

# V24 - Stochastic Dynamics simulations of a photosynthetic vesicle

## where bioinformatics meets biophysics

I Introduction: prelude photosynthesis

II Process view and geometric model of a chromatophore vesicle  
Tihamér Geyer & V. Helms (Biophys. J. 2006a, 2006b)

III Stochastic dynamics simulations  
T. Geyer, Florian Lauck & V. Helms (J. Biotechnol. 2007)

IV Parameter fit through evolutionary algorithm  
T. Geyer, X. Mol, S. Blaß & V. Helms (PLoS ONE 2010)

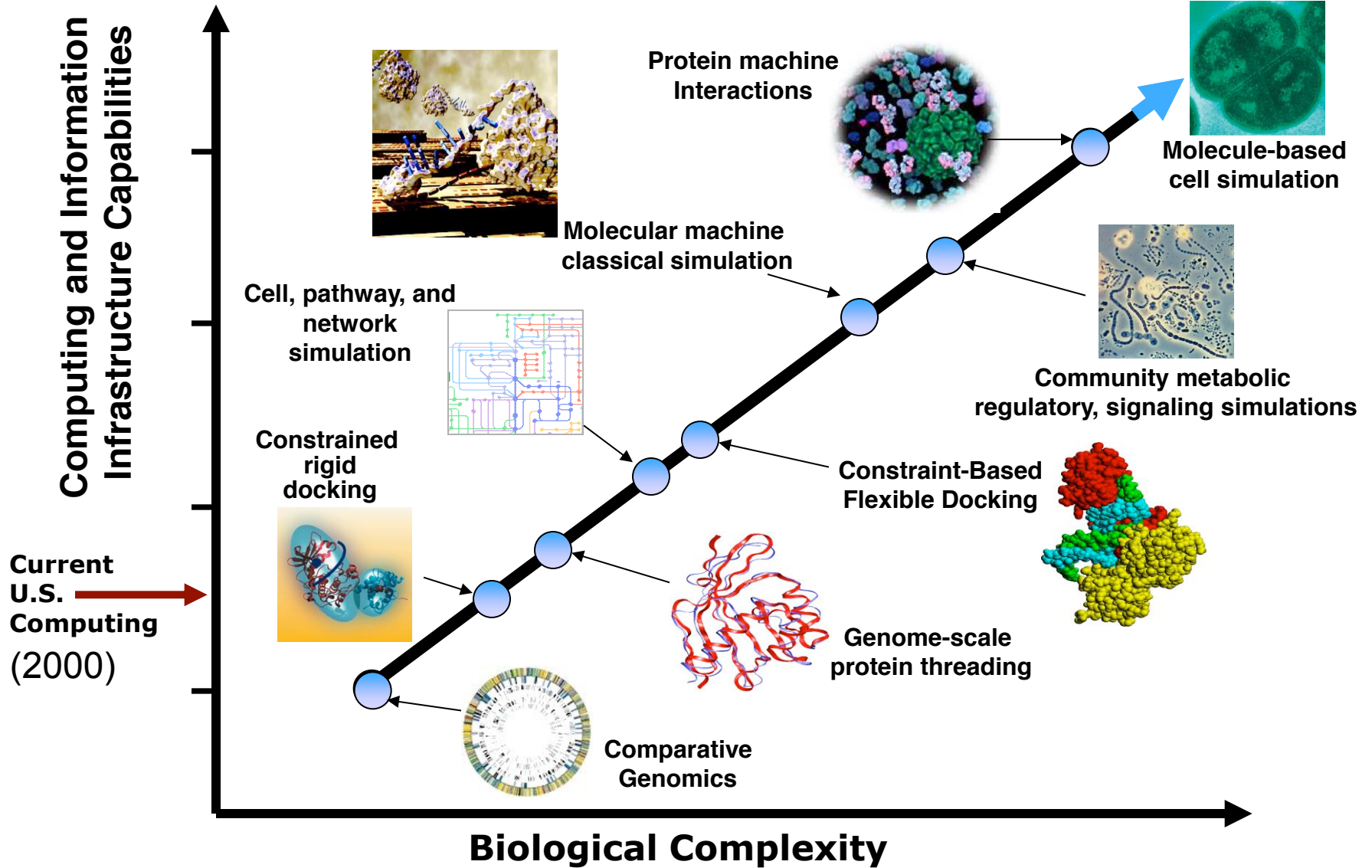
## Content of final exam (March 7, 2016)

Lecture	Slides relevant for exam
1	18-24
2	1-14, 18-20
3	17-26
4	All
5	1-32
6	None
7	None
8	All
9	None
10	5-17
11	8-27
12	The main ideas

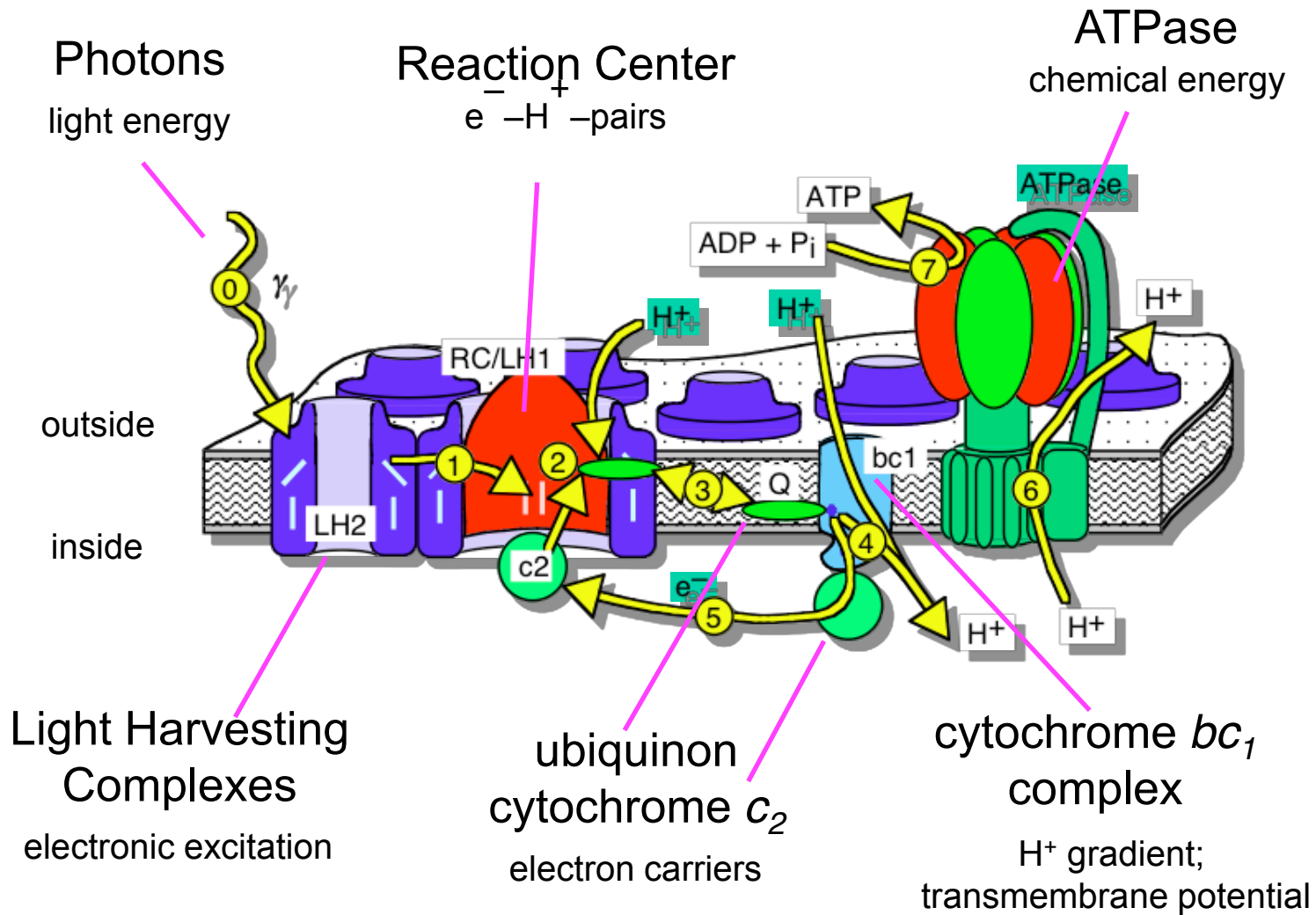
Lecture	Slides relevant for exam
13	The main ideas
14	1-18
15	1-22
16	1-11, 14
17	41-48
18	1-14
19	18-41
20	None
21	None
22	None
23	None
24	None

**Relevant are also the assignments !  
(theoretical parts, not the programming parts)**

# “Genomes To Life” Computing Roadmap (NIH/DOE)

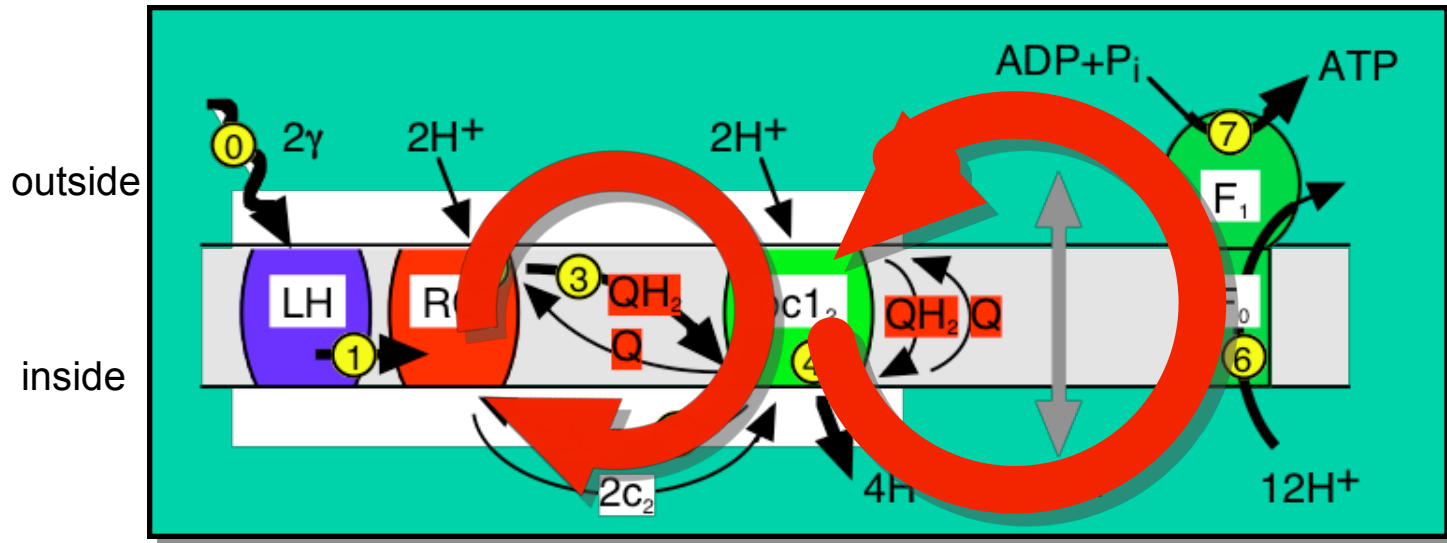
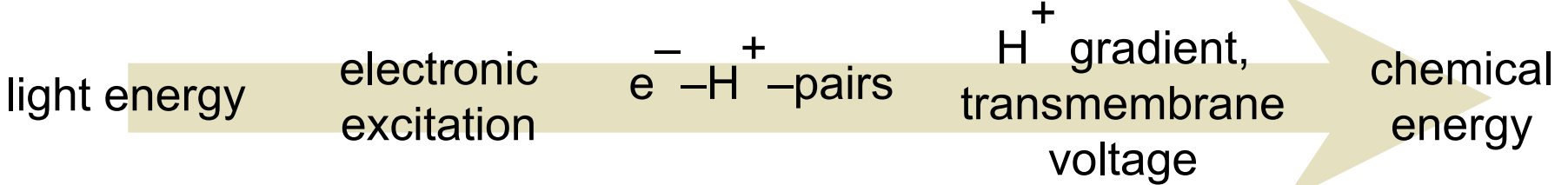


# Bacterial Photosynthesis 101



# Photosynthesis – cycle view

The conversion chain: stoichiometries must match turnovers!



