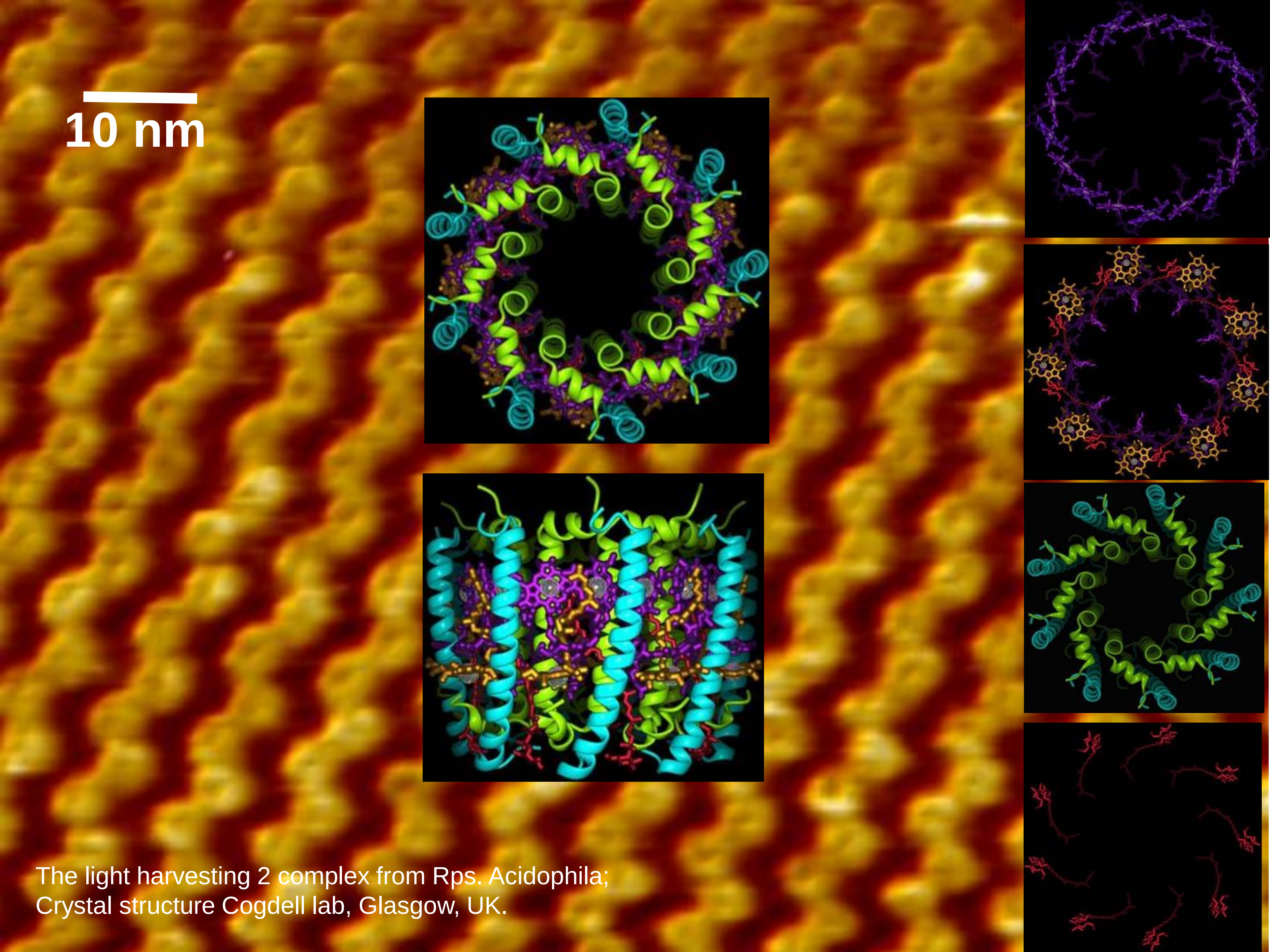
Photosynthaics:

photosynthetic biohybrid devices for solar energy harvesting

Phototech school Athens 2013
Raoul Frese, VU University Amsterdam-FEW/biophysics

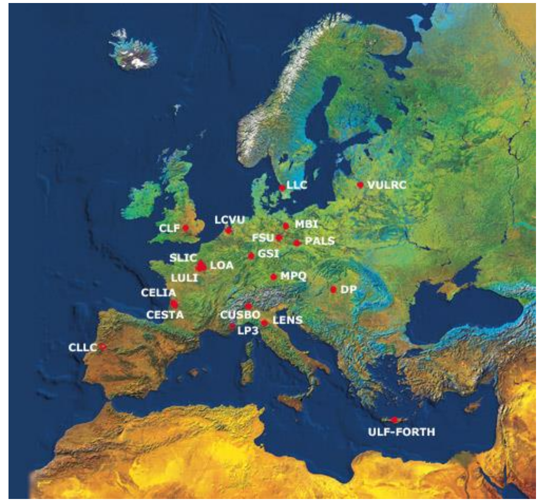


The light harvesting 2 complex from Rps. Acidophila;
Crystal structure Cogdell lab, Glasgow, UK.

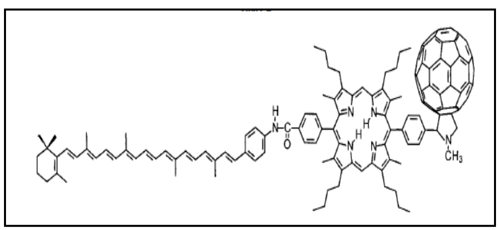
Biophysics of photosynthesis

Laserlab Europe

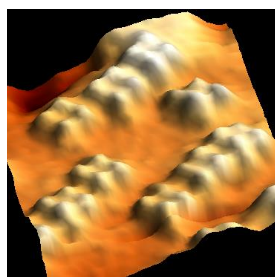
LaserLab Amsterdam



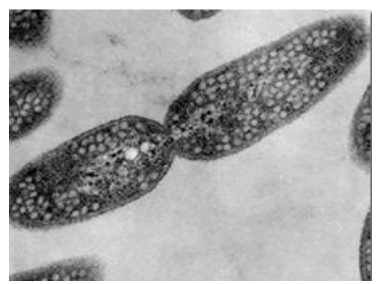
vrije Universiteit amsterdam



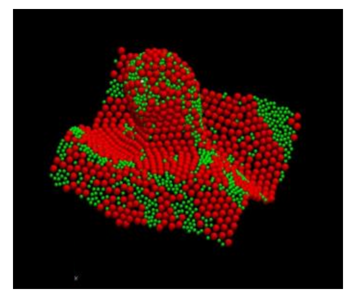
Artificial photosynthesis



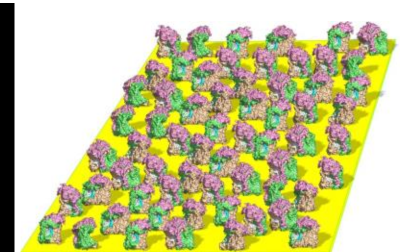
Atomic force Microscopy



Membrane biogenesis and function



Membrane simulations



Biosensors / Biosolar cells

This lecture: biophysics of assemblies, basic performance of devices

When you study functioning of devices you also study biophysics of assemblies.

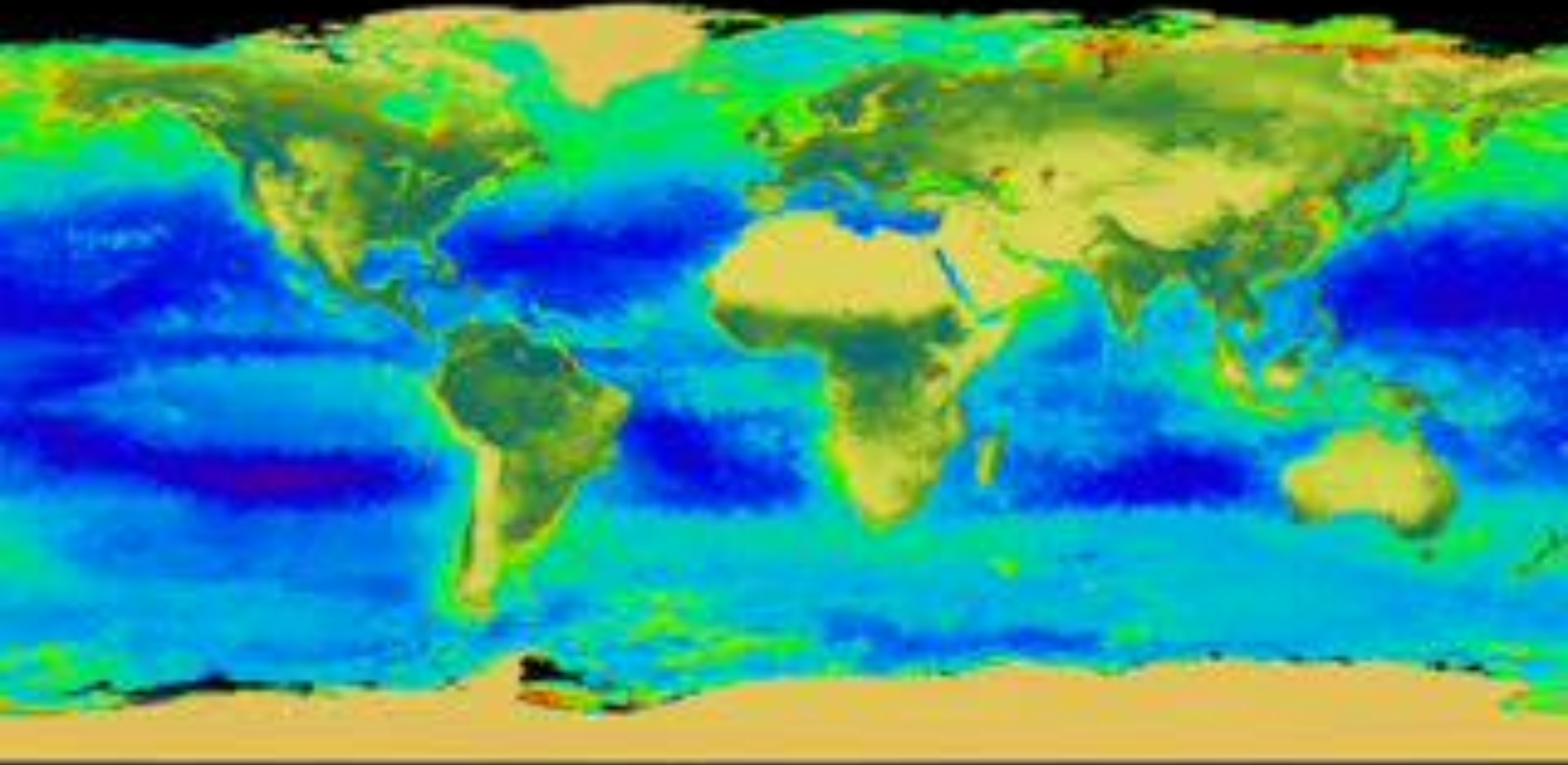
What basic performance issues should be addressed?

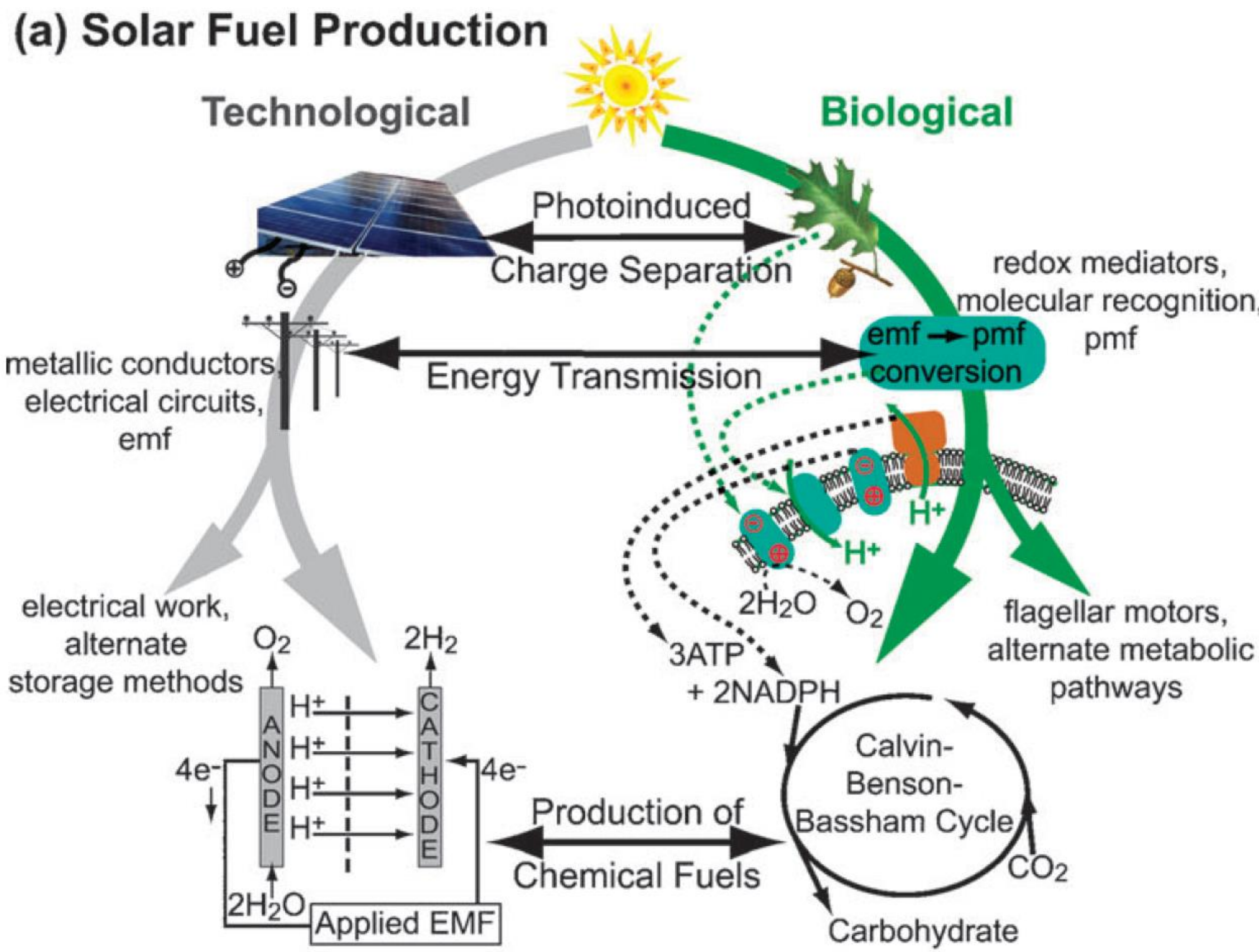
Part 1 biophysics

- Why photosynthetic proteins?
- Atomic force microscopy: 2d crystals and membranes
- Organization of membranes

Part 2 hybrid devices

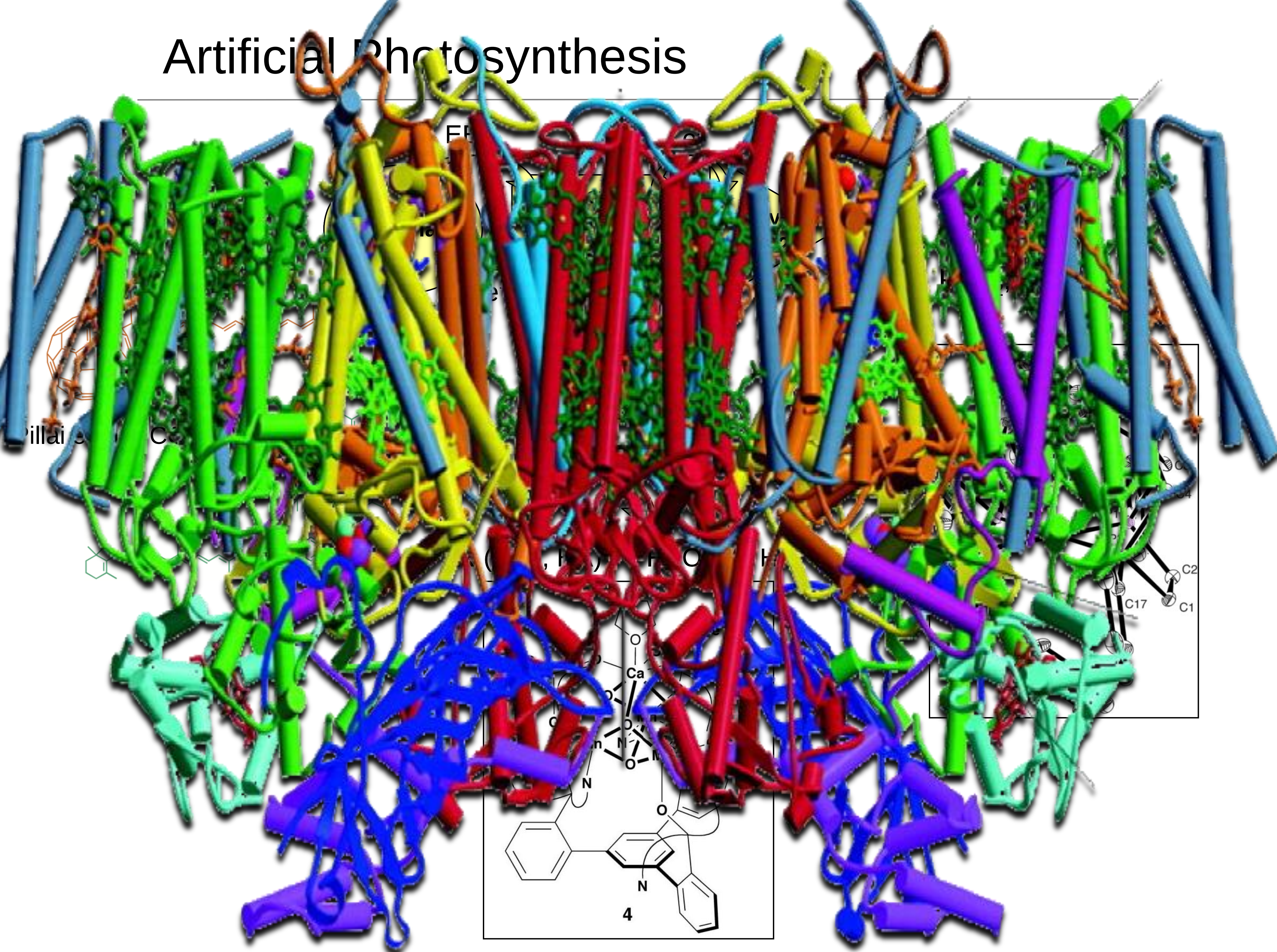
- Photoelectrochemistry
- Solar cells



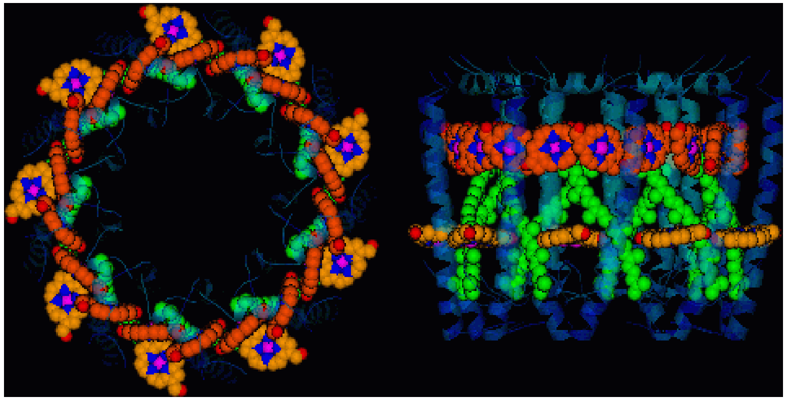


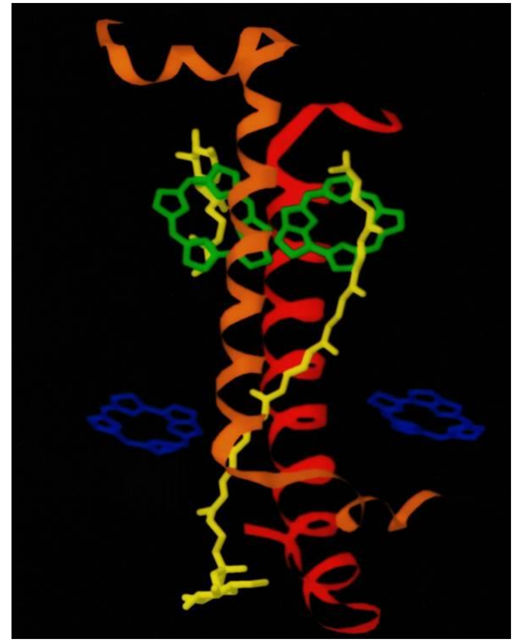
Slide Prof. Moore

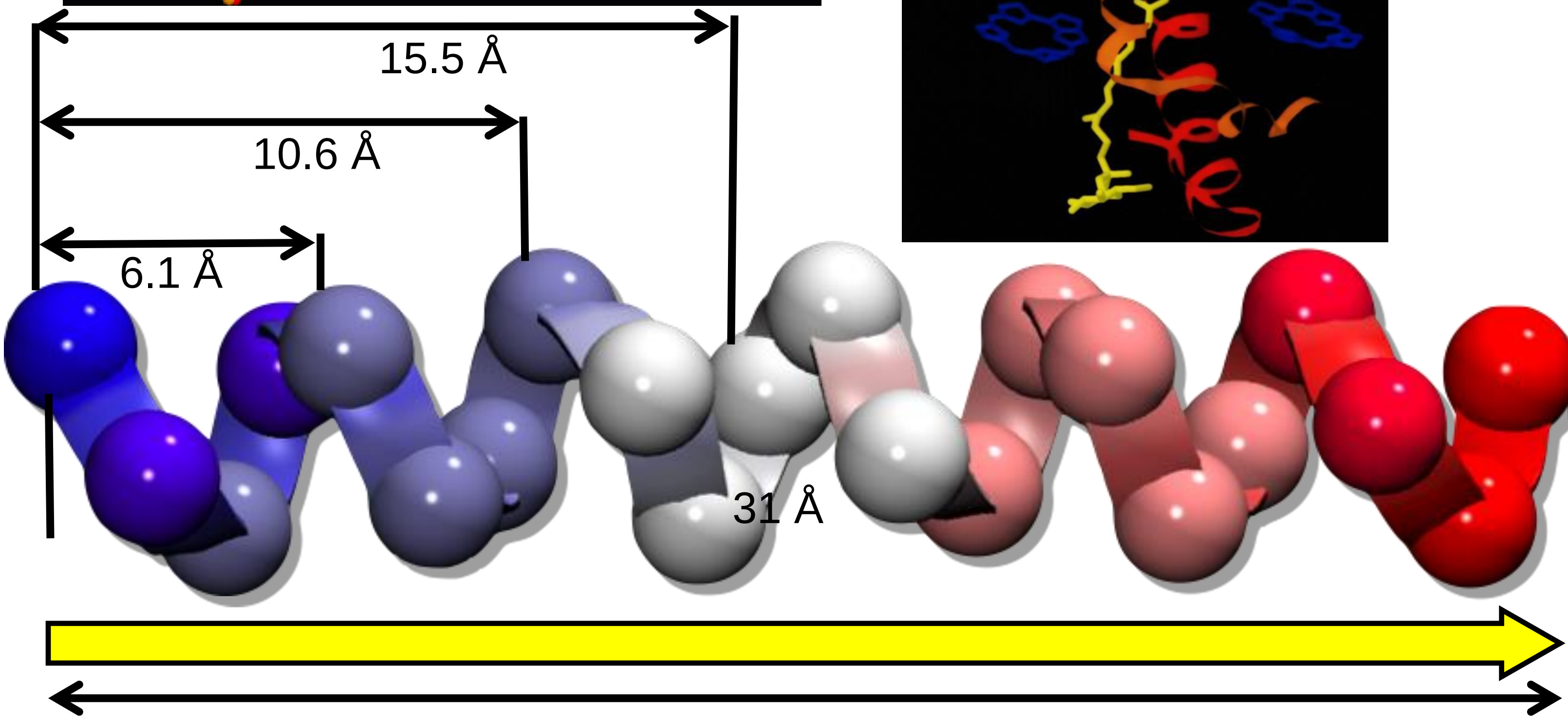
Artificial Photosynthesis



Proteins allow nanometric control



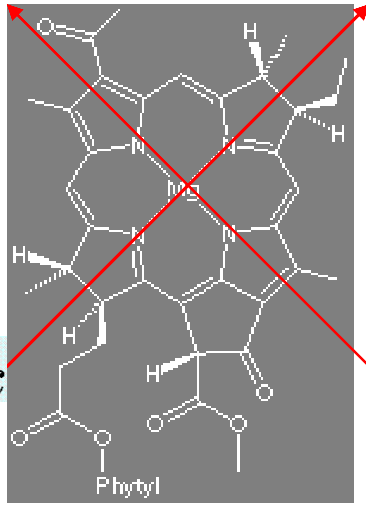




Energy transfer 10 **ps**

From dror noy

(bacterio-) chlorophyll structure, absorption, and model



$\vec{\mu}_x$ $\vec{\mu}_y$

ππ* excited state

Pigment Hamiltonian:

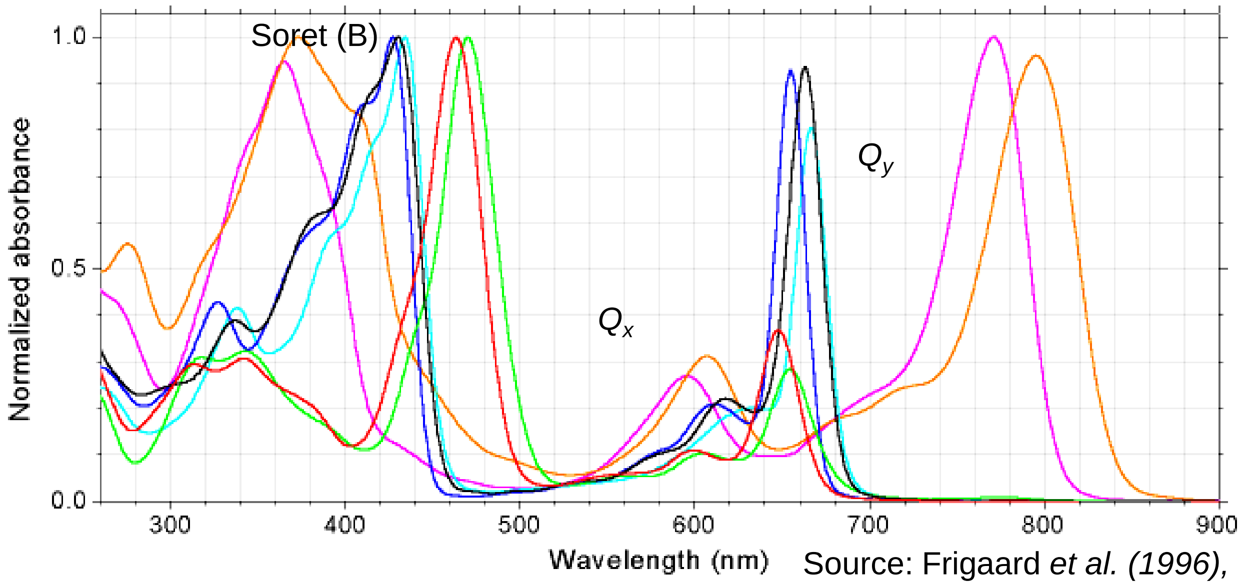
$$\mathcal{H} = \sum_i \epsilon_i |i><i|$$

Interaction with external fields:

$$\mathcal{H}_{int} = -\sum_i \vec{\mu}_i \cdot \vec{E}_{ext}$$

Absorption spectra of various chlorophylls in methanol

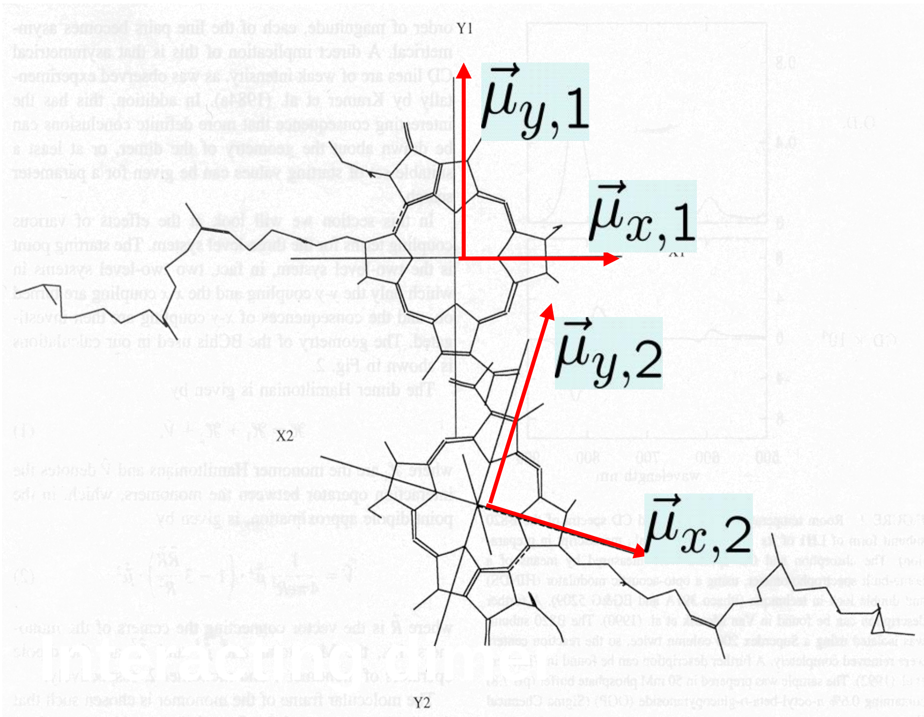
Chla: black
Chlb: red
BChla: magenta
BChlb: orange
BChlc: cyan
BChld: bue
BChle: green

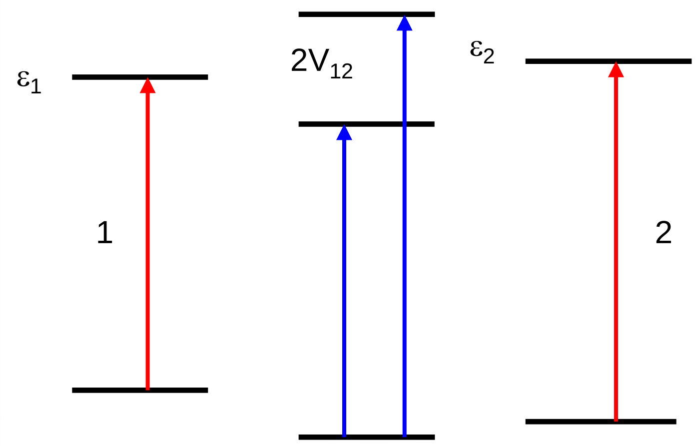


Source: Frigaard *et al. (1996), FEMS Microbiol. Ecol. 20: 69-77*

Design: tune absorbance characteristics, extended molecular frame

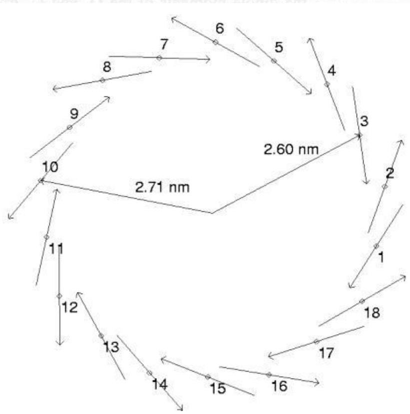
Excitonic pigment-pigment interaction:



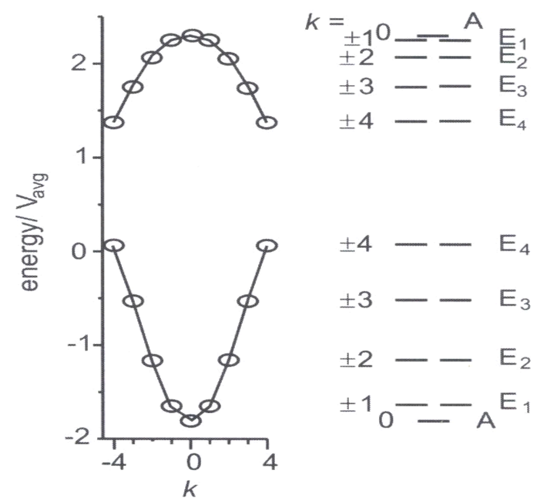


energy level scheme

LH2:
ring
of
dimers

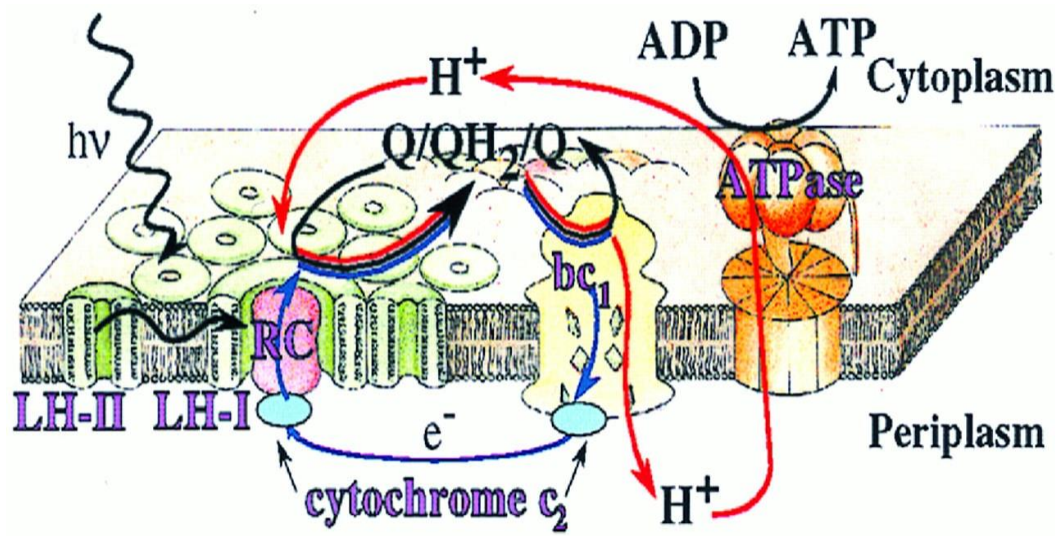


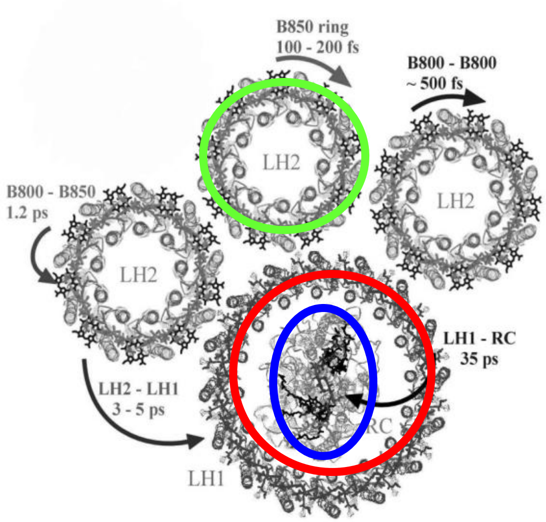
- No disorder
- α, β different
- C_9 symmetry
- Davidov splitting



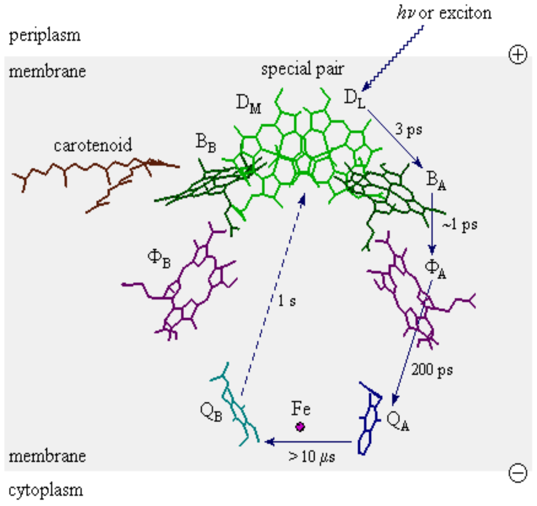
Design: tune absorbance, directed ET and EET, Charge Transfer States

