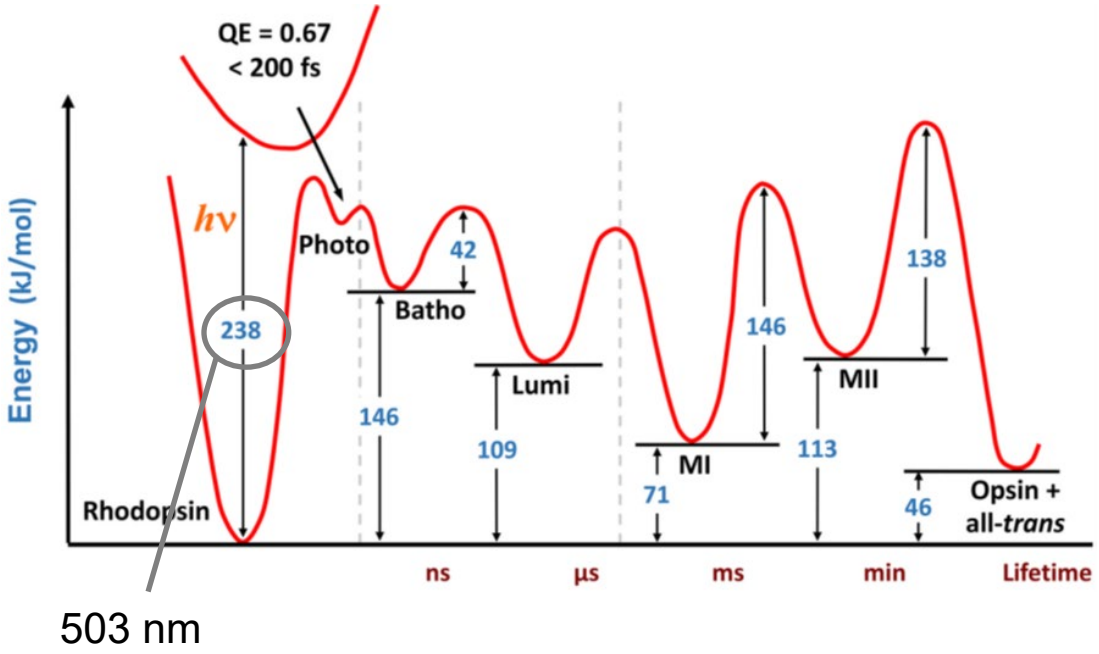
Process	General form	Example
Ionization	$A^* \rightarrow A^+ + e^-$	$NO^* \xrightarrow{134\ nm} NO^+ + e^-$
Electron transfer	$A^* + B \rightarrow A^+ + B^-$ or $A^- + B^+$	$[Ru(bpy)_3]^{2+*} + Fe^{3+} \xrightarrow{452\ nm} [Ru(bpy)_3]^{3+} + Fe^{2+}$
Dissociation	$A^* \rightarrow B + C$	$O_3^* \xrightarrow{1180\ nm} O_2 + O$
	$A^* + B{-}C \rightarrow A + B + C$	$Hg^* + CH_4 \xrightarrow{254\ nm} Hg + CH_3 + H$
Addition	$2\ A^* \rightarrow B$	
	$A^* + B \rightarrow AB$	
Abstraction	$A^* + B{-}C \rightarrow A{-}B + C$	$Hg^* + H_2 \xrightarrow{254\ nm} HgH + H$
Isomerization or rearrangement	$A^* \rightarrow A'$	

* Excited state.

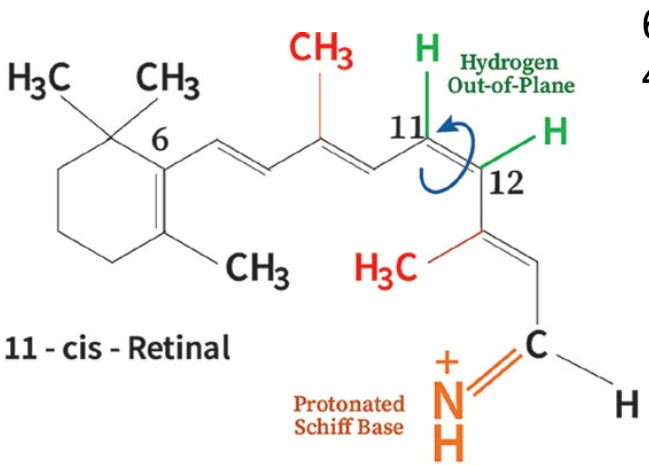
Fotosiomerisering av 11-*cis*-retinal

conical intersection/
molecular funnel





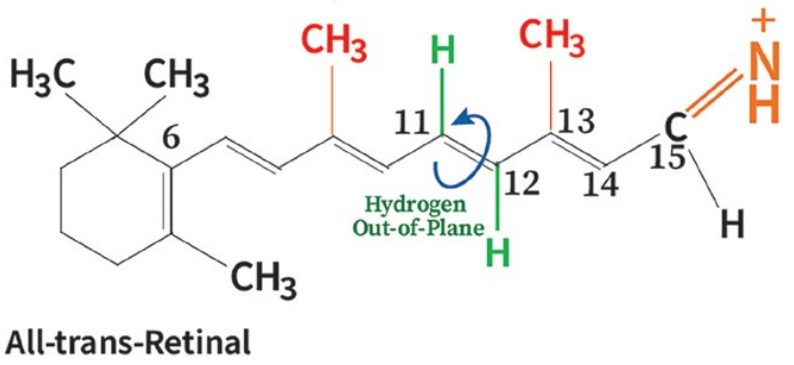
503 nm



11 - cis - Retinal

610 nm i gasfas
440 nm i MeOH

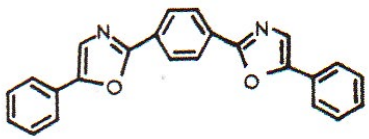
$\pi \rightarrow \pi^*$



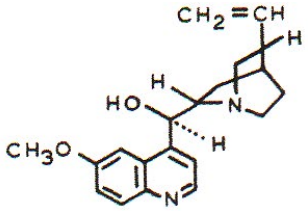
All-trans-Retinal

⇨ Aktivering av rhodopsin (GPCR)

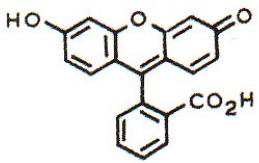
Några exempel på fluoroforer...