2 cycles:

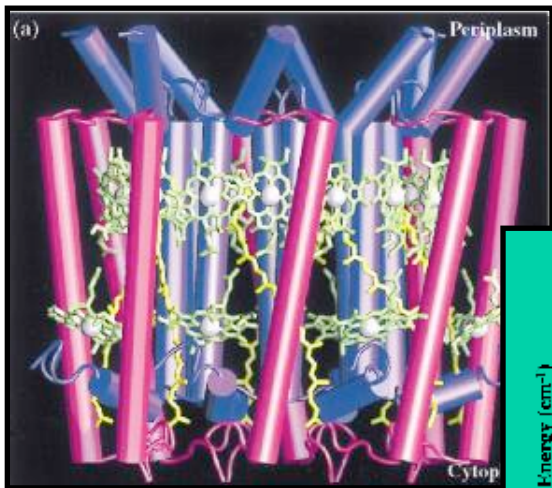
electrons

protons

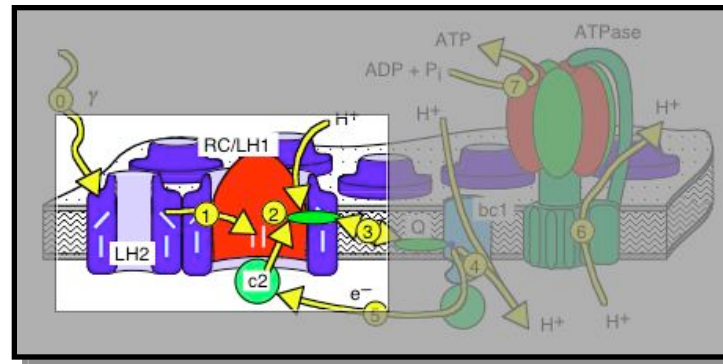
# LH1 / LH2 / RC — a la textbook

Collecting photons

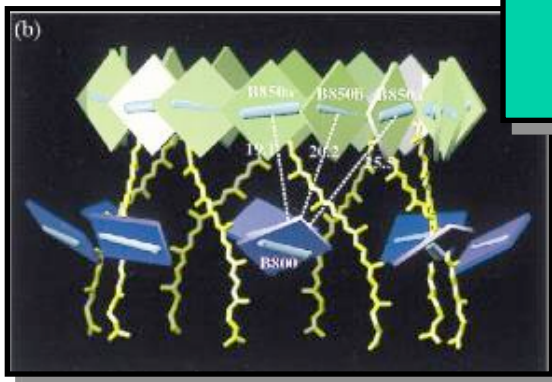
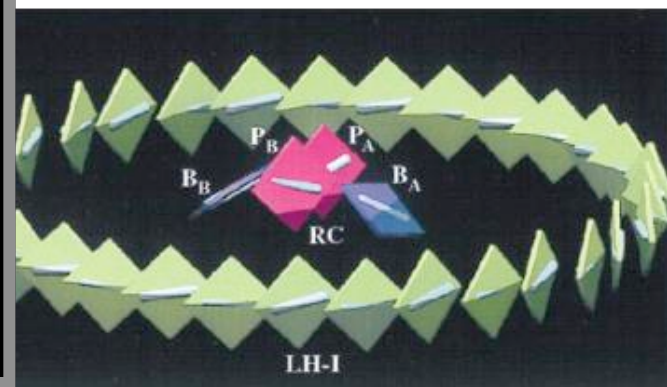
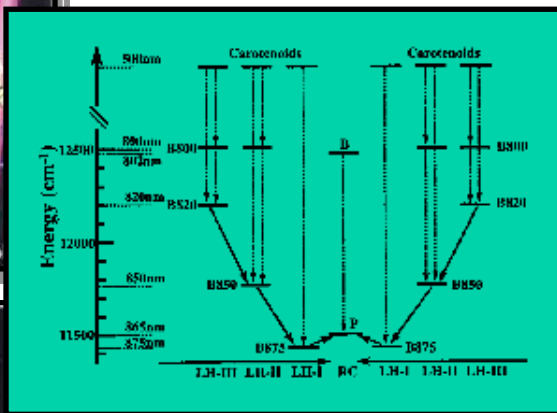
LH2: 8  $\alpha\beta$  dimers



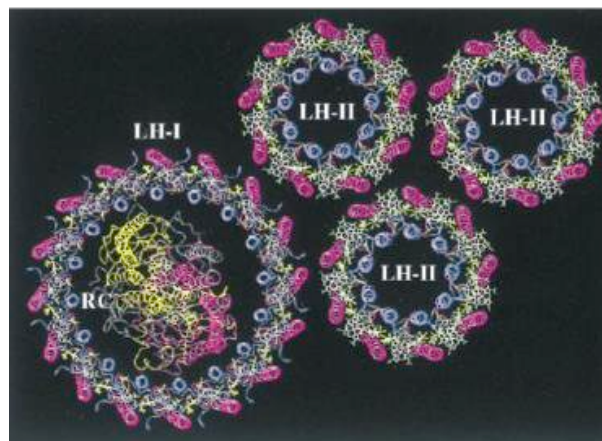
downhill transport  
of excitons  
LH2  $\rightarrow$  LH1  $\rightarrow$  RC



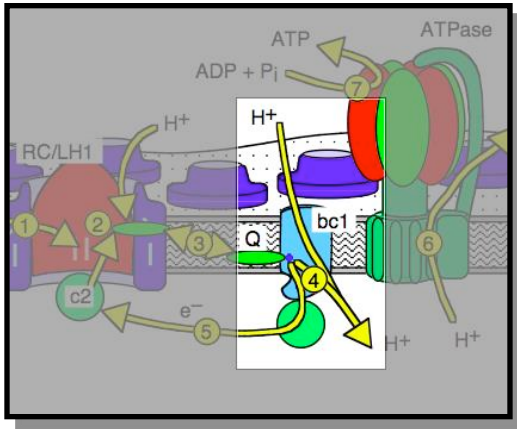
LH1: 16  $\alpha\beta$  dimers



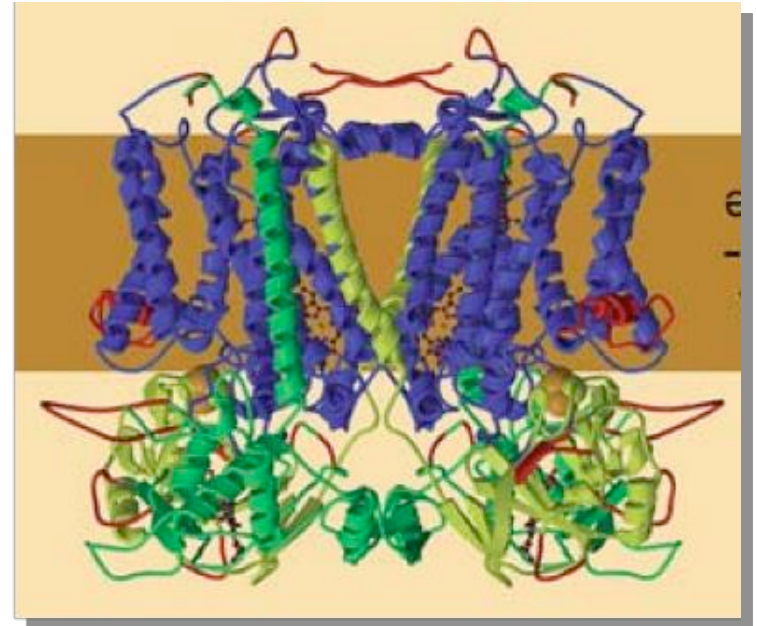
B800, B850, Car.



# The Cytochrome *bc<sub>1</sub>* complex

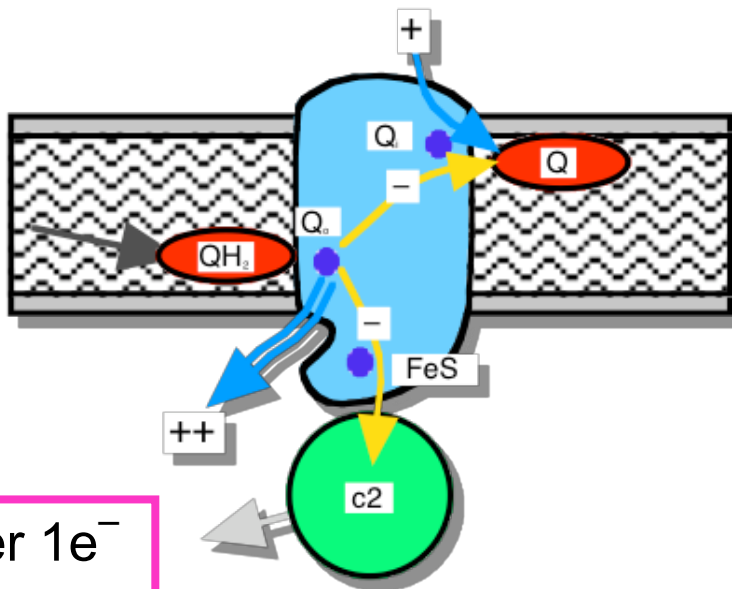


the "proton pump"



Berry, etal, 2004

Q-cycle:

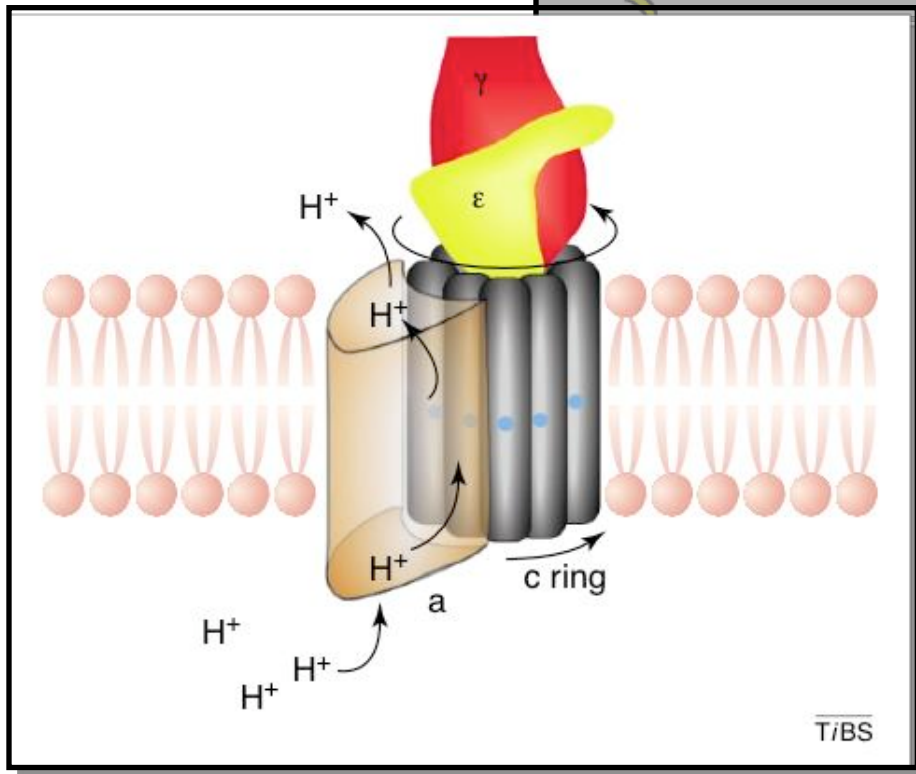


$2\text{H}^+$  per  $1\text{e}^-$

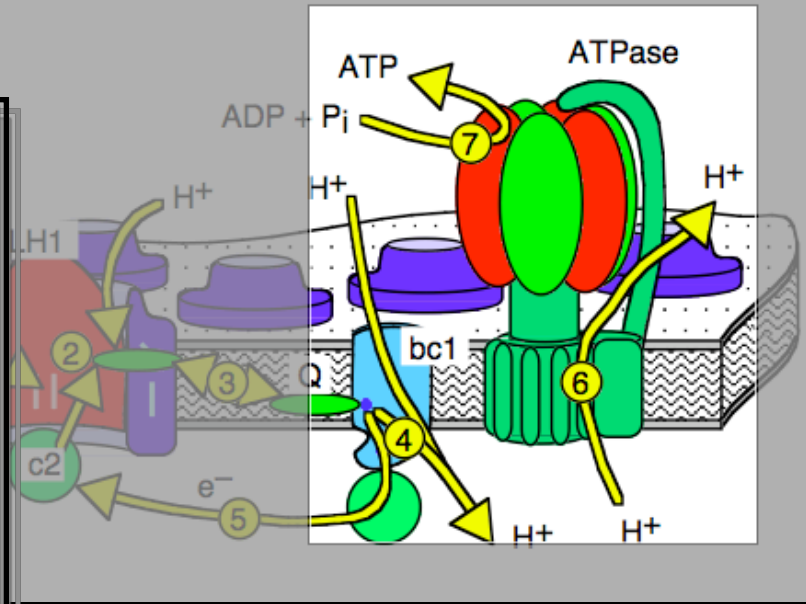
X-ray structures known  
always forms a dimer

# The $F_0F_1$ -ATP synthase I

at the end of the chain: producing ATP from the  $H^+$  gradient



Capaldi, Aggeler, 2002



per turn:

$10-14 H^+ \rightarrow 3 ATP$

$1 ATP \triangleq 4 H^+$

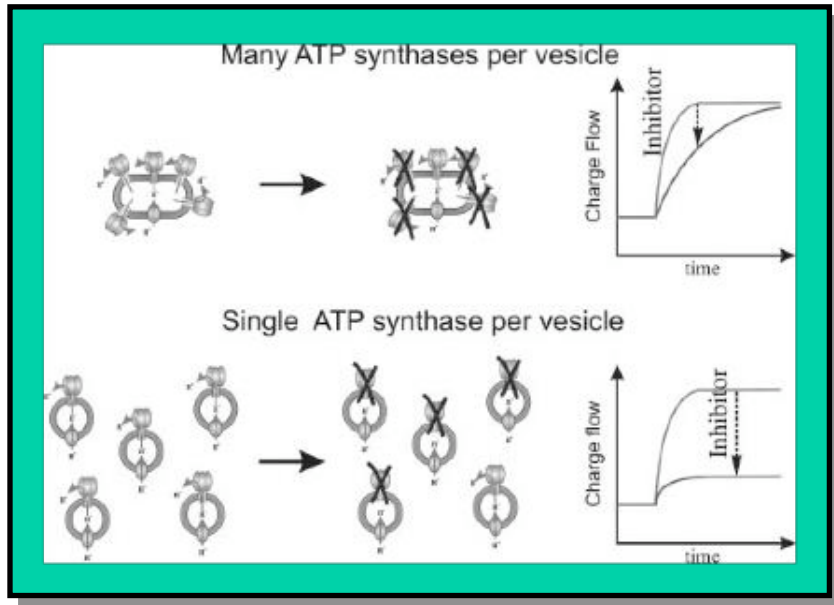


# The $F_1F_0$ -ATP synthase

"...mushroom like structures observed in AFM images..."

→ ATPase is "visible"

1 ATPase per vesicle

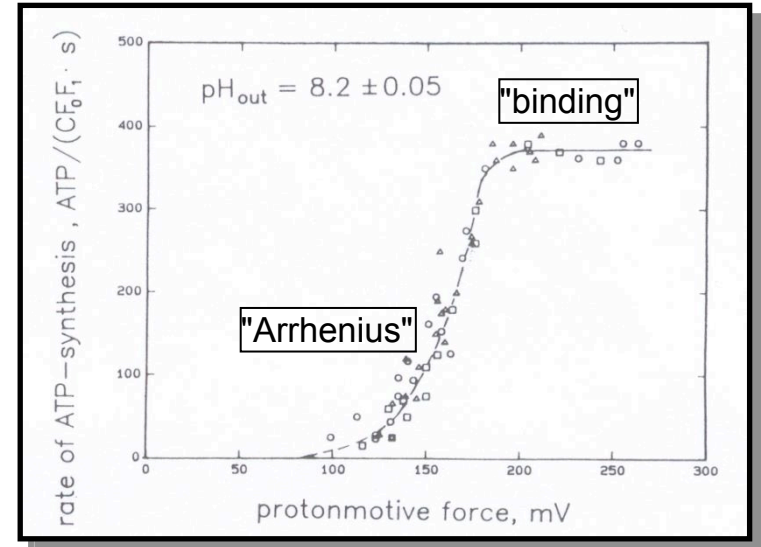


Feniouk et al, 2002

per turn: 10–14  $H^+$  per 3 ATP

→ 1 ATP  $\cong$  4  $H^+$

limited throughput of the ATPase



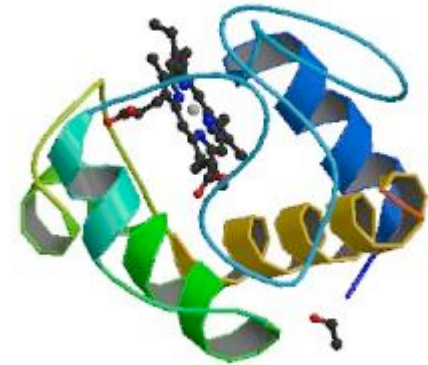
ATPase from	ATP/s	$H^+$ /s
chloroplasts	<400	1600
E. coli	<100	400

Gräber et al, 1991, 1999

# The electron carriers

**Cytochrome *c***: carries electrons from  $bc_1$  to RC

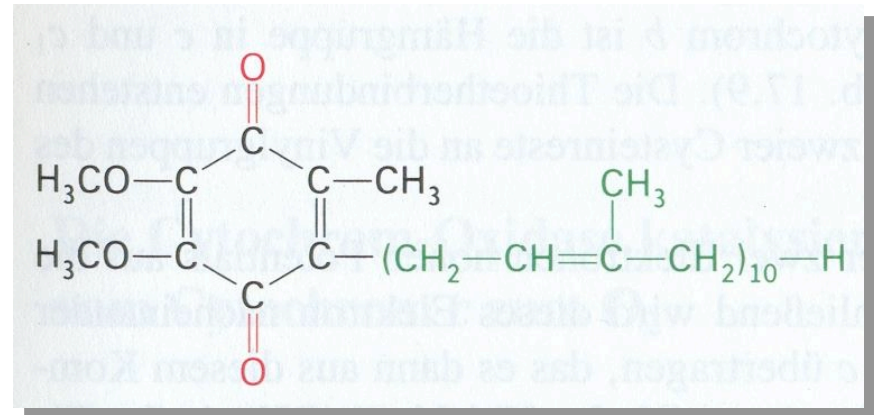
- heme in a hydrophilic protein shell
- 3.3 nm diameter, water-soluble



**Ubiquinone UQ10**:

carries electron–proton pairs  
from RC to  $bc_1$

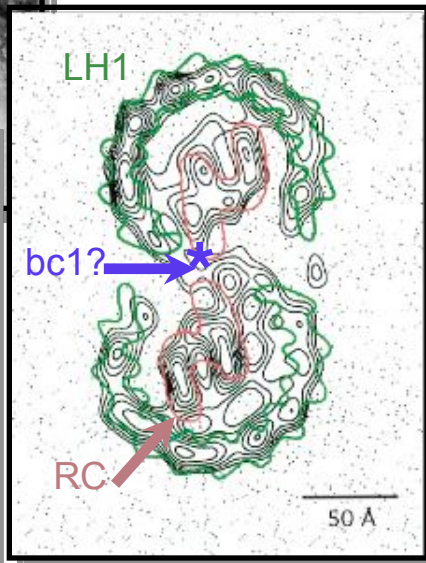
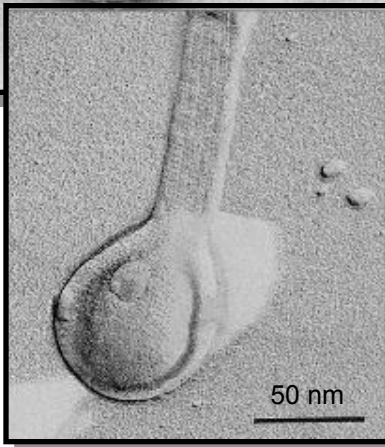
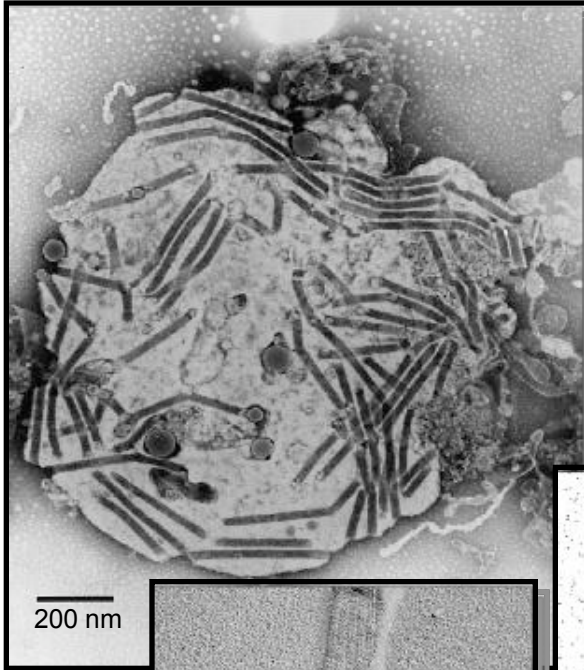
- long (2.4 nm)  
hydrophobic  
isoprenoid tail,  
membrane-soluble