Photosynthesis is a solarcell

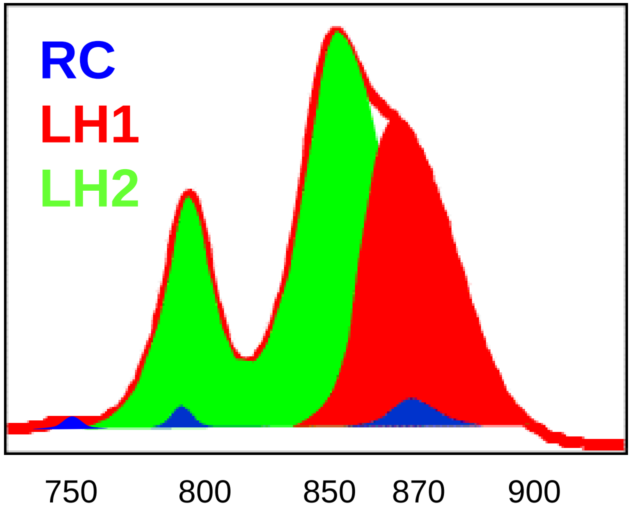




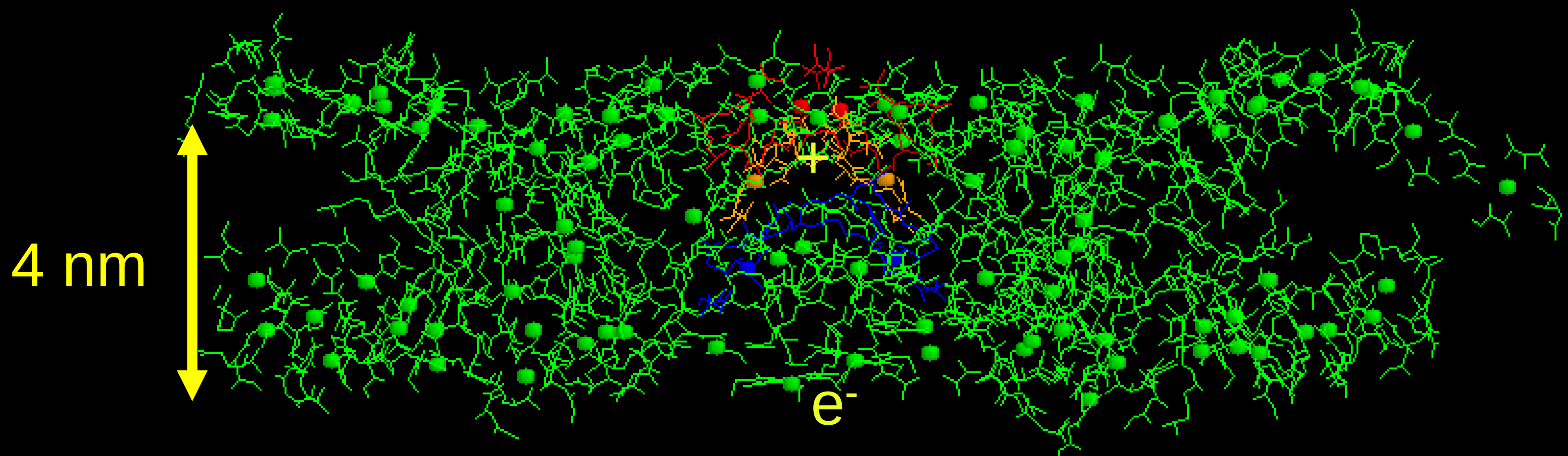
Energy transfer ~$\mathbf{R}^{-6}$



Electron transfer ~$\mathbf{e}^{-\mathbf{R}}$

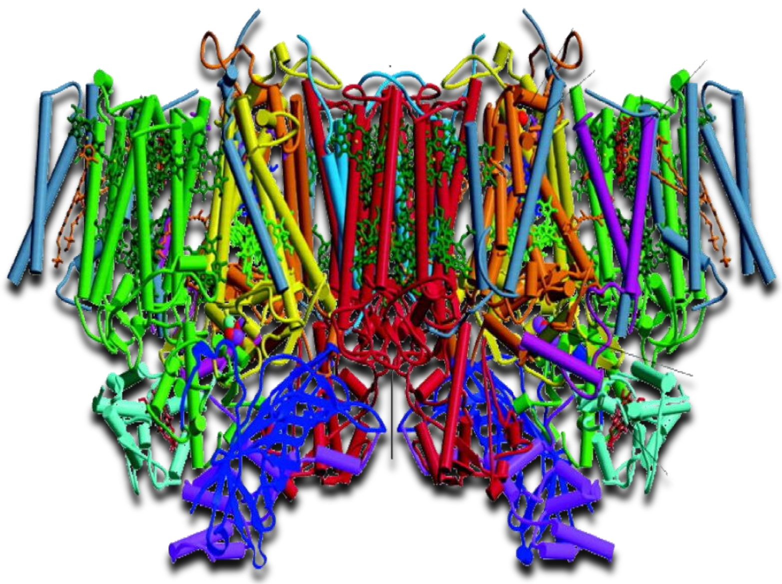


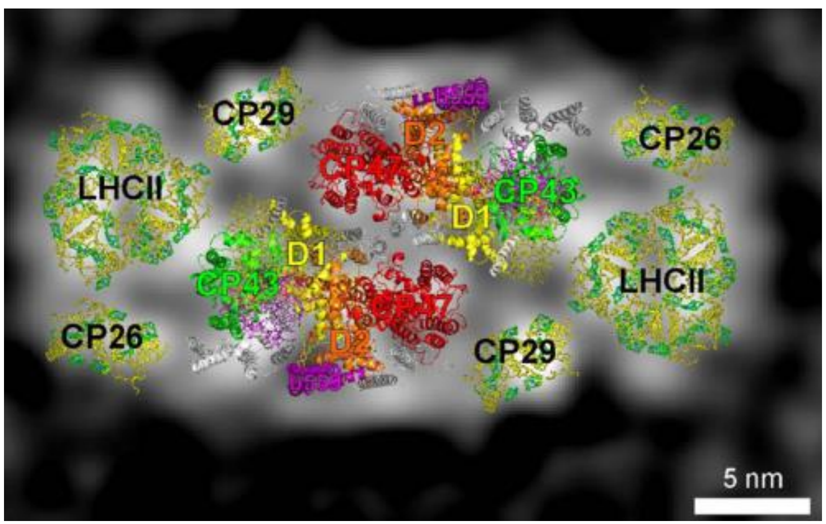
First steps in photosynthesis: 100% quantum effciency

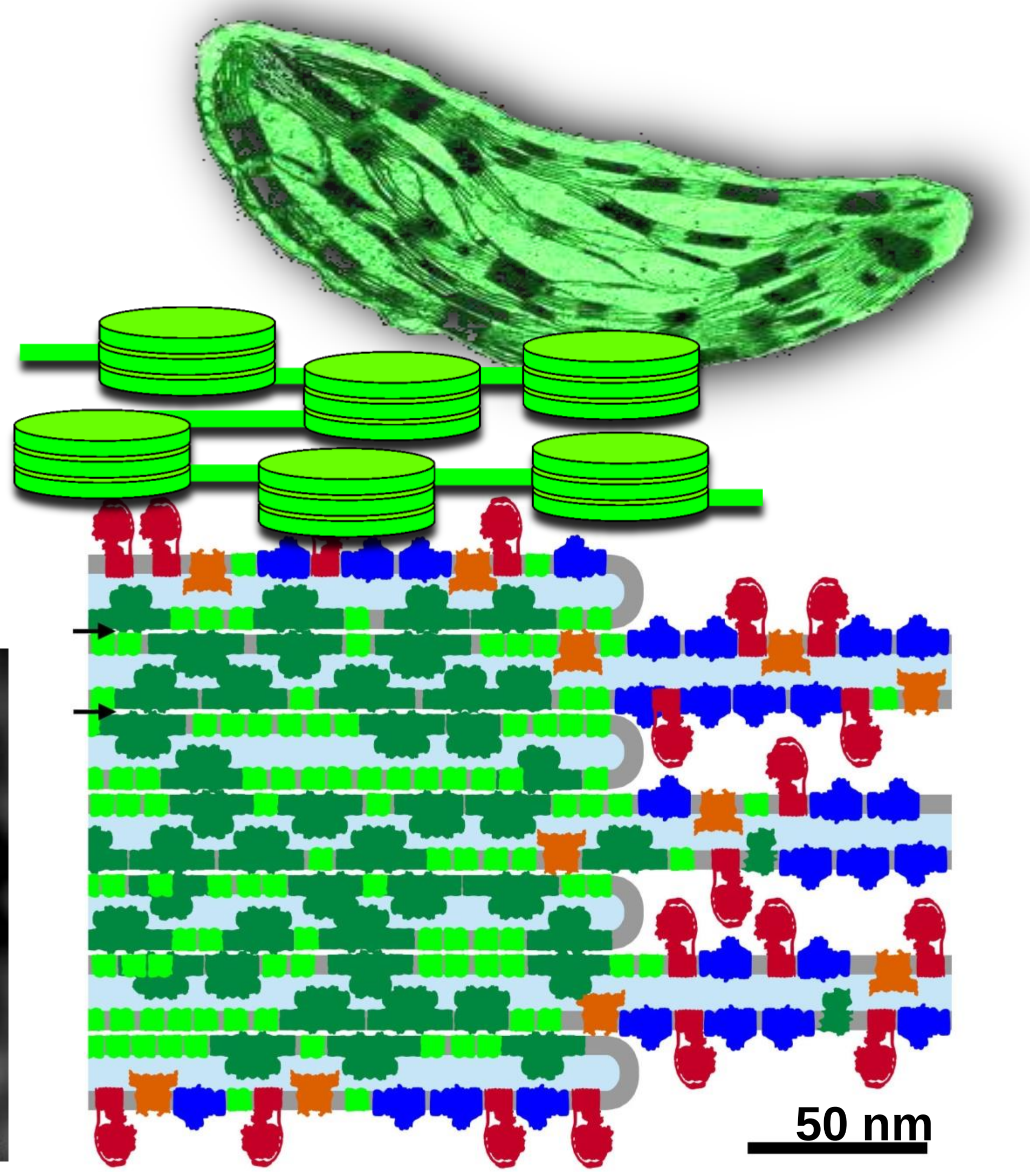


Finished in 20 picoseconds

Network of protein complexes





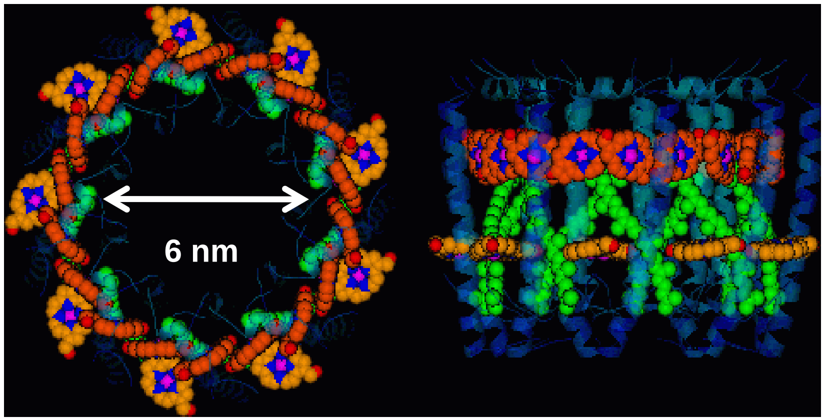


Dekker, Boekema

Protein complexes are building blocks

Excitonic building block

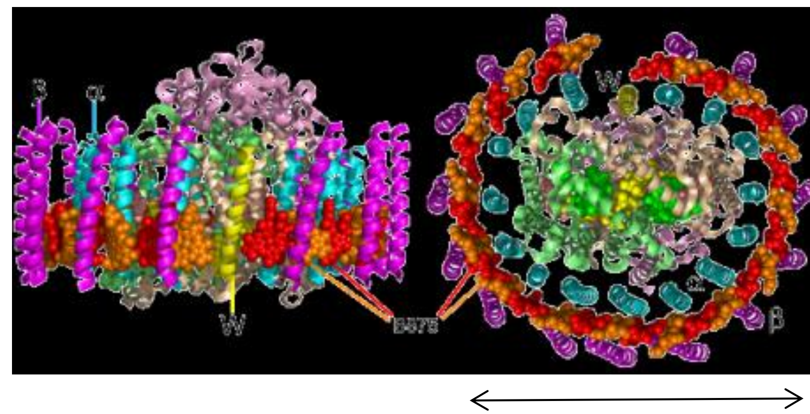
Light harvesting 2 complex



$$\psi_k = \Sigma\, c_{ki}\, \varphi i$$

Photovoltaic building block

Reaction center - Light harvesting 1 complex

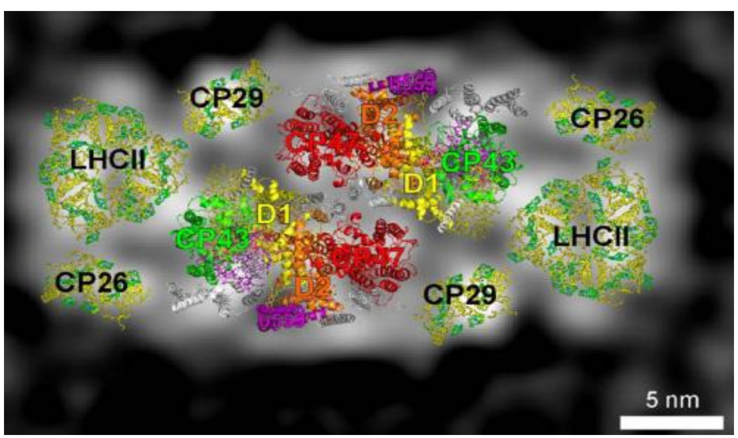


$$FF = \frac{Vmp \times Imp}{Voc \times Isc}$$

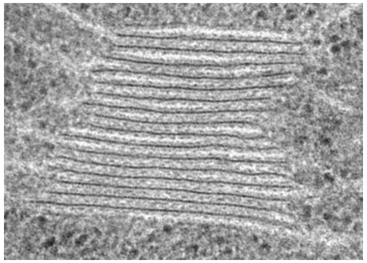
Catalytic building block

Photosystem 2 supercomplex

$$\frac{\eta,in}{\eta,out}$$

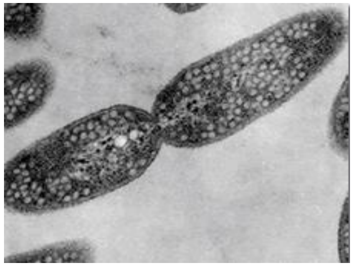


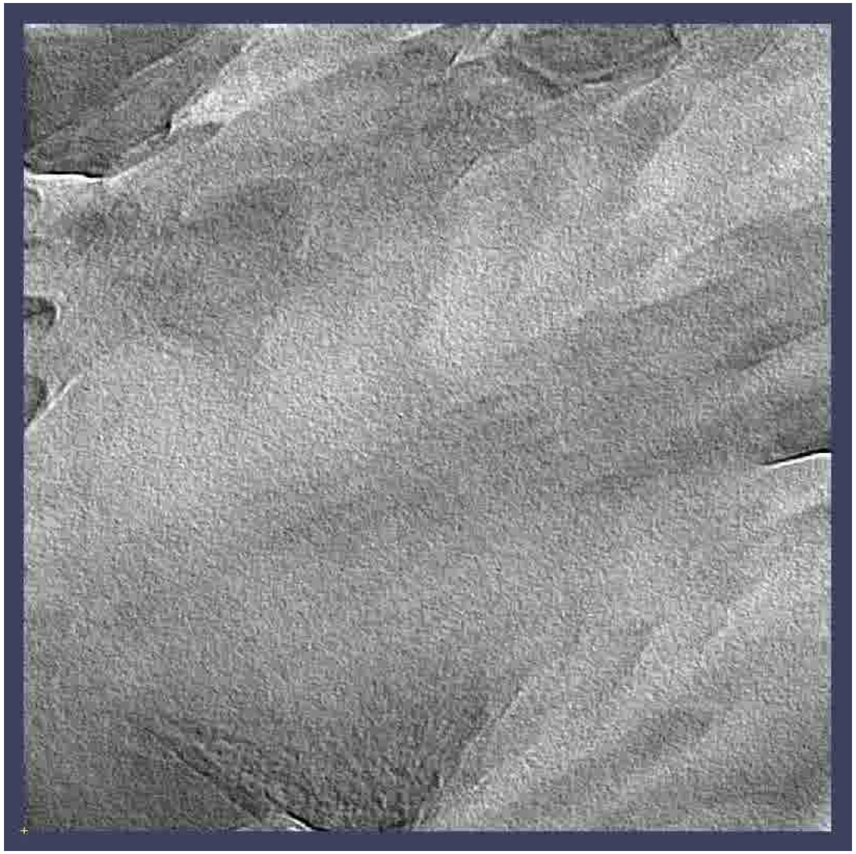
Ultrastructures: cryo electron tomography



<u>Plant Grana membrane</u>
Daum et al. Plant Cell 2010

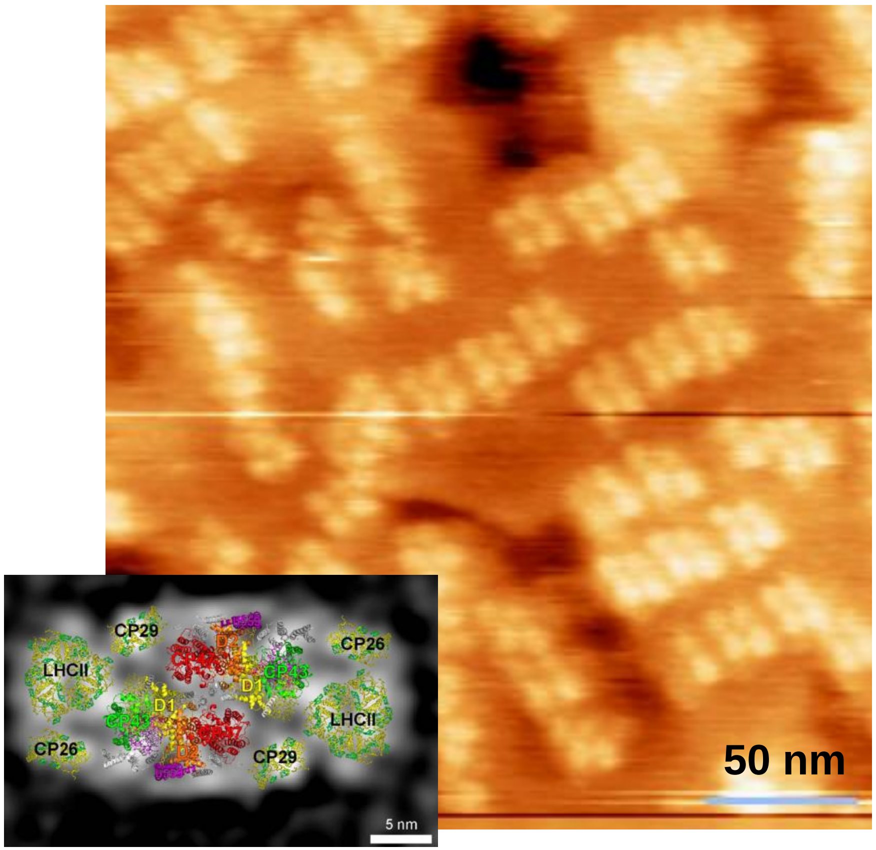
<u>Bacterial chromatophores</u>
Tucker et al. Molecular Microbiology 2010





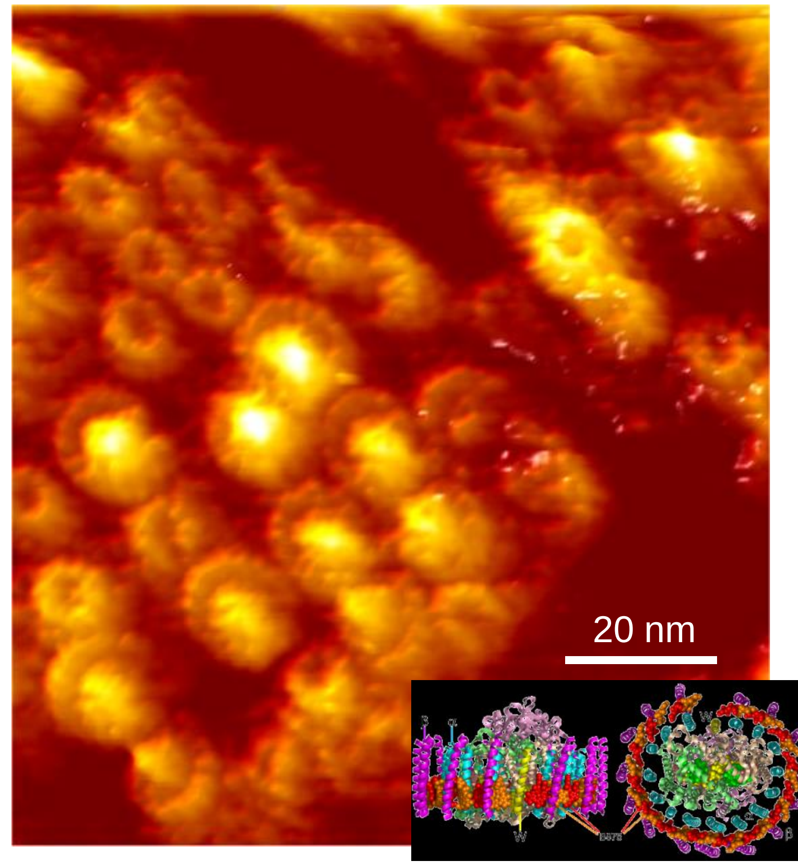
Molecular view: Atomic Force Microscopy

Plant Grana membrane



Sznee et al. JBC 2012

Purple Bacterial membrane



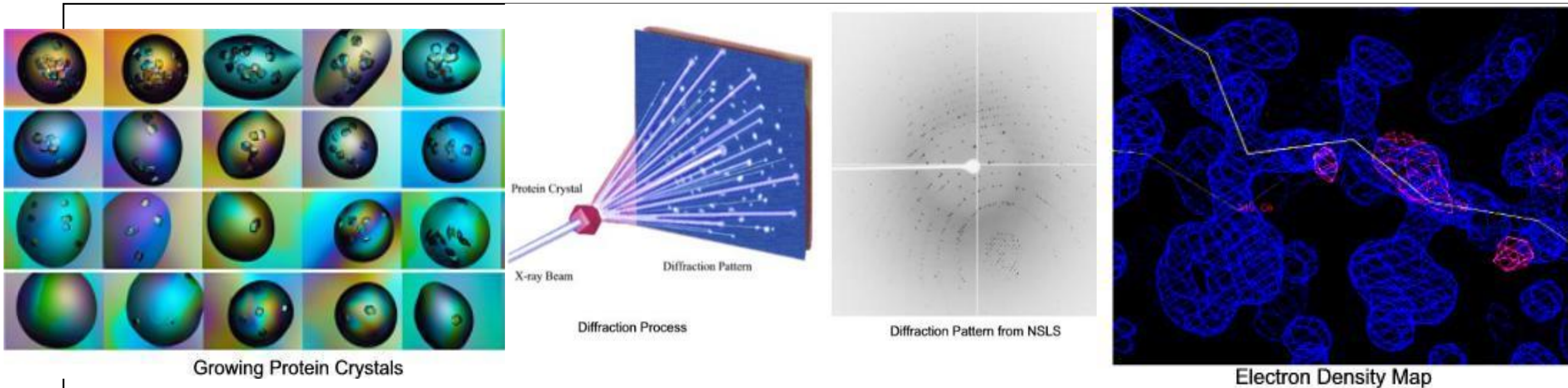
Bahatyrova et al. Nature 2004

3-D crystals of membrane protein-complexes:

three steps:

- ***growth***
- ***x-ray diffraction***
- ***fitting protein density maps***

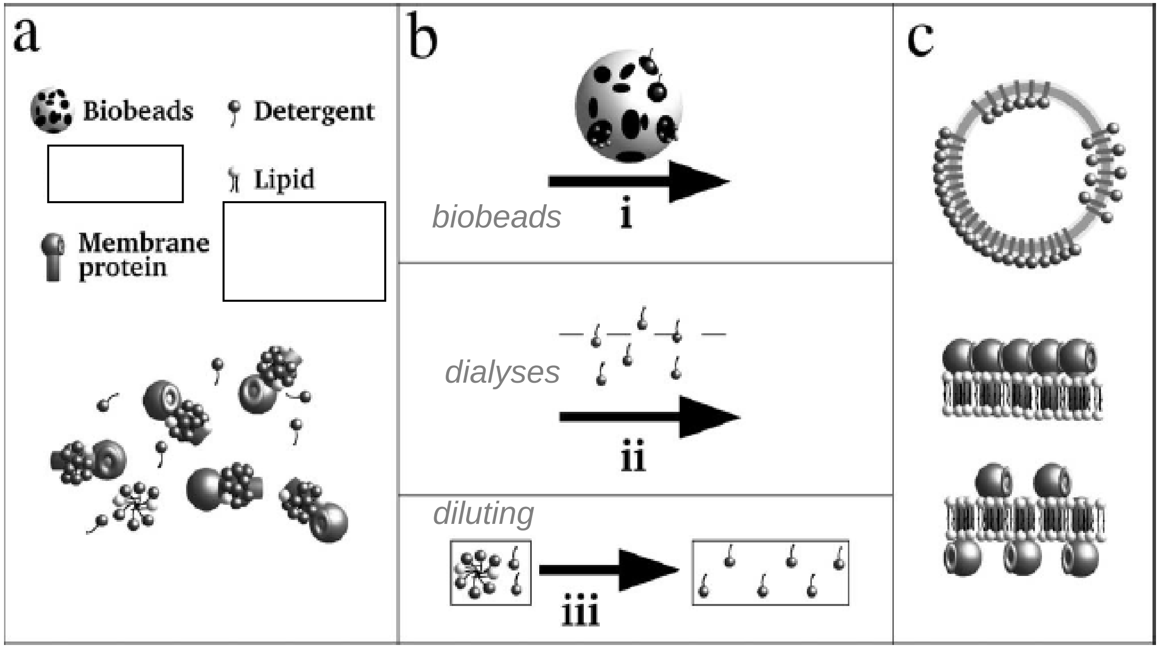
Total count: (2012): 379
Total number of membrane proteins: 15,000
http://blanco.biomol.uci.edu/Membrane_Proteins_xtal.html



Growing Protein Crystals
Diffraction Process
Diffraction Pattern from NSLS
Electron Density Map

2-D crystals of membrane protein-complexes:

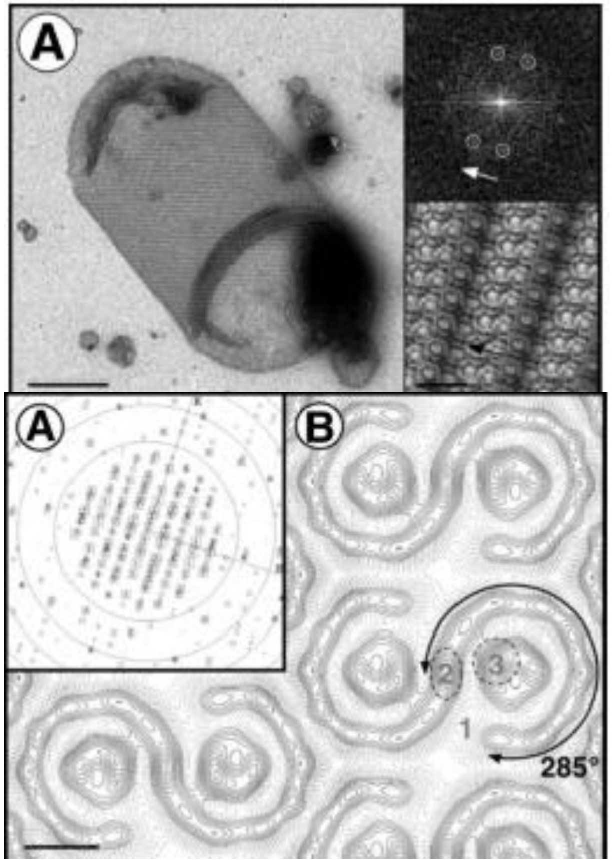
P.J.L. Werten et al./FEBS Letters 529 (2002) 65–72



Vary:

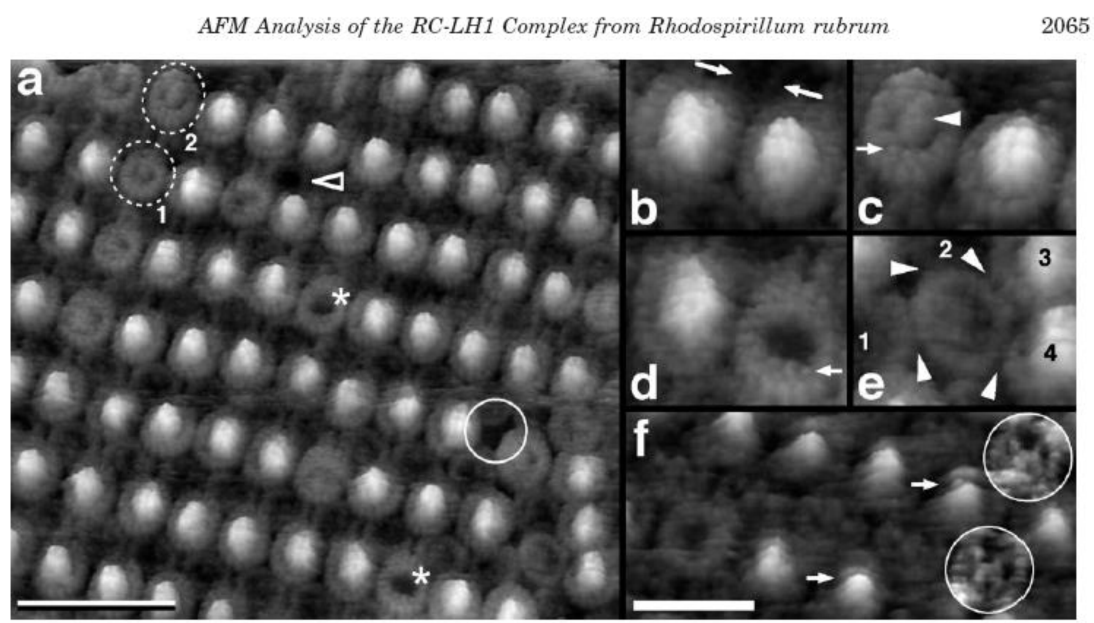
purification procedure, pH, ions, detergent type & concentration

2-D crystals of membrane protein-complexes:



Scheuring et al. JBC 2004

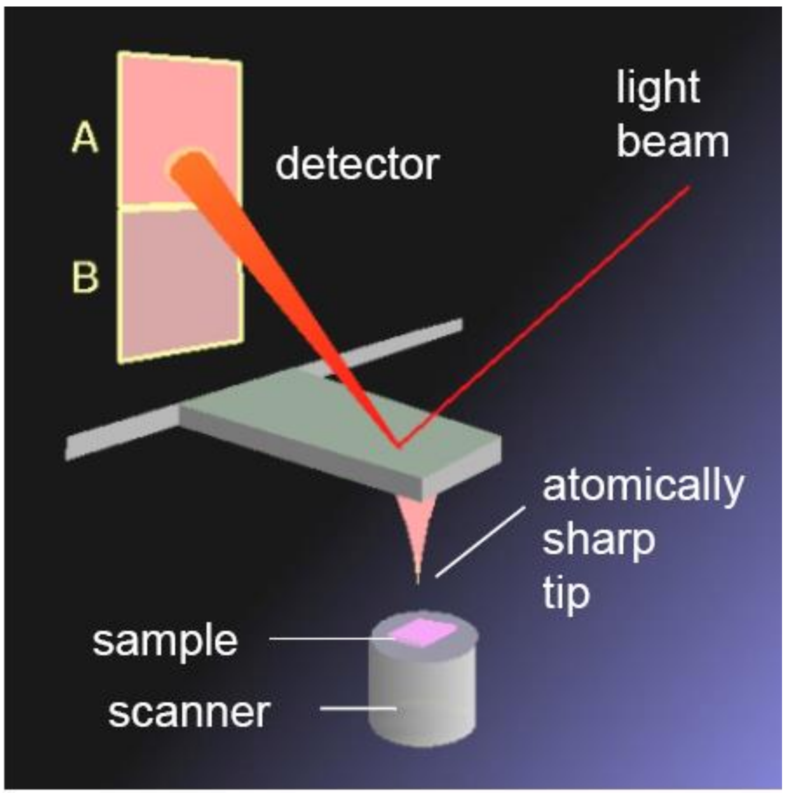
RCLH1 complexes



Fotiadis et al. JBC 2004

Investigation of LH1 and LH2 2-D crystals with AFM:

<u>2D-crystallization</u> of proteins by detergent removal from a lipid-protein-detergent micellar solution



- Cantilever is oscillated at resonance
- Feedback on constant amplitude
- Tip is raster scanned over the surface by piezo

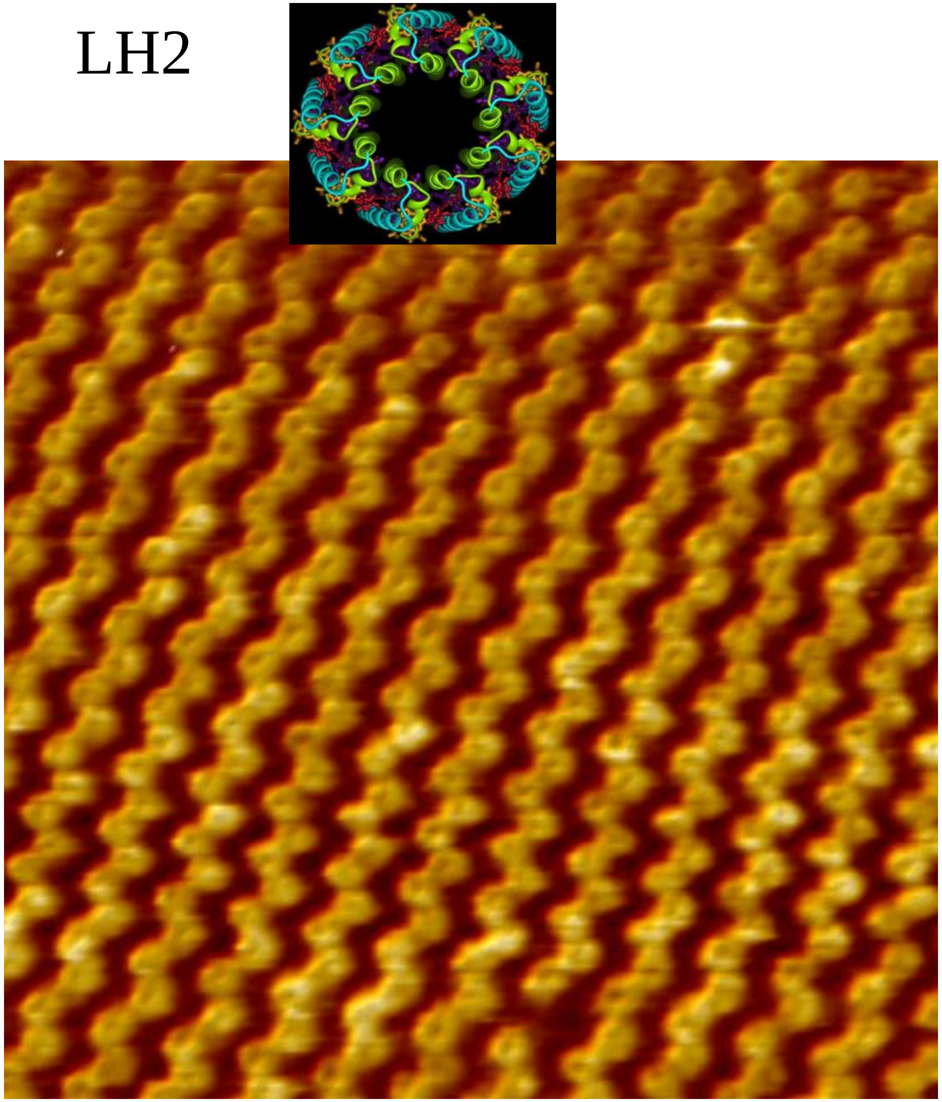
Tapping mode in liquid

Silicon nitride cantilevers - resonance frequencies ~30 kHz (liquid); spring constant 0,5 N/m; tip radius of curvature ~20-30 nm