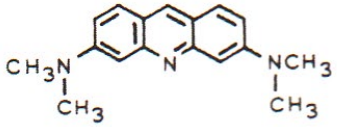
POPOP



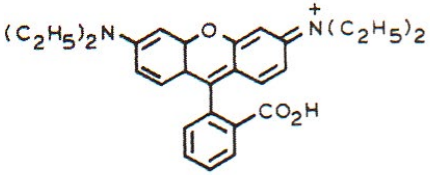
Quinine



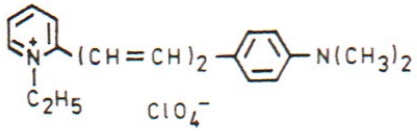
Fluorescein



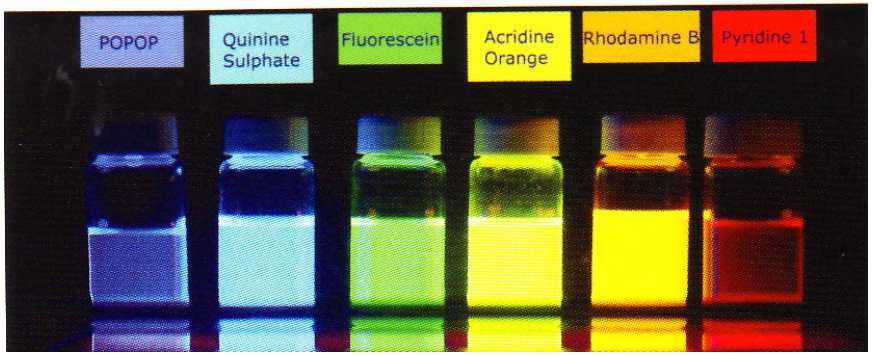
Acridine Orange

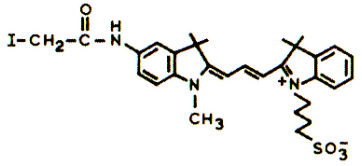


Rhodamine B



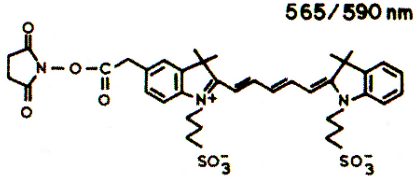
Pyridine 1





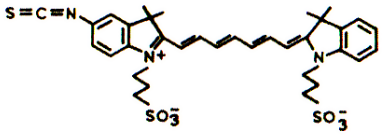
Cy-3 Iodo Acetamide

565/590 nm QY≈0.07



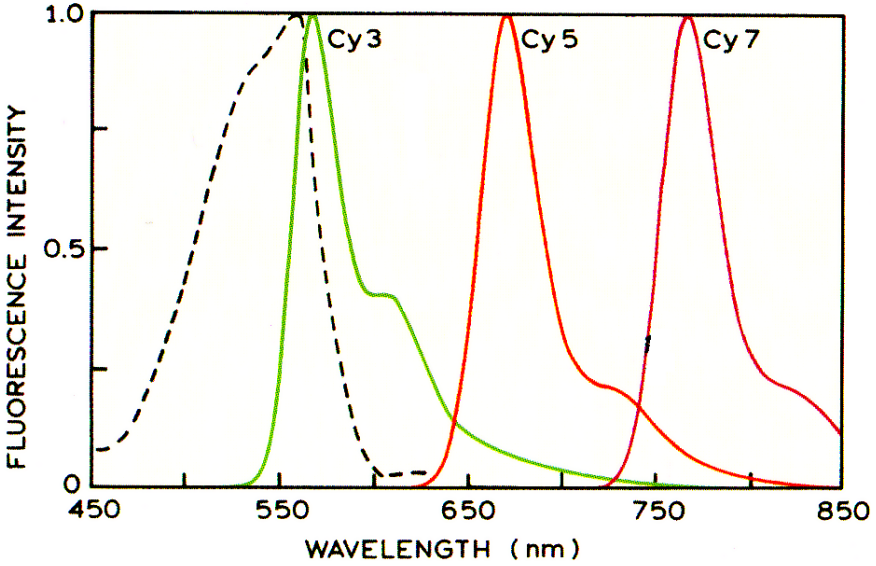
Cy-5-N-Hydroxysuccinimide

648/669 nm QY≈0.10

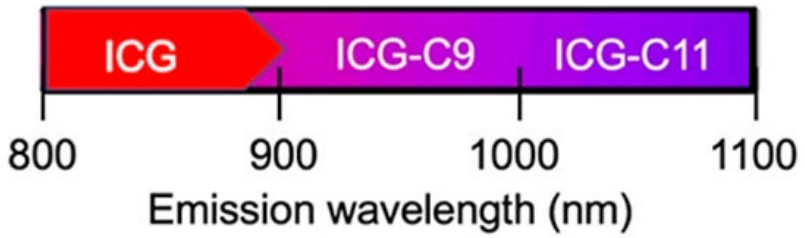


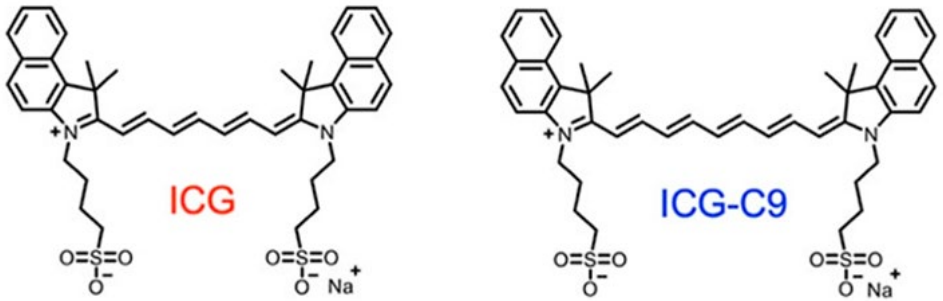
Cy-7-Isothiocyanate

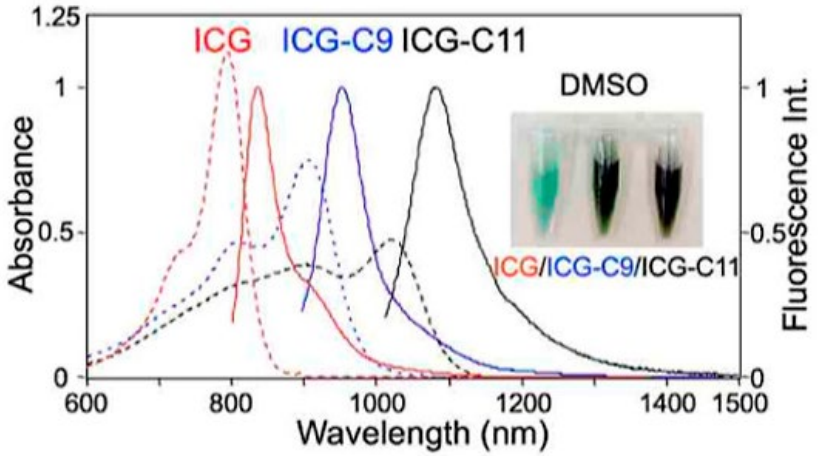
750/777 nm QY≈0.10

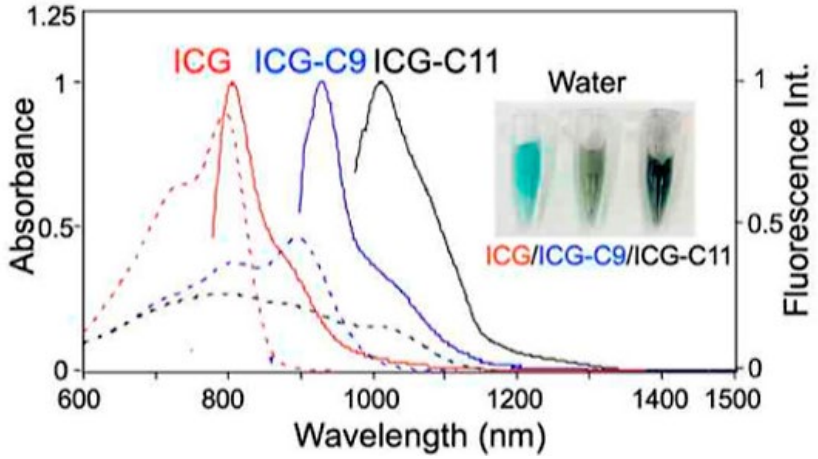


Homologer av *indocyanine green* (ICG) – partikeln i lådan i praktiken





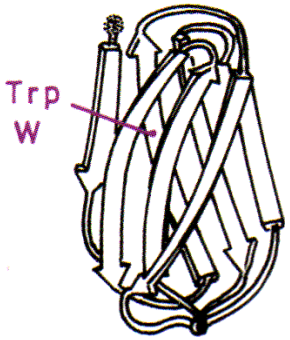


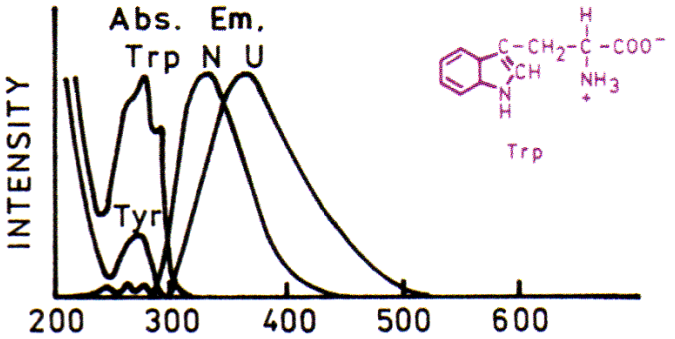


ACS Appl. Mater. Interfaces **16,** 17253 (2024), DOI: 10.1021/acsami.4c01000

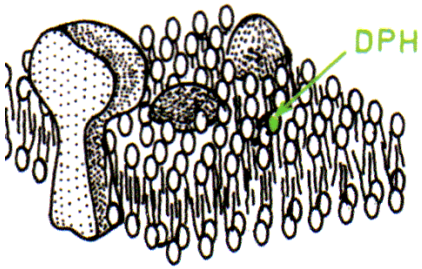
Prober för biomolekyler

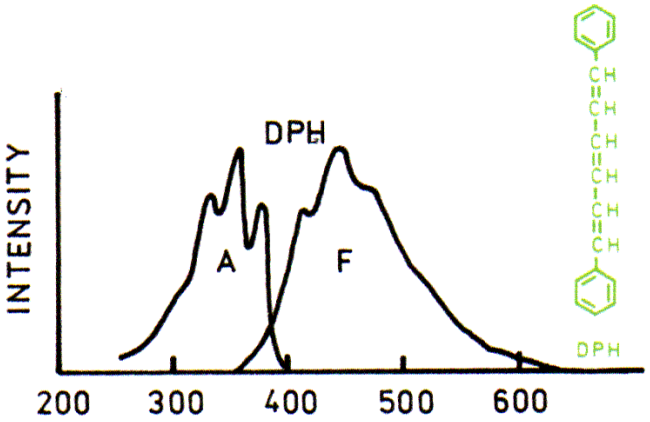
Tryptofan är fluorescent och utnyttjas för koncentrations-bestämning eller för att studera den lokala omgivningen.