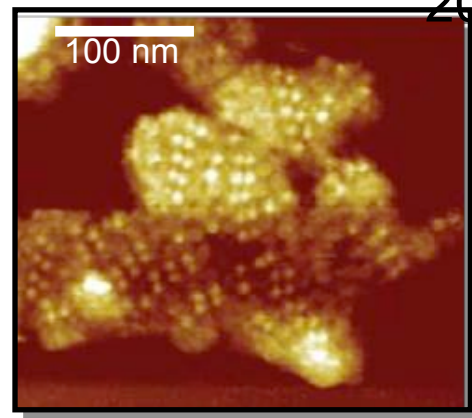
taken from Stryer

# Tubular membranes – photosynthetic vesicles where are the $bc_1$ complexes and the ATPase?

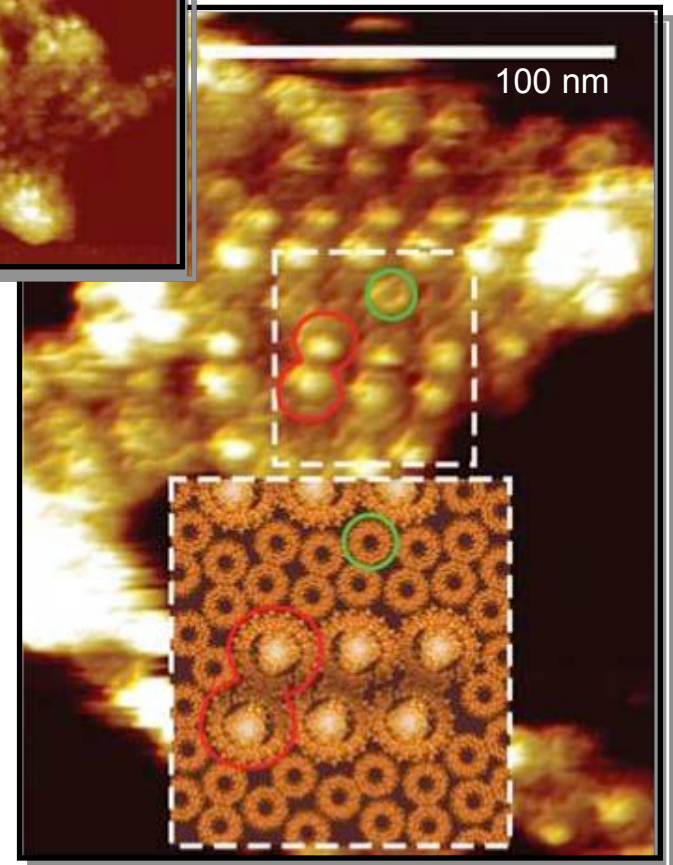
Jungas *et al.*, 1999



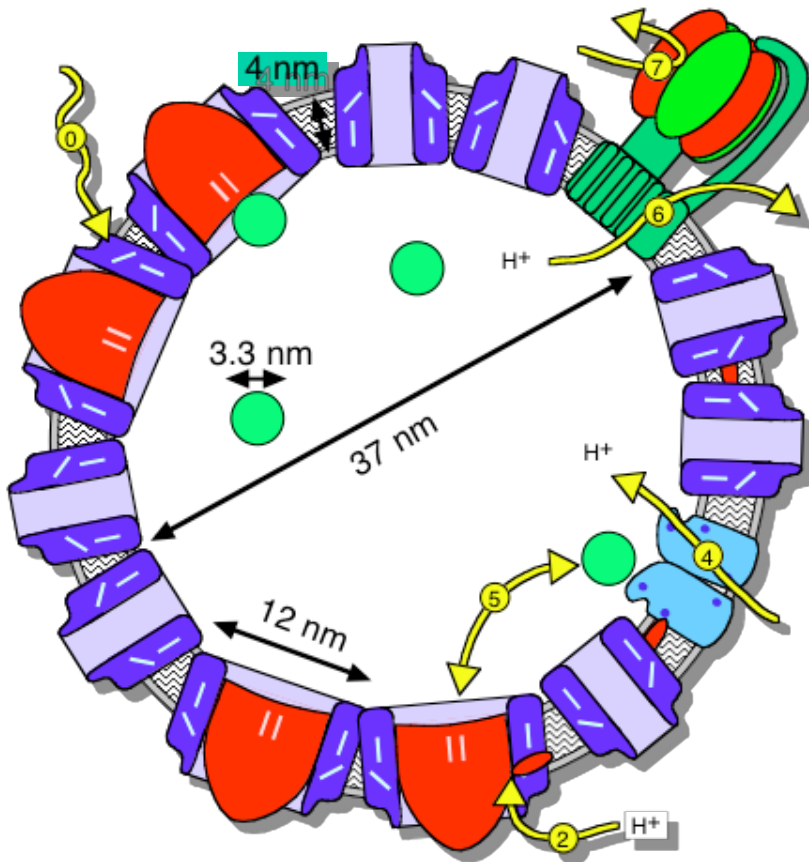
Bahatyrova *et al.*,  
2004



no  $bc_1$   
found!



# Chromatophore vesicle: typical form in *Rh. sphaeroides*



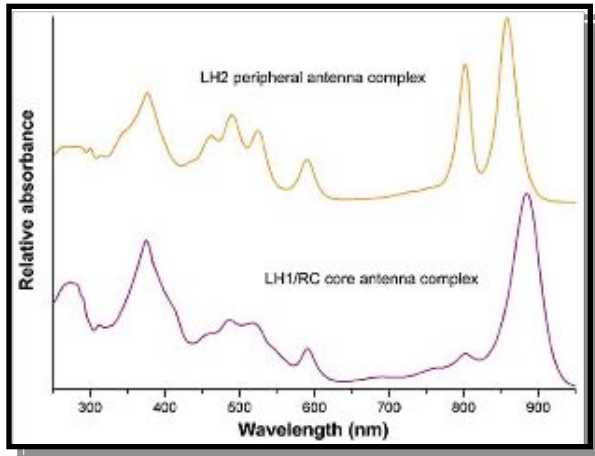
Lipid vesicles  
30–60 nm diameter  
 $H^+$  and cyt *c* inside

average  
chromatophore surface  
vesicle, 45 nm  $\varnothing$ : 6300 nm<sup>2</sup>

Vesicles are really small!

# Photon capture rate of LHC's

relative absorption spectrum  
of LH1/RC and LH2

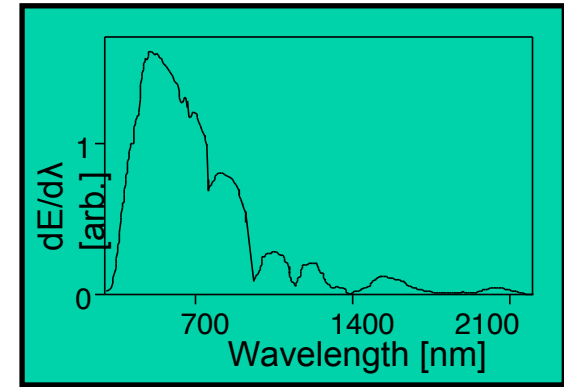


Cogdell et al, 2003

+ Bchl extinction coeff.  
normalization ( $\sigma_{\text{Bchl}} = 2.3 \text{ \AA}^2$ )

Franke, Amesz, 1995

sun's spectrum at ground  
(total: 1 kW/m<sup>2</sup>)



Gerthsen, 1985

multiply

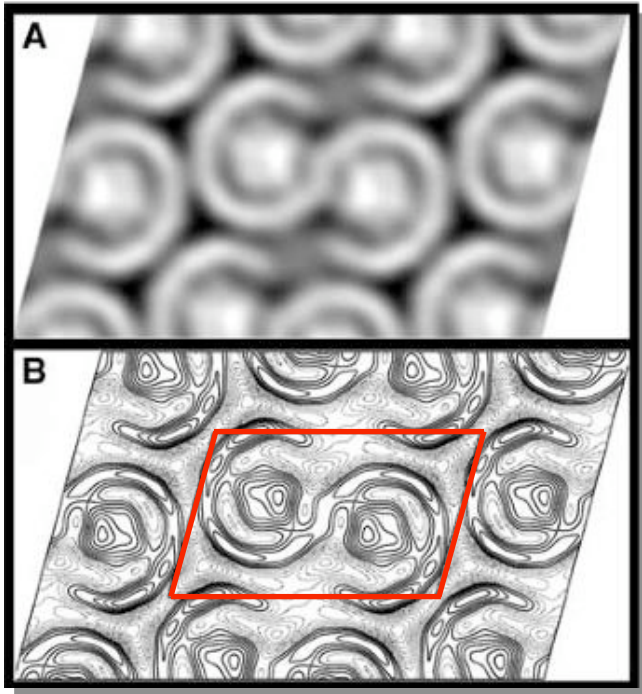
capture rate:  $0.1 \frac{\gamma}{\text{s kW Bchl}}$