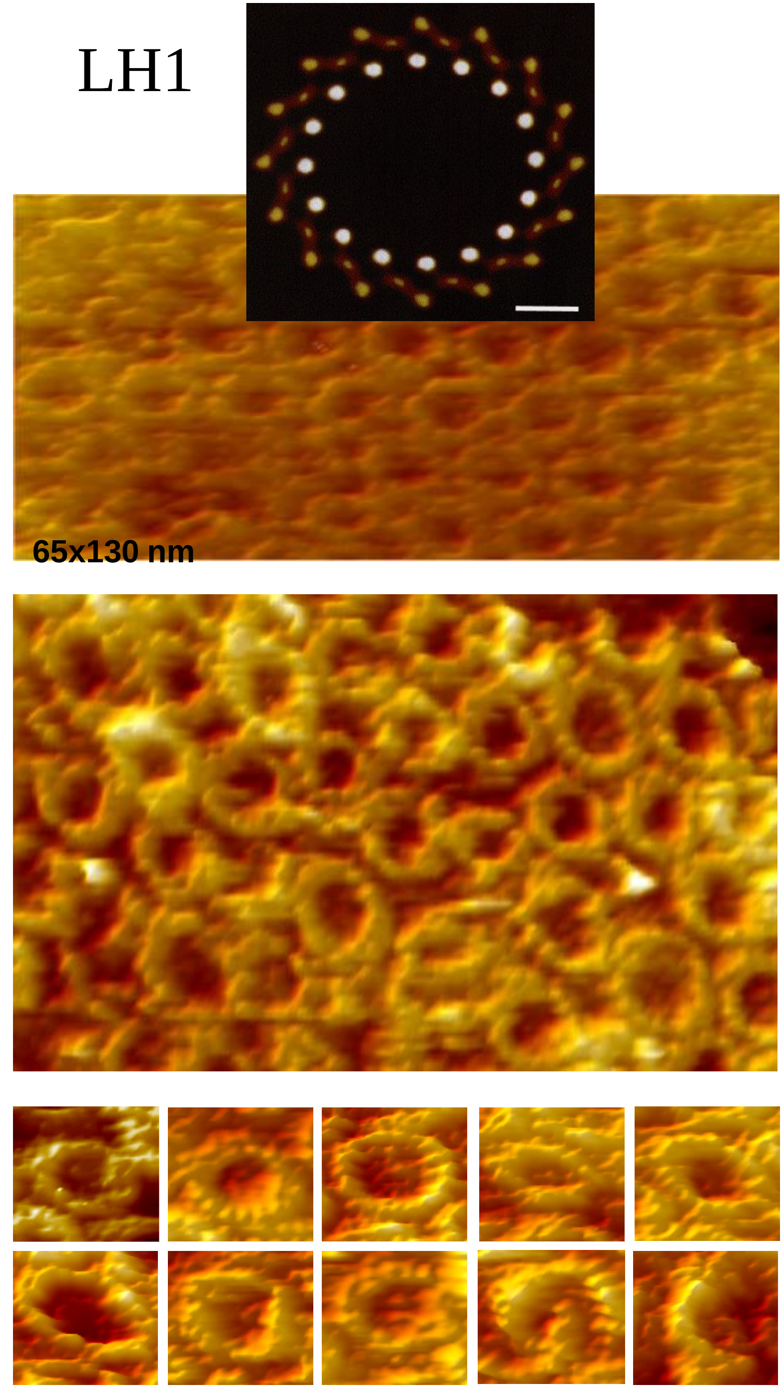
scan range 100÷2500 nm; 256 pixels; line frequency ~2-4 Hz;

tapping amplitude 2-5 nm; damping of the free oscillation 10%

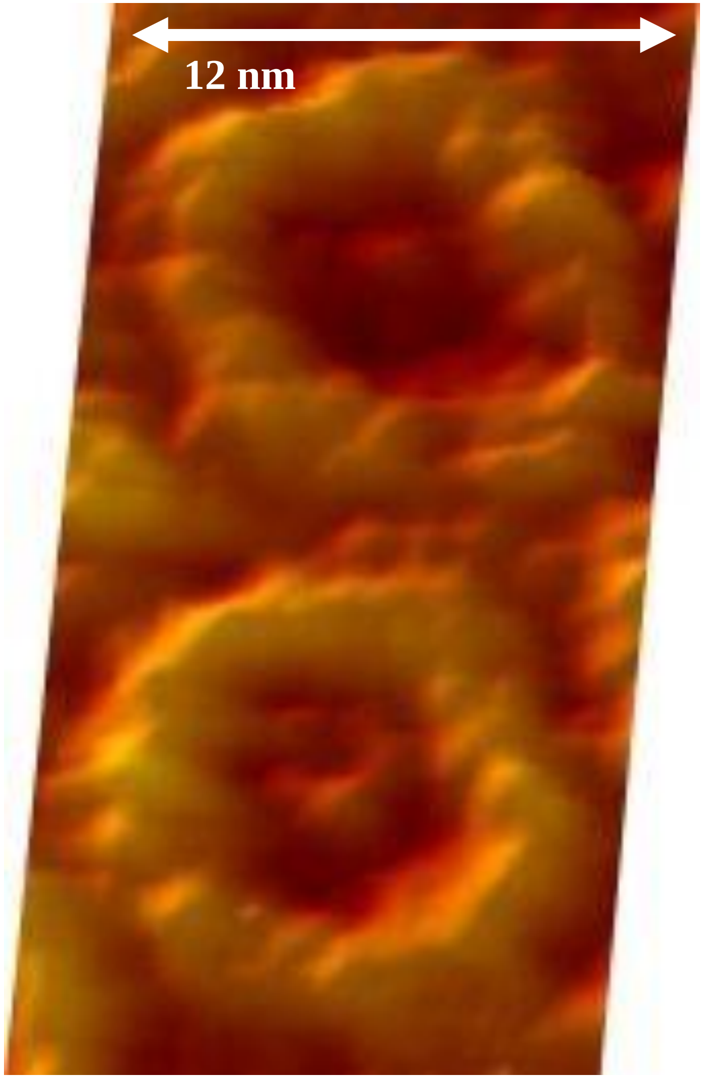
LH2



LH1



Investigation of LH1 2-D crystals with AFM:



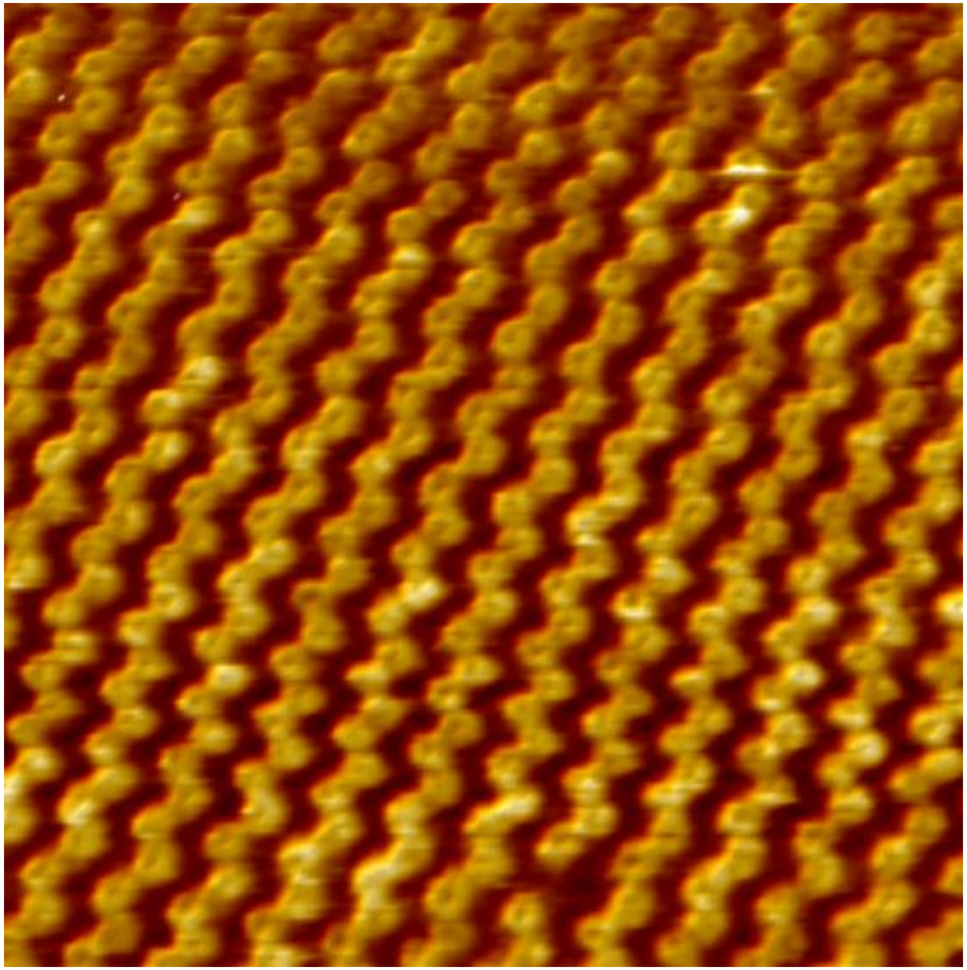
LH1 rings show high degree of flexibility.

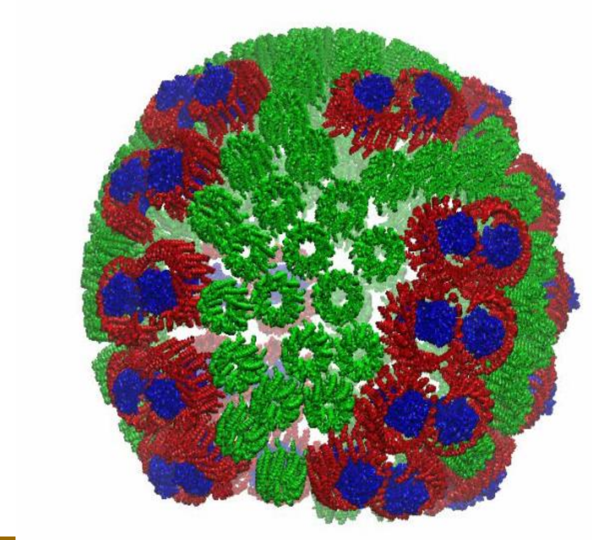
Hypothesis:

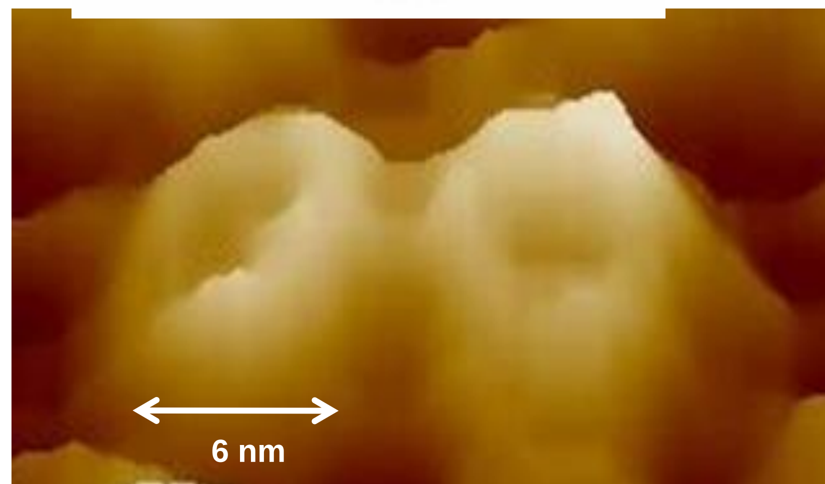
this flexibility allows the LH1 ring to be distorted, allowing

1. ***dense packing***
2. ***quinon exchange and***
3. ***specific RC-LH1 interactions***

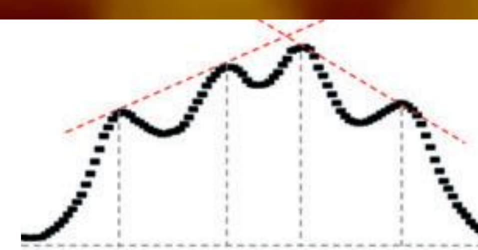
2D crystals ⟷ natural systems





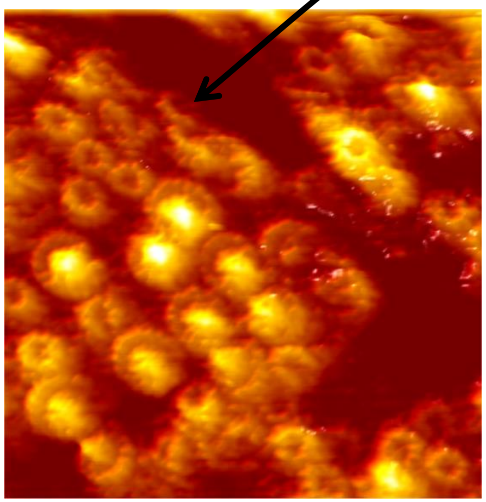


Lu Ning-Liu, Thijs J. Aartsma and Raoul N. Frese; *FEBS J.* (2008).

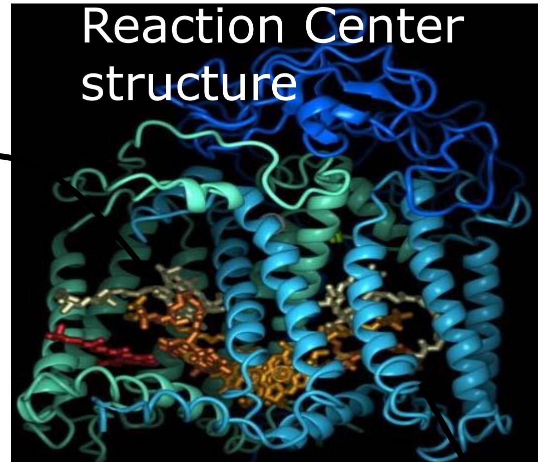


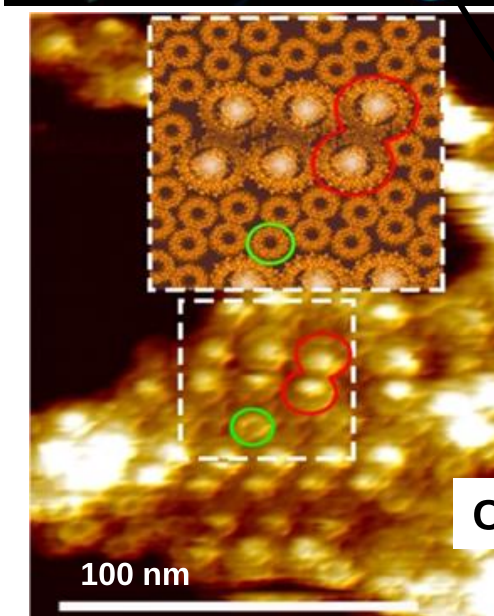
From 2D to 3D to dynamics: a special case

AFM



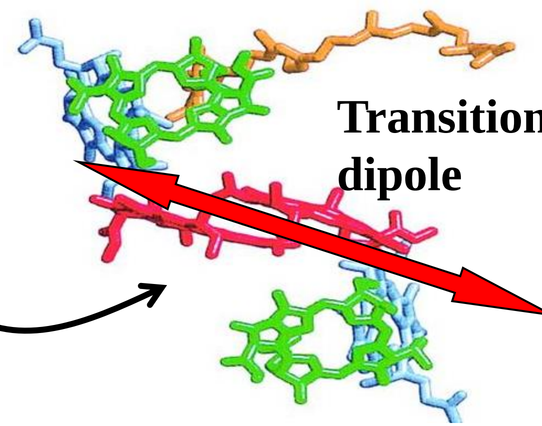
Topography





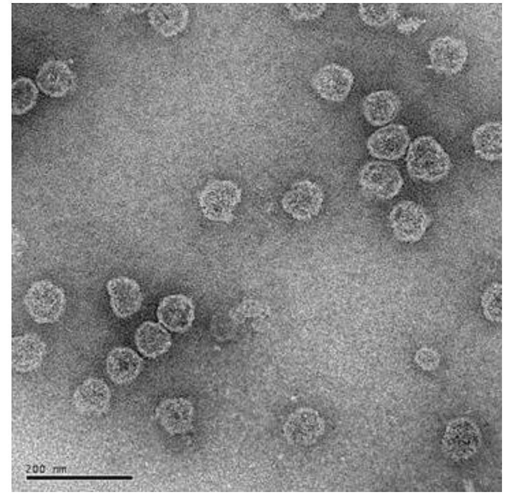
Correct AFM image

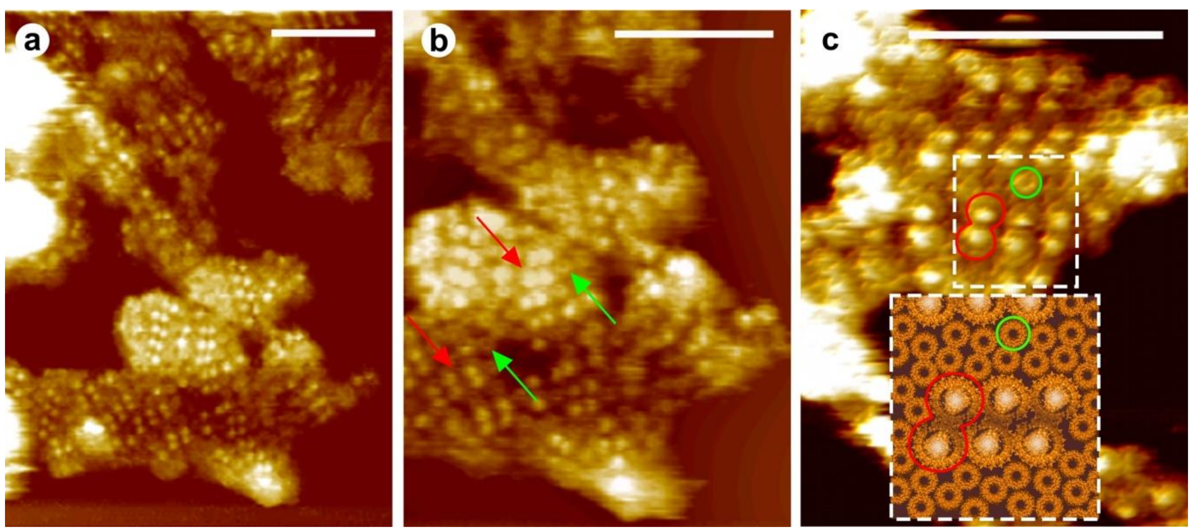
spectroscopy



Orientation
chlorophylls

Natural systems on surfaces

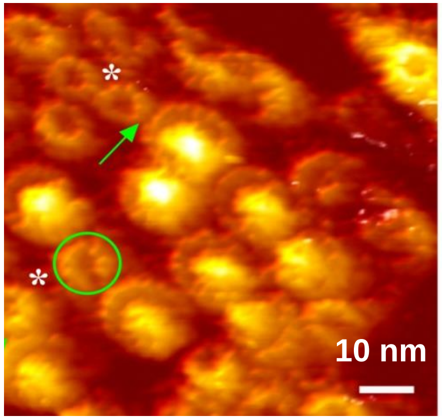


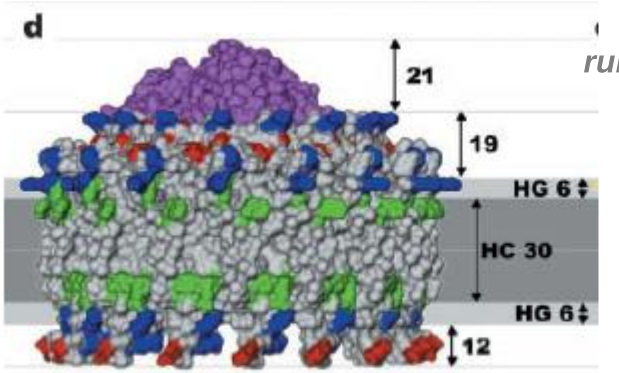


Svetlana Bahatyrova, Raoul N. Frese C. Alistair Siebert, John D. Olsen, Kees van der Werf, Rienk van Grondelle, Robert Niederman, Per A. Bullough, Cees Otto and Neil C. Hunter *Nature* (2004).

Accuracy of height measurements reveals soft tapping regime:

sphaeroides



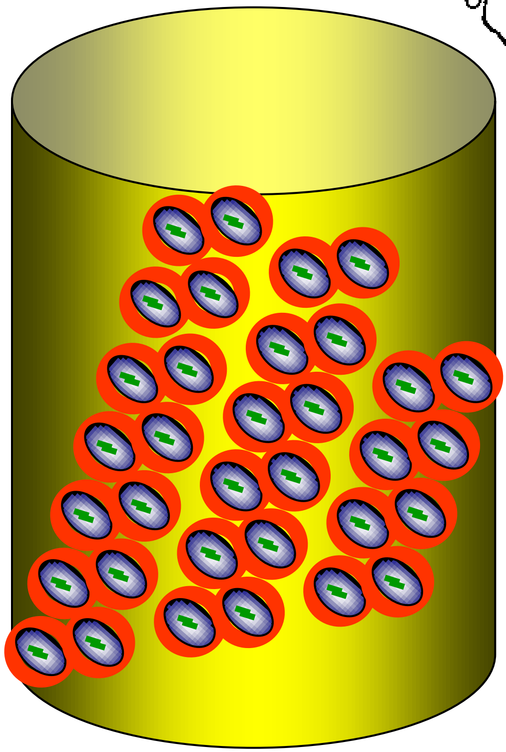


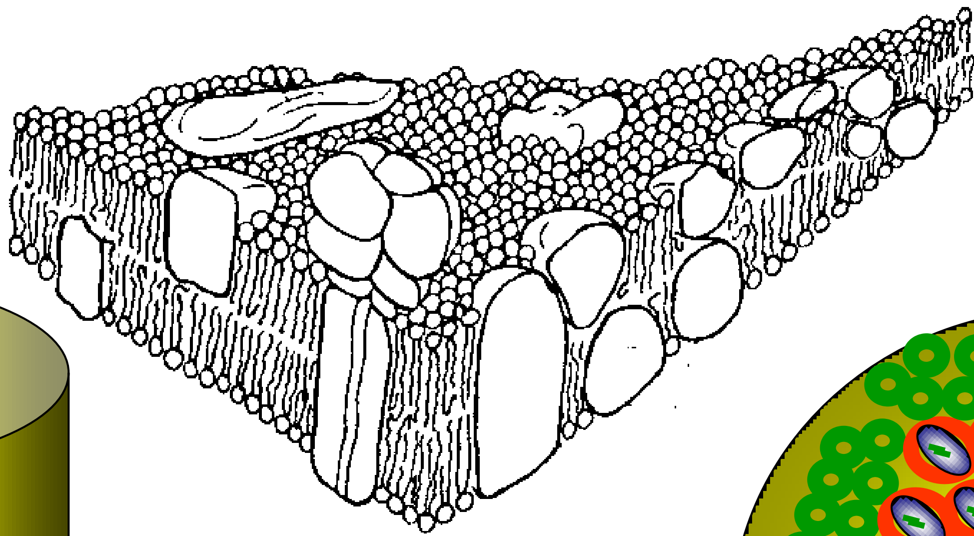
rubrum

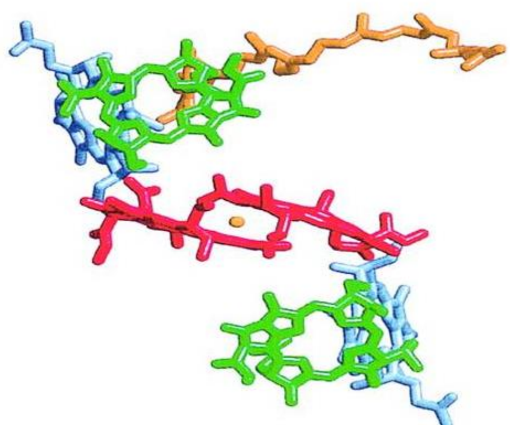
Fotiadis et al. JBC 2004

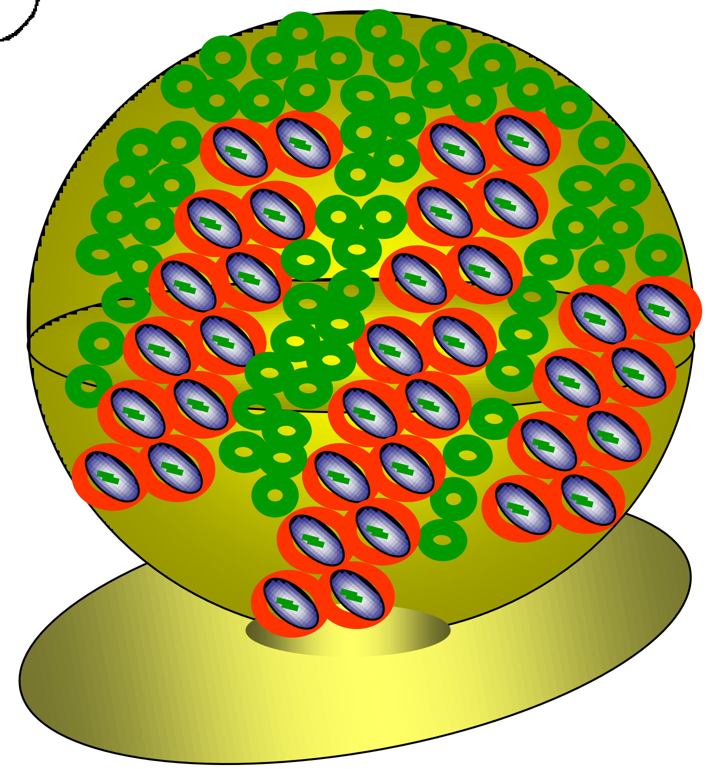
Measured topography in nm:		***rubrum* model:**
Full height above surface:	**8.5**	**(9.4)**
LH1 above lipid:	**1.4**	**(1.9)**
H-unit above LH1:	**3.5**	**(4.0)**
LH1 cytoplasmic side:	**0.8**	**(1.2)**

Protein membranes





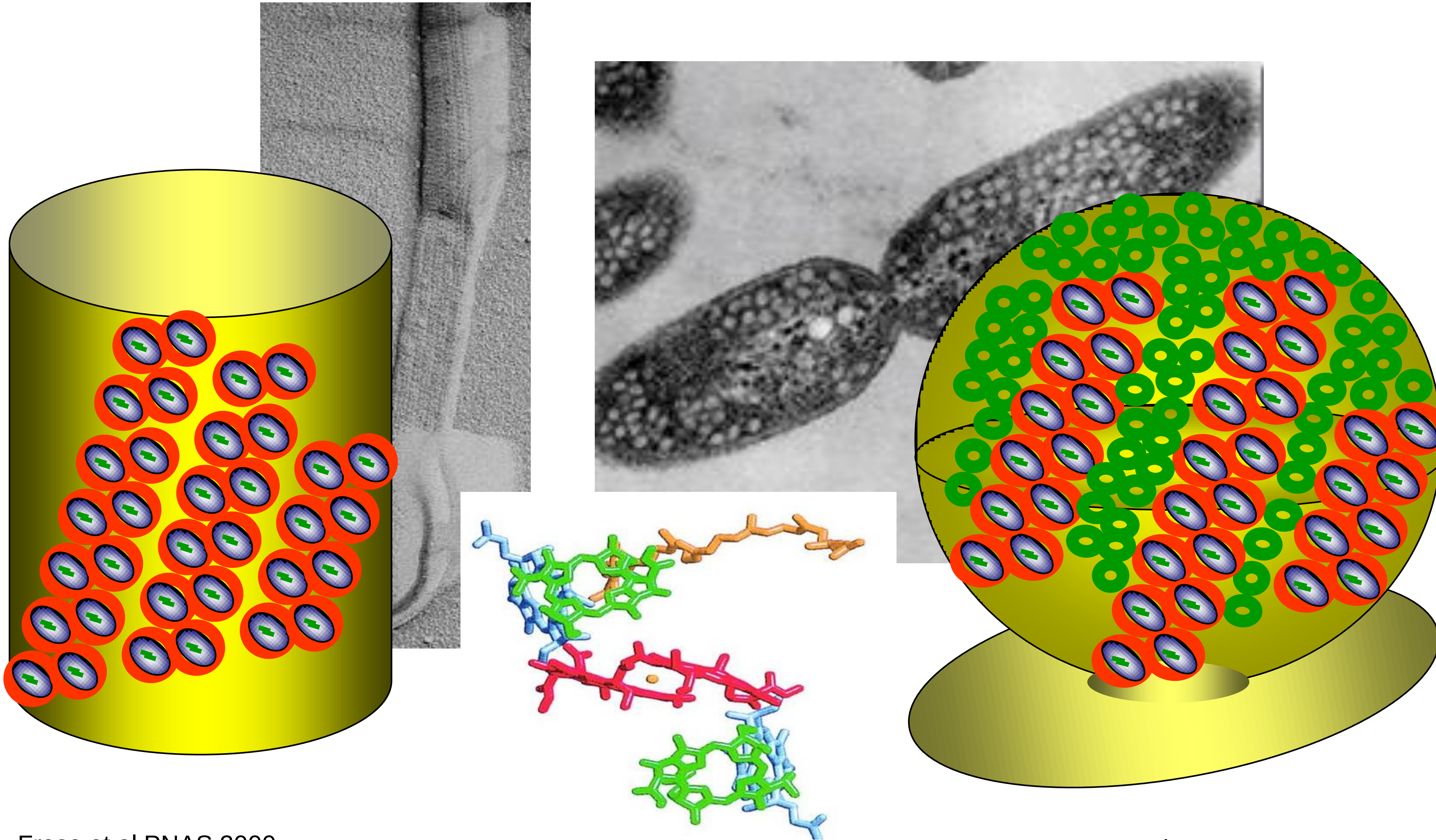




Raoul N. Frese, John D. Olsen, Rikard Branvall, Willem H. J. Westerhuis, C. Neil Hunter and Rienk van Grondelle. *Proc. Natl. Acad. Sci* USA (2000)

Raoul N. Frese, C. Alistair Siebert, Robert A. Niederman, C. Neil Hunter, C. Otto and Rienk van Grondelle; *Proc. Natl. Acad. Sci.* USA (2004)

Protein ordering on a cylinder and a sphere

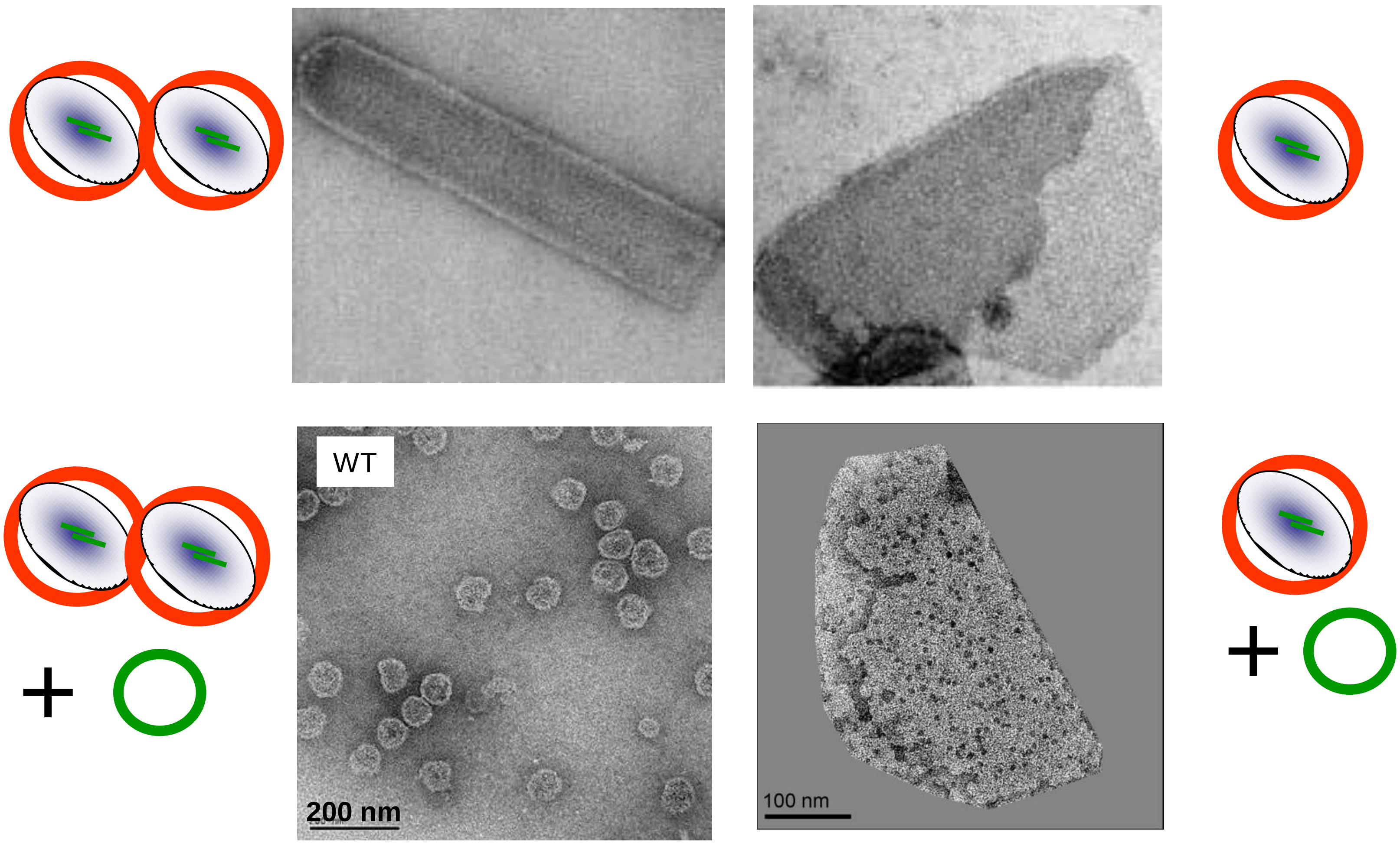


Frese et al PNAS 2000

Frese et al PNAS 2004

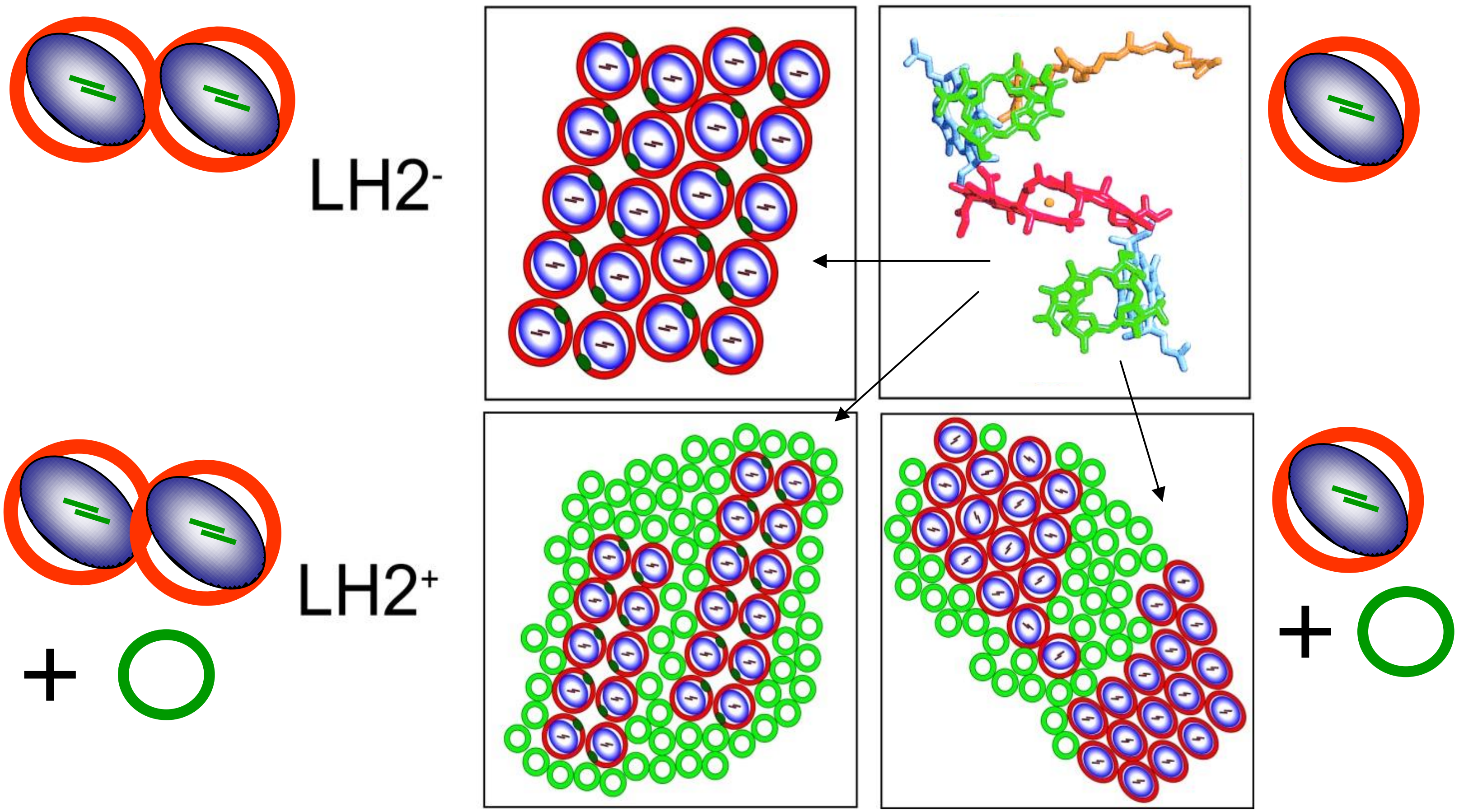
What drives the organization?