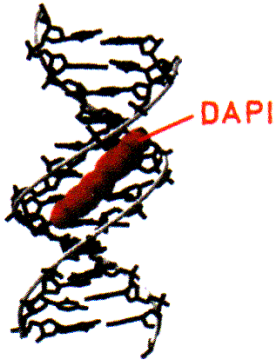


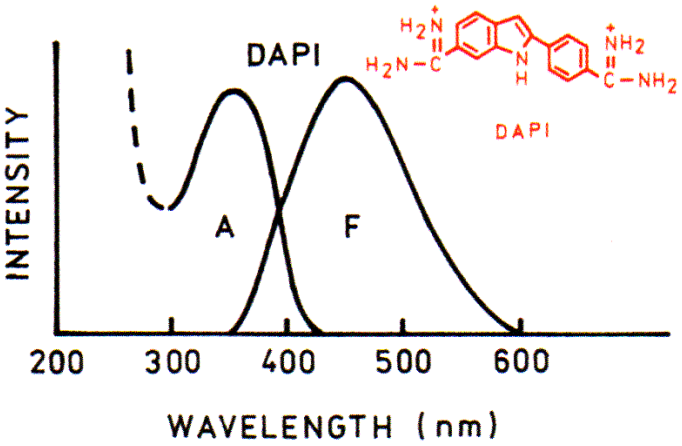
Difenylhexatrien (DPH) används för att studera fluiditet i membran, och ger information om fastillstånd och fasövergångar.





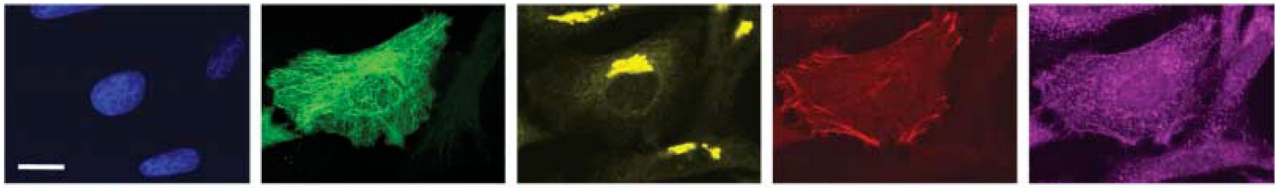
DAPI är cell-permeabel och binder specifikt till DNA; används bl.a. för att indikera DNA och studera ändringar i DNA-struktur.



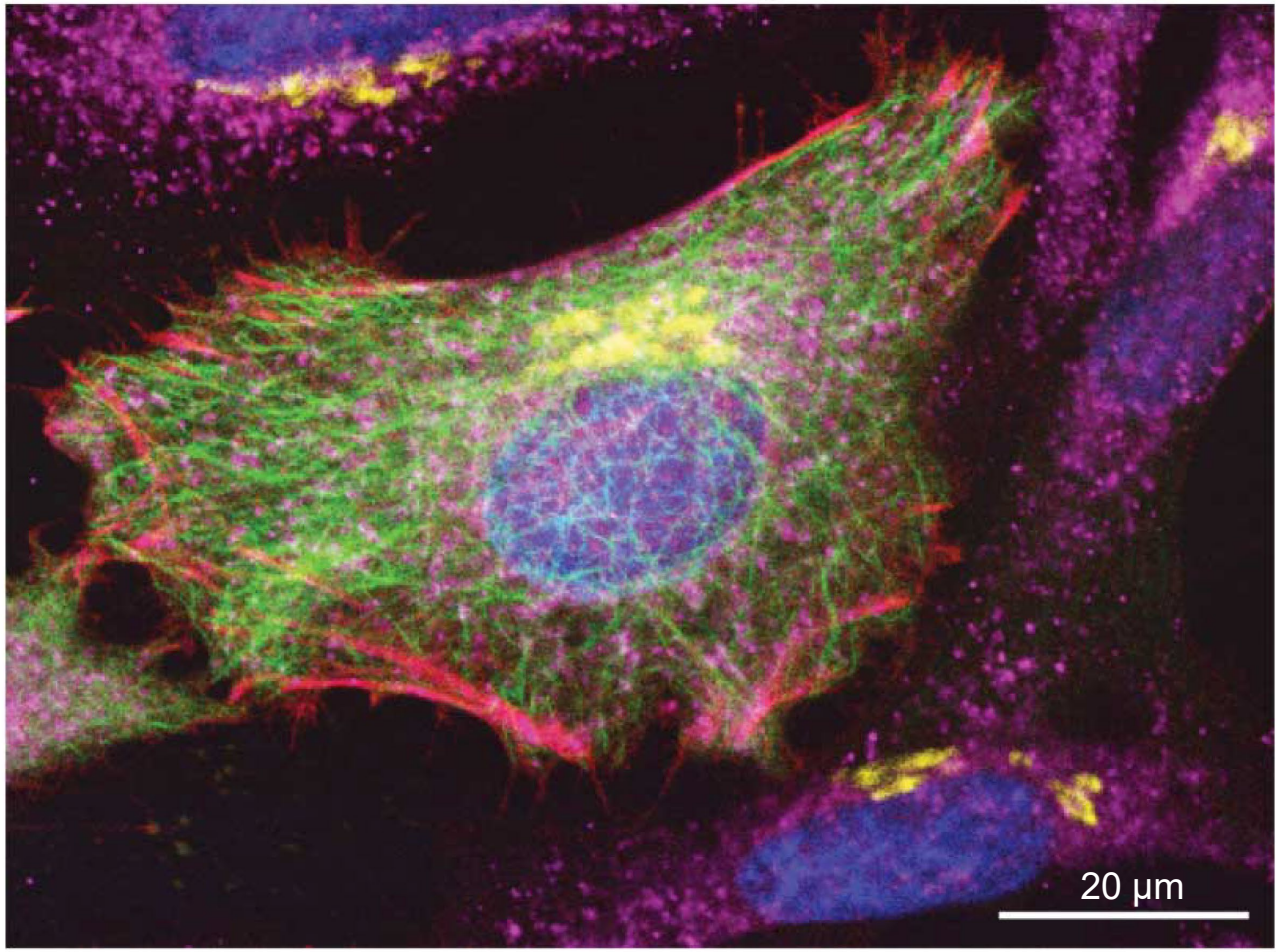


Excitation (nm):	800 (2 photon)	488	432	568	637
Emission (nm):	410-490	500-530	555-565	580-620	>660
Fluorophore:	Hoechst	GFP	QD565	ReAsH	Cy5
Targeting:	direct affinity	genetic	immuno	genetic	immuno
Target:	DNA	α-tubulin	giantin	β-actin	Cytochrome c
Structure:	nuclei	microtubules	golgi	stress fibers	mitochondria

’false-colour’ images



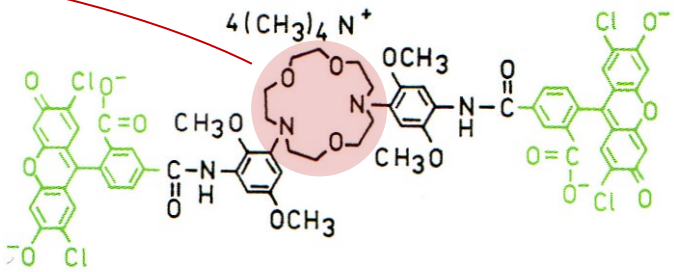
Parallell användning av olika fluoroforer och metoder för inmärkning i en HeLa cell.



Giepmans, ‘The Fluorescent Toolbox for Assessing Protein Location and Function’, Science **312**, 217 (2006). DOI: 10.1126/science.1124618

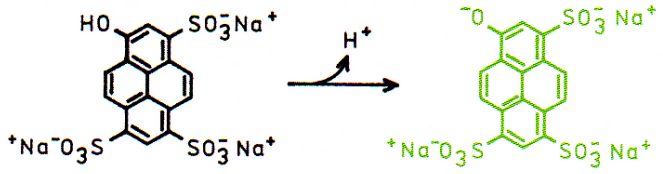
Fluorescenta indikatorer

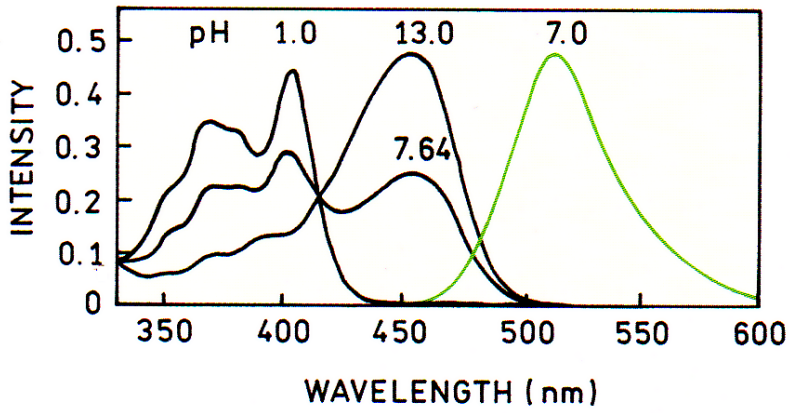
Den centrala kronetern i natriumgrönt binder Na^+, och emissionen beror av natrium-koncentrationen i lösningen (t.h.).