typical growth condition:  
18 W/m<sup>2</sup> Feniouk et al, 2002

LH1: 16 * 3 Bchl	➔	14 γ/s
LH2: 10 * 3 Bchl	➔	10 γ/s

# LH1 / LH2 / RC — native

electron micrograph  
and density map



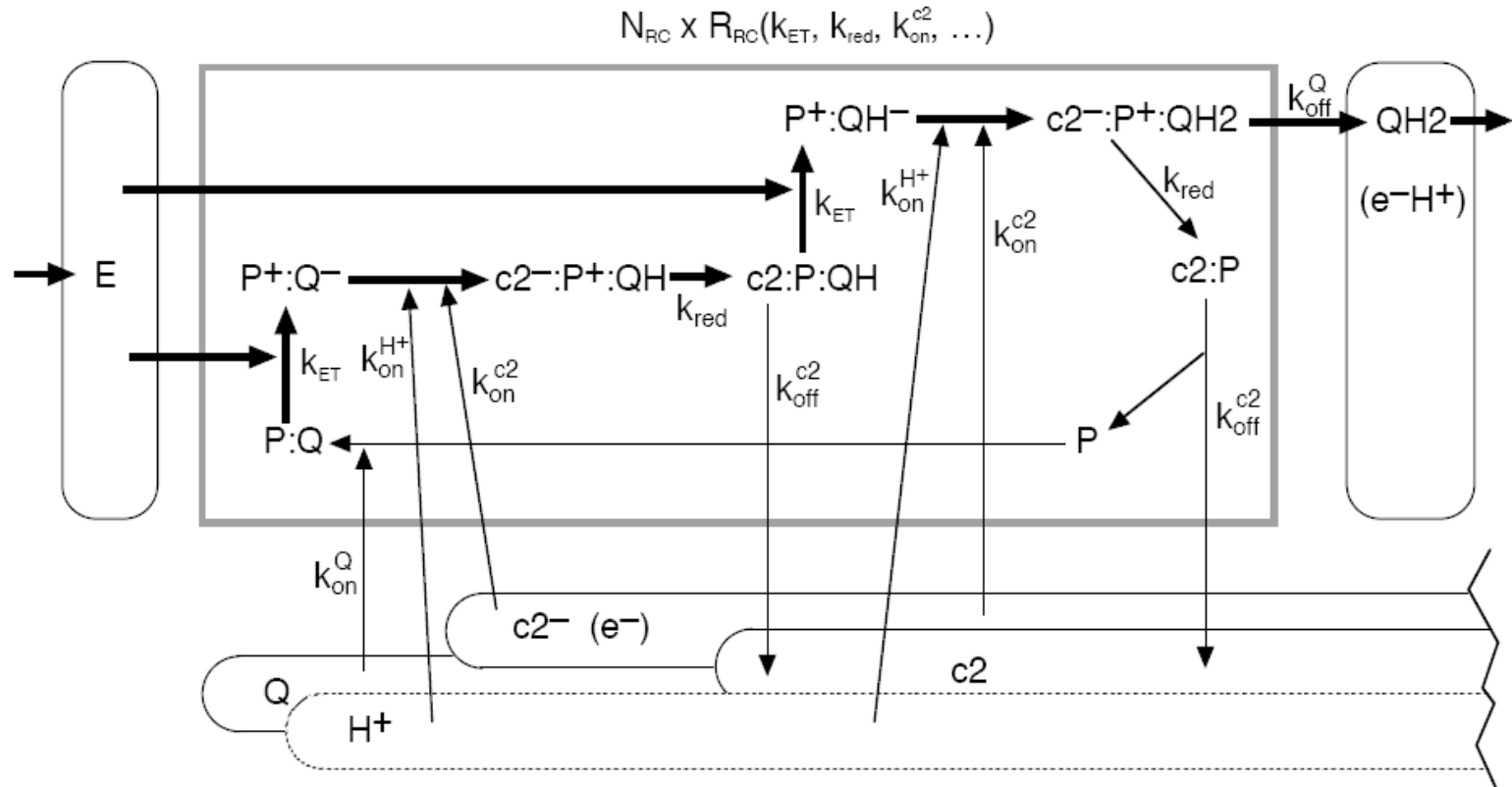
Siebert et al, 2004  
 $125 * 195 \text{ \AA}^2$ ,  $\gamma = 106^\circ$

	Area per:	per vesicle (45 nm)
LH1 monomer (hexagonal)	146 nm <sup>2</sup>	
LH1 dimer	234 nm <sup>2</sup>	
LH2 monomer	37 nm <sup>2</sup>	
LH1 <sub>2</sub> + 6 LH2	456 nm <sup>2</sup>	11

Chromatophore surface vesicle, 45 nm Ø: 6300 nm<sup>2</sup>



# Modelling of internal processes at reaction center



All individual reactions with their individual rates  $k$  together determine the overall conversion rate  $R_{RC}$  of a single RC.

Thick arrows : flow of the energy from the excitons through the cyclic charge state changes of the special pair Bchl (P) of the RC.

Rounded rectangles : reservoirs



# *bc*<sub>1</sub> Placement — Diffusional limits?

Roundtrip times  
maximal capacity of the carriers:

$$T = T_{RC} + T_{bc1} + T_{Diff}$$

## Cytochrome *c*<sub>2</sub>:

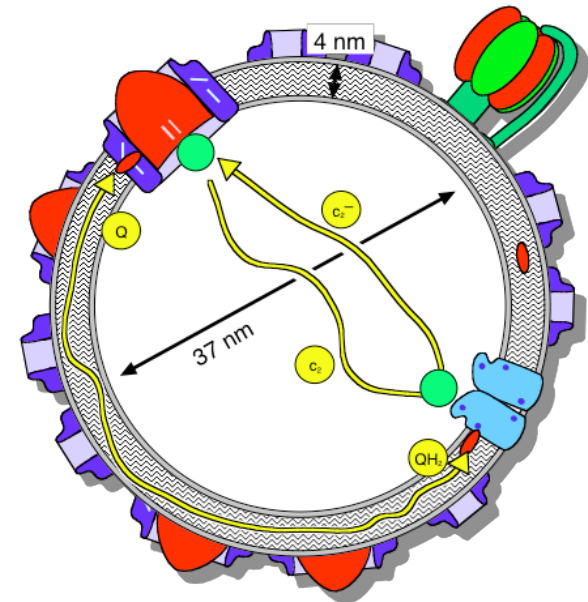
$$T_{RC} \approx 1 \text{ ms} \quad T_{bc1} \approx 12 \text{ ms} \quad T_{Diff} \approx 3 \mu\text{s}$$

$T_{\text{round-trip}} = 13 \text{ ms} \rightarrow \leq 3 \text{ cyt } c \text{ per vesicle}$   
sufficient to carry  $e^-$ 's  
available: 22 cyt *c* per vesicle

## Quinol:

$$T_{RC} \approx 50 \text{ ms} \quad T_{bc1} \approx 23 \text{ ms} \quad T_{Diff} \approx 1 \text{ ms}$$

$T_{\text{round-trip}} = 75 \text{ ms} \rightarrow \leq 7 \text{ Q per vesicle}$   
sufficient to carry  $e^-$ 's.  
available: 100 Q per vesicle

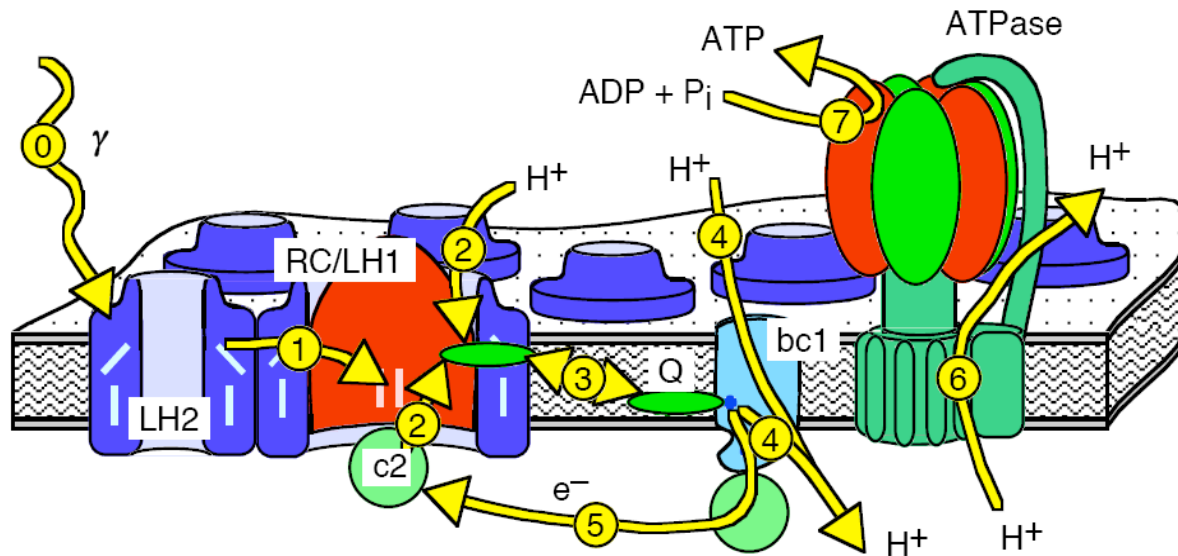


Diffusion is not limiting

$\rightarrow$  poses no constraints  
on the position of *bc*<sub>1</sub>

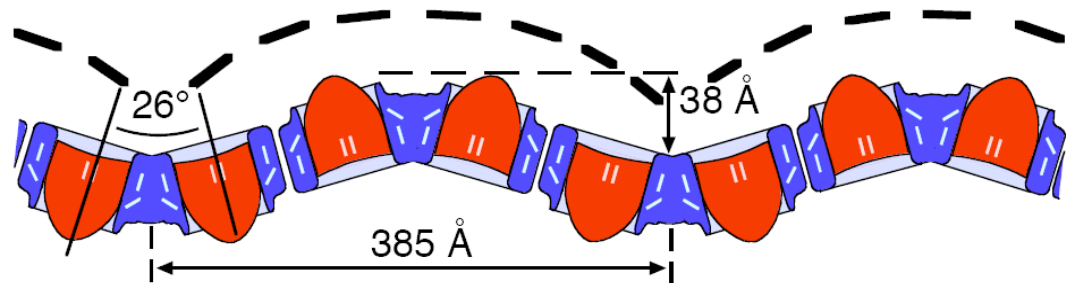
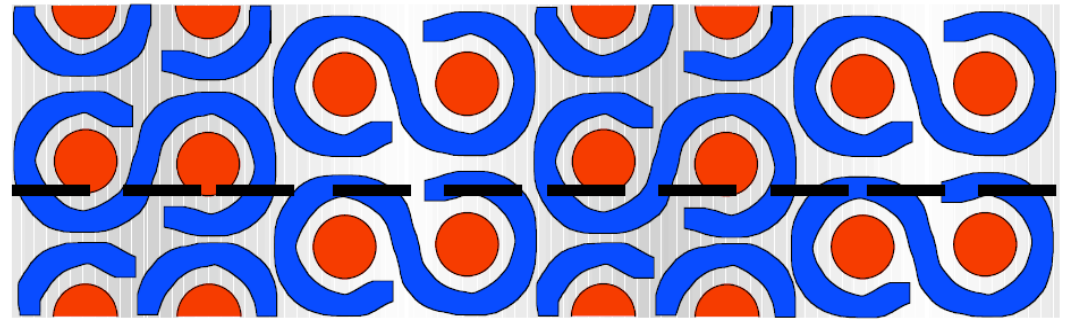
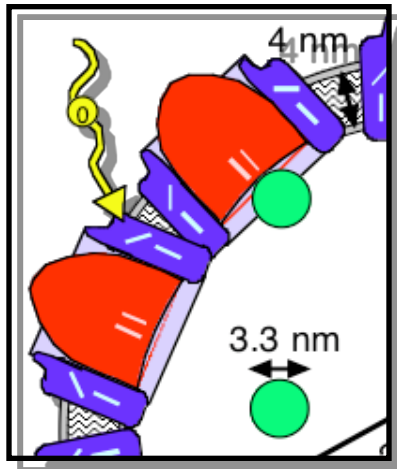
# Parameters

protein	throughput per protein (natural units)	H <sup>+</sup> equivalents per protein [1/s]	total number per avg. vesicle of 45 nm diameter	rate determined from	explained in section
LH2	10 $\gamma/s$	20	60	absorption spectra +	III A
LH1 dimer	2 $\times$ 14 $\gamma/s$	56	10	+ light intensity of 18 W/m <sup>2</sup>	III A
RC	22 QH2/s	88	20	QH2 (un)binding	III B
bc1 dimer	$\leq$ 2 $\times$ 42 c2/s	168	3 (... 10)	measured activity at $\Delta$ pH = 0	III C
ATPase	$\leq$ 100 ATP/s	400	1	measured throughput	III D
cytochrome <i>c</i> <sub>2</sub>	80 e <sup>-</sup> /s	160	20	(un)binding at the bc1	V A (III B, III C)
ubiquinone	10 $\times$ 2(e <sup>-</sup> H <sup>+</sup> )/s	40	100	(un)binding at the RC and the bc1	V A (III B, III C)



# reconstituted LH1 dimers in planar lipid membranes explain intrinsic curvature of vesicles

Drawn after AFM images of Scheuring *et al* of LH1 dimers reconstituted into planar lipid membranes.