Genetically altered membranes



CA. Siebert, PhD thesis, Sheffield 2005

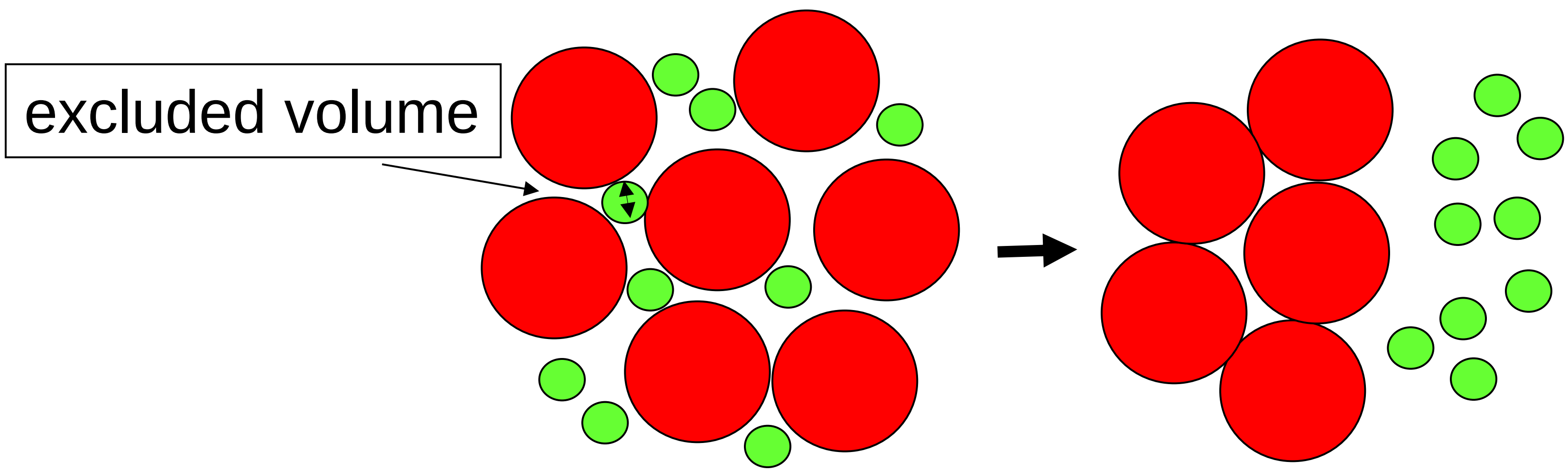
Four packing lattices,
different surface interactions, same features



ordering and domain formation cannot be due to specific interactions

What drives domain formation and order?

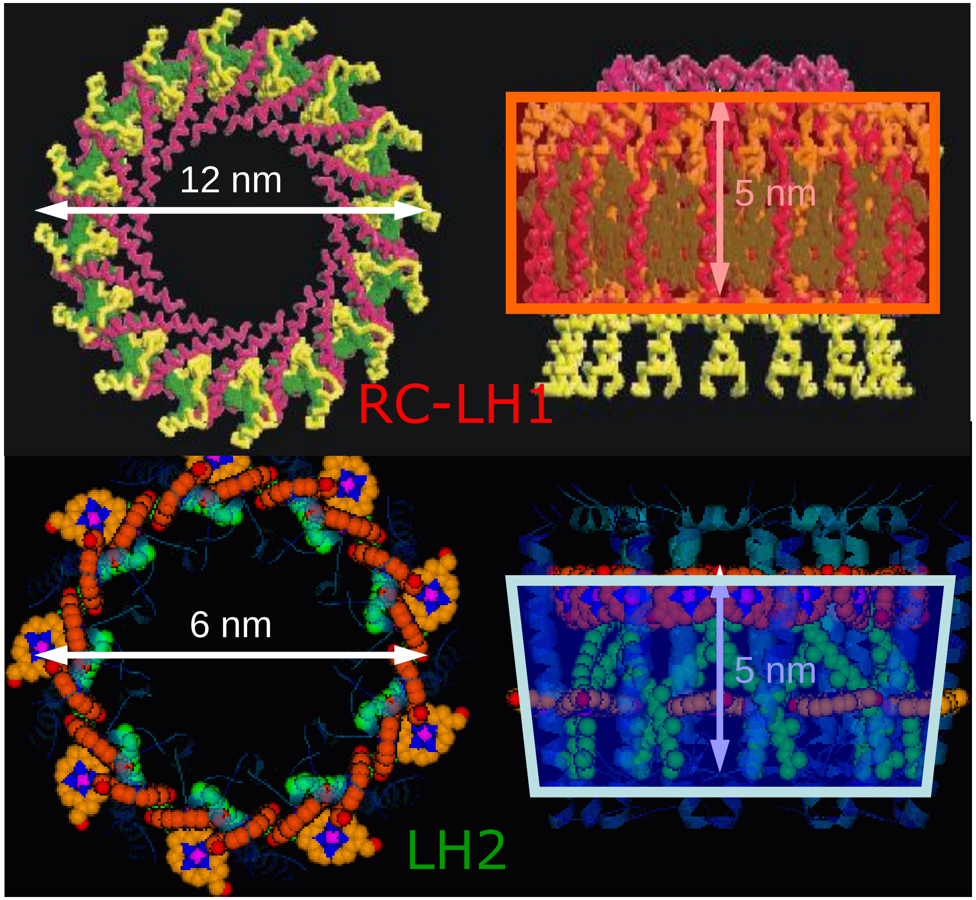
Phase behaviour in dense mixtures:
minimizing free energy: **F= U-TS**, U = 0, ***only entropy remains***



depletion-induced attraction or
macromolecular crowding

Membranes as a binary system of non-interacting particles,
include only intrinsic protein curvature and size

Proteins as colloidal particles: no attractive forces



Input: shapes and sizes only

Size ratio

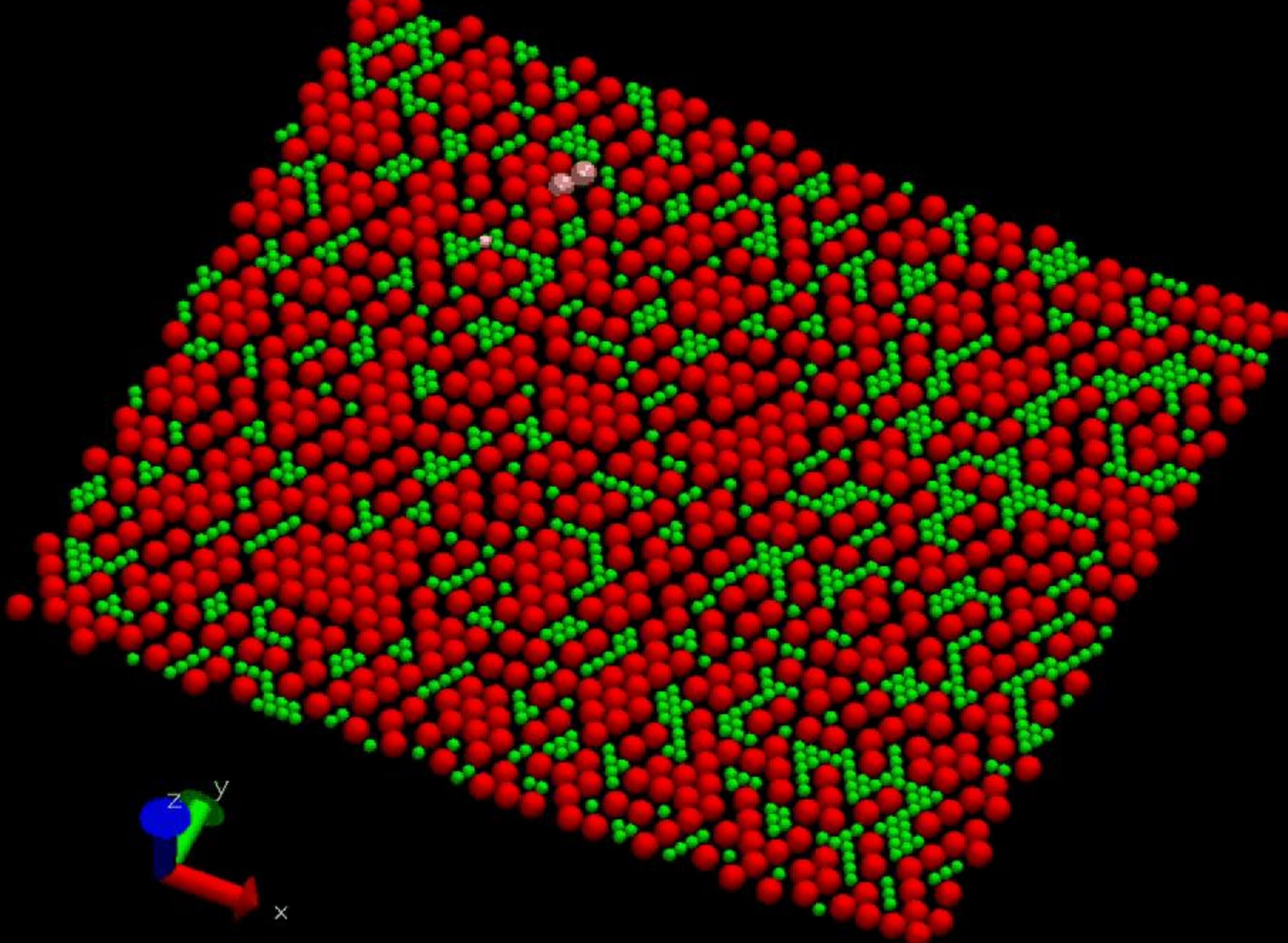
LH2/RC-LH_1 → q = 0.5

LH2 has a non-zero spontaneous curvature

RCLH1 in contact to LH2

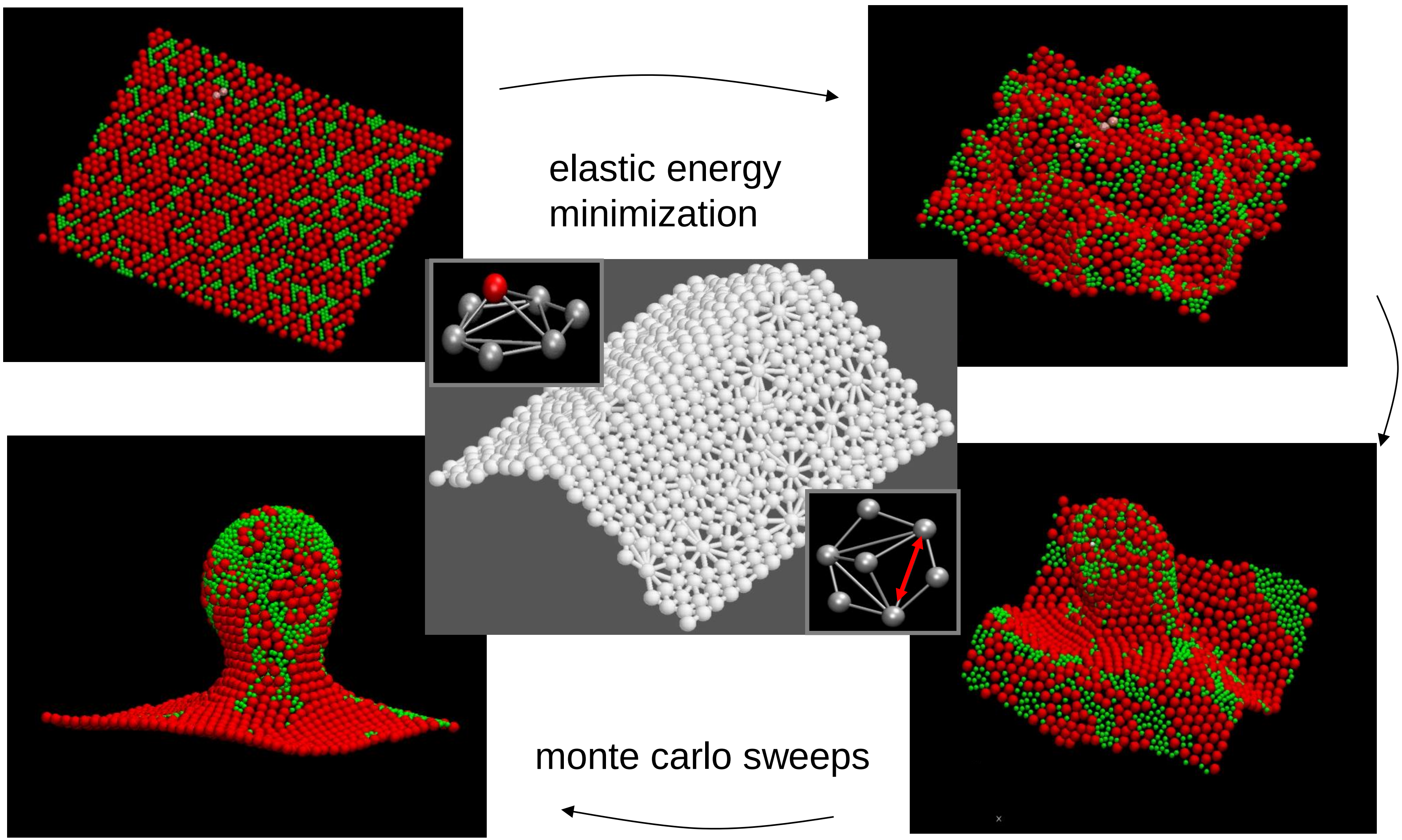
is deformable: adapts to

LH2-curvature



z
y
x

Membranes as a binary system



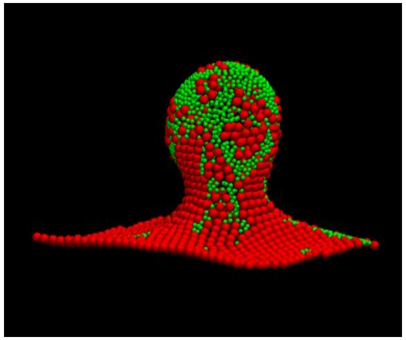
RN. Frese, JC. Pàmies, JD. Olsen, S Bahatyrova, CD. de Wit, T Aartsma, C Otto, C Hunter, D Frenkel, R van Grondelle; *Biophysical Journal (2008)*

Supramolecular self-organization and ultrastructure

1. *Domains can be formed without special interactions*

2. *Size differences can create fluid phases*

3. *Shape differences create budded membranes*

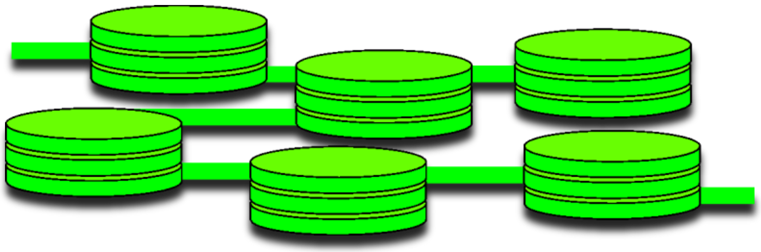


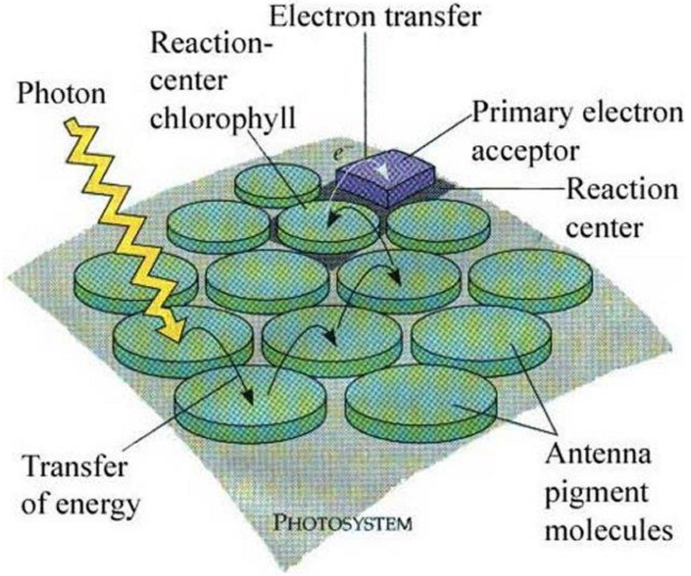
Photosynthesis: colloidal networks

1. *Network of networks lead to ultrastructure*

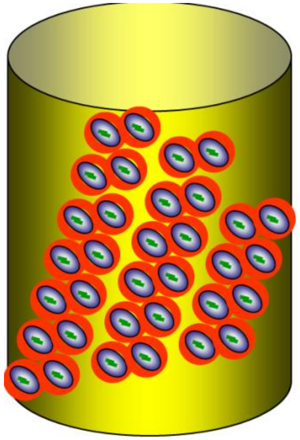
2. *Networks are designed for close packing and diffusion*

3. *Flexibility in networks induced by flexibility in size/shape*



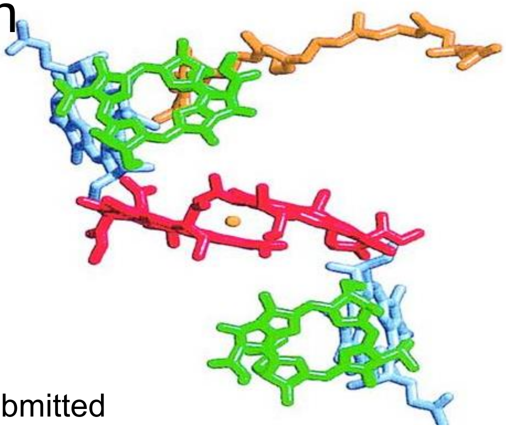


Higher hierarchical order

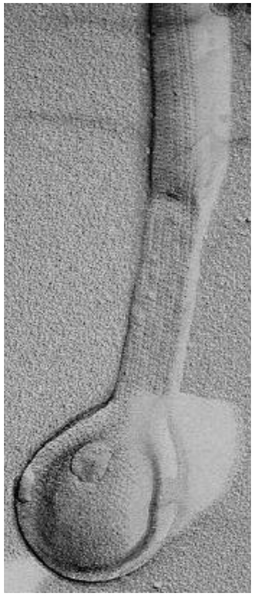


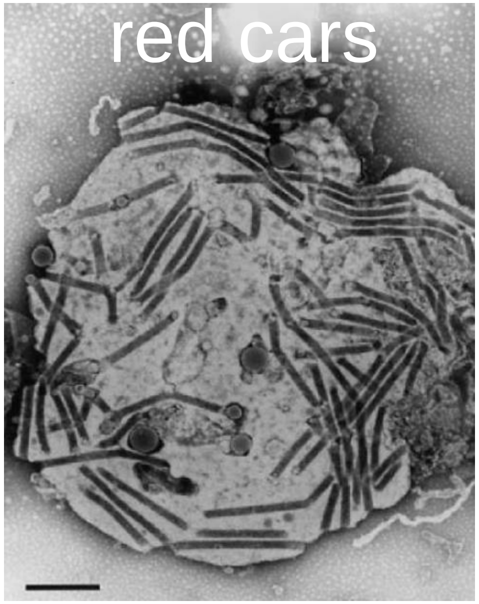
Membranes span the entire cell length with

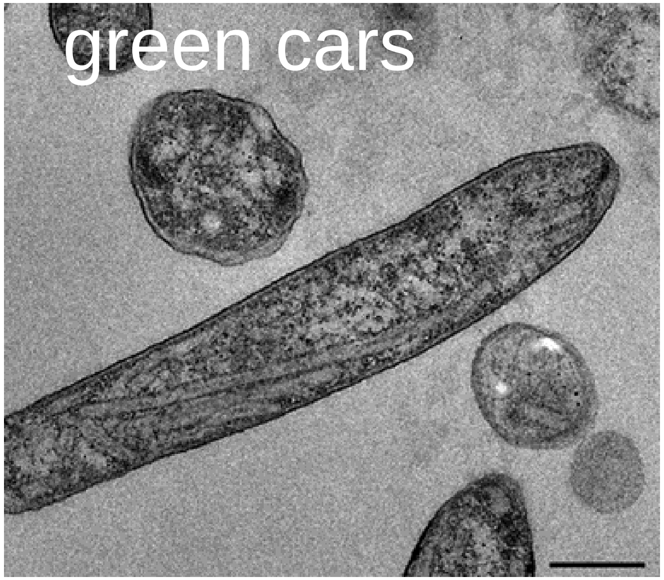
- neurosporene as carotenoid,
- grown in the dark.



D'Haene et al. submitted



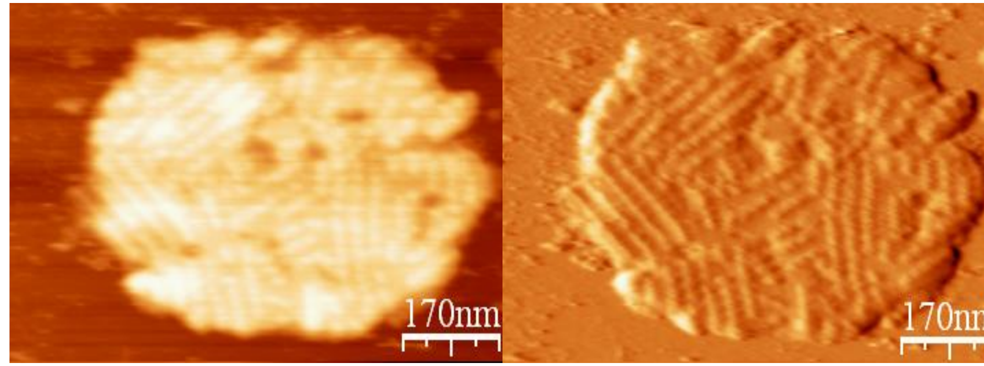


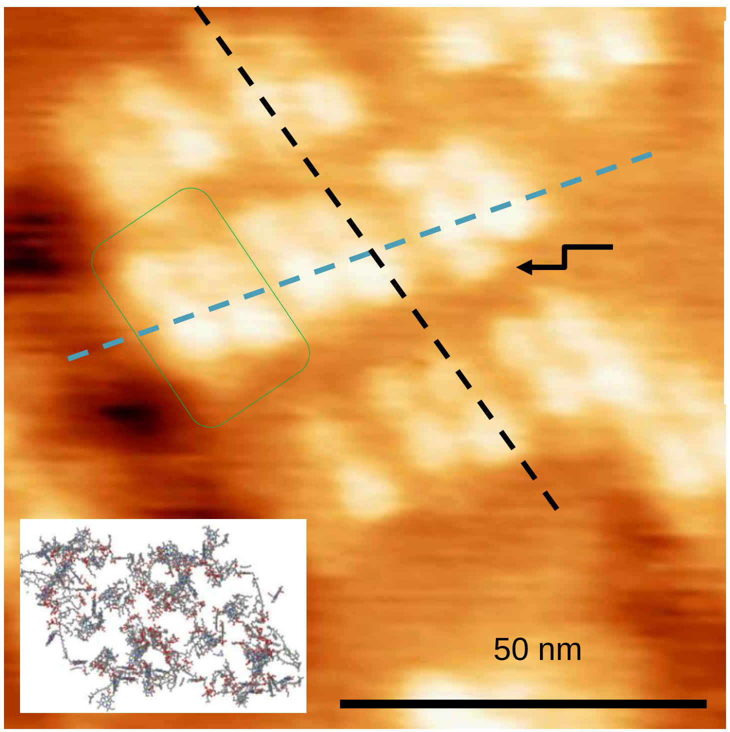


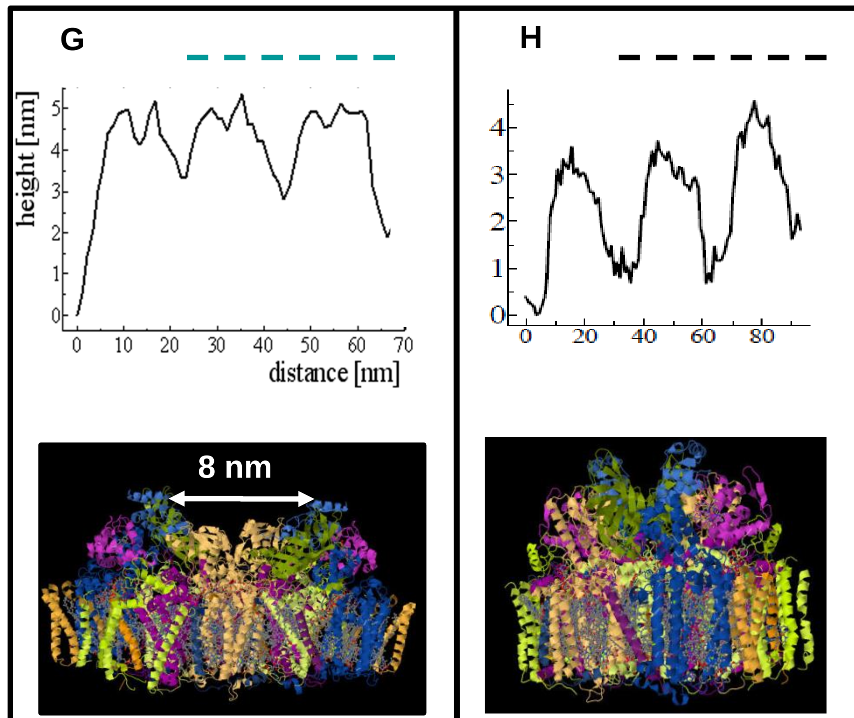
EM: Jungas et al. 99; Siebert et al. 2004

Molecular view grana membrane spinach

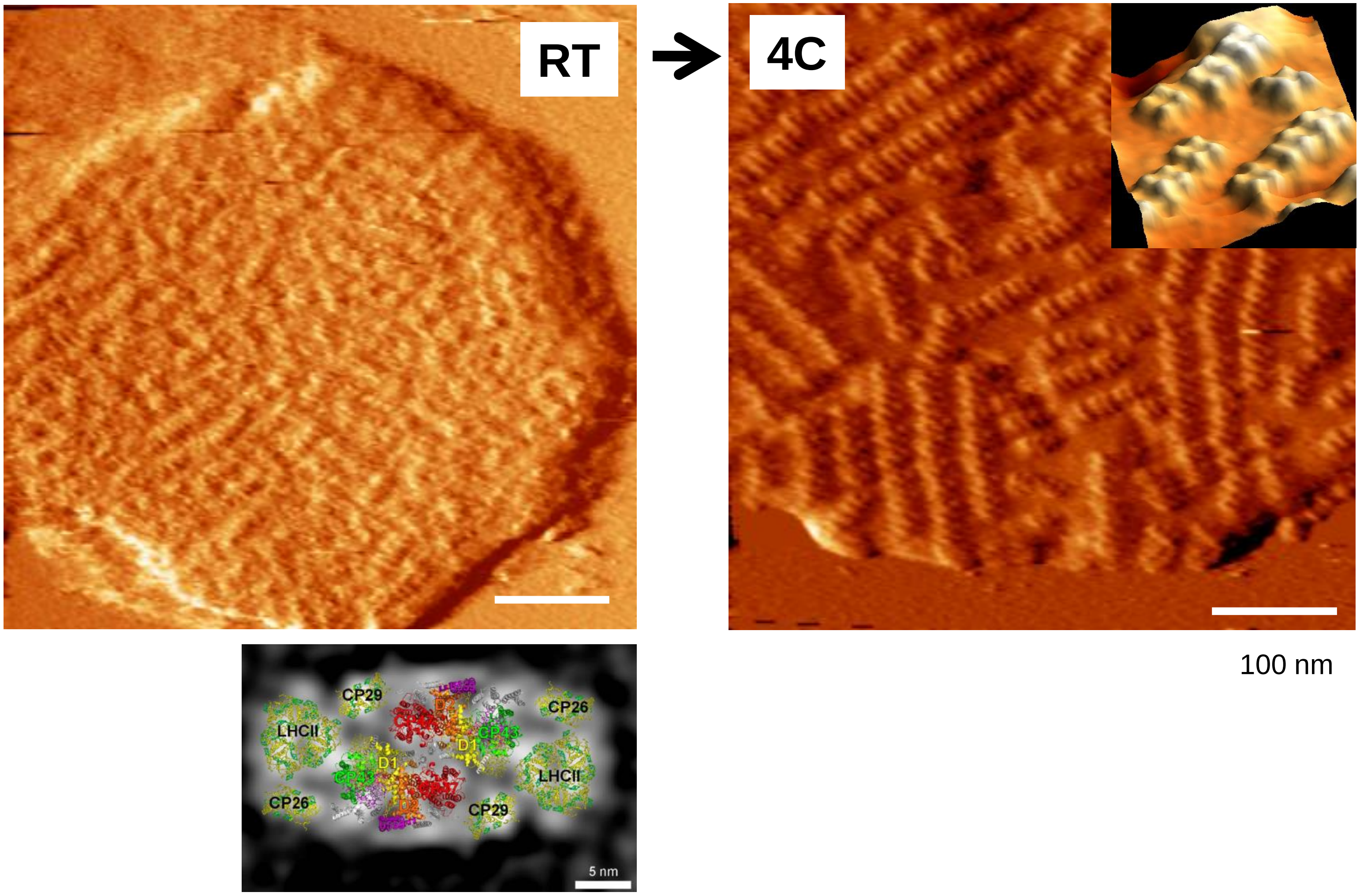
High resolution jumping mode AFM







Dynamics on the surface: phase transitions



Kinga Sznee, Jan P. Dekker, Remus T. Dame, Henny van Roon, Gijs J. L. Wuite, Raoul N. Frese; (2011) Journal of Biological Chemistry

End of Part 1

Investigating photosynthetic assemblies indicates versatility of biological properties.

Membranes are more gel like then fluid, more crystalline then disordered.

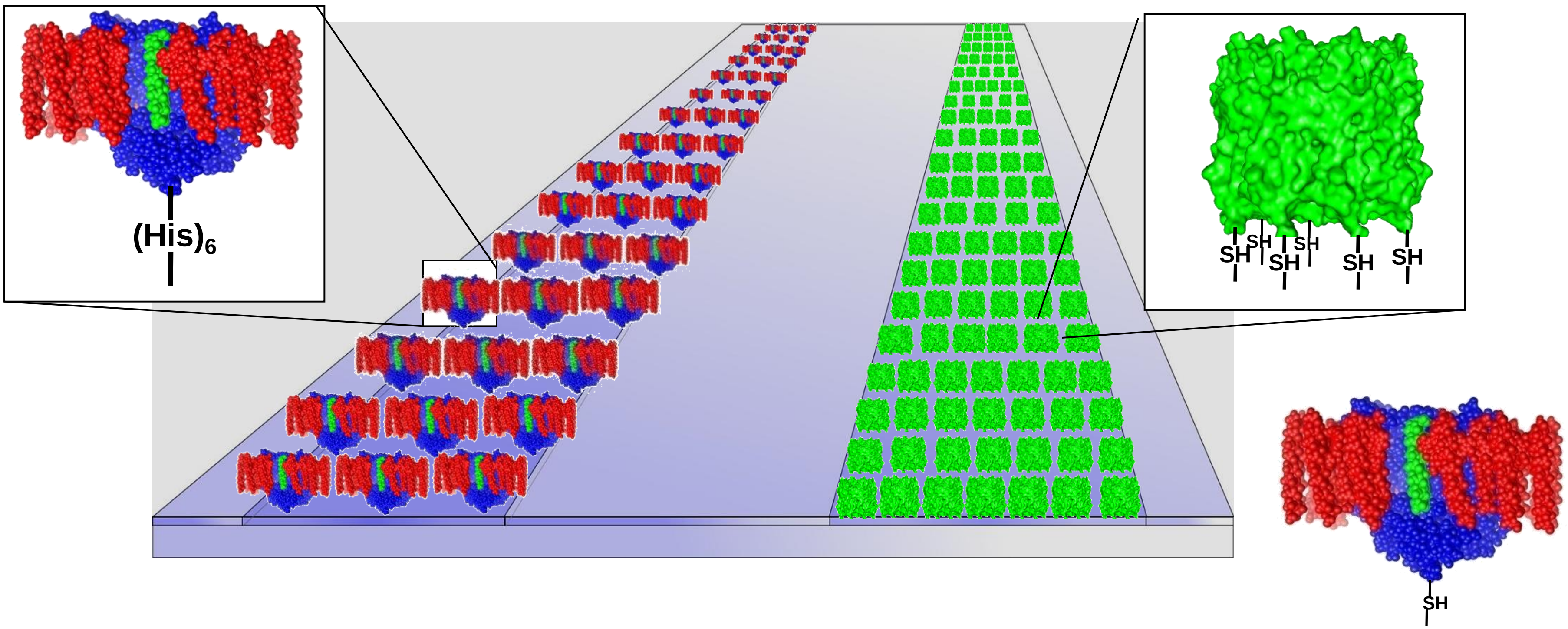
Nature is playing with building blocks, can we do too?

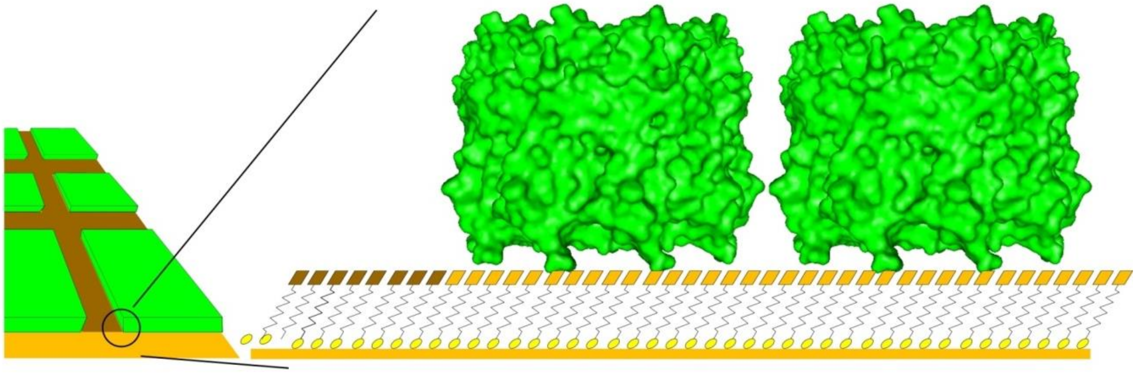
Can we retain dynamics within devices?

Arent proteins just molecules and why should they be more labile than other molecules? (especially in a semi liquid environment)…

In fact: keep an eye out for improved performance of proteins outside the cell

Part 2: photosynthesis on gold electrodes





Why (on earth)?