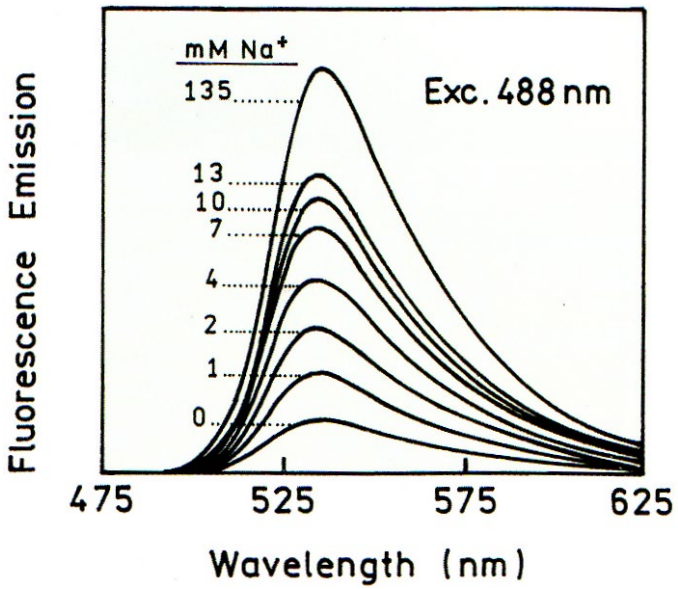


Indikatorer finns för t.ex.:

Na^+, Cl^-, Ca^{2+}, Mg^{2+}, pH (nedan), O_2 m.m.







Och nu till … solkräm?!

Innehåll:

Aktiva ingredienser
Själva solskyddet – se nästa bild!

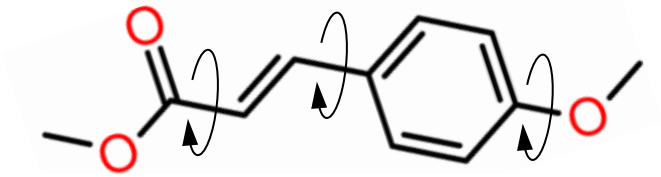
Stabiliseringsmedel
Lösningsmedel, förtjockningsmedel, emulgeringsmedel (Mer om dessa i TFYA47 Ytor och gränsskikt!)
Konserveringsmedel

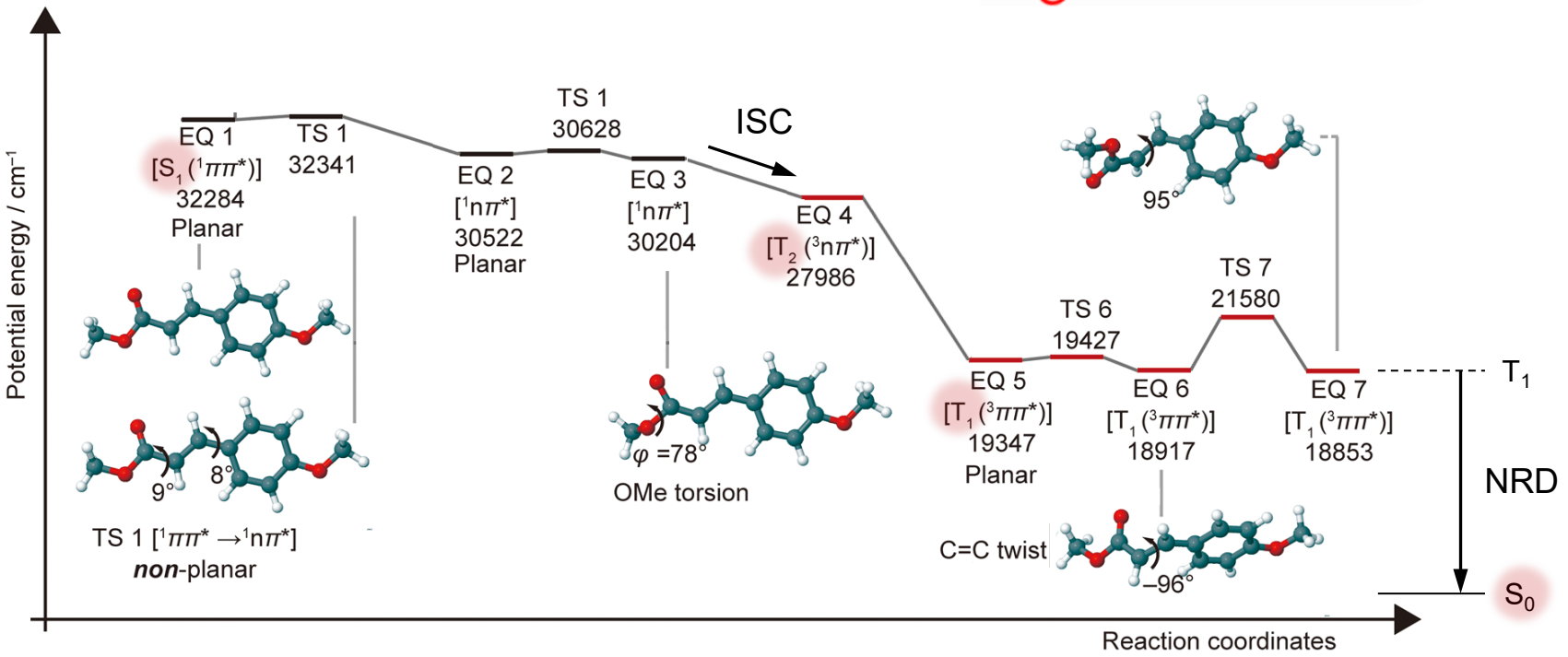
Sensoriska tillsatser
Parfym, mjukgörande medel

Fotofysik hos solskydd

Två typer av solskydd: Fysikaliskt – reflekterar UV-ljus (ZnO eller TiO_2-partiklar)
Kemiskt – absorberar UV-ljus, med strålningsfri deexcitation

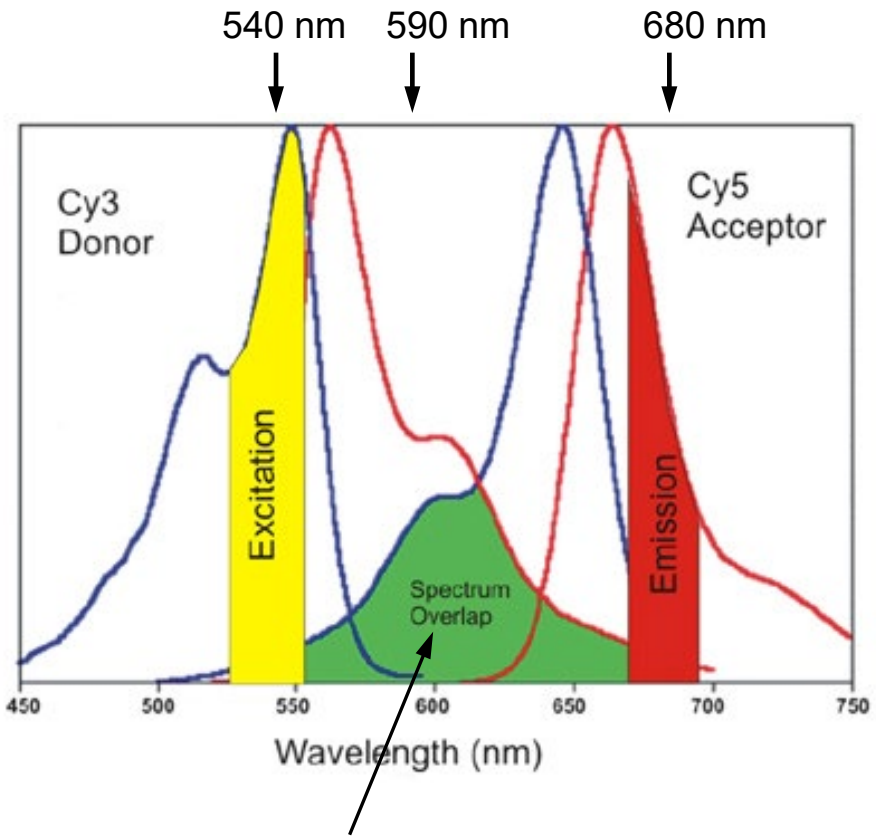
Para-metoxymetylcinnamat (p-MMC) absorberar UV-strålning, och deexciteras via strålningsfria övergångar (nonradiative decay, NRD) som involverar *trans* → *cis*-isomerisering.