Values fit nicely to the proposed arrangement of LH1 dimers, when one assumes that they are stiff enough to retain the bending angle of  $26^\circ$  that they would have on a spherical vesicle of 45 nm diameter and taking into account the length of a single LH1 dimer of about 19.5 nm.

# Proposed setup of a chromatophore vesicle

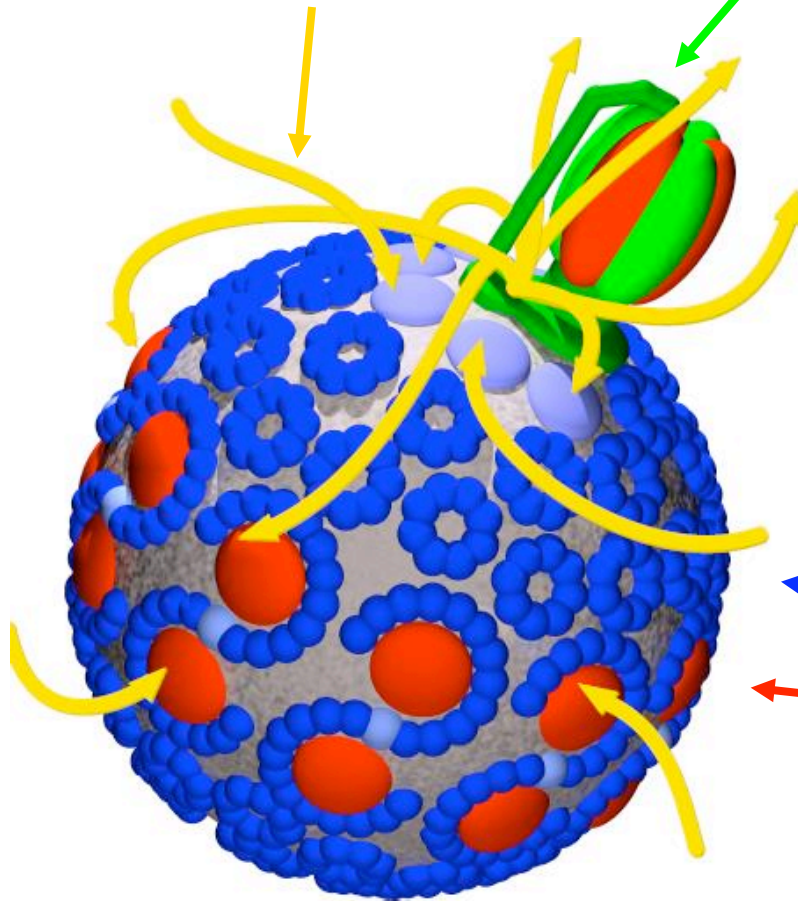
yellow arrows: diffusion of the protons out of the vesicle via the ATPase and to the RCs and bc1s.

At the „poles“

green/red: the ATPase

light blue: the bc1 complexes

Increased proton density close to the ATPase suggests close proximity of ATPase and  $bc_1$  complexes.



blue: small LH2 rings (blue)

blue/red: Z-shaped LH1/RC dimers form a linear array around the “equator” of the vesicle, determining the vesicle’s diameter by their intrinsic curvature.

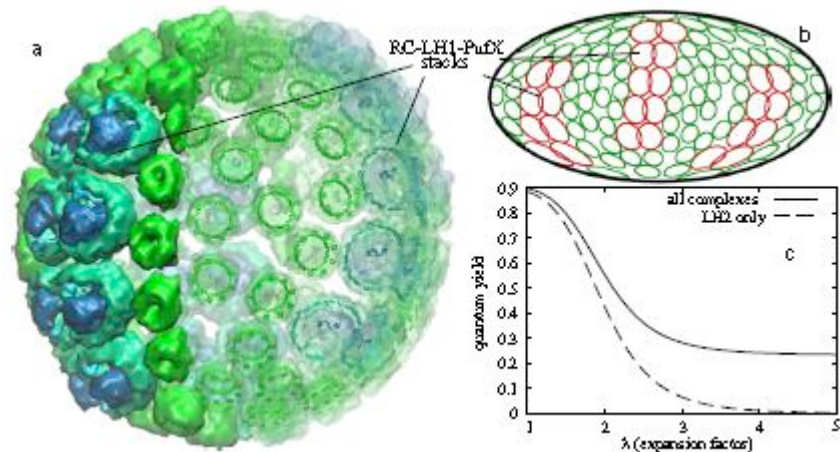
# Summary

Integrated model of binding + photophysical + redox processes  
inside of chromatophore vesicles

Various experimental data  
fit well together

Equilibrium state.

How to model  
non-equilibrium processes?



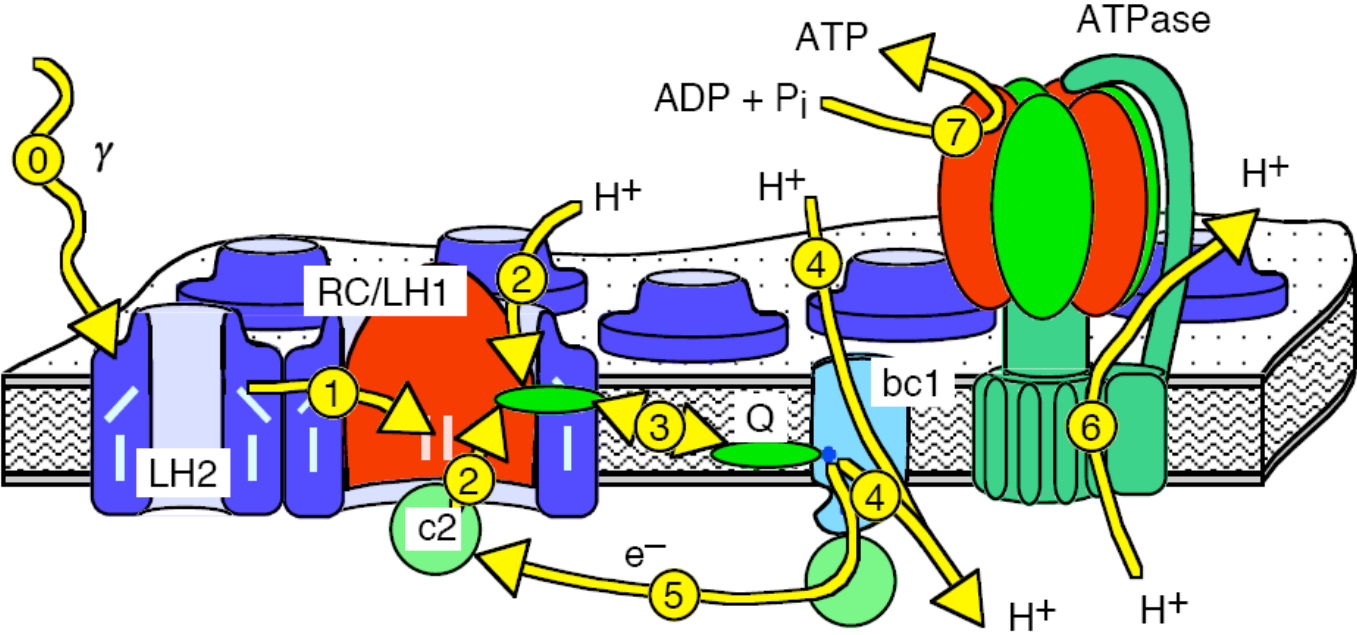
Biophysical Journal Volume 99 July 2010 67-75

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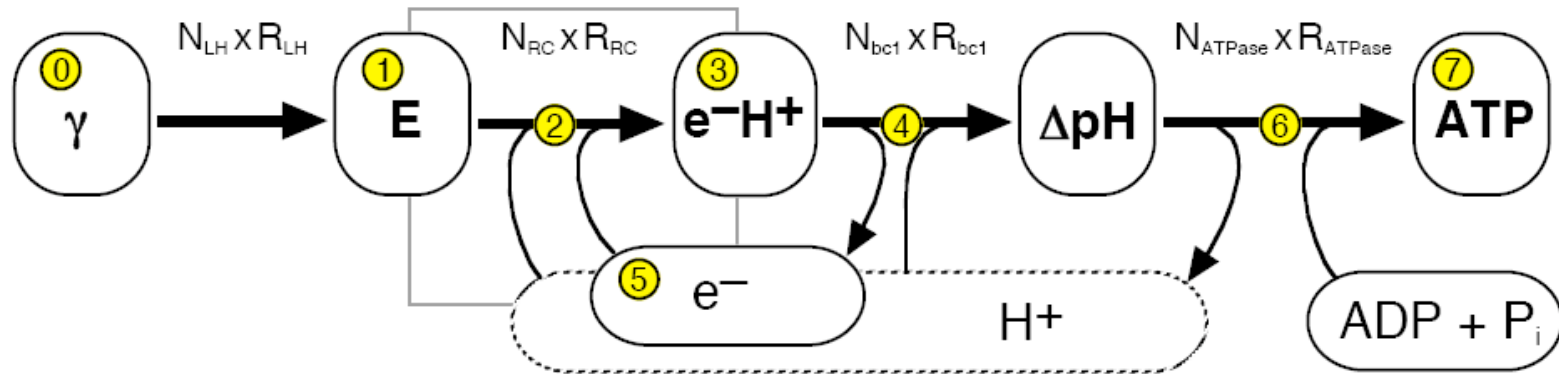
## Photosynthetic Vesicle Architecture and Constraints on Efficient Energy Harvesting

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<sup>†</sup>Beckman Institute for Advanced Science and Technology, <sup>‡</sup>Department of Physics, and <sup>§</sup>Center for Biophysics and Computational Biology, University of Illinois at Urbana-Champaign, Urbana, Illinois; <sup>¶</sup>Department of Molecular Biology and Biotechnology, University of Sheffield, Sheffield, United Kingdom; and <sup>||</sup>Institute of Physics and <sup>\*\*</sup>Institute of Molecular and Cell Biology, University of Tartu, Tartu, Estonia

# Photosynthesis: textbook view



# Viewing the photosynthetic apparatus as a conversion chain



Thick arrows : path through which the photon energy is converted into chemical energy stored in ATP via the intermediate stages (rounded rectangles).

Each conversion step takes place in parallelly working proteins. Their number  $N$  times the conversion rate of a single protein  $R$  determines the total throughput of this step.