Biophysics of photosynthesis

- Direct measurement of photosynthetic performance

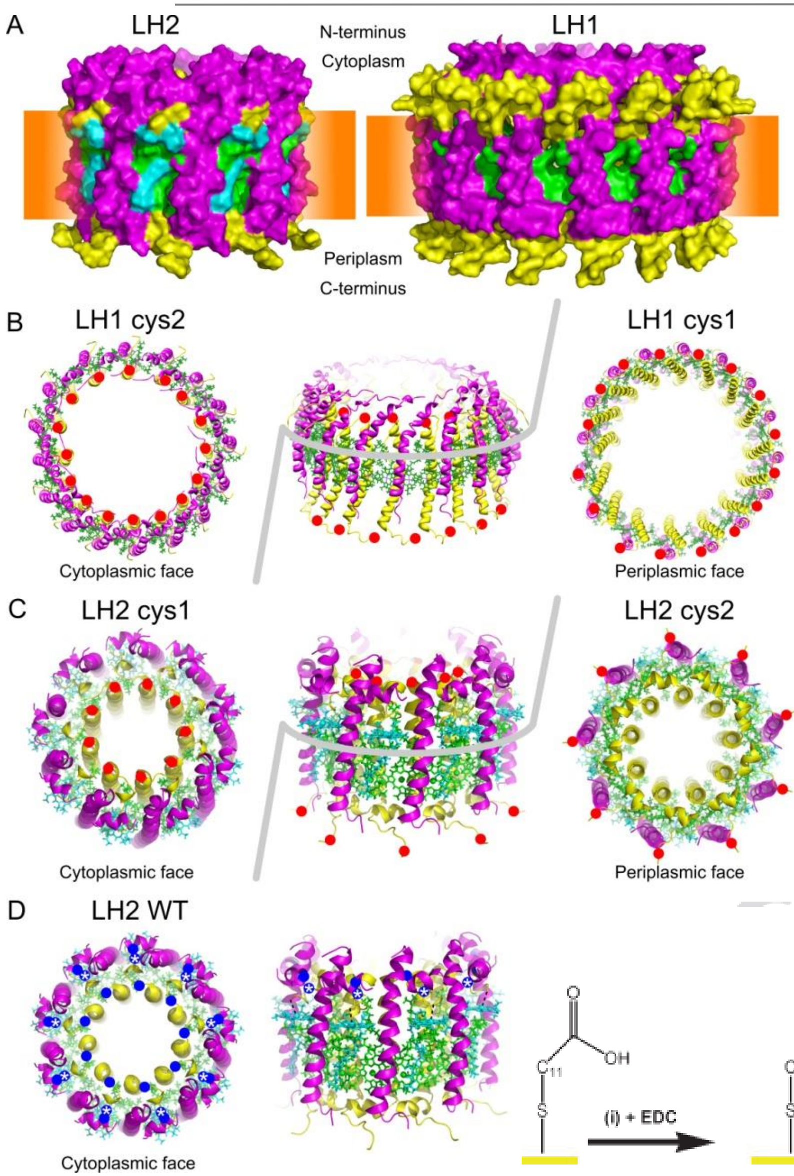
Biosensors

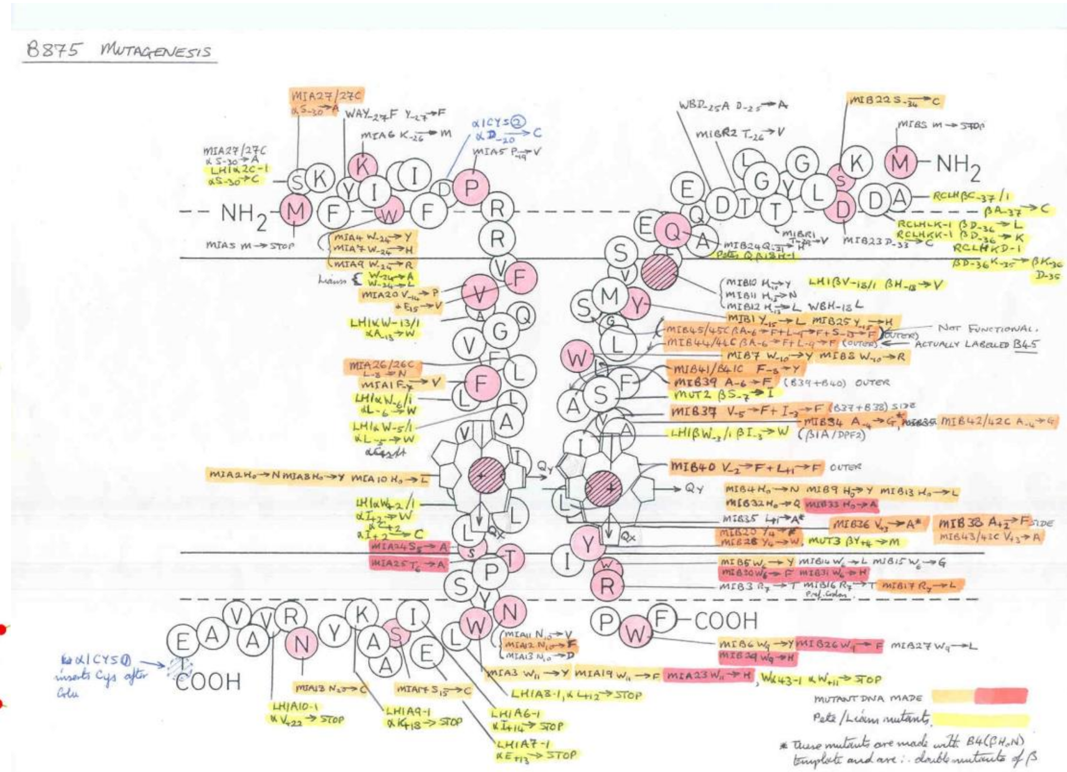
- Photosynthaic sensors for pollutant detection
- as template for other (medical) biosensors

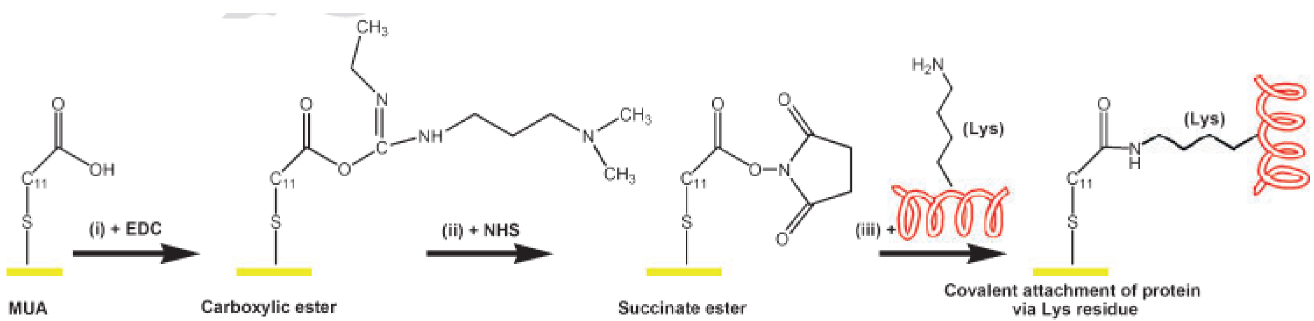
Biophotovoltaics

- Photosynthetic material as cheap active layer; note: photosynthetic material is **made** by solar energy!
- Material properties as novel **dynamic** solar cells.
- As template for photoactive catalysts **with a matrix.**

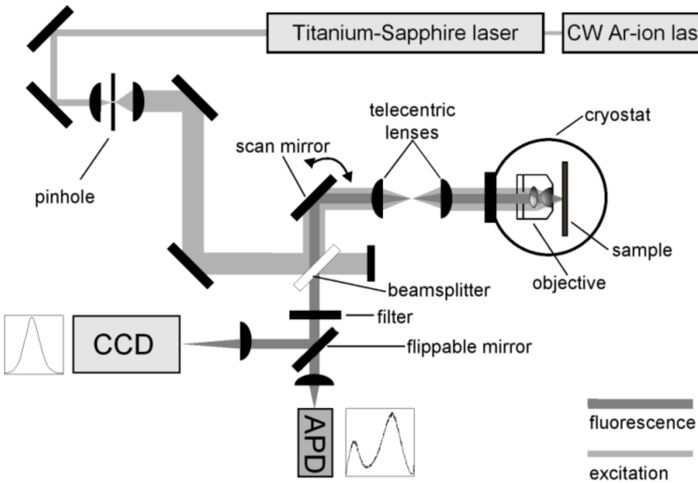
Controlled binding of LH1 and LH2 with cys and lys

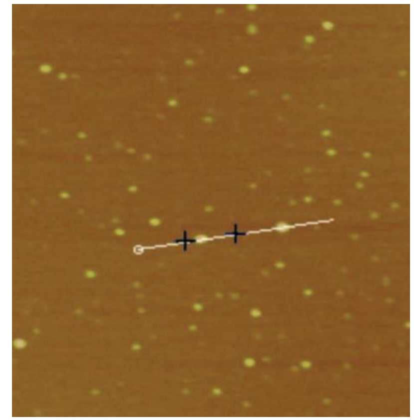


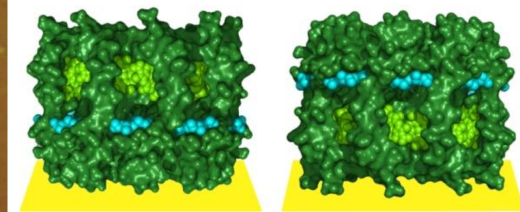


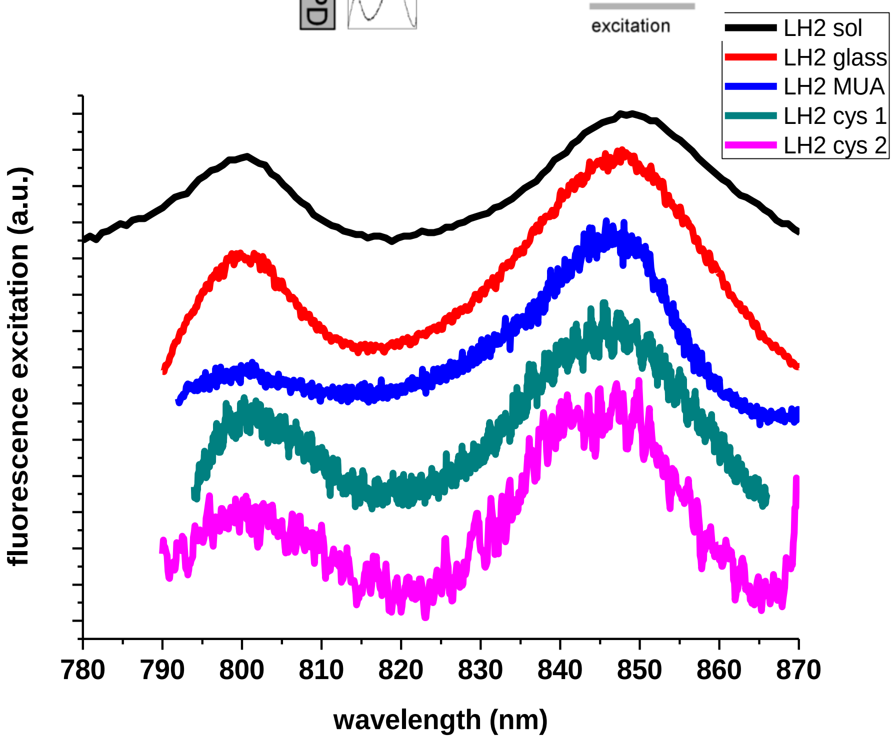


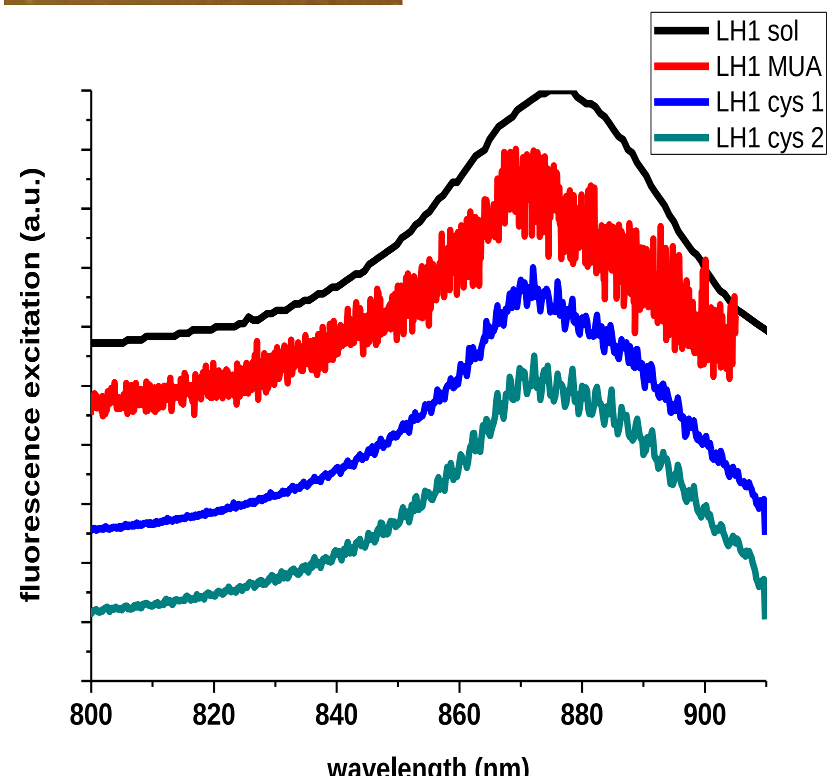
LH1 and LH2 on gold







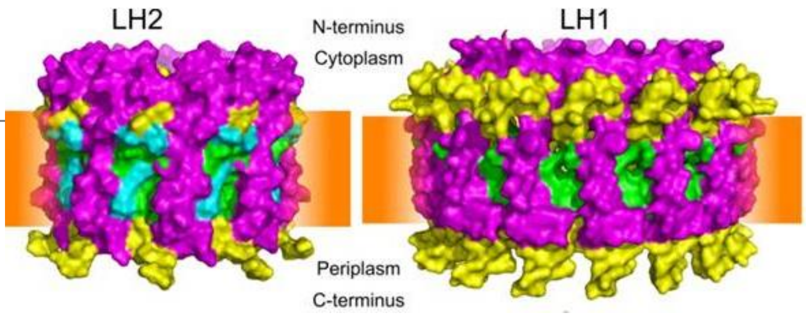


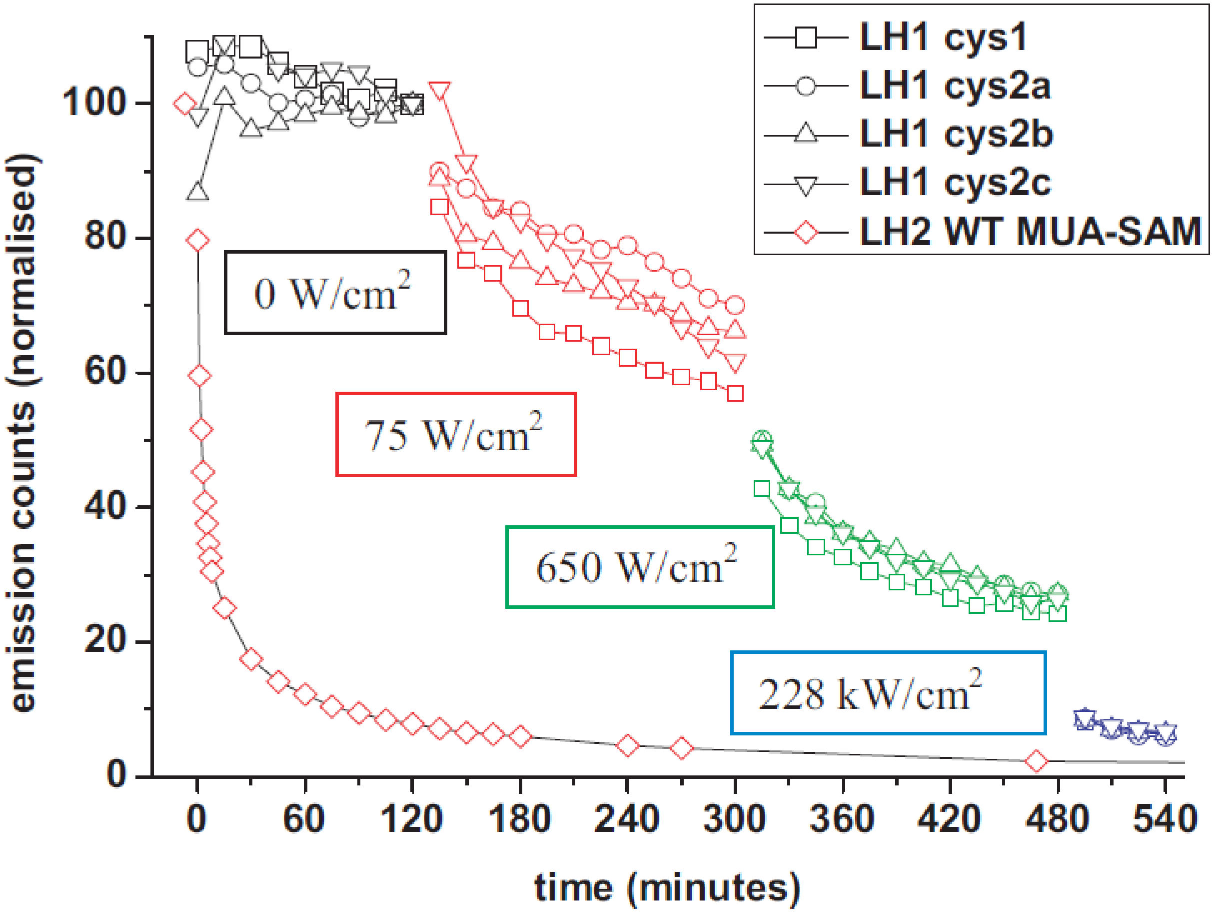


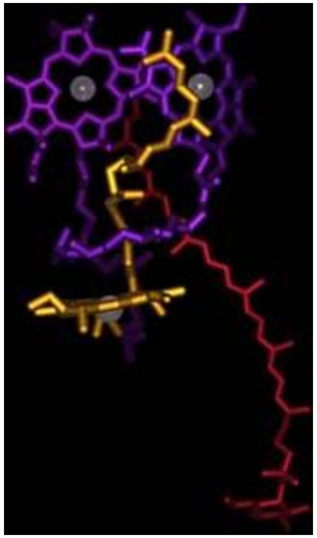
Magis et al. PCPB 2011

Stability towards oxygen

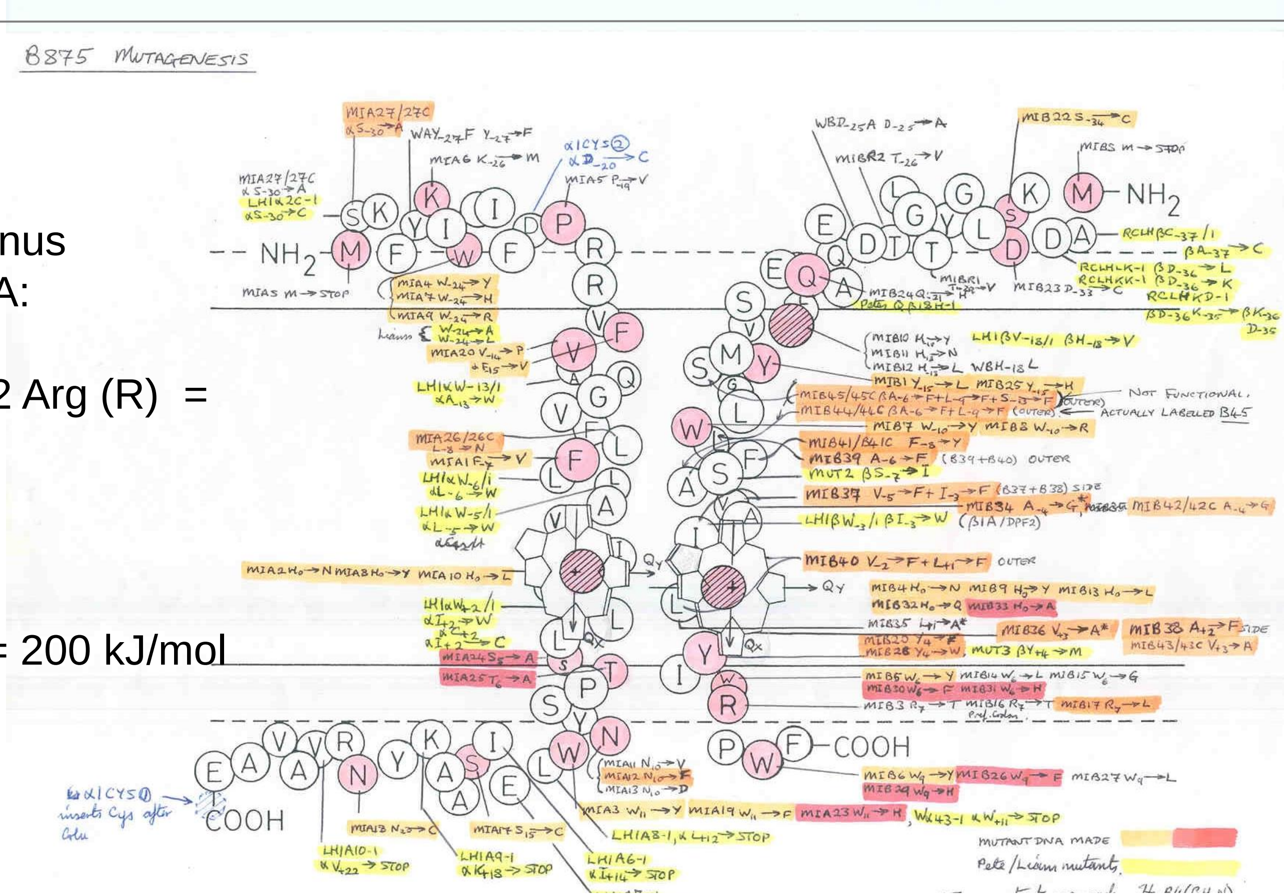
and illumination power







Proteins can stick to gold



LH1 N terminus
Accesible AA:

3 Lys (K) + 2 Arg (R) =

~160 kJ/mol

Au-S bond = 200 kJ/mol

Table 1. Absolute Value of the Interaction Free Energy in kJ/mol of the Noncovalent Association of the Amino Acids with a Gold (111) Surface

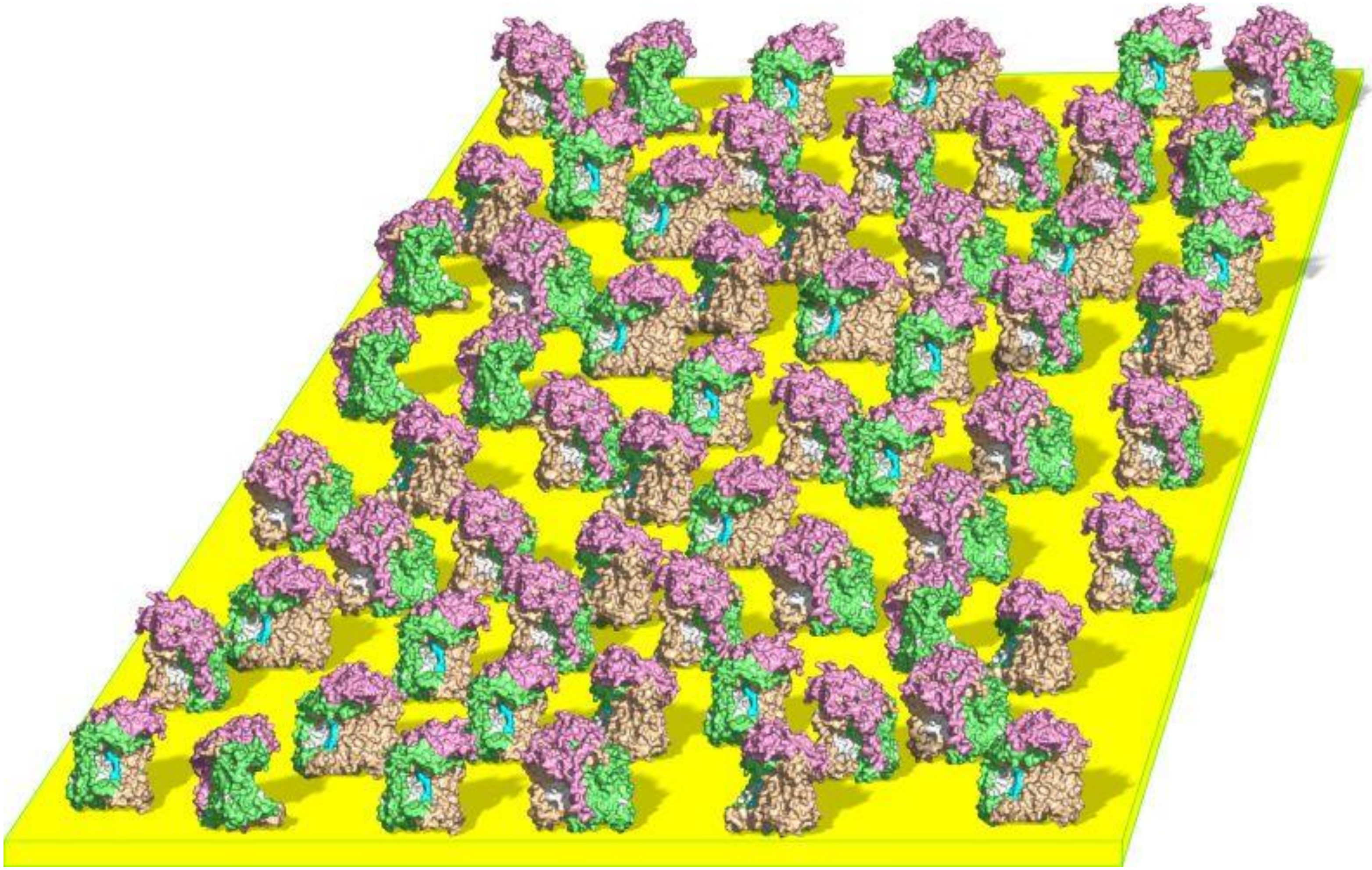
Ala	Arg	Asn	Asp	Cys	Gln	Glu	Gly	His	Ile	Leu	Lys	Met	Phe	Pro	Ser	Thr	Trp	Tyr	Val
21.9	36.3	26.1	25.5	37.7	28.6	17.5	23.6	34.0	25.1	25.4	30.0	39.3	43.6	26.0	23.1	28.9	40.2	44.2	24.8

Interaction of Amino Acids with the Au(111) Surface: Adsorption Free Energies from Molecular Dynamics Simulations

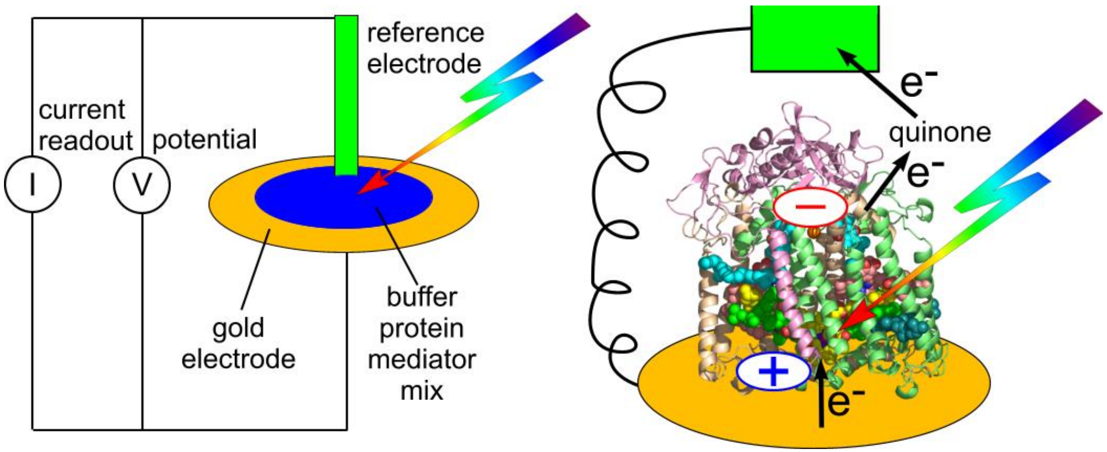
Langmuir **2010**, *26(11)*, 8347–8351

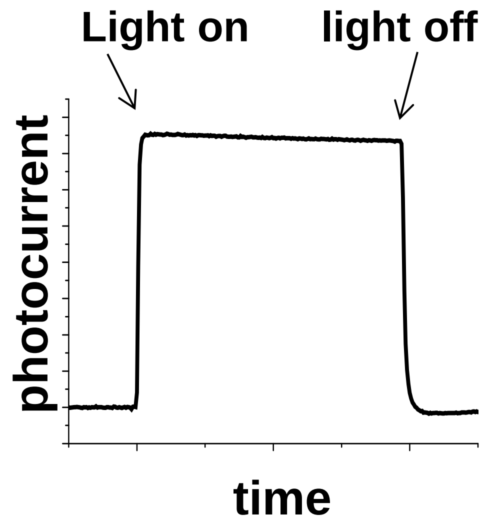
Martin Hoefling,[†] Francesco Iori,[‡] Stefano Corni,[‡] and Kay-Eberhard Gottschalk*[,†,§]

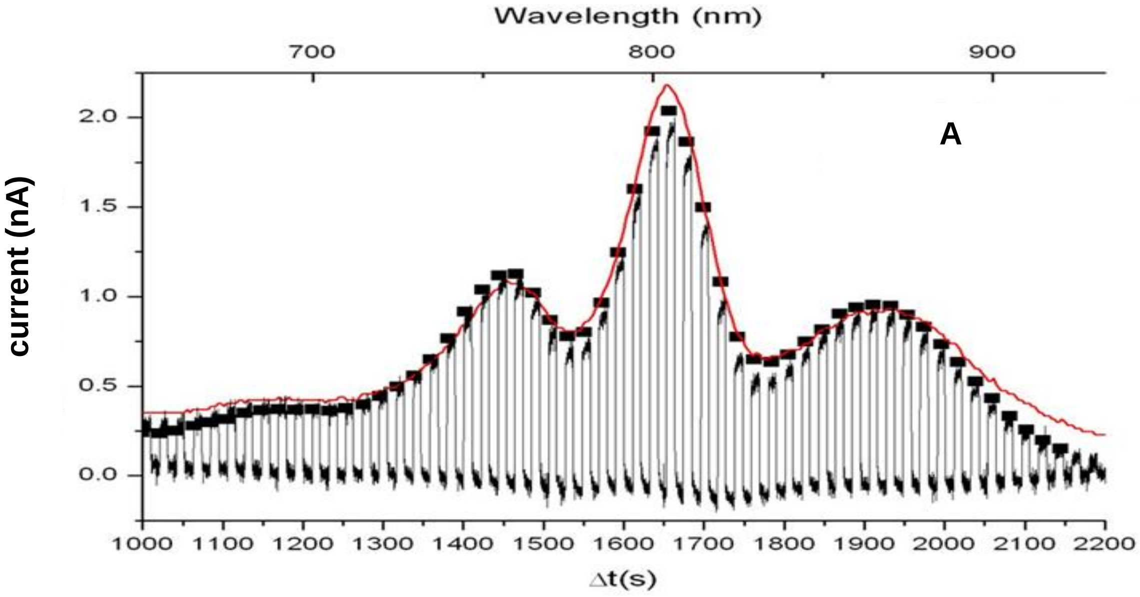
Reaction Centers directly adhered to gold

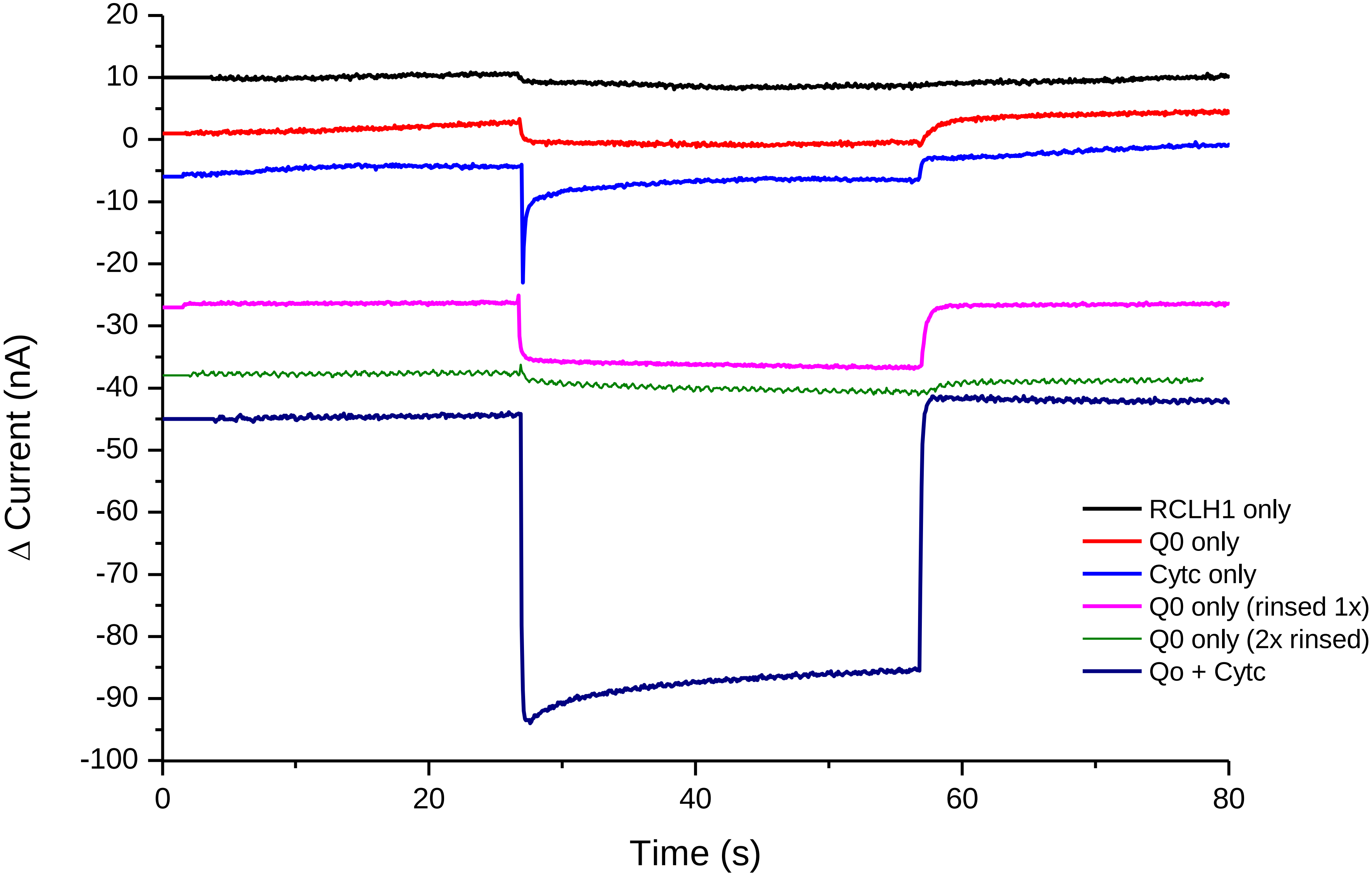


Photoelectrochemistry



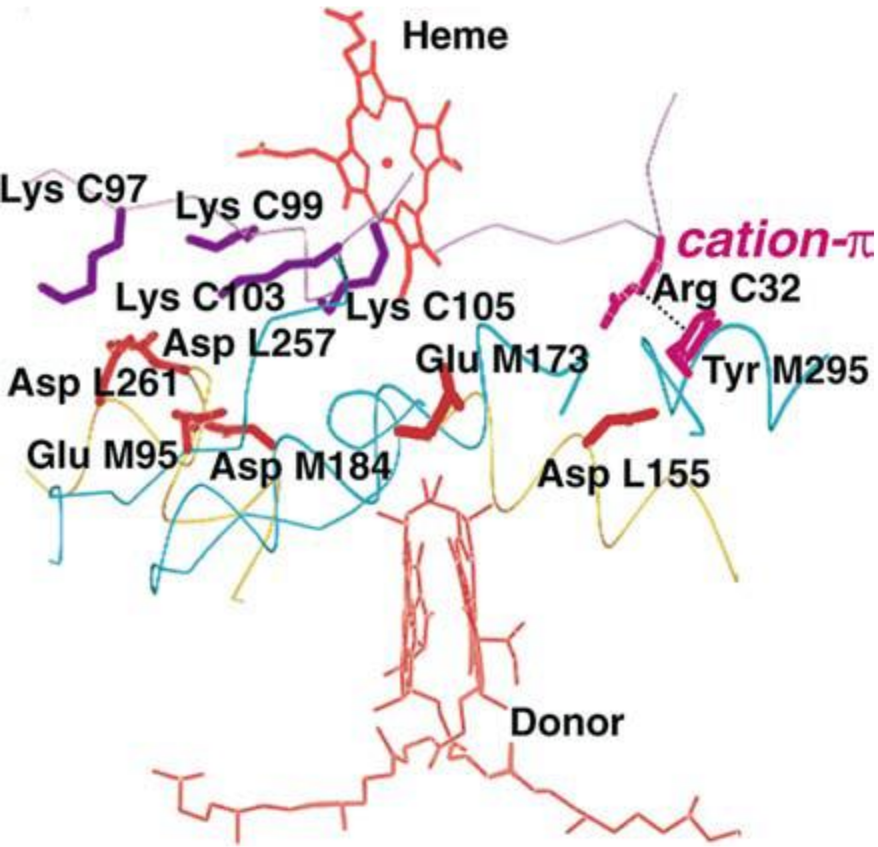




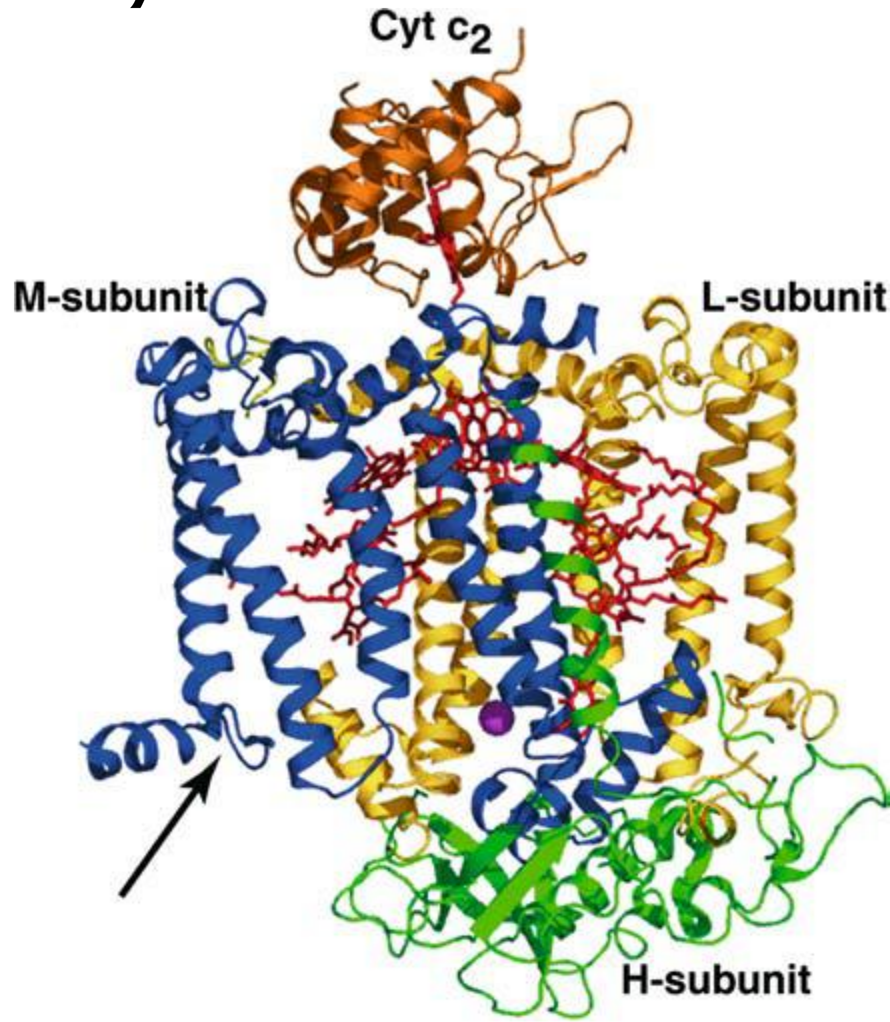


20
10
0
-10
-20
-30
-40
-50
-60
-70
-80
-90
-100
Δ Current (nA)
0
20
40
60
80
Time (s)
RCLH1 only
Q0 only
Cytc only
Q0 only (rinsed 1x)
Q0 only (2x rinsed)
Qo + Cytc