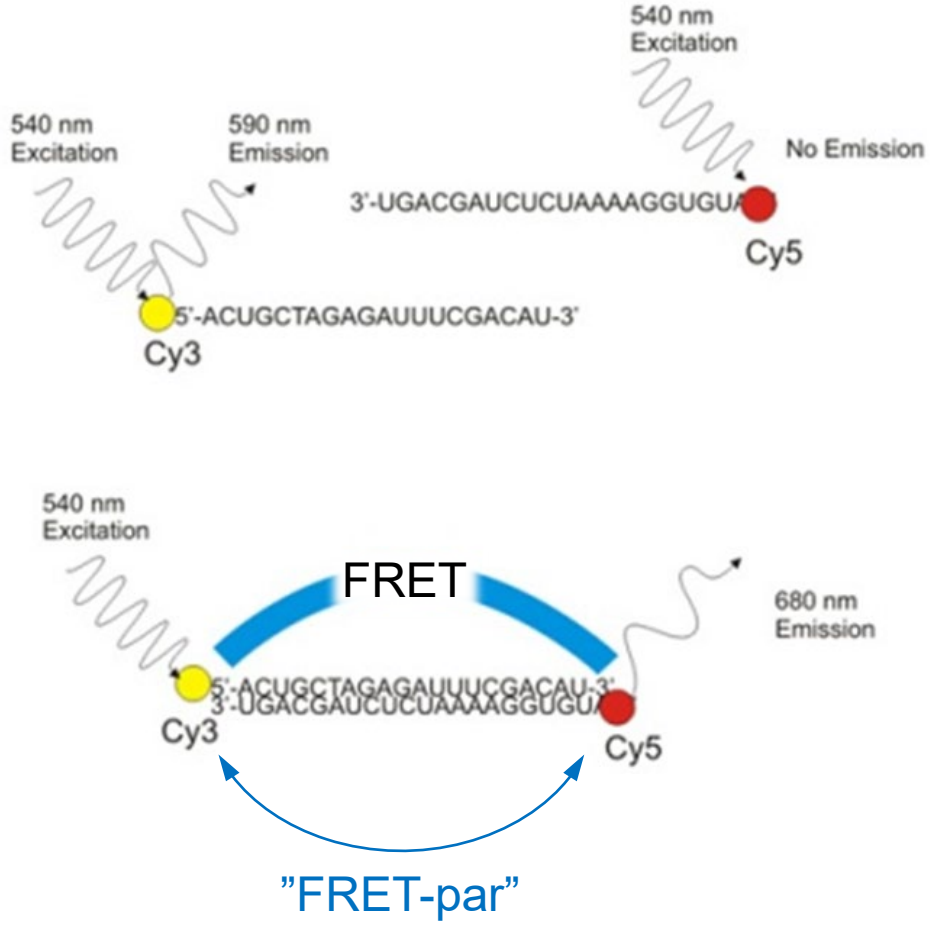




Yamazaki, *J. Phys. Chem. Lett.*, **7**, 4001 (2016), DOI: 10.1021/acs.jpclett.6b01643

Fluorescence Resonance Energy Transfer (FRET)

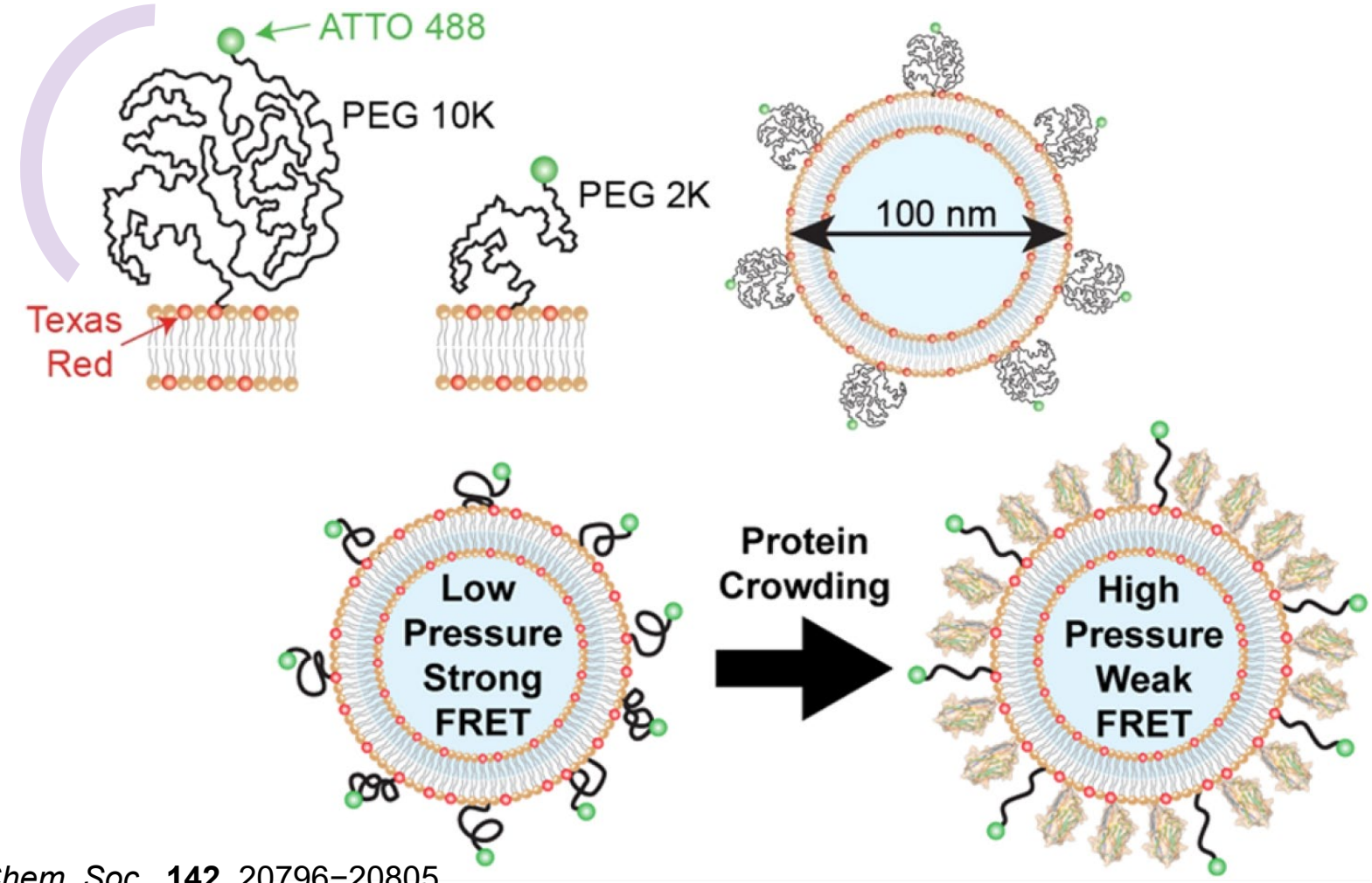




”Förster resonance”; överföring av excitationsenergi mellan olika molekyler.

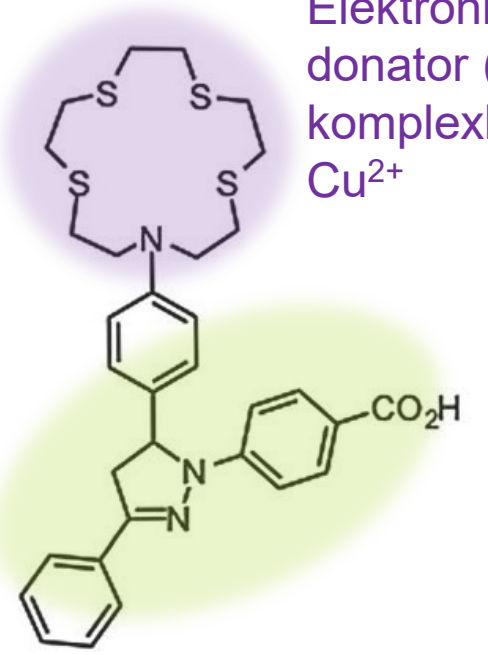
A Förster Resonance Energy Transfer-Based Sensor of Steric Pressure on Membrane Surfaces

Avstånds-beroende FRET



Houser, *J. Am. Chem. Soc.*, **142**, 20796−20805 (2020). DOI: 10.1021/jacs.0c09802