$\gamma$  : incoming photons collected in the LHCs

$E$  : excitons in the LHCs and in the RC

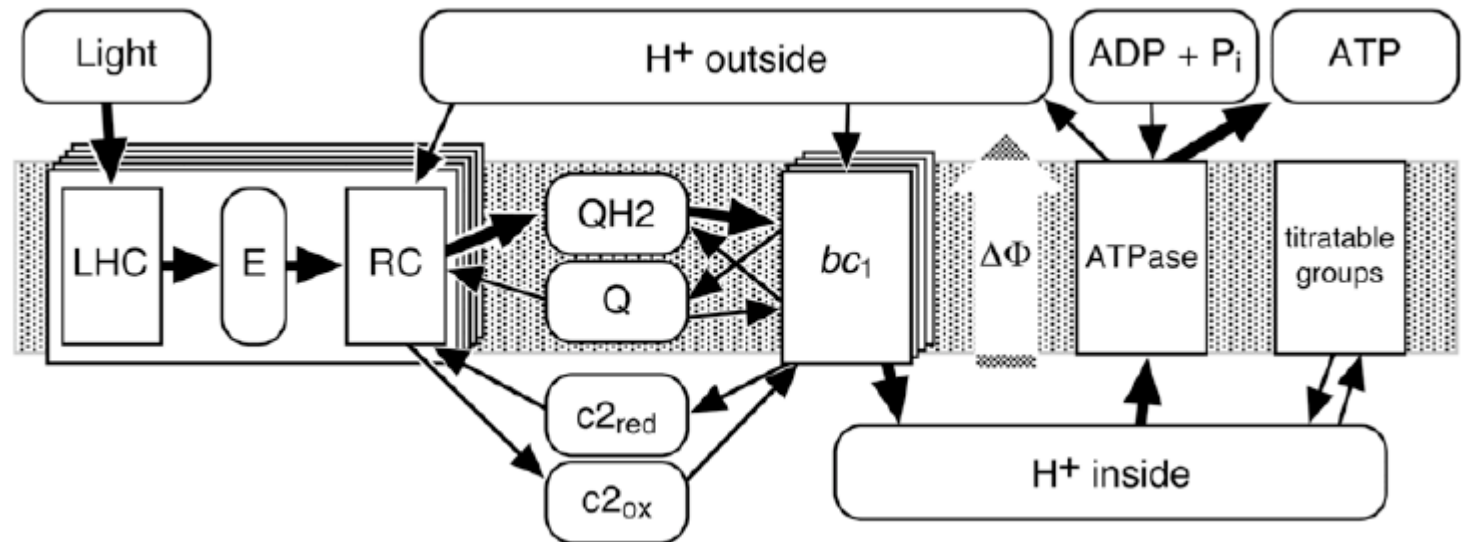
$e^-H^+$  electron–proton pairs stored on the quinols

$e^-$  for the electrons on the cytochrome  $c_2$

$pH$  : transmembrane proton gradient

$H^+$  : protons outside of the vesicle (broken outline of the respective reservoir).

# Stochastic dynamics simulations: Molecules & Pools model



Round edges: **pools** for metabolite molecules

Rectangles: protein machines are modeled explicitly as multiple copies

fixed set of parameters

integrate rate equations with stochastic algorithm



# Stochastic simulations of cellular signalling

**Traditional computational approach** to chemical/biochemical kinetics:

- (a) start with a set of coupled **ODEs** (reaction rate equations) that describe the time-dependent concentration of chemical species,
- (b) use some **integrator** to calculate the concentrations as a function of time given the rate constants and a set of initial concentrations.

Successful **applications** : studies of yeast cell cycle, metabolic engineering, whole-cell scale models of metabolic pathways (E-cell), ...

**Major problem**: cellular processes occur in very small volumes and frequently involve **very small number of molecules**.

E.g. in gene expression processes a few TF molecules may interact with a single gene regulatory region.

*E.coli* cells contain on average only 10 molecules of Lac repressor.

## Include stochastic effects

(Consequence1) → modeling of reactions as continuous fluxes of matter is no longer correct.

(Consequence2) Significant **stochastic fluctuations** occur.

To study the stochastic effects in biochemical reactions, stochastic formulations of chemical kinetics and Monte Carlo computer simulations have been used.

**Daniel Gillespie** (J Comput Phys 22, 403 (1976); J Chem Phys 81, 2340 (1977)) introduced the exact **Dynamic Monte Carlo (DMC)** method that connects the traditional chemical kinetics and stochastic approaches.

# Basic outline of the direct method of Gillespie

**(Step i)** generate a list of the components/species and define the initial distribution at time  $t = 0$ .

**(Step ii)** generate a list of possible events  $E_j$  (chemical reactions as well as physical processes).

**(Step iii)** using the current component/species distribution, prepare a probability table  $P(E_j)$  of all the events that can take place.

Compute the total probability

$$P_{tot} = \sum P(E_j)$$

$P(E_j)$  : probability of event  $E_j$  .

**(Step iv)** Pick two random numbers  $r_1$  and  $r_2 \in [0...1]$  to decide which event  $E_\mu$  will occur next and the amount of time  $\tau$  after which  $E_\mu$  will occur.

# Basic outline of the direct method of Gillespie

Using the random number  $r_1$  and the probability table, the event  $E_\mu$  is determined by finding the event that satisfies the relation

$$\sum_{i=1}^{\mu-1} P(E_i) < r_1 P_{tot} \leq \sum_{i=1}^{\mu} P(E_i)$$

The second random number  $r_2$  is used to obtain the amount of time  $\tau$  between the reactions

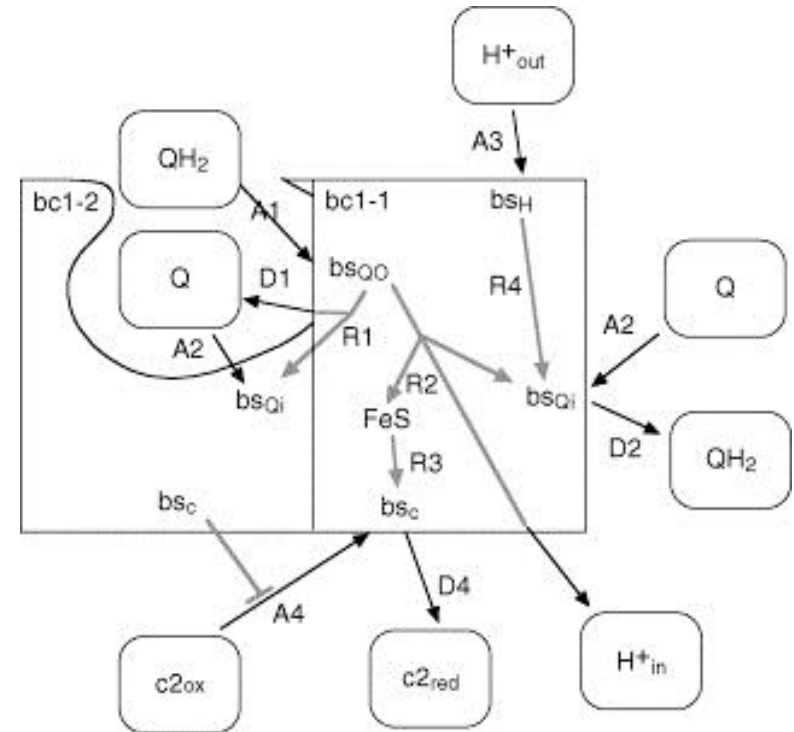
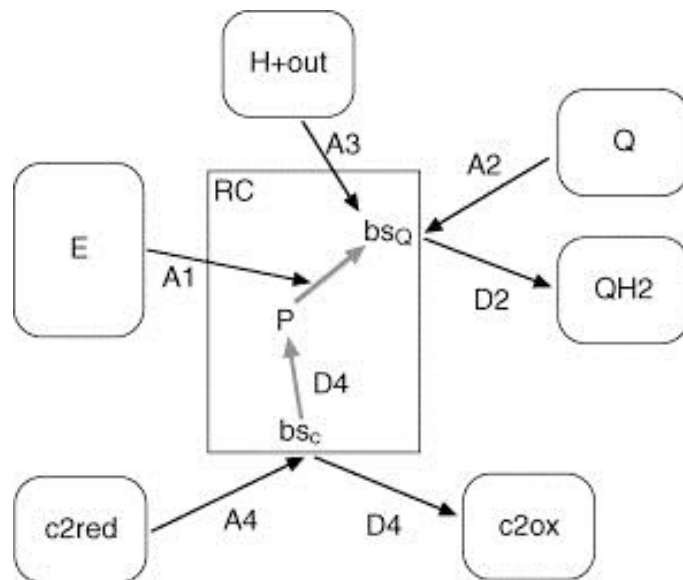
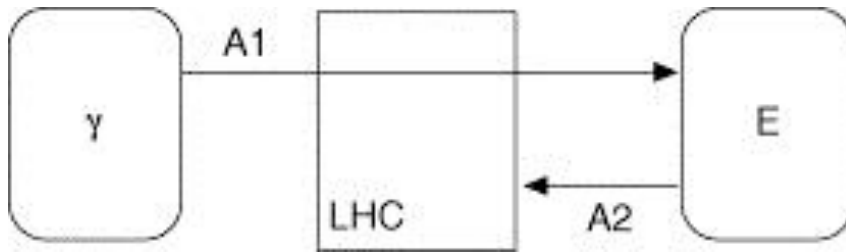
$$\tau = -\frac{1}{P_{tot}} \ln(r_2)$$

As the total probability of the events changes in time, the time step between occurring steps varies.

Steps (iii) and (iv) are repeated at each step of the simulation.

The necessary number of runs depends on the inherent noise of the system and on the desired statistical accuracy.

# reactions included in stochastic model of chromatophore



# Stochastic simulations of a complete vesicle

Model vesicle: 12 LH1/RC-monomers  
1-6  $bc_1$  complexes  
1 ATPase

120 quinones  
20 cytochrome  $c_2$

integrate rate equations with:

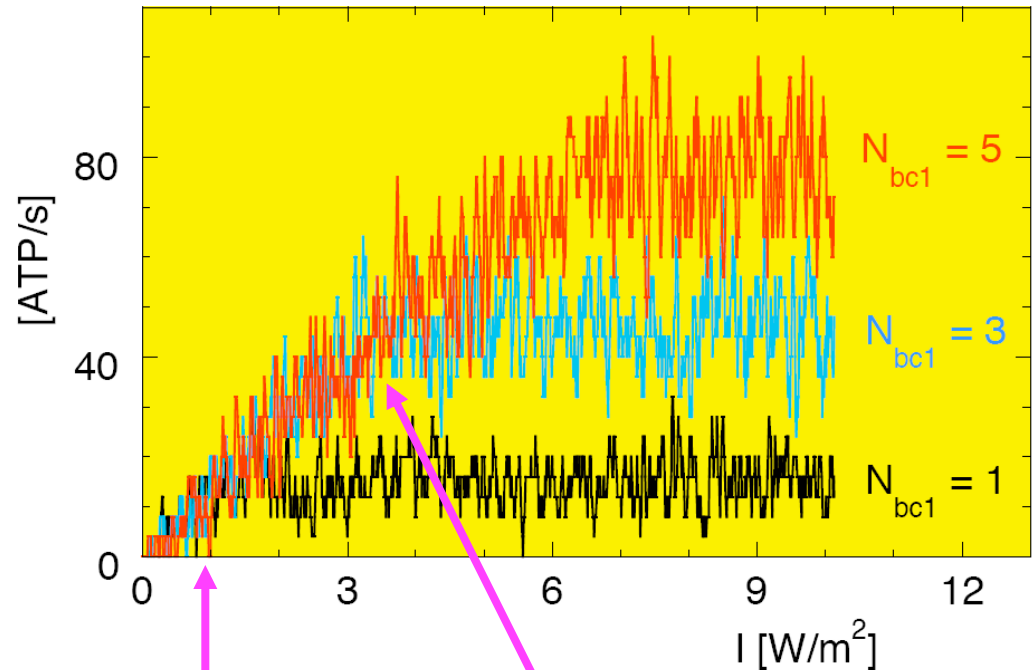
- Gillespie algorithm (associations)
- Timer algorithm (reactions); 1 random number determines when reaction occurs

simulating 1 minute real time requires 1.5 minute on one opteron 2.4 GHz proc

# simulate increase of light intensity (sunrise)

during 1 minute,  
light intensity is slowly  
increased from 0 to 10 W/m<sup>2</sup>  
(quasi steady state)