(sphaeroides) RC: cytc Interface



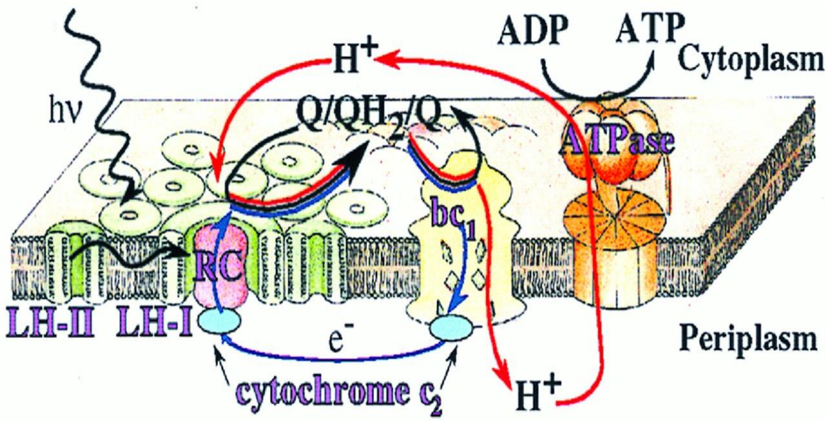
Charged residues at the interface of the cyt c2:RC complex. Acidic residues on the RC surface interact with basic residues on cyt c2. Most of the interactions are on the left (M subunit side) of the RC. In addition, a cation-p interaction is shown. Modified from Axelrod et al. 2002

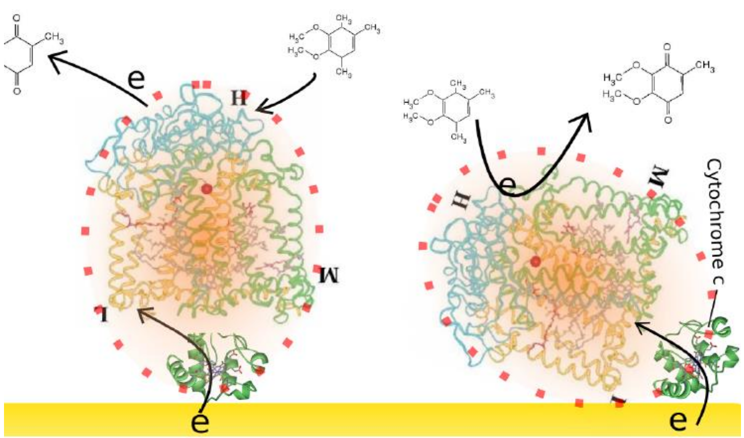


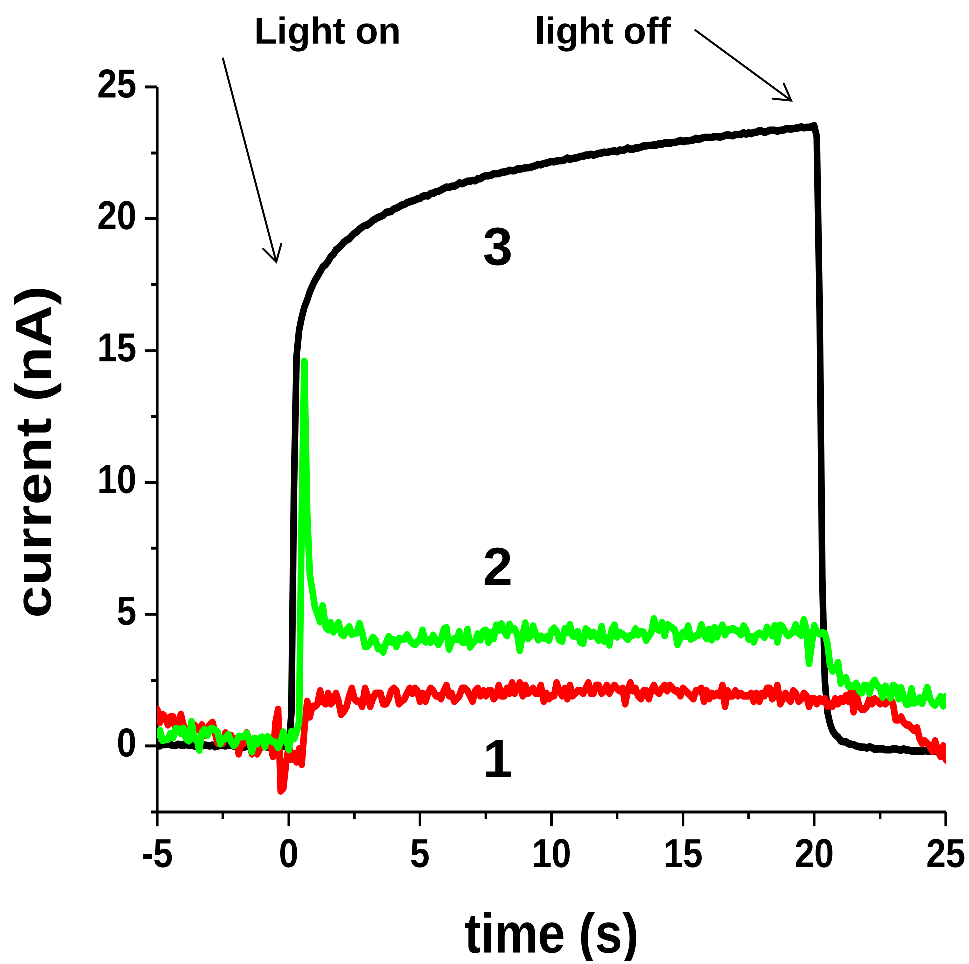
Structure of the cyt c2:RC complex from Rb. sphaeroides showing the position of the bound cyt c2 (orange) and the heme prosthetic group (red), the RC L-subunit (yellow), the RC Msubunit (blue), and theRCHsubunit (green). The location of a conformational change in the co-crystal at the N-terminal end of the M subunit is indicated by an arrow.

Modified from Axelrod et al. 2002.

Molecular wiring RCs and cytochrome-c



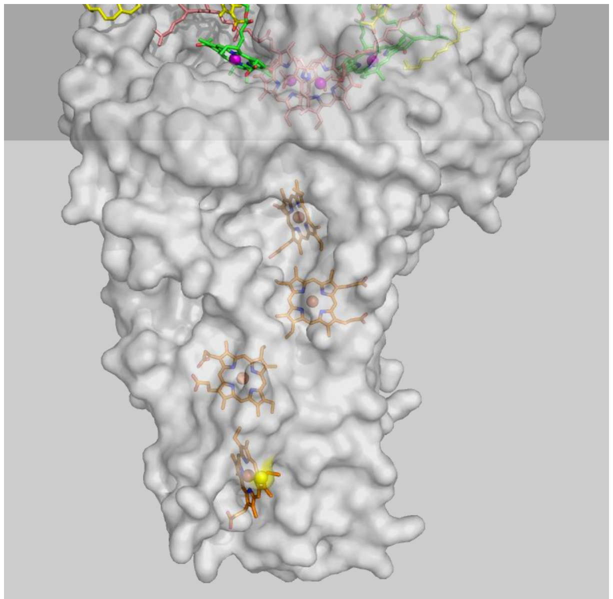


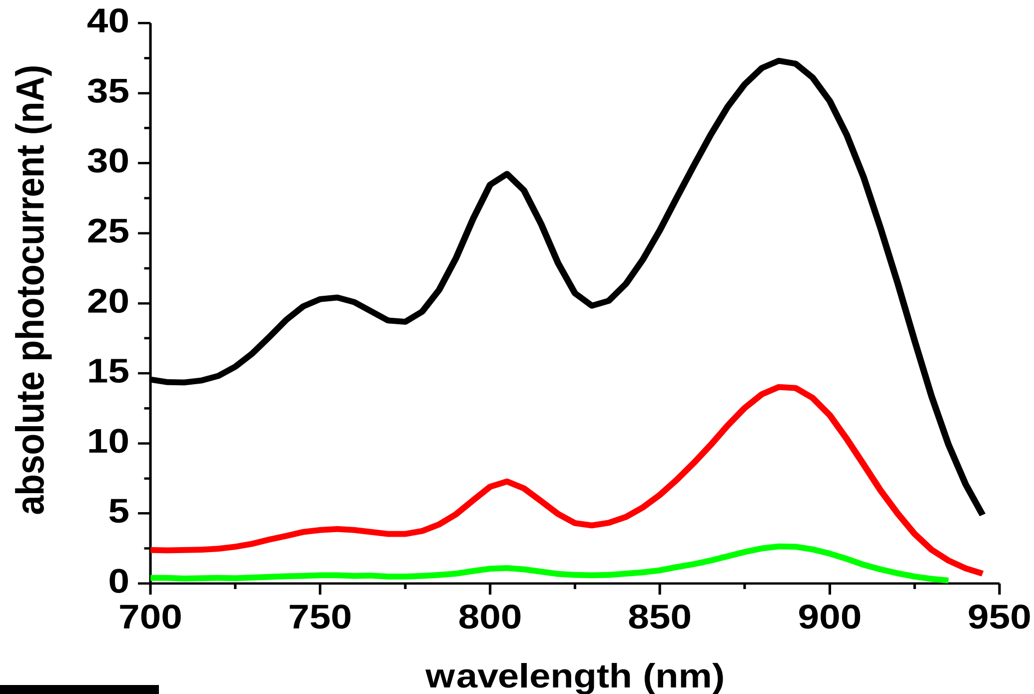


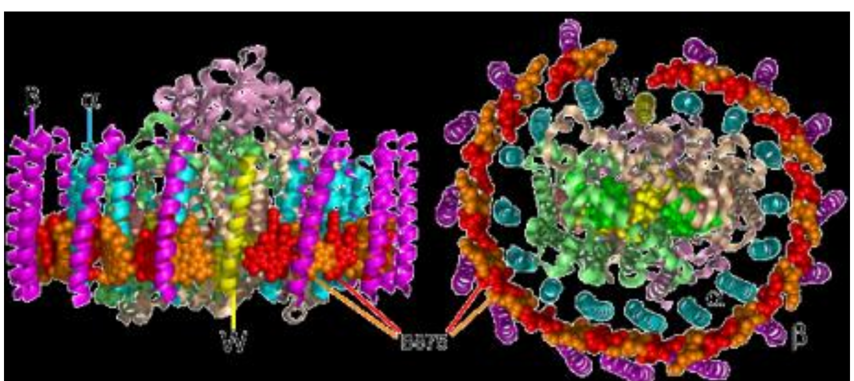
Method: Trammell and Lebedev

1. Quinone only, current x10
2. Replace buffer, cyt-c2 only, current x10
3. Replace buffer, quinone only, current not scaled

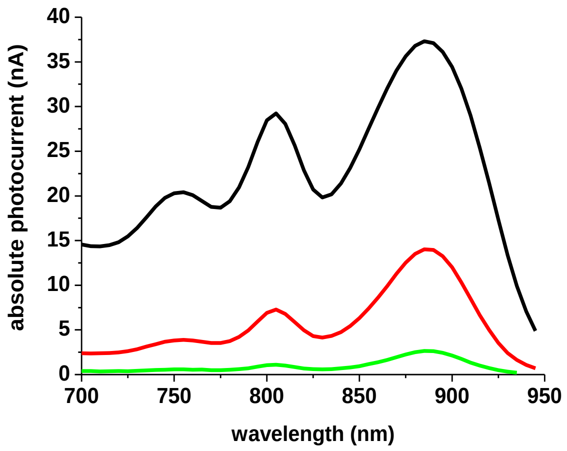
RCLH1 complexes directly adhered onto gold

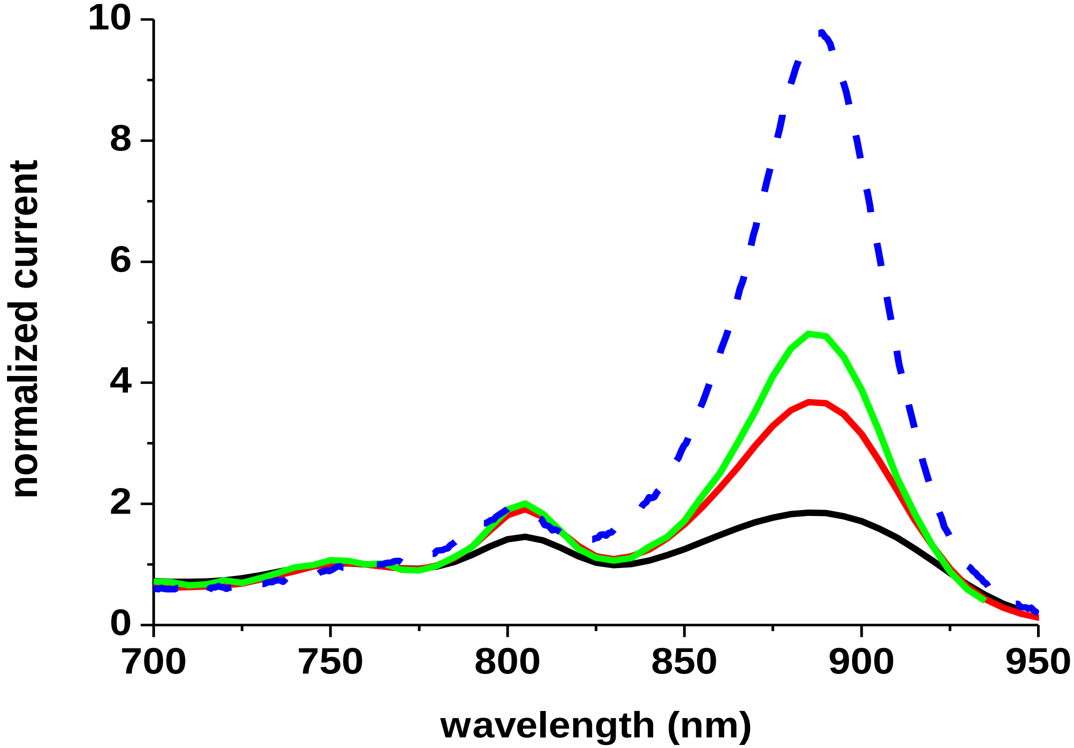




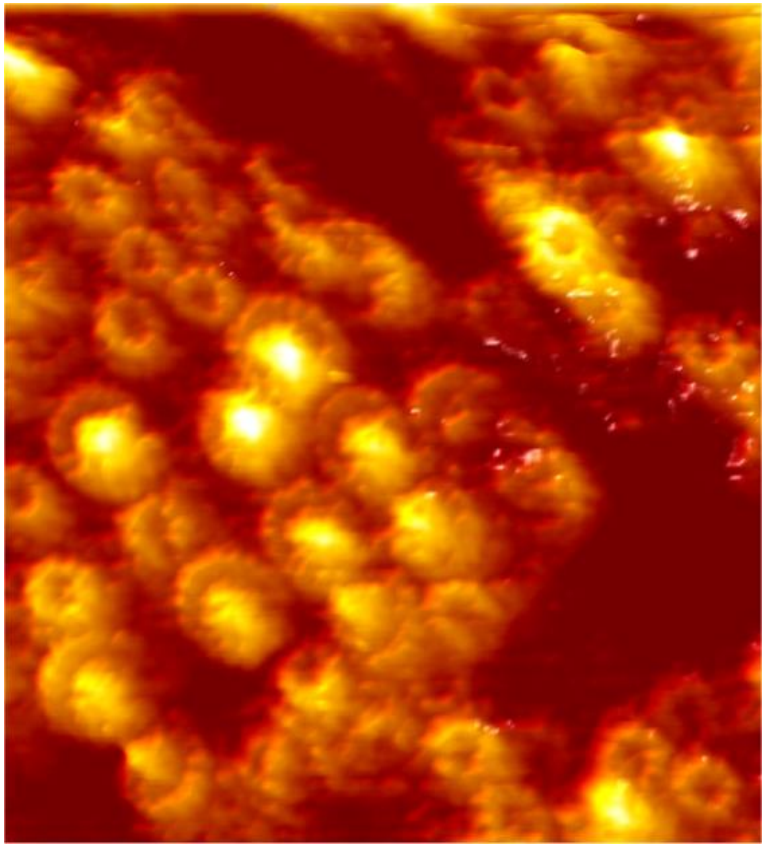


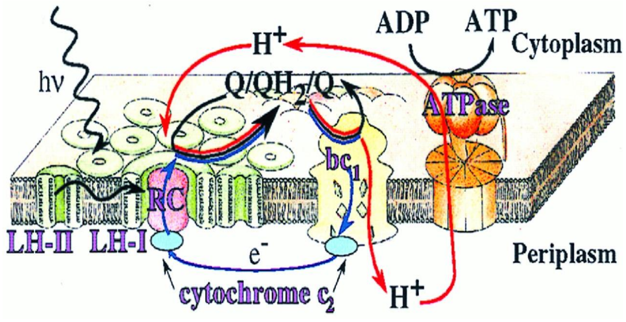
LH1 does not contribute to photocurrent





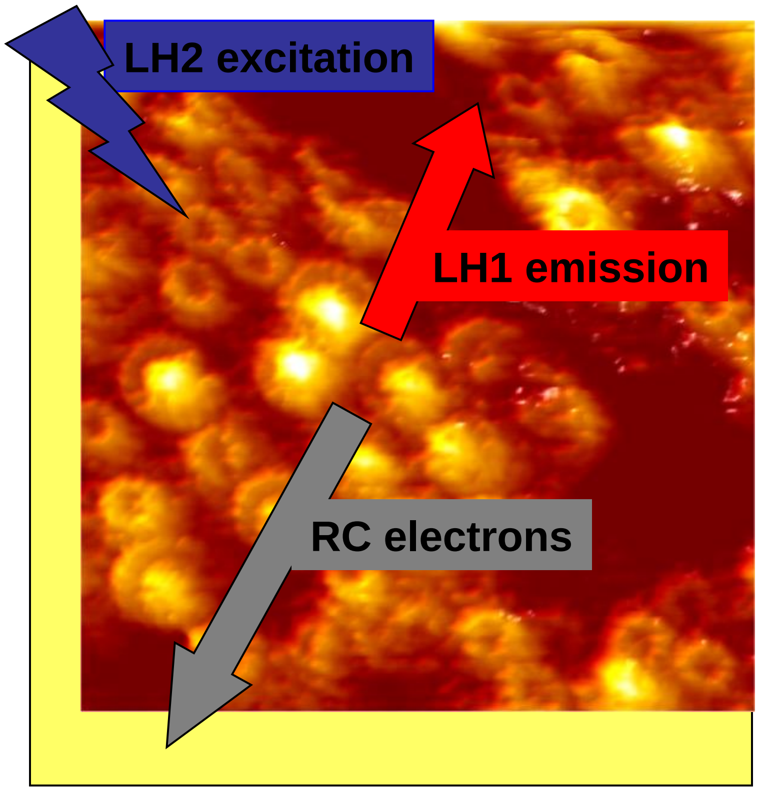
Whole membranes on gold electrodes



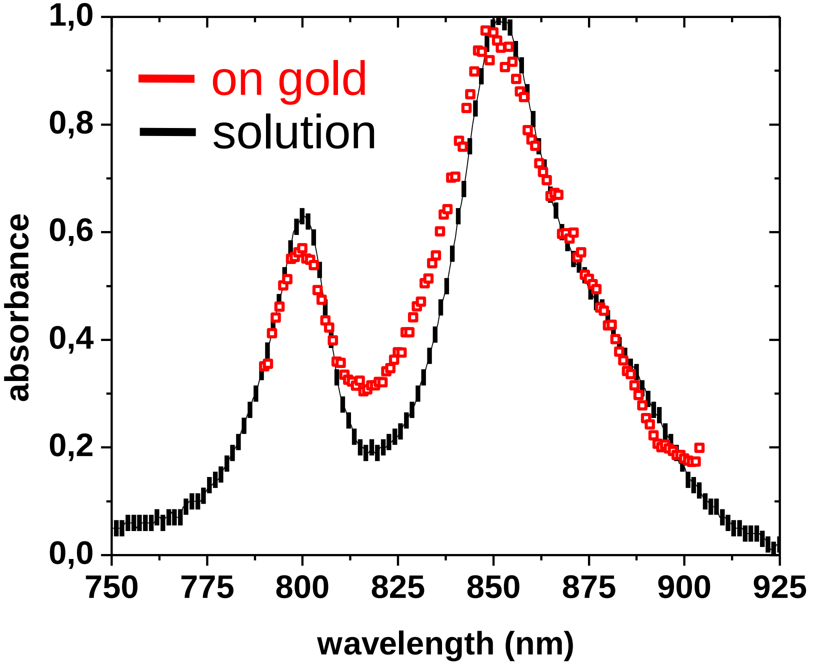


Let biology synthesise desired assemblies and apply these directly

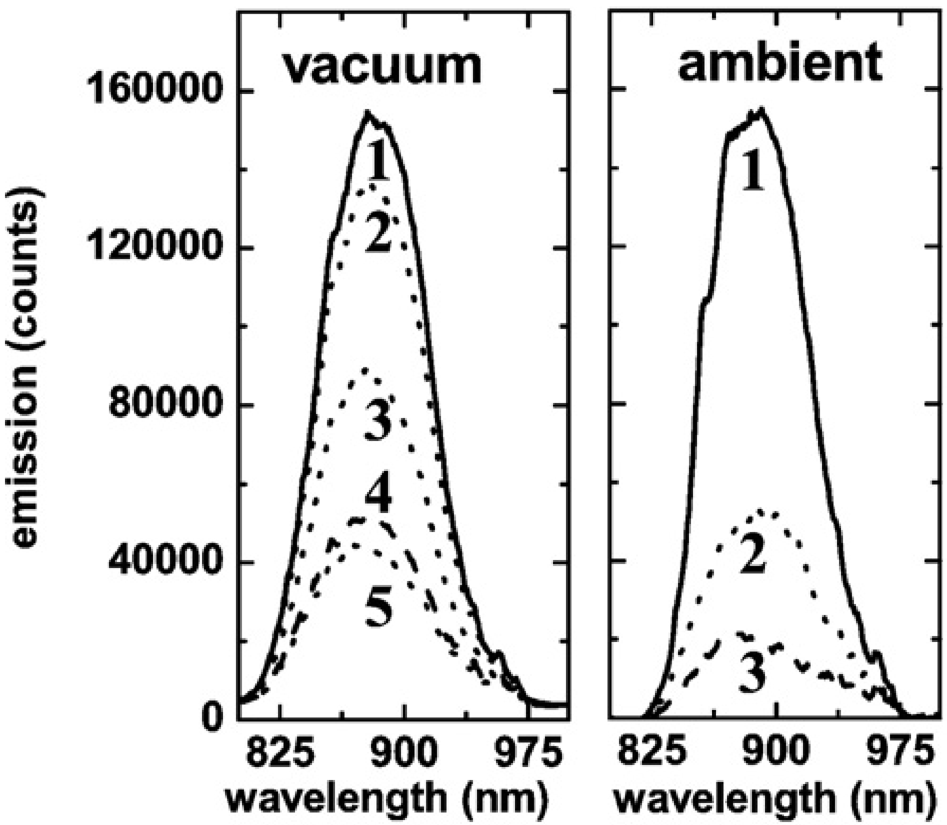
Whole membranes on gold electrodes



Fluorescence excitation follows absorbance

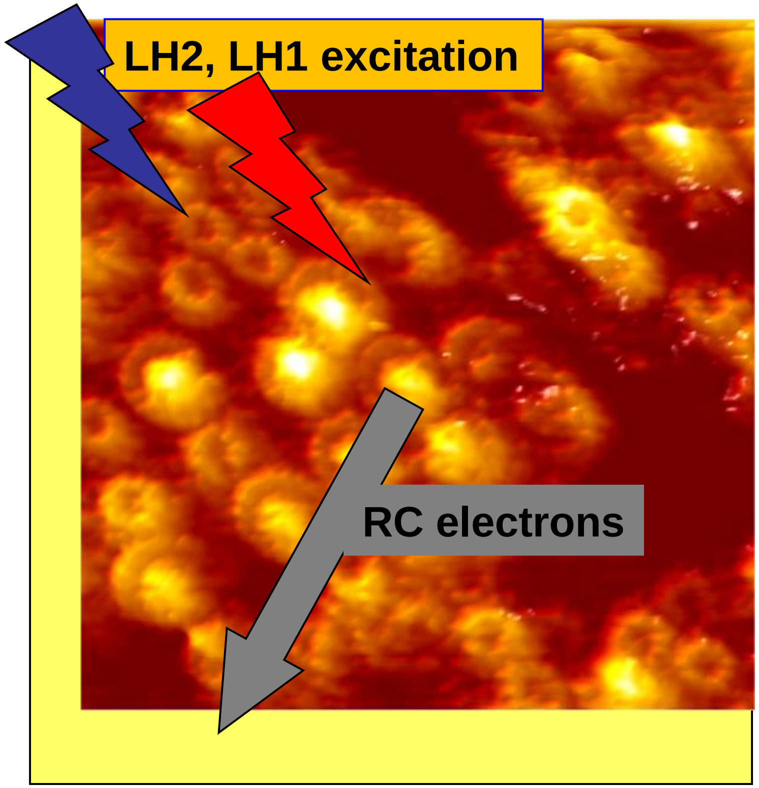


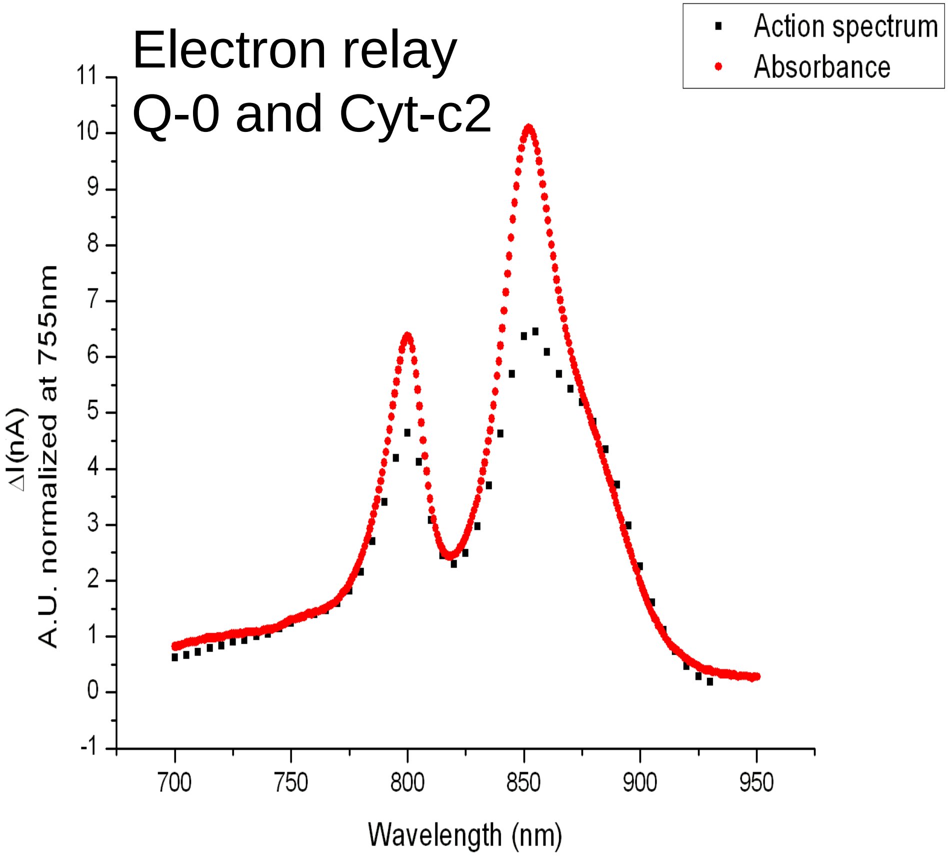
Stability of energy transferring system



Illumination time dependency of fluorescence,
30 minute intervals, varying (high) powers

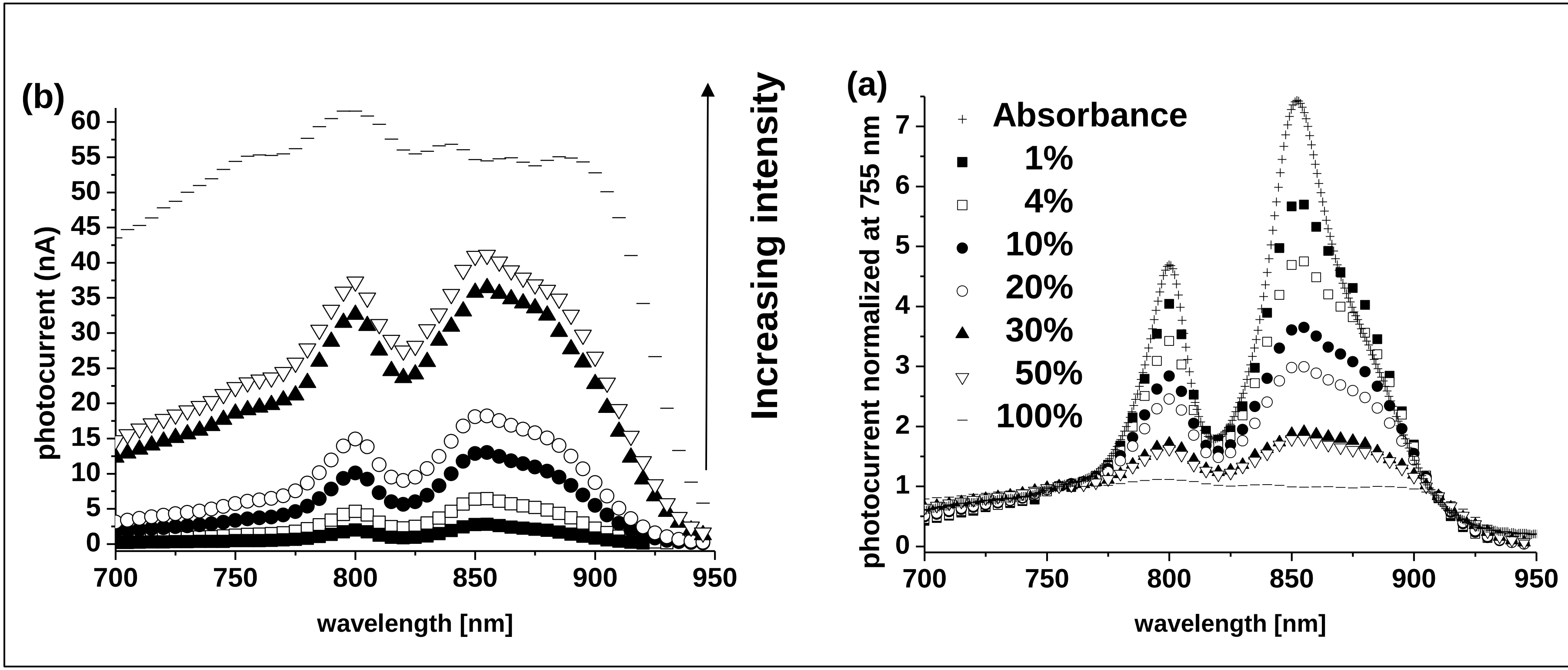
Photoelectrochemistry on membranes





Light induced current action spectrum follows solution absorbance spectrum

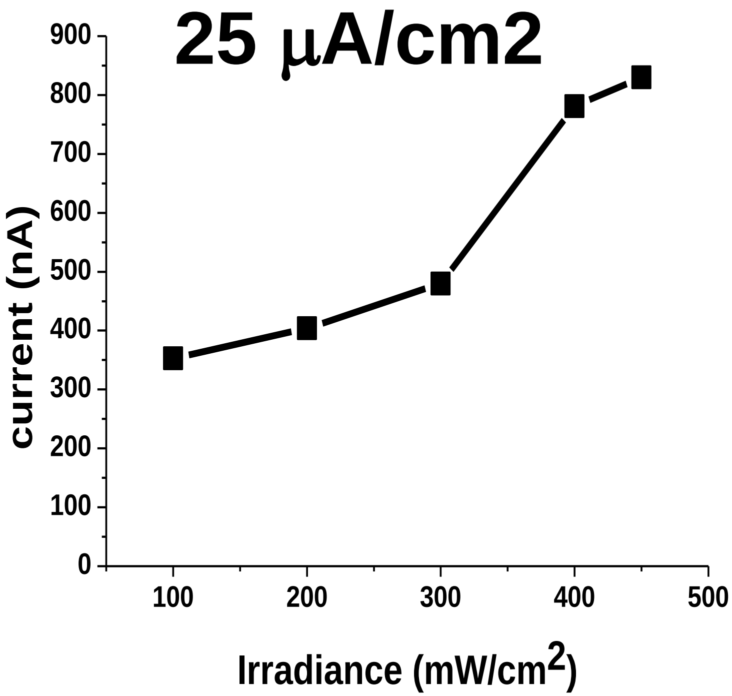
Light intensity variation

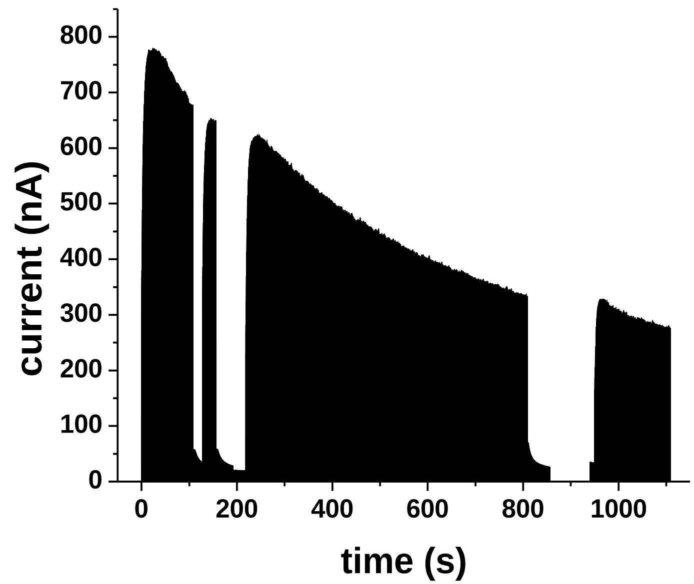


Higher intensity of light: strong reduction LH2 to photocurrent

Maximum current: 10 microA/cm2

No limit?