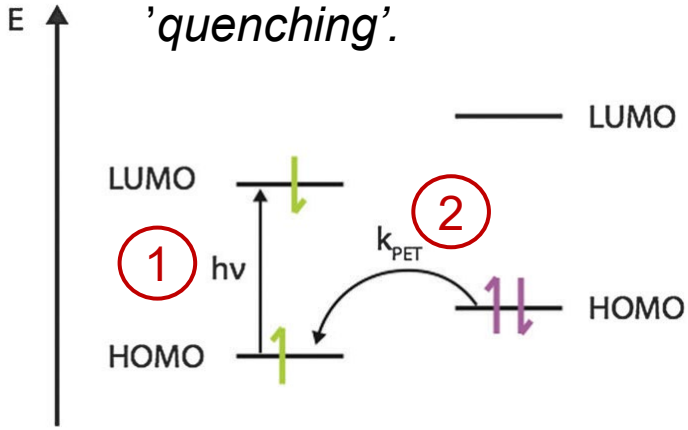
Prober för detektion av Cu^{2+} - *Quenching*

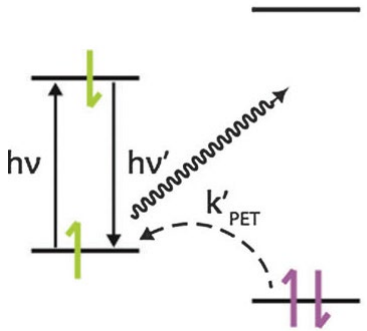


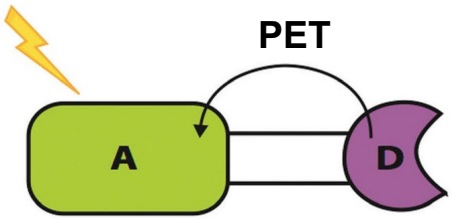
Elektronrik donator (D), komplexbinder Cu^{2+}

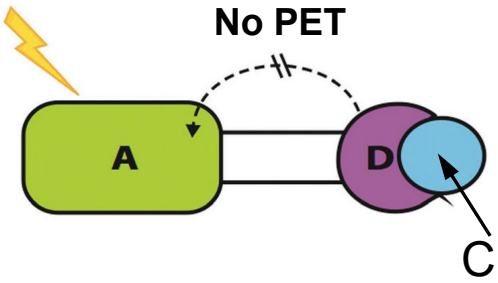
I frånvaro av Cu-jon:
Vid fotoexcitation (1) fylls acceptorns HOMO från donatorn (2), deexcitation (fluorescens) förhindras, ’*quenching’.*

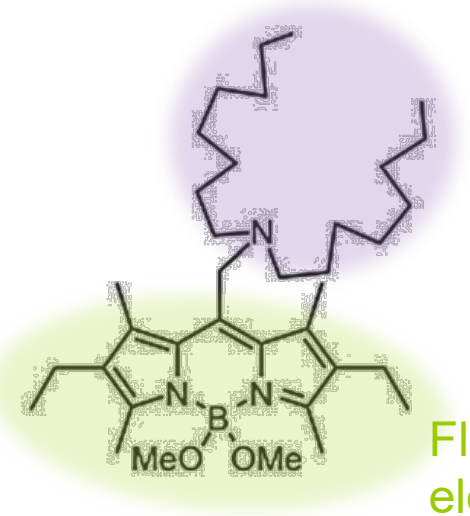
Med Cu-jon:
Donatorns HOMO sänks och elektronöverföring till acceptorn förhindras, så att fluorescens är möjlig.







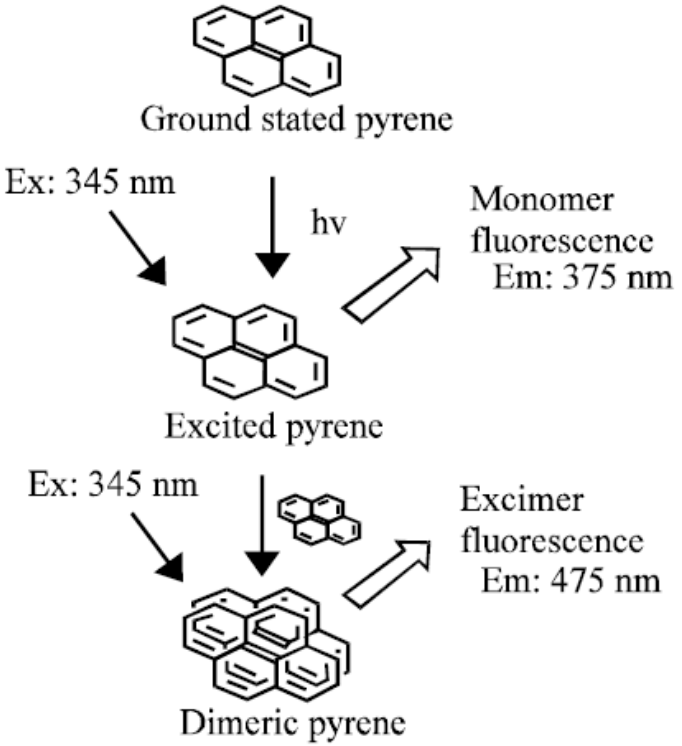


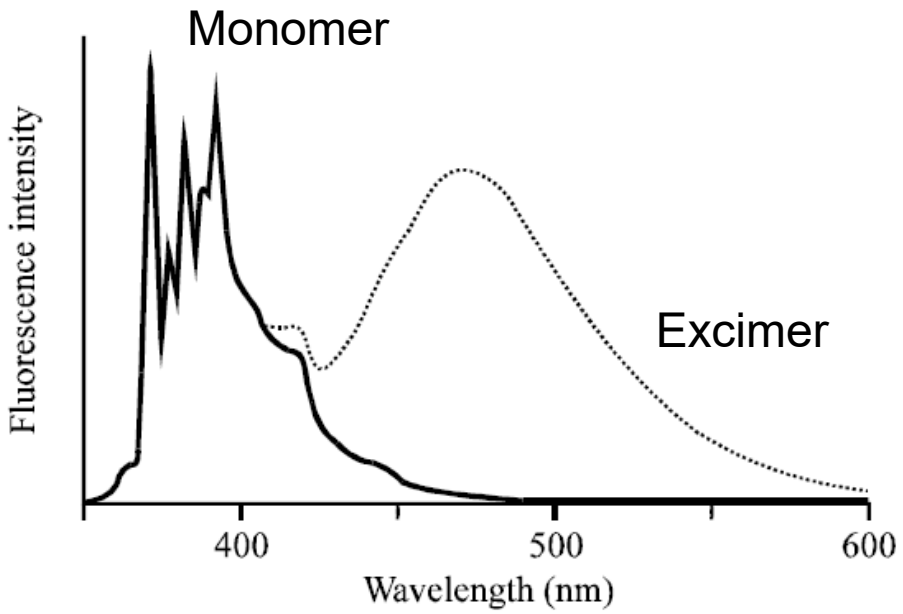


Fluorescent elektronacceptor (A)