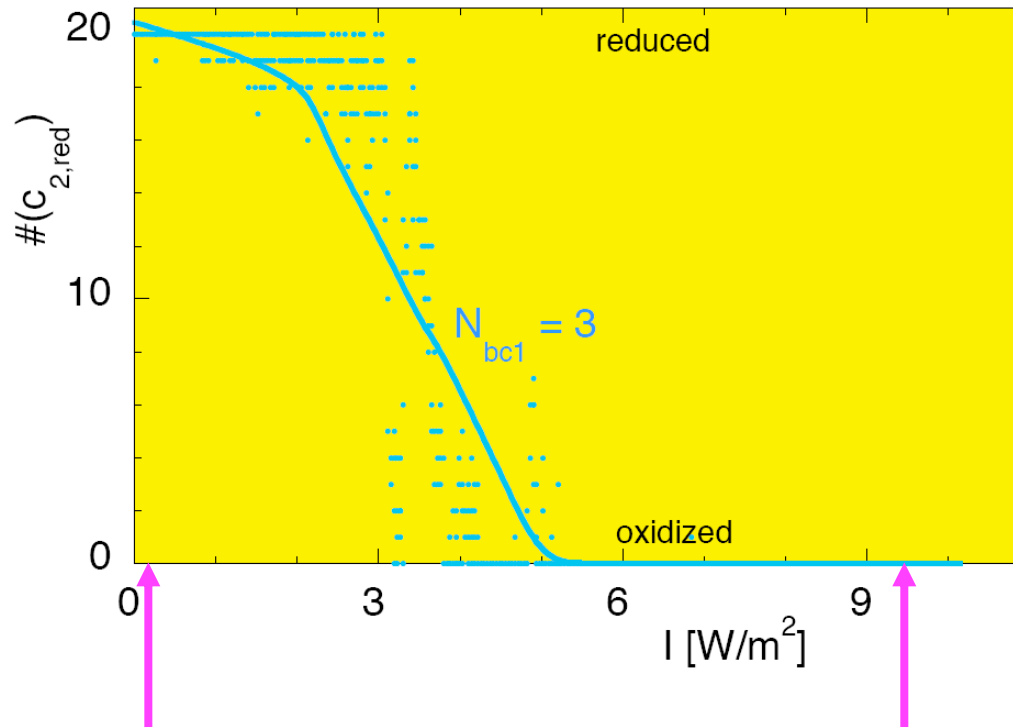
- there are two regimes
- one limited by available light
  - one limited by bc<sub>1</sub> throughput



**low light intensity:**  
linear increase of  
ATP production  
with light intensity

**high light intensity:**  
saturation is reached  
the later the higher the  
number of bc<sub>1</sub> complexes

# oxidation state of cytochrome $c_2$ pool

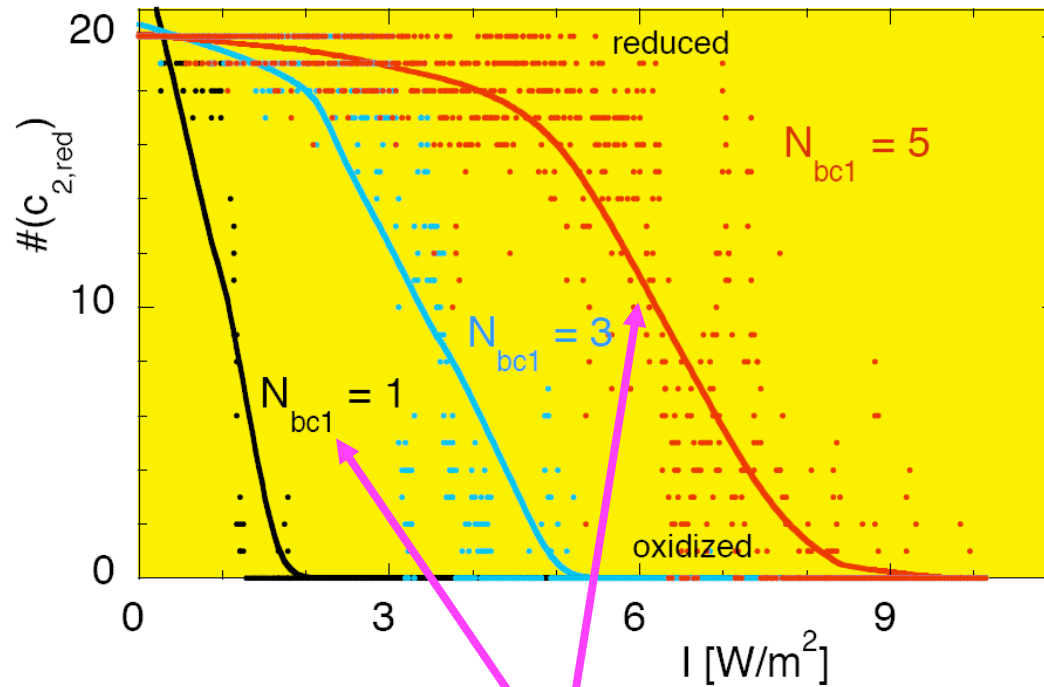


**low light intensity:**  
all 20 cytochrome  $c_2$   
are reduced by  $bc_1$

**high light intensity**  
RCs are faster than  $bc_1$ ,  
 $c_2$ s wait for electrons



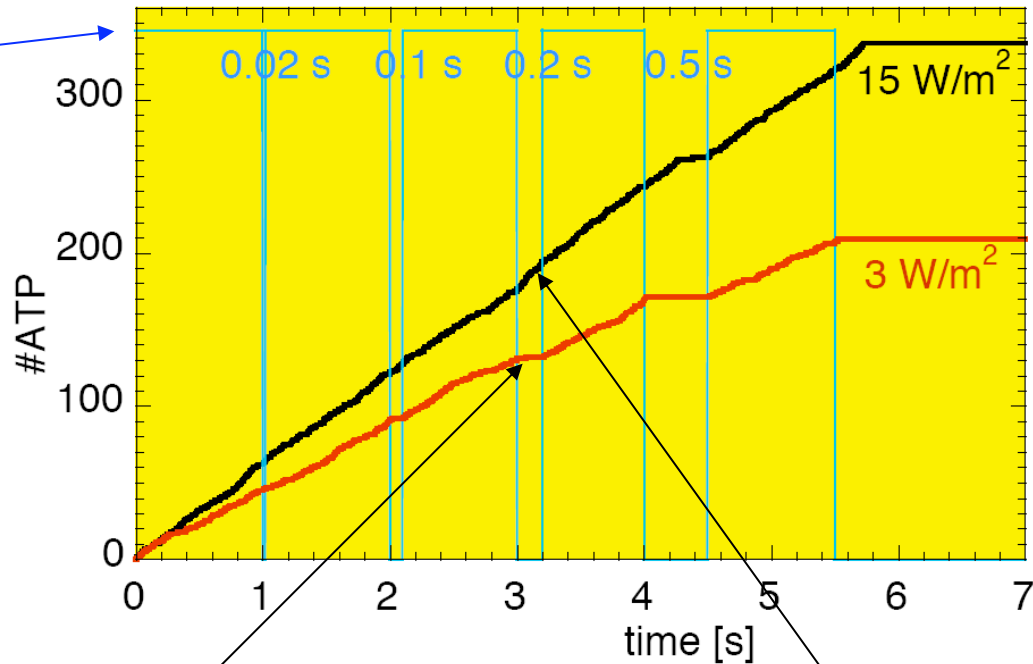
# oxidation state of cytochrome $c_2$ pool



more  $bc_1$  complexes  
can load more  
cytochrome  $c_2$ s

# total number of produced ATP

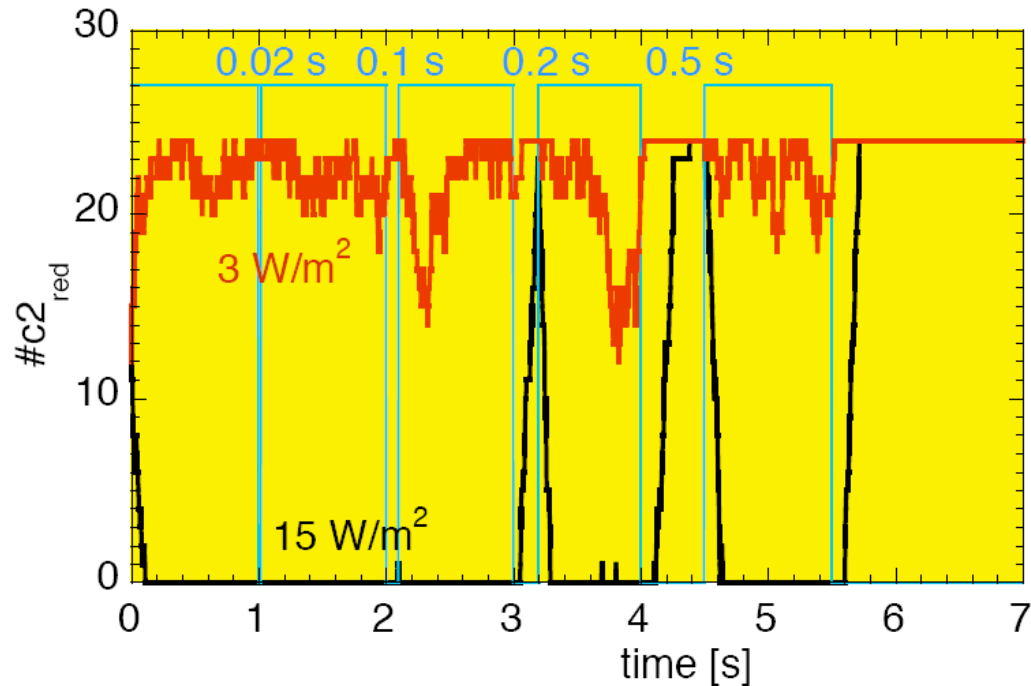
blue line:  
illumination



low light intensity: any interruption stops ATP production

high light intensity: interruptions are buffered up to 0.3 s duration

## c<sub>2</sub> pool acts as buffer



At high light intensity, c<sub>2</sub> pool is mainly oxidized.

If light is turned off, bc<sub>1</sub> can continue to work (load c<sub>2</sub>s, pump protons, let ATPase produce ATP) until c<sub>2</sub> pool is fully reduced.

# What if parameters are/were unknown ?

## Bridging the Gap: Linking Molecular Simulations and Systemic Descriptions of Cellular Compartments