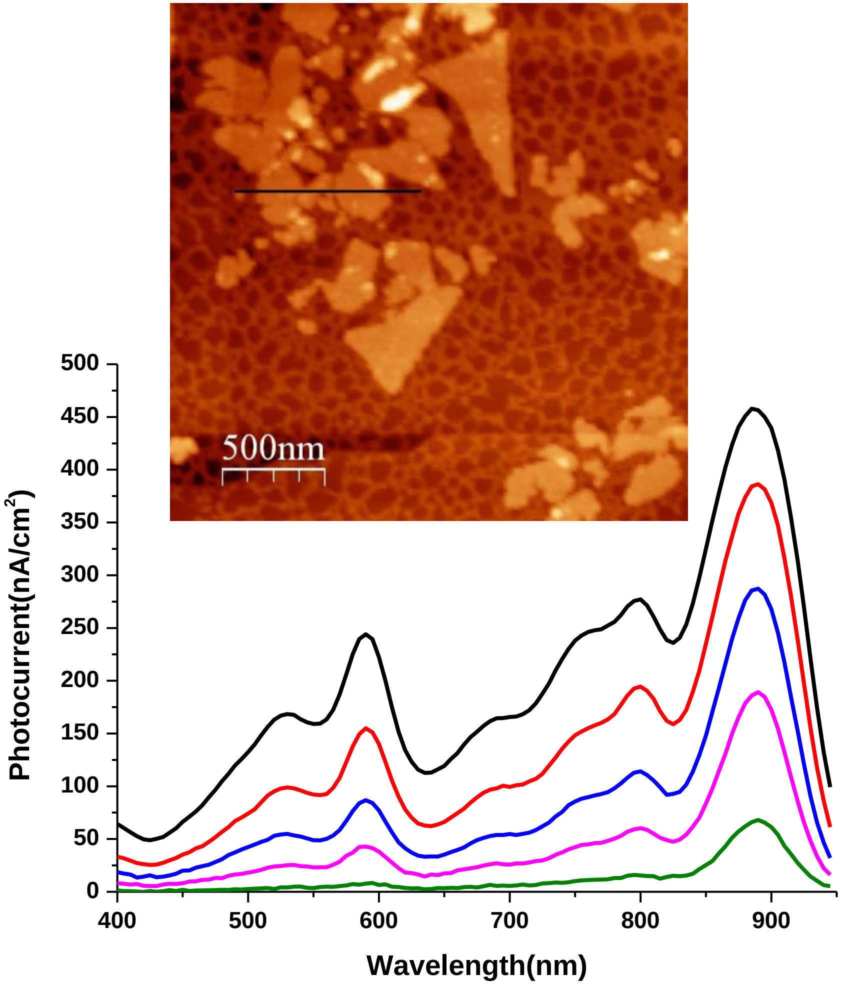




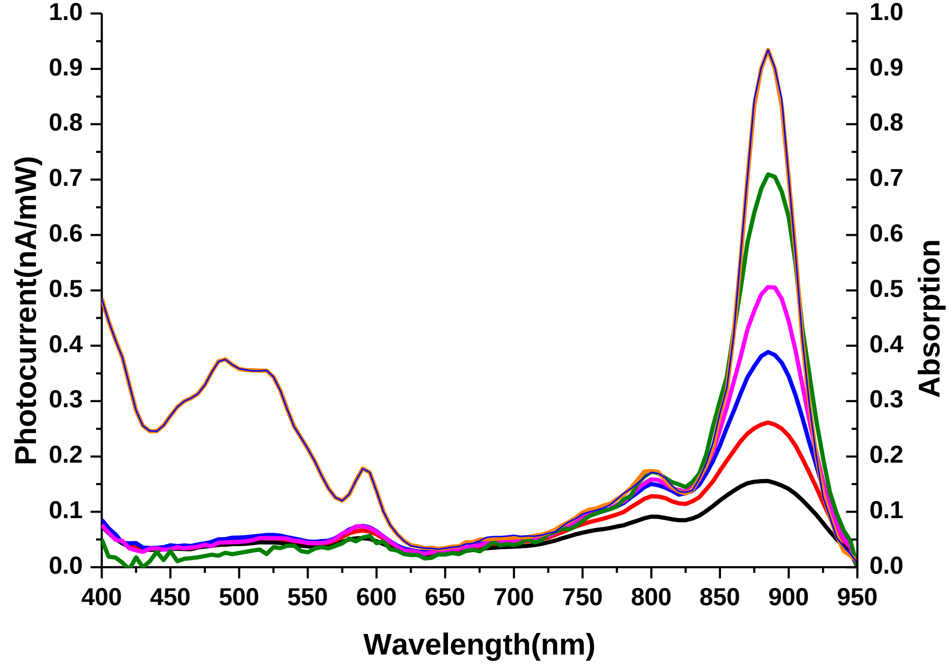
- Three days: minimally10 million cycles per RC
- Total charge: ~10 mC
- Ambient conditions

Magis et al.
BBA 2010)

High packed assemblies: 2D crystals of RC-LH1



Action spectra of 2D crystals of RC-LH1 complex deposited on gold electrode as function of light intensity.

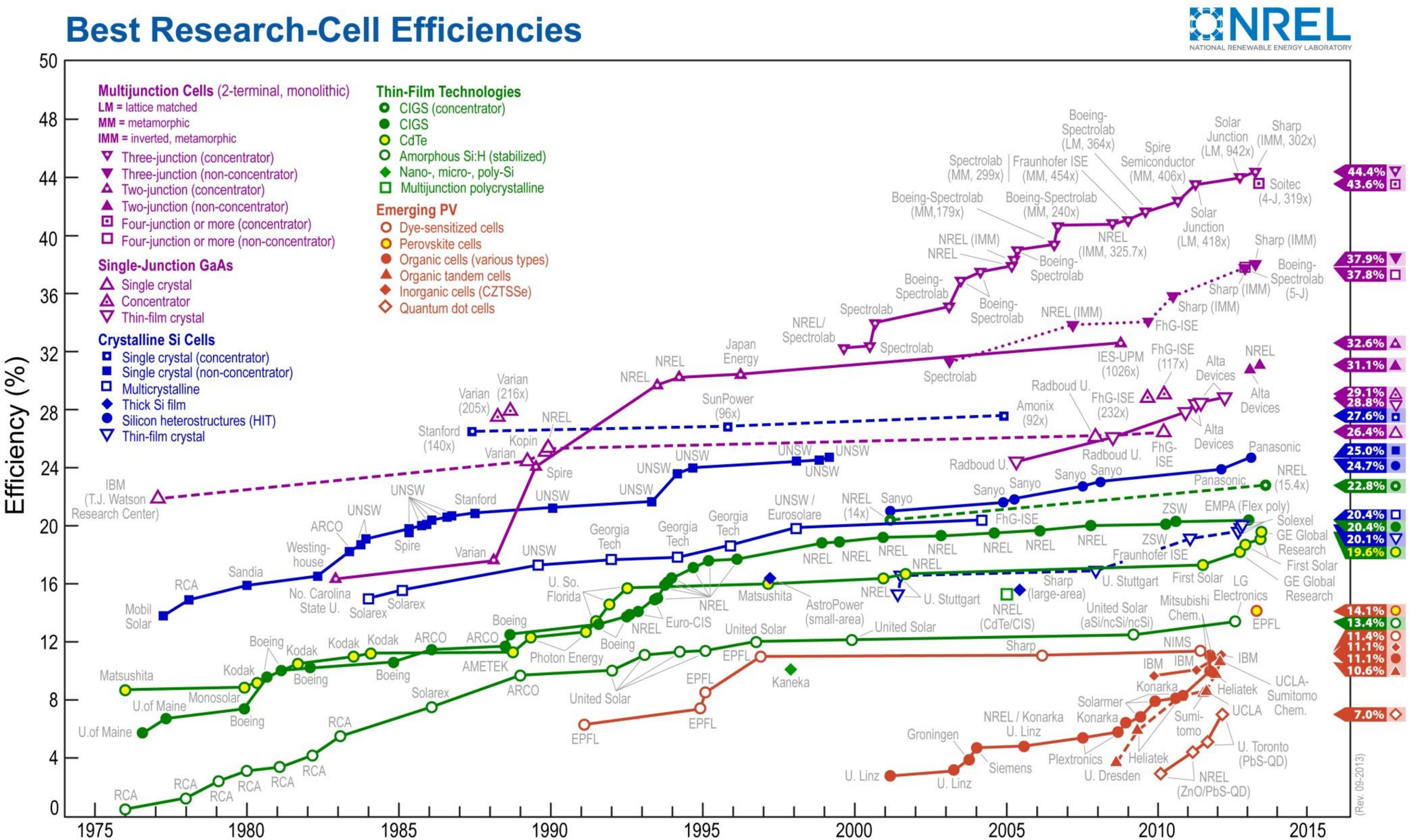


Normalized action spectra of 2d crystals of RC-LH1 adsorbed on gold electrode, absorption spectra of 2d crystals of RC-LH1 and absorption spectrum of isolated RC-LH1.

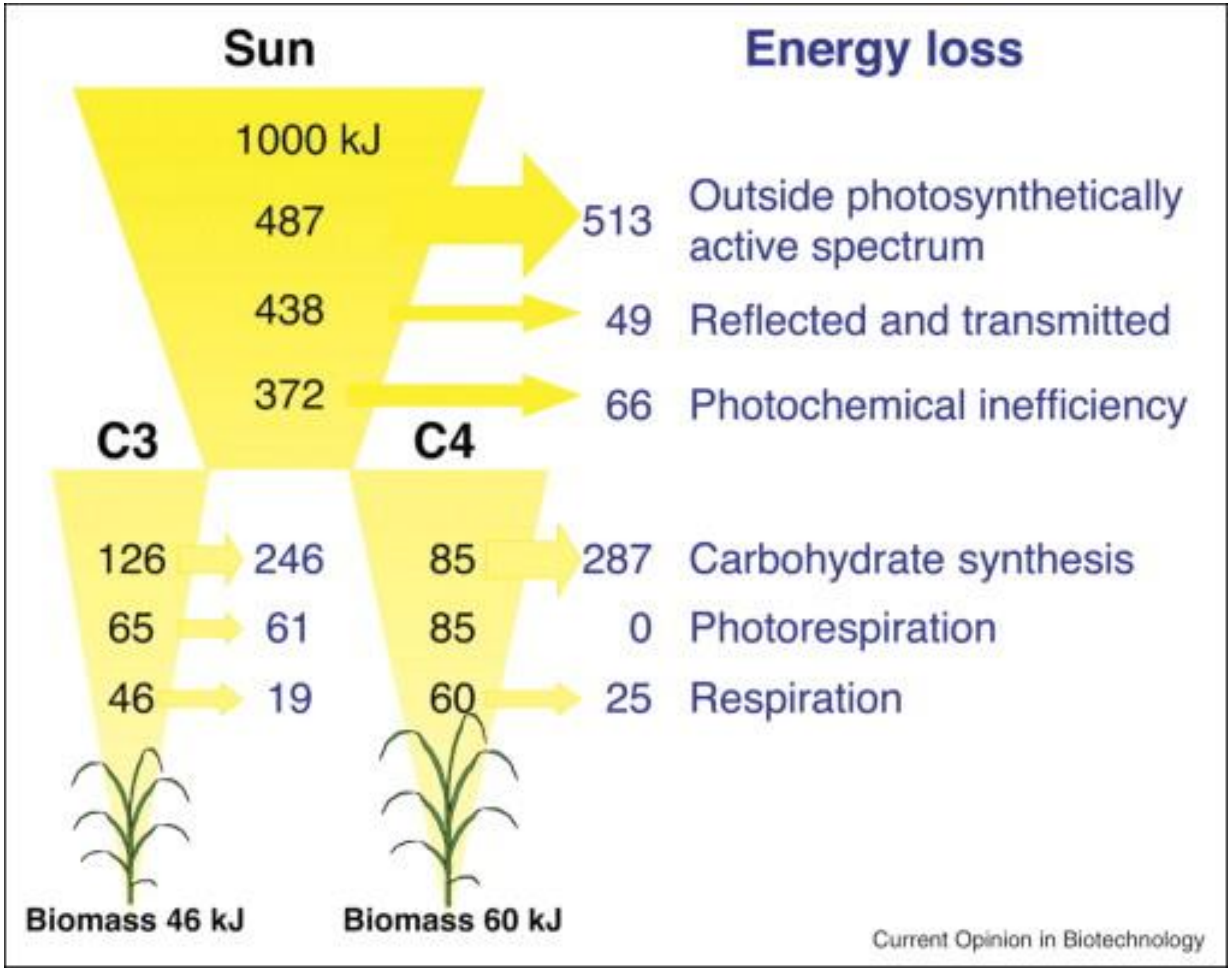
Proteins can function on bare gold

- Enhances electron tunneling
- Should have been a control many years ago
- Finally out of the nano ampere range
- LH systems influenced by gold quenching
- Enhanced energy transfer by plasmons
- RC systems too?
- Membranes also function, let nature do the work?

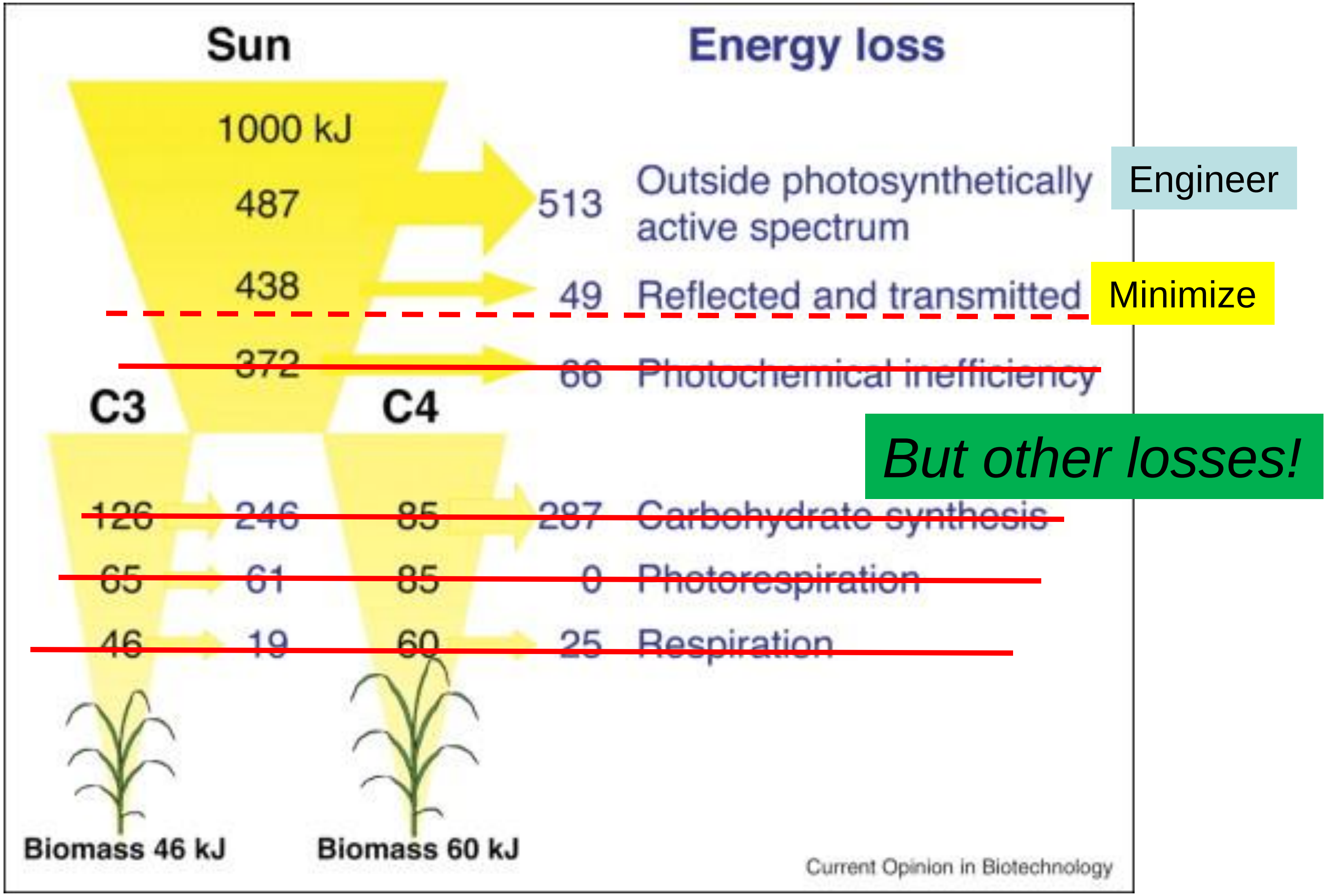
Biohybrid solar cells



efficiencies



biohybrid



Sun
Energy loss
1000 kJ
487
513
Outside photosynthetically active spectrum
Engineer
438
49
Reflected and transmitted
Minimize
372
66
Photochemical inefficiency
C3
C4
But other losses!
126
246
85
287
Carbohydrate synthesis
65
61
85
0
Photorespiration
46
19
60
25
Respiration
Biomass 46 kJ
Biomass 60 kJ
Current Opinion in Biotechnology

Biohybrid for photovoltaics? a stepping stone for biohybrid photocatalyses? For artifical photosynthesis?

Self-assembled photosystem-I biophotovoltaics on nanostructured TiO_2 and ZnO

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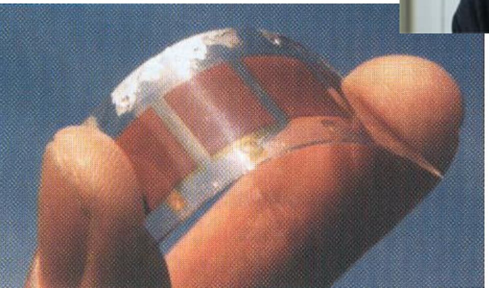
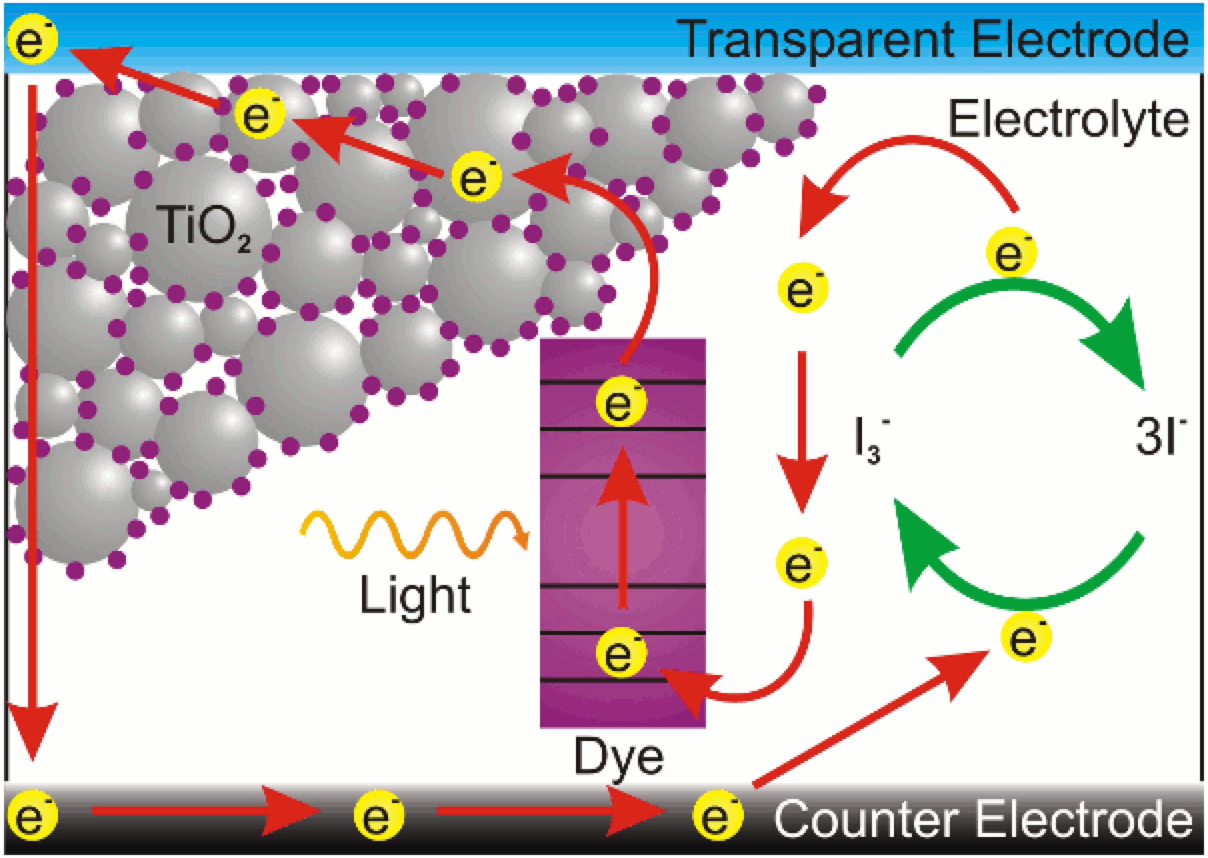
Daoyong Yu,[2]
Michael Vaughn,[3]
Md. K. Nazeeruddin,[4]
Barry D. Bruce,[3]
Michael Graetzel[4]
& Shuguang Zhang[2]





Dye sensitized solar cell or Graetzel Cell

Dye sensitized solar cell or Graetzel Cell

Same design as electrochemical photosynthesis based solar cell

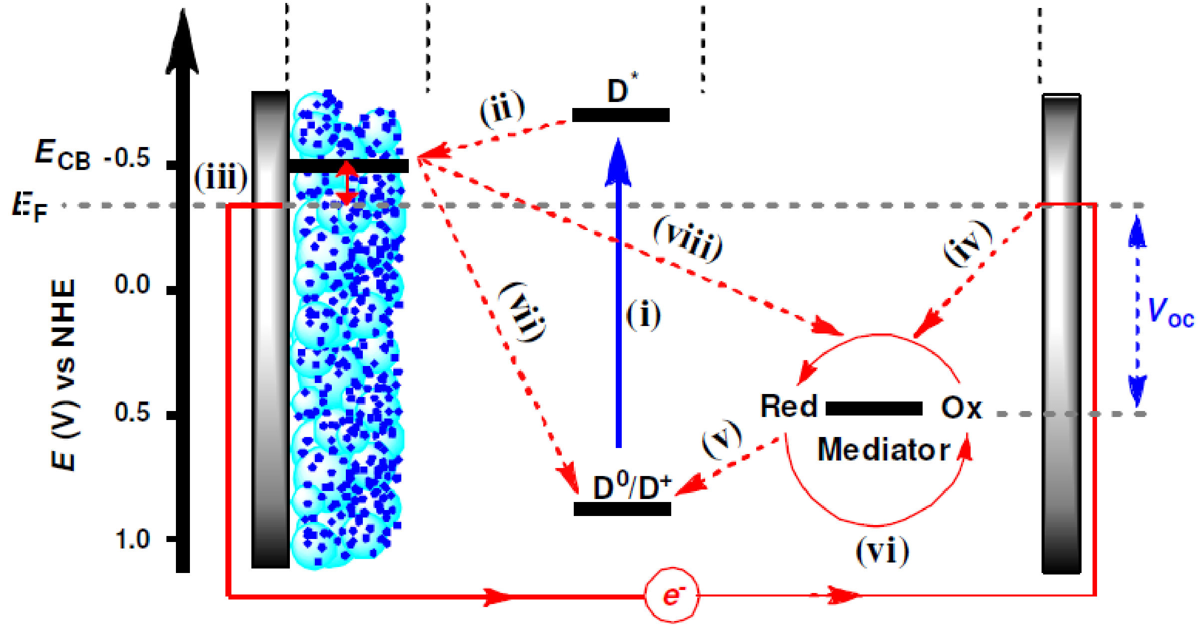


Figure 2. Energy level alignment of components in a conventional *n*-type DSSC device (kinetics shown in bold):

(i) dye electronic excitation (**fs**)

(ii) charge injection to semiconductor conduction band (**150 ps**; energy loss; limits I_{SC})

(iii) electron equilibration to semiconductor Fermi level (energy loss) and diffusion to FTO (100 **µs**)

(iv) redox mediator reduction at counter electrode (limits V_{OC})

(v) dye regeneration/mediator oxidation (**1 µs**; energy loss)

(vi) mass-transfer diffusion of redox mediator (limits I_{SC})

(vii) $TiO_2^{\bullet-} \rightarrow Dye^+$ charge recombination (**3 µs**; limits I_{SC} and V_{OC})

(viii) $TiO_2^{\bullet-} \rightarrow Mediator^+$ charge recombination (**1 ms**; limits I_{SC} and V_{OC})

But only two electrodes and measure photocurrent over variable resistance and varying potential

Quantum efficiency of photovoltaics

internal (*h_{PV}*int) and **external (*h_{PV}*ext)** quantum efficiency:

photovoltaic **material** photovoltaic **device**

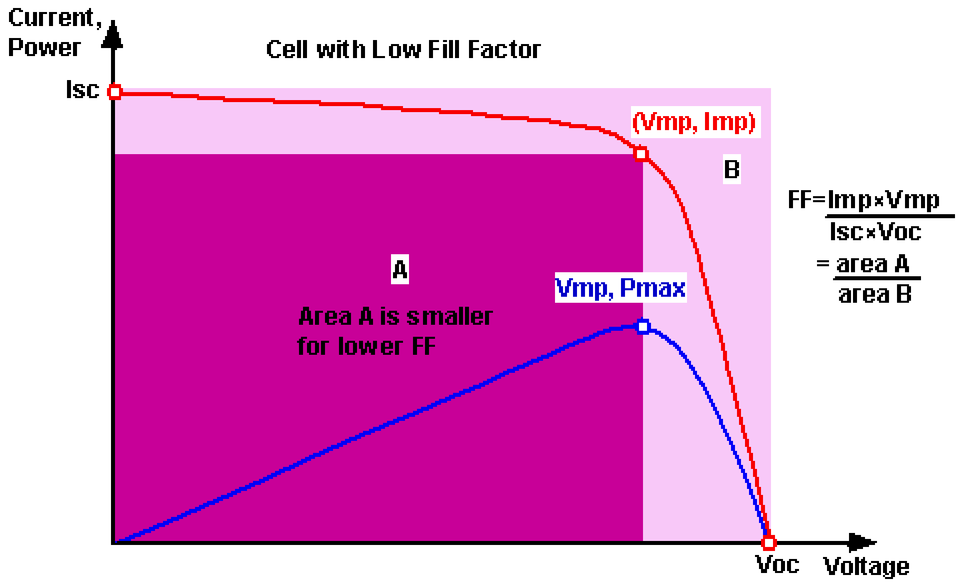
DSSC:
incident photon to current efficiency (**IPCE**),
absorbed photon to current efficiency (**APCE**),
light harvesting efficiency (**LHE**)

Internal quantum efficiency (*h_{PV}***int**) is
a # of electrons penetrating in an external circuit / # of **absorbed** photons.

product of exciton diffusion efficiency (*h_{ED}*),
charge transfer efficiency (*h_{CT}*), and
charge collection efficiency (*h_{CC}*).

Exciton diffusion efficiency (*h_{ED}*) is a fraction of photogenerated excitons that reaches p-n-junction: problem is taken care of by nature!

The "fill factor", "FF"

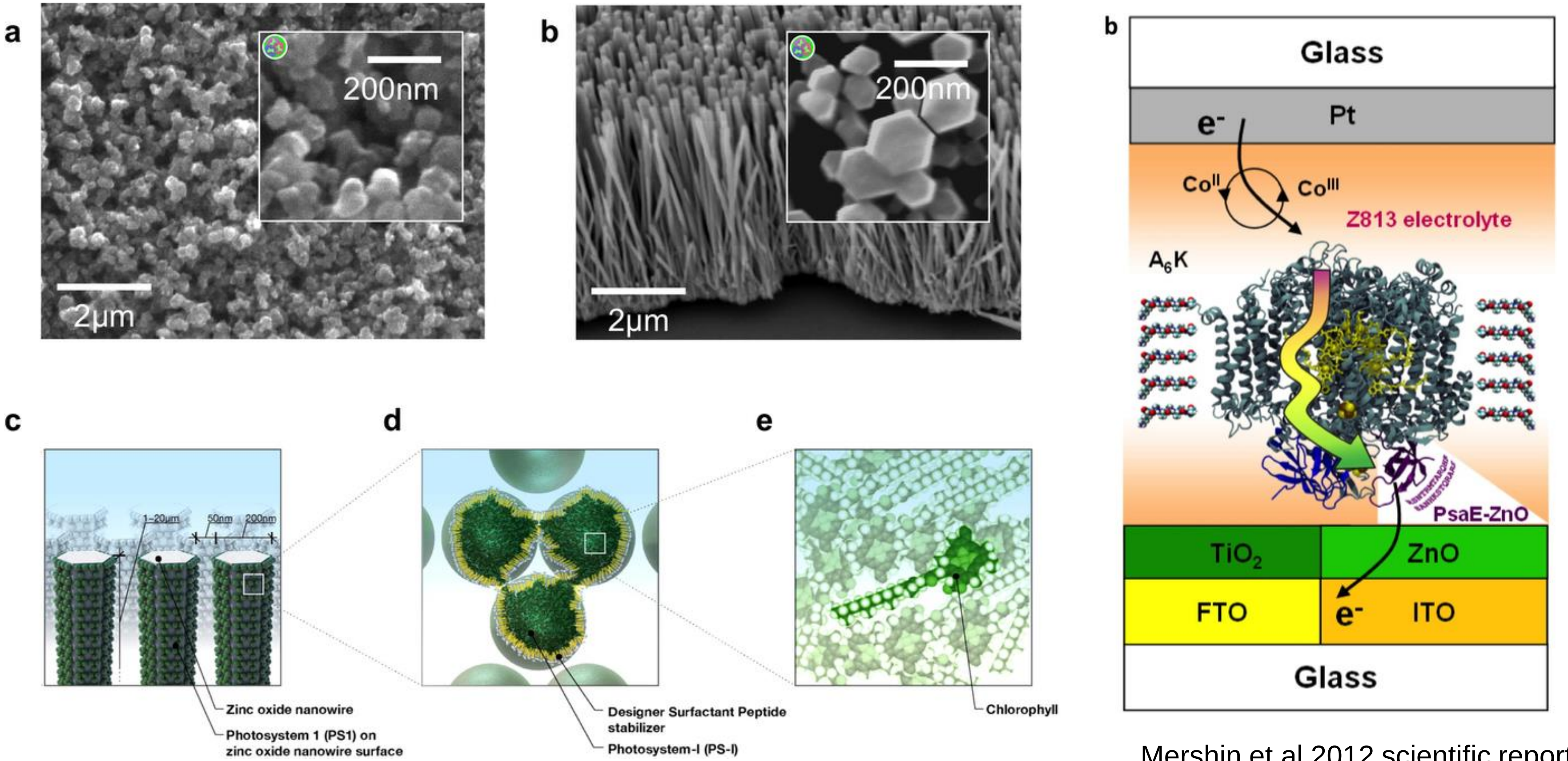


The short-circuit current and the open-circuit voltage are the maximum current and voltage respectively from a solar cell.

The "fill factor“ is a parameter which, in conjunction with V_{oc} and I_{sc}, determines the maximum power from a solar cell. The FF is defined as the ratio of the maximum power from the solar cell to the product of V_{oc} and I_{sc}. Graphically, the FF is a measure of the "squareness" of the solar cell and is also the area of the largest rectangle which will fit in the IV curve.

All about PV, go to: http://pveducation.org/

Self-assembled photosystem-I biophotovoltaics on nanostructured TiO_2 and ZnO; Mershin et al. scientific reports 2012



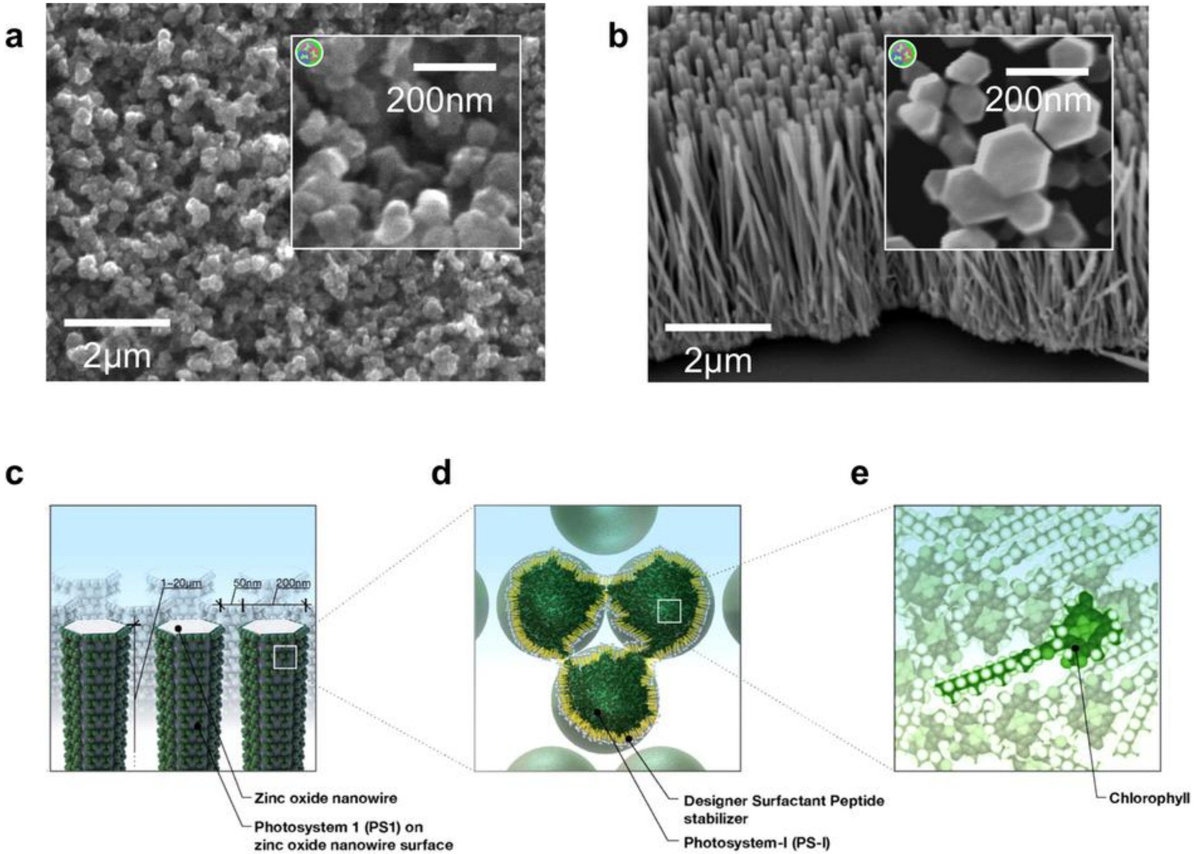
Mershin et al 2012 scientific reports

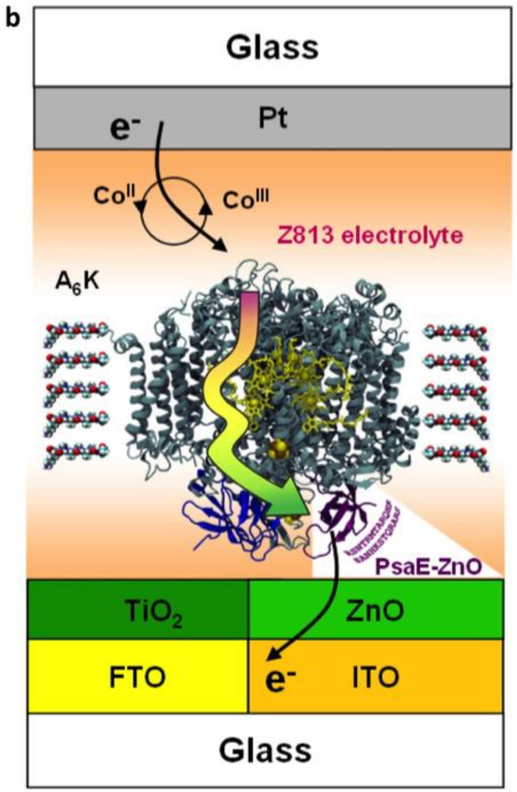
yielding open circuit photovoltage of 0.5 V, fill factor of 71%, electrical power density of 81 µW/cm^2 and photocurrent density of 362 µA/cm^2

1.3 µA/cm^2

But note: area given is for illumination area!
For area occupied by PS1 it is 200 times less!