PET = Photoinduced electron transfer

Excimerer





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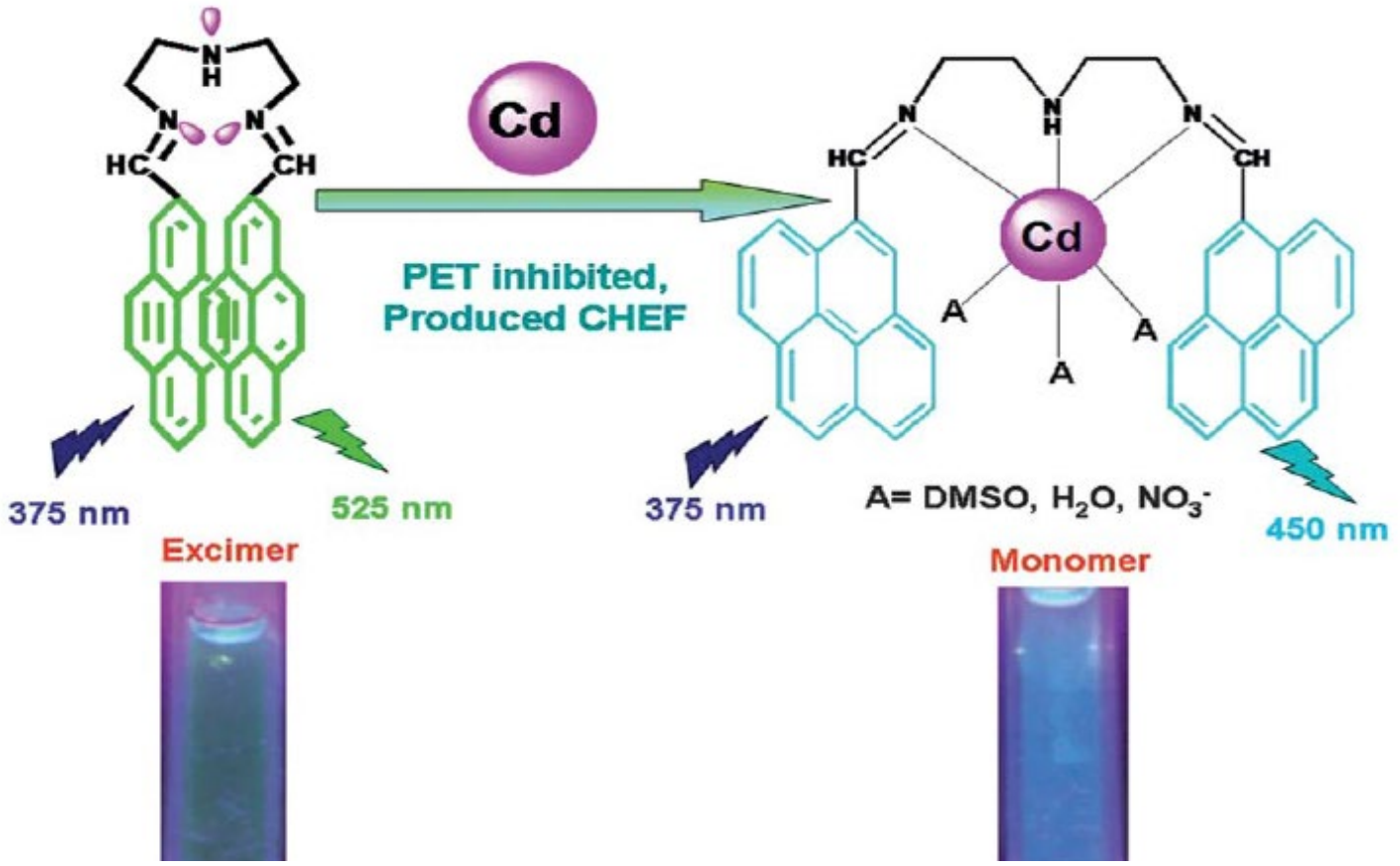
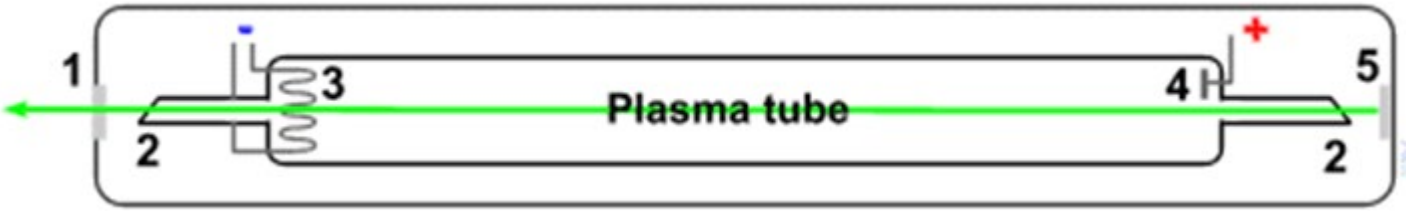
Cd(II)-triggered excimer–monomer conversion of a pyrene derivative: time dependent red-shift of monomer emission with cell staining application†



Selektiv detektion av Cd^{2+} i närvaro av andra vanliga metalljoner, med en detektionsgräns på 20 nM.

Shana et al., *Analyst*, **137**, 3910 (2012), DOI: 10.1039/c2an35731c

HeNe – en vanlig ’gaslaser’



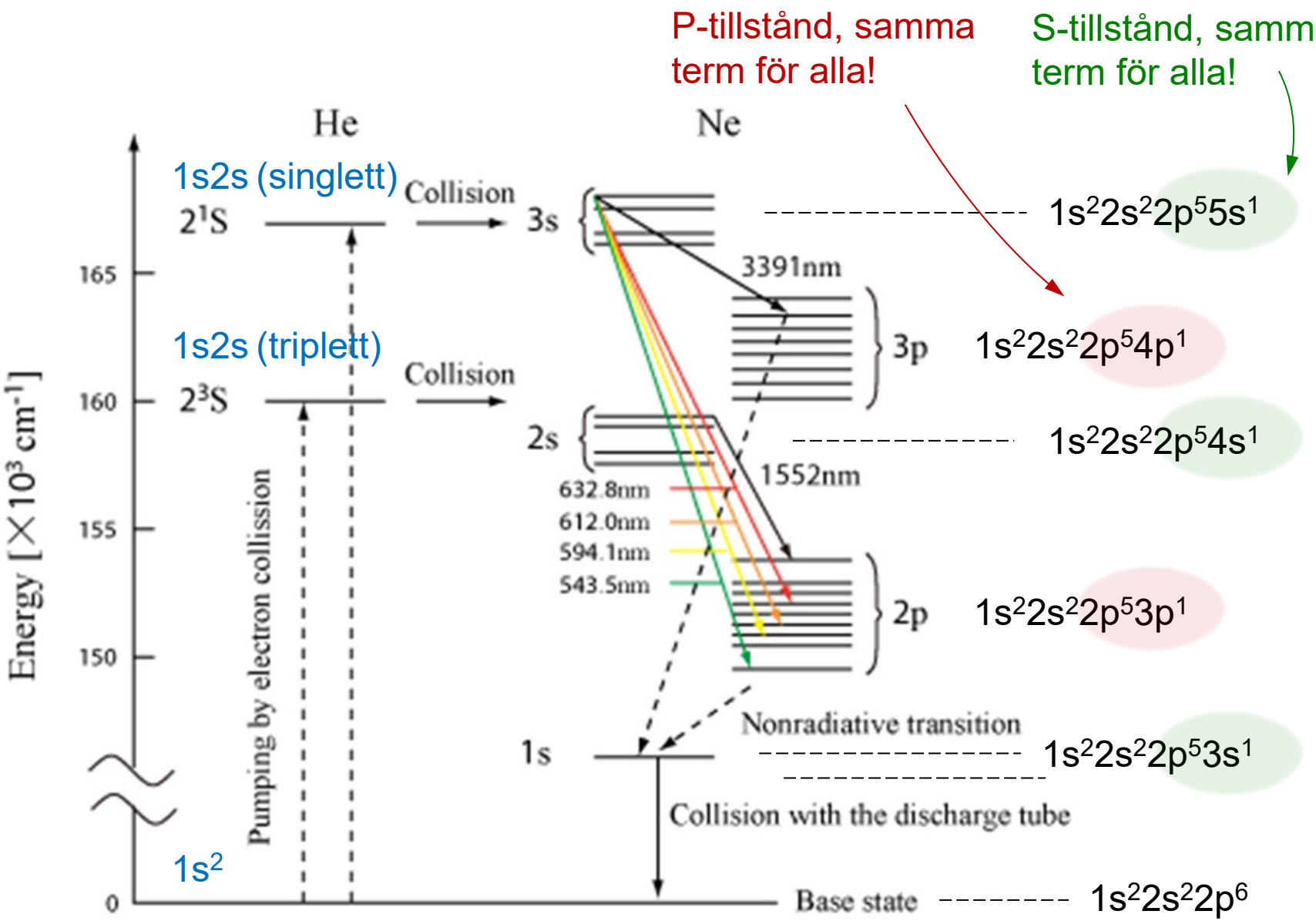
1 Utgång – delvis genomskinlig (~1 %) spegel
2 Brewster-fönster
3 Katod (filament)
4 Anod
5 Spegel

Vanligen använd för emission vid 632.8 nm (rött).