Self-assembled photosystem-I biophotovoltaics on nanostructured TiO_2 and ZnO



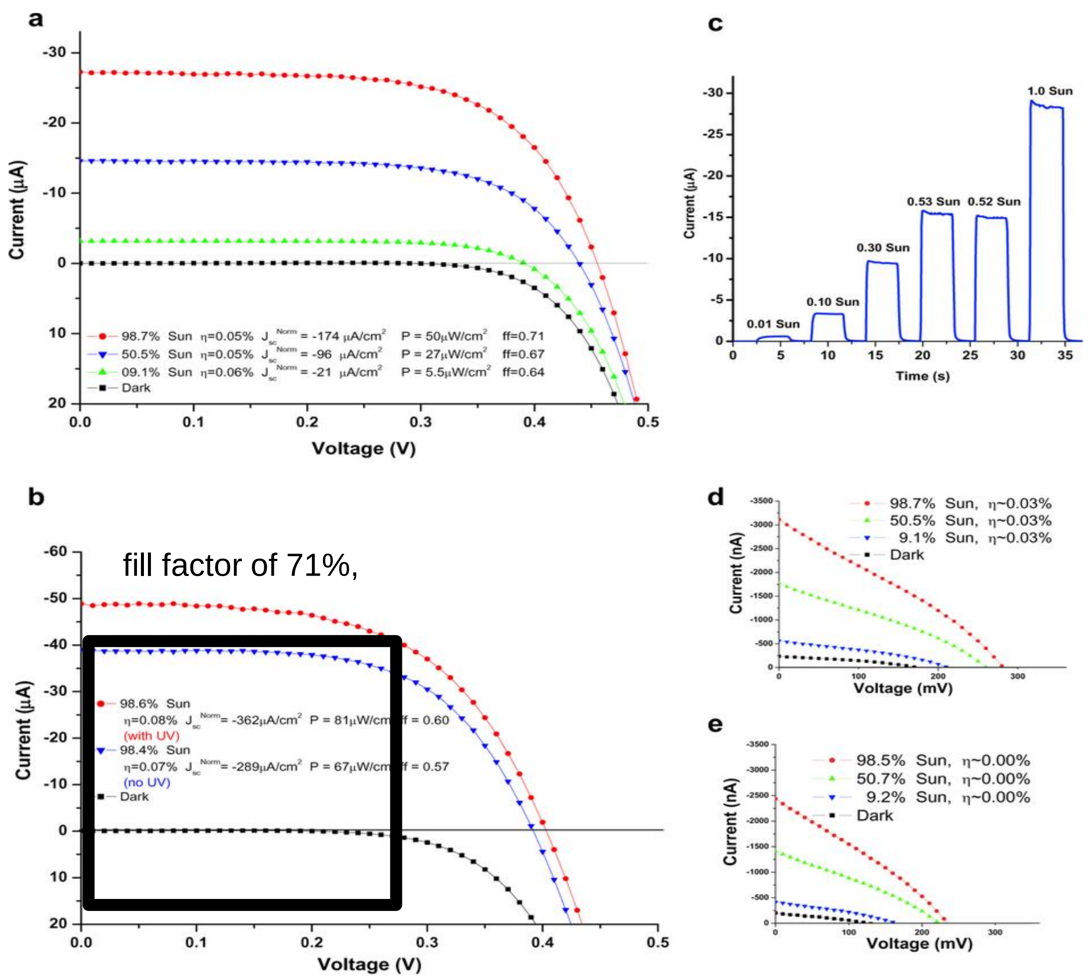


Mershin et al 2012 scientific reports

- ***Stability not reported, is this a photovoltaic biosensor?***

- ***Maybe an ideal photosynthaic biosensor is powered by sunlight?***

For a DSSC very poor behaviour



Mershin et al 2012 scientific reports

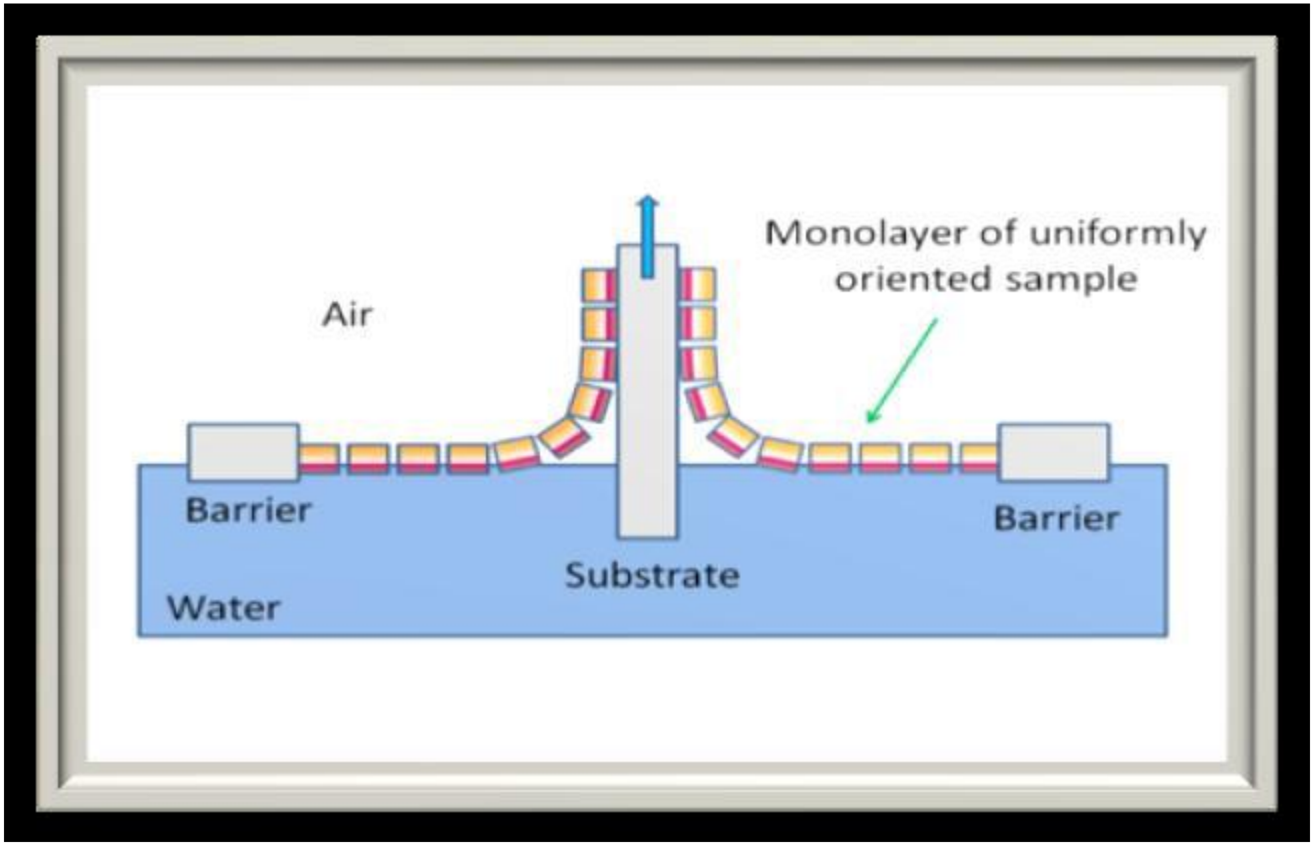
Applying DSSC does not show materials properties of PS1,

But much more materials properties of TiO2

The first and most basic question that needs to be answered is:

<u>Can we retain the quantum efficiency of photosynthesis?</u>

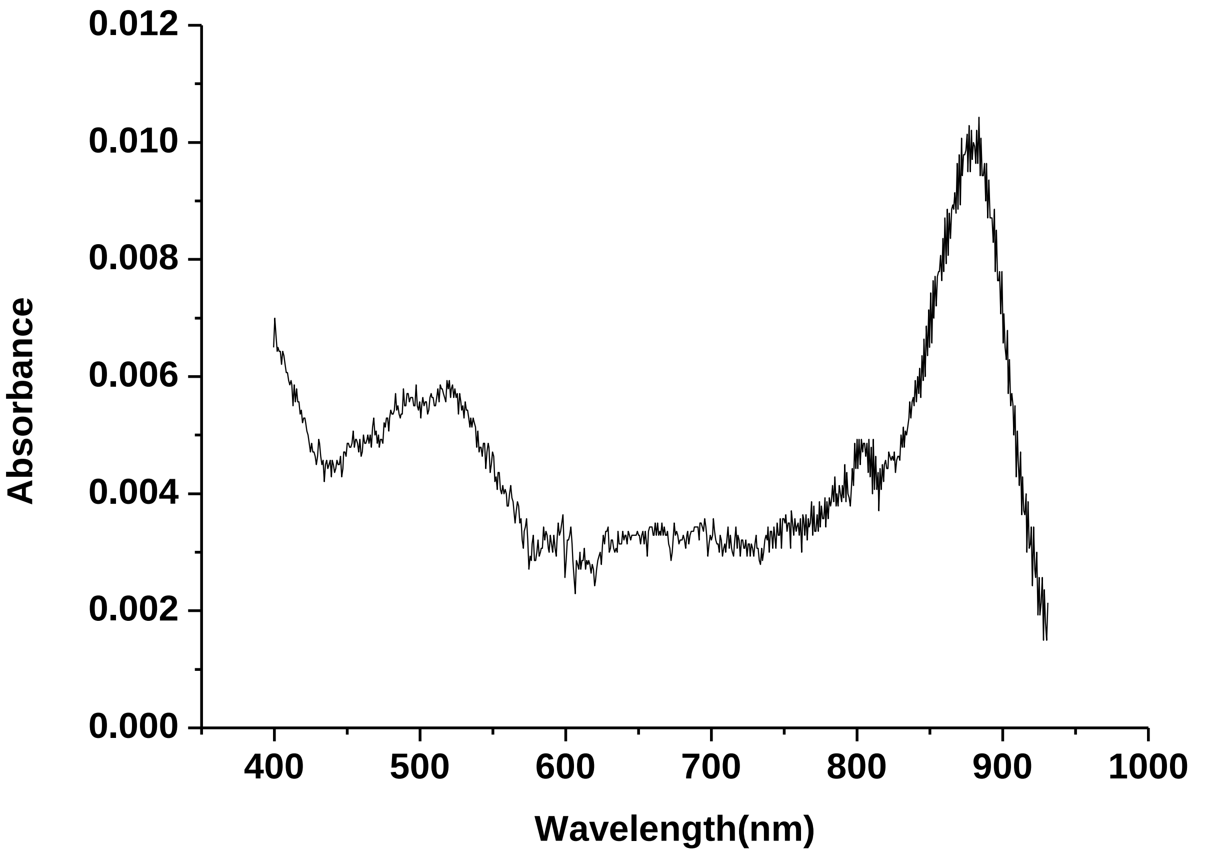
Retaining the quantum efficiency



Langmuir Blodgett films

‘

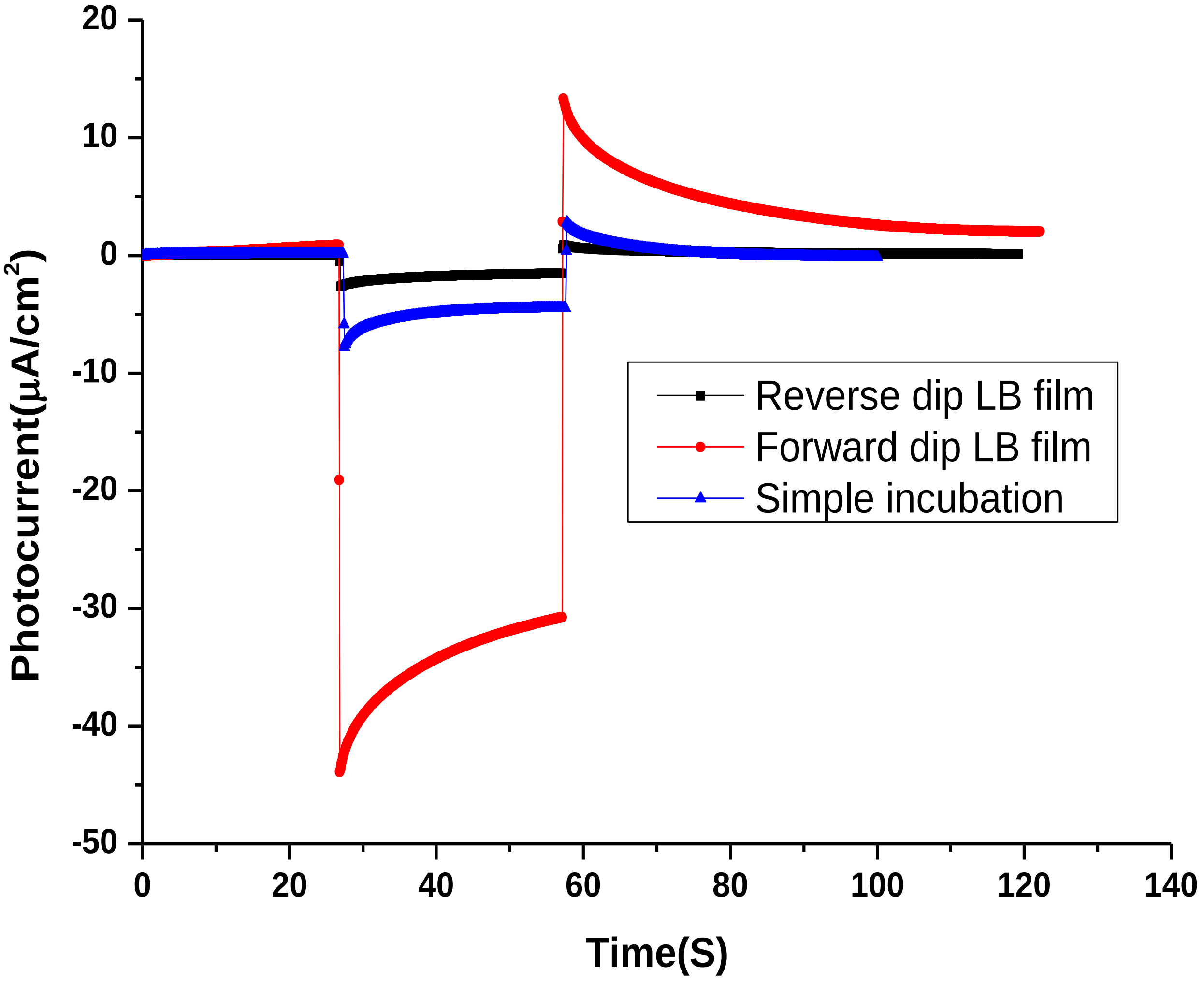
Absorbance of LB film



OD max = 0.007 but for RCLH1 on both sides of the slide

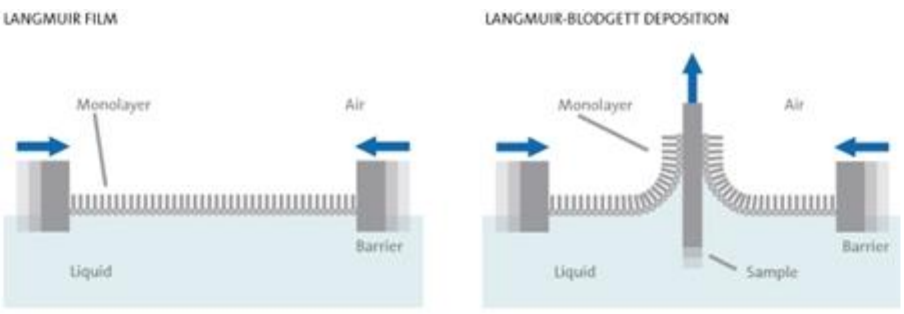
OD max 1 side, active layer = 0.0035

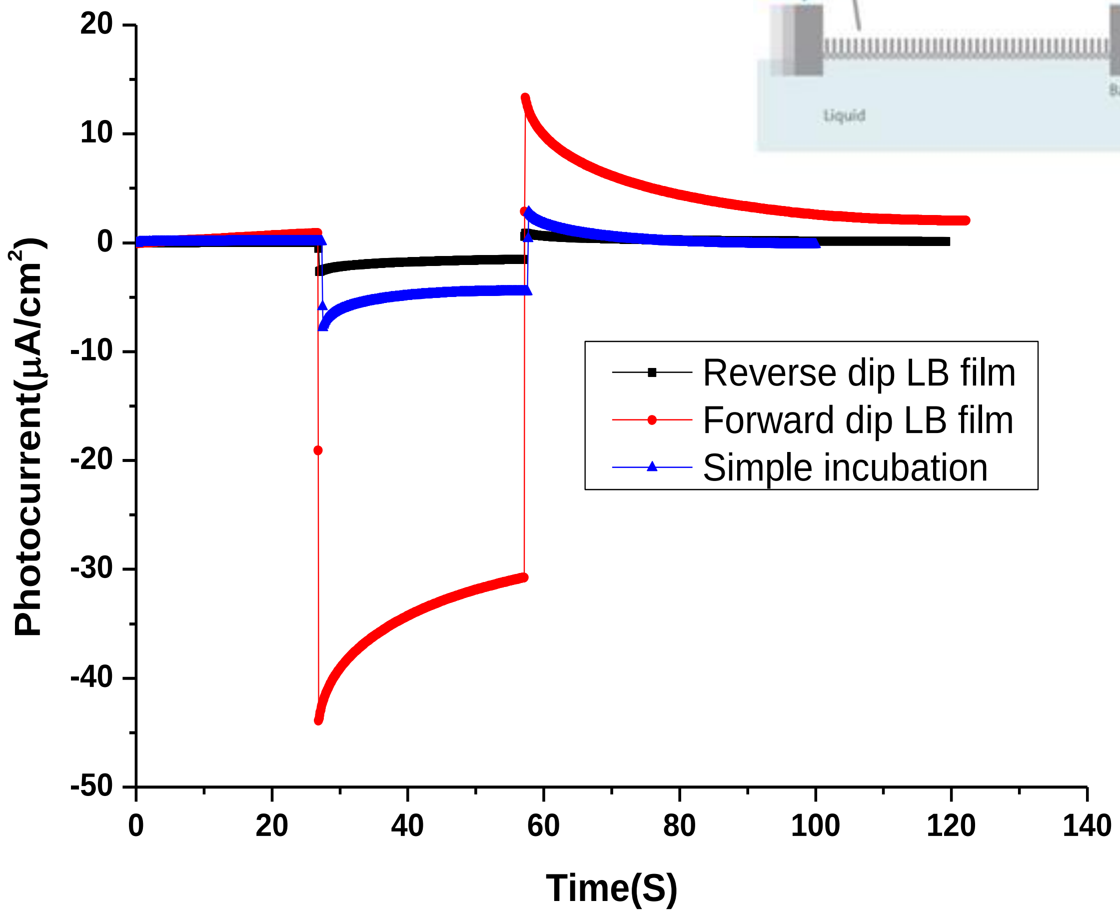
Comparison with adsorption



-175 mV vs SCE

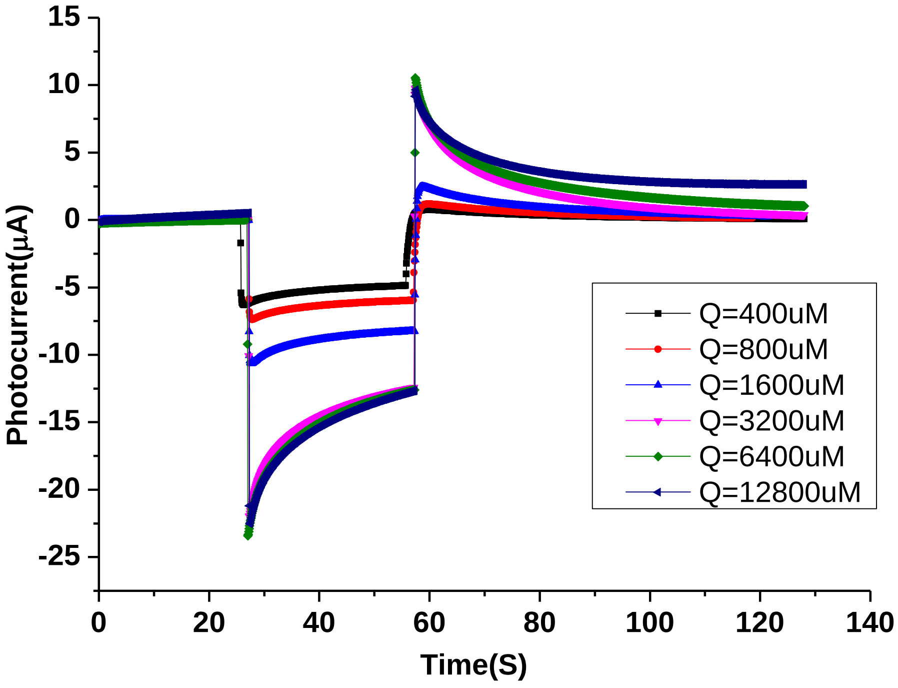
Controlling the orientation





Q only

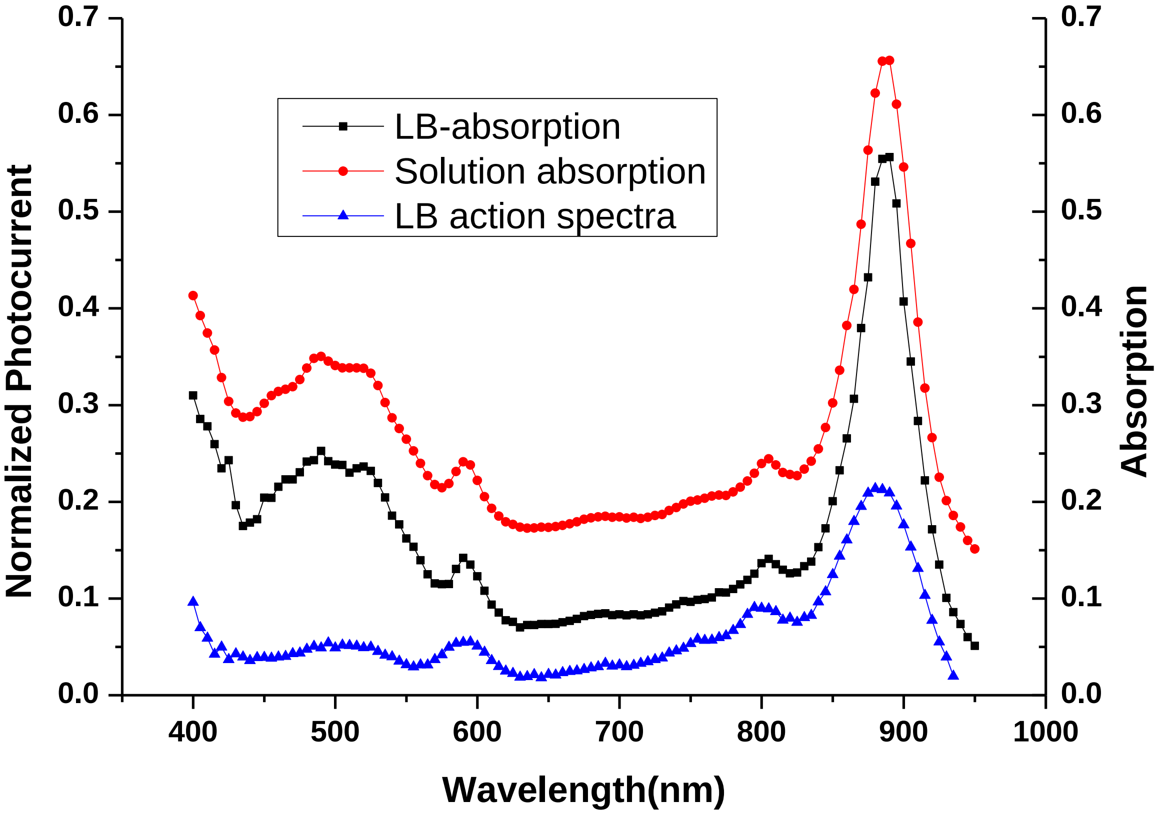
A significant amount of photocurrent was generated by using Quinones-only as mediators in the measuring buffer.



Only 1 mediator but high photocurrents.

Tight packing leads to improved connectivity P and gold.

Wavelength dependency of current generation



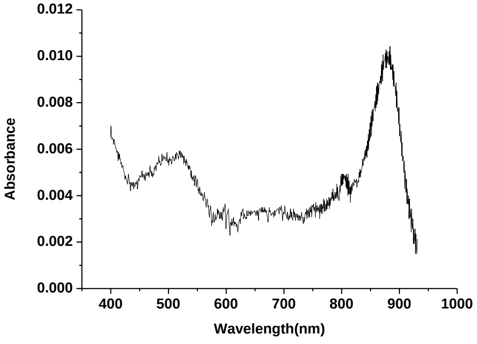
LH1 excitations partly quenched

Quantum efficiency 1

Quantum efficiency = $N_{el}/N_{abs} = N_{el}/[(1 – T)\ N_0]$

• Number of electrons per second per number of photons absorbed

With



• Transmission: T (from OD)

• Incident number of photons/s: N_0 (from power illumination)

• Number of absorbed photons/s: N_{abs}

• absorption cross-section : σ (calculate from the extinction coefficient)

• number if electrons per second: N_{el} (calculate from the current)

Quantum efficiency 2

Quantum efficiency =
max photocurrent/measured photocurrent

✓Assume 100% surface coverage,
✓Calculate number of complexes
✓Calculate number of absorbed photons from extinction coefficient

✓Assume every photon absorbed = 1 electron transferred> gives maximum photocurrent

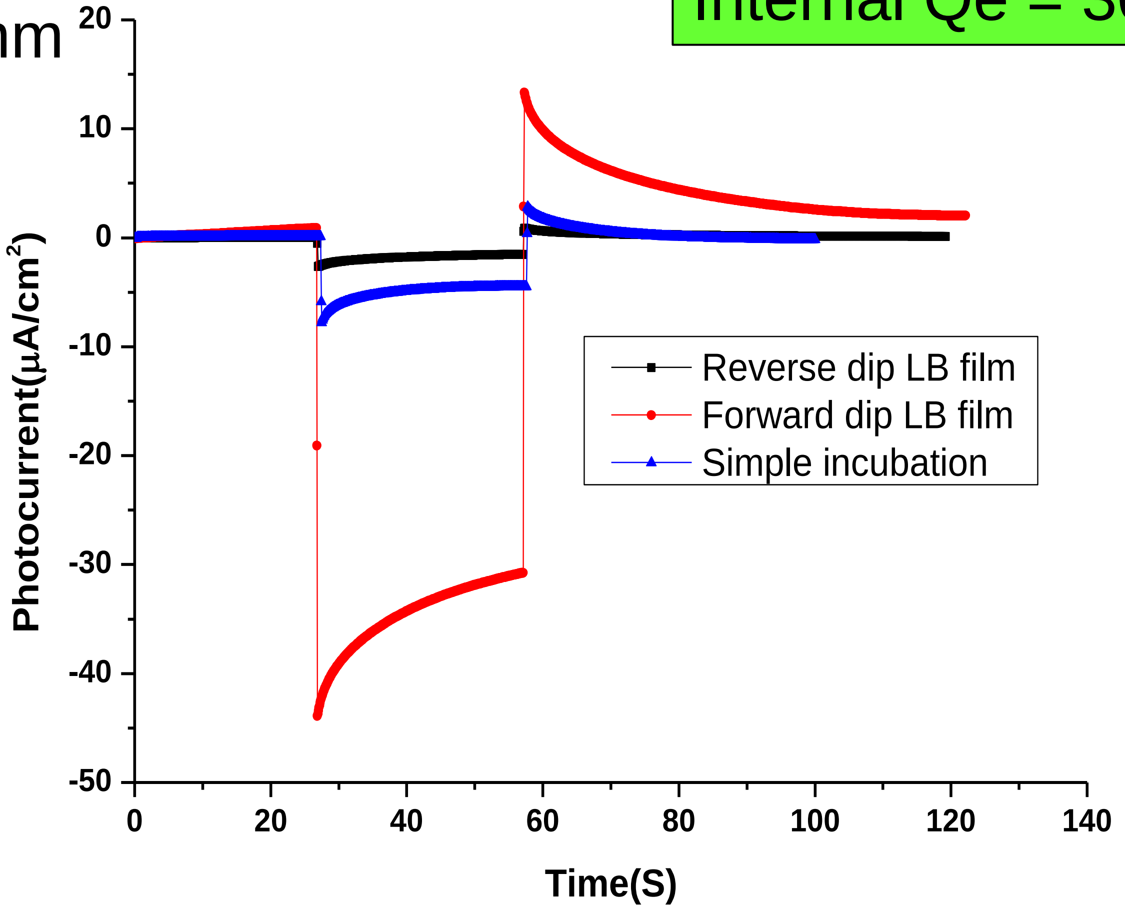
✓Measure surface coverage: AFM
✓Max photocurrent for this coverage/measured photocurrent

photocurrent RCLH1 monolayer = 45 $\mu A/cm^2$

With 23 mW/cm^2

@ 880 nm

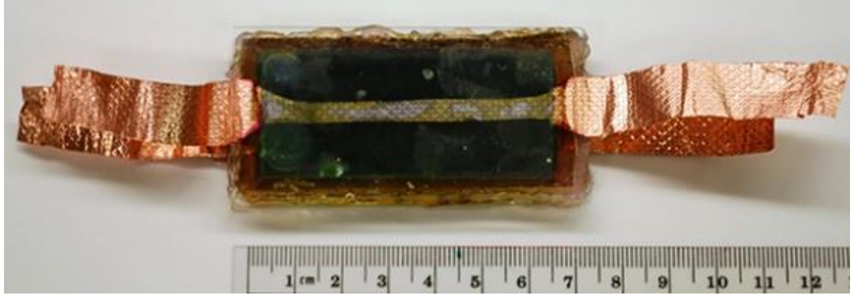
Internal Qe = 30(10)%

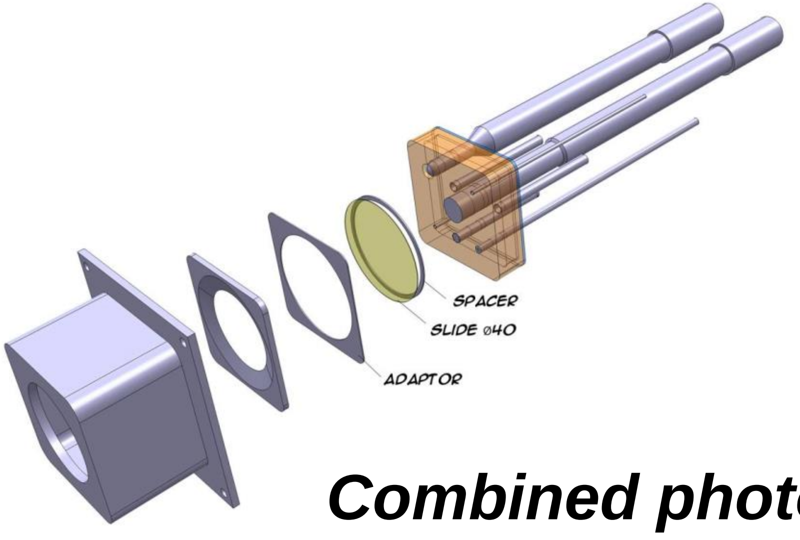


Whats next?

Techniques:

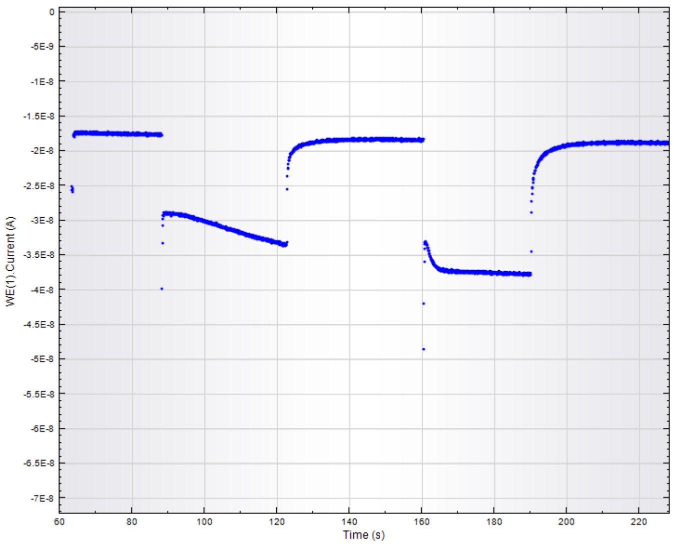
Photovoltaic Solar cells





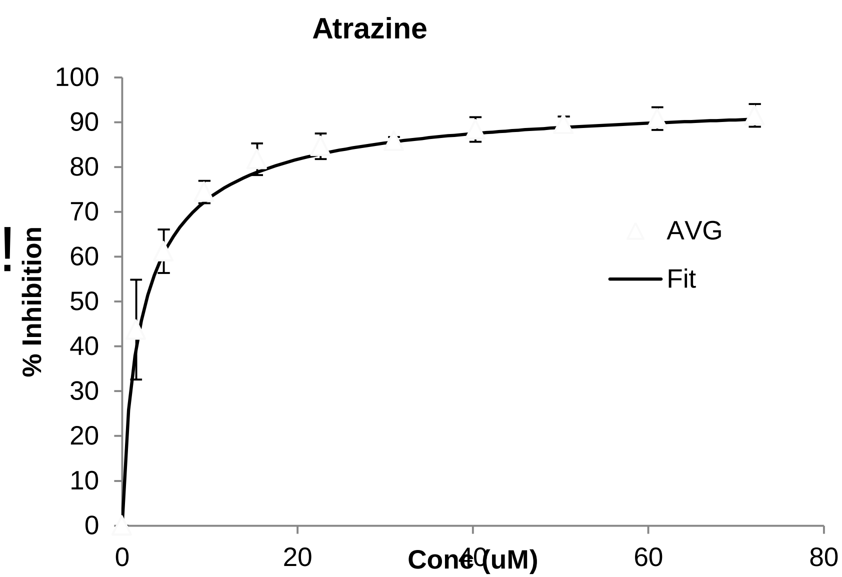
Combined photo electrochemistry and fluorescence

Whats next?



NO Tech solar cells: direct application of crude photosynthetic materials as solar cell material

And yes: ***biosensors***!



Work done by:

- Gerhard Magis, Mart Jan den Hollander, Muhammad Kamran (Leiden U.)
- Josep Pamies (Amolf, Cambridge UK)
- Vincent Friebe, David Delgado, Kinga Sznee (VU Univ)

Collaborators: Neil Hunter (Sheffield),

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