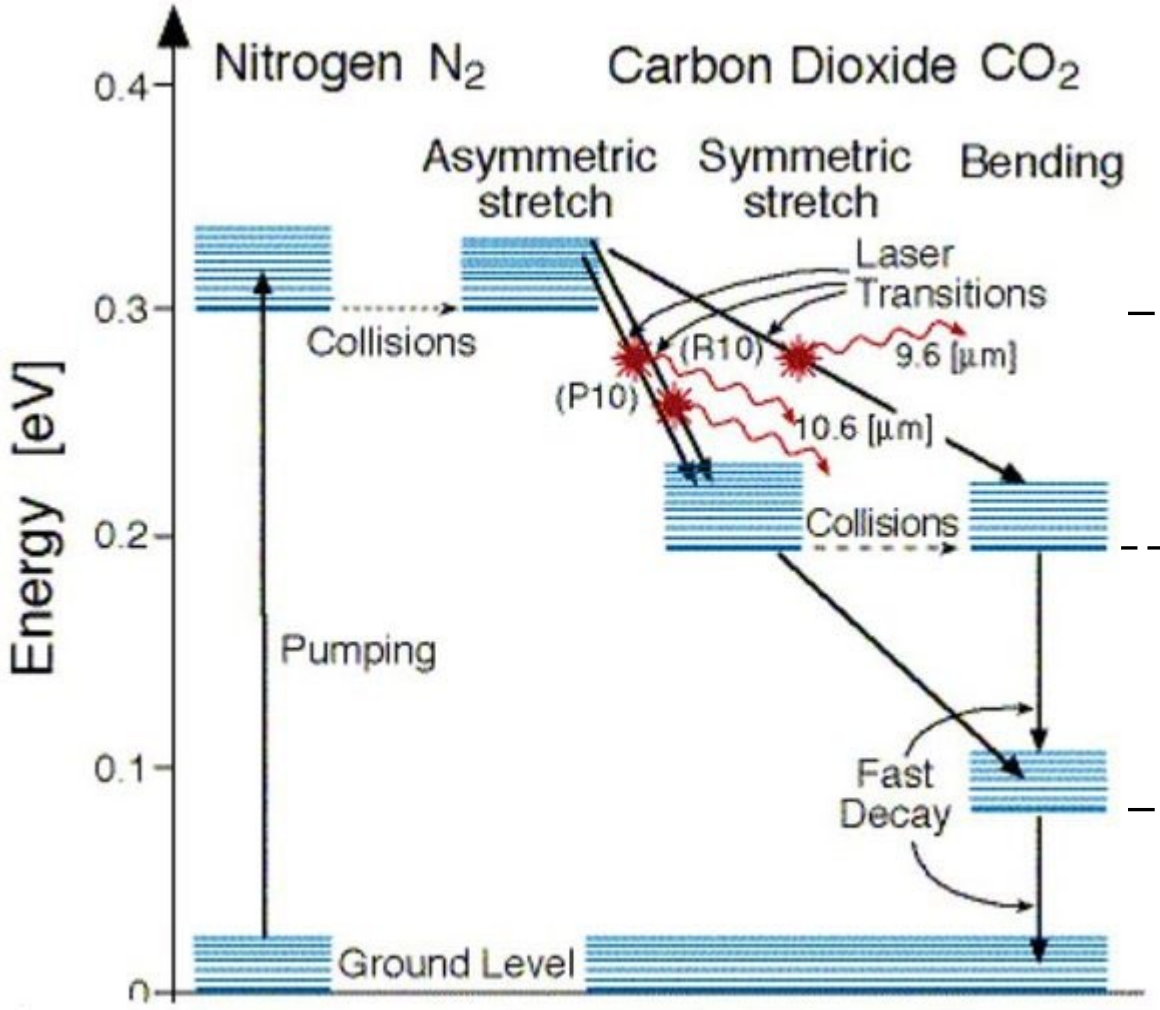
Medium: mellan 5:1 och 20:1 He och Ne vid lågt tryck.
Excitation: Urladdning vid hög spänning (~kV)

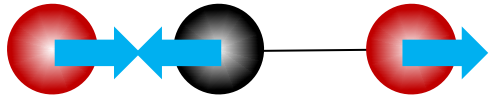
Laserprocess: elektroner från urladdningen exciterar He via kollisioner till metastabila tillstånd med lång livstid. Dessa exciterar i sin tur Ne-atomer via kollisioner, dessa emitterar strålning vid de-excitation.

Tillstånd och övergångar i HeNe-lasern

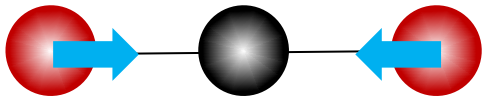


CO_2-laser – gaslaser med IR-emission

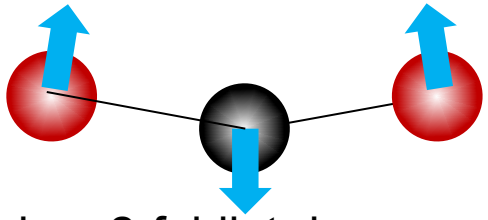




Asymmetrisk sträckning
2349 cm^{-1} (IR-aktiv)



Symmetrisk sträckning
1388 cm^{-1} (Raman-aktiv)

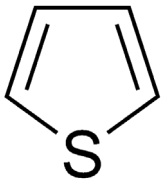


Böjning, 2-faldigt degenererad
667 cm^{-1} (IR-aktiv)
(ν_2 vid 1334 cm^{-1})

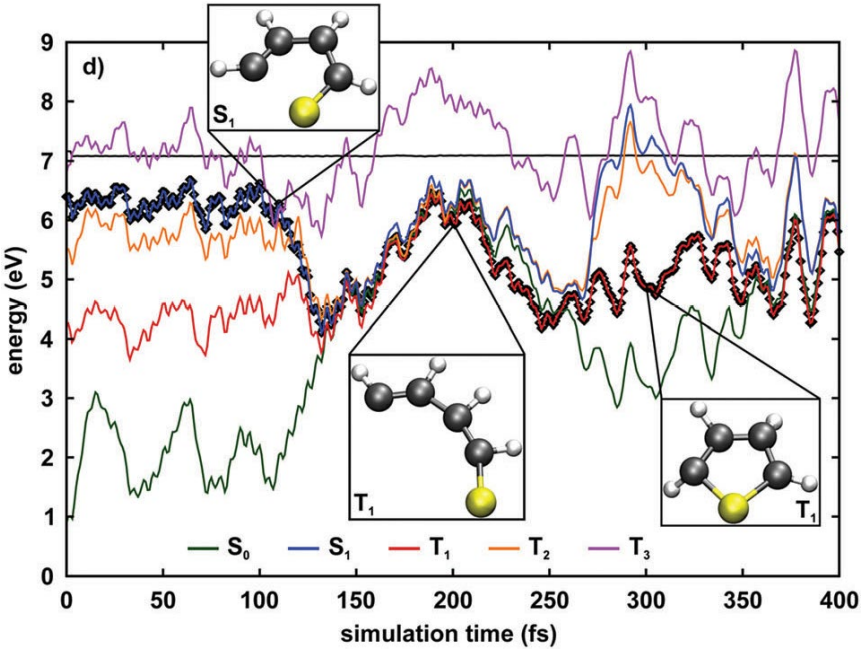
http://repairfaq.cis.upenn.edu/sam/laserhen.htm

Simulering av fotoprocesser

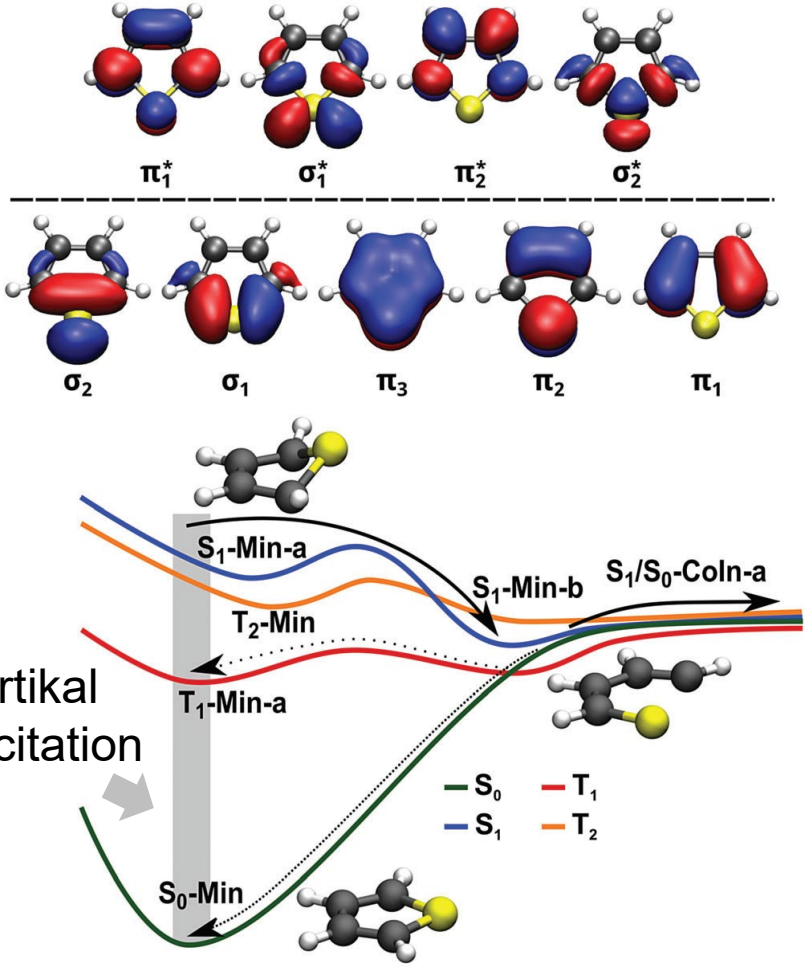
- Ett oumbärligt verktyg för molekylär förståelse!



Fotoexcitation of tiofen ger långlivade tillstånd med öppen ring. Varför?



Simulering av de olika tillstånden på en S_1-potential.



Både det involverade triplett-tillståndet och de grunda potentialerna orsakar långsam återgång till en sluten ring i grundtillståndet.

Schnappinger, *Phys. Chem. Chem. Phys.*, **19**, 25662 (2017), DOI: 10.1039/c7cp05061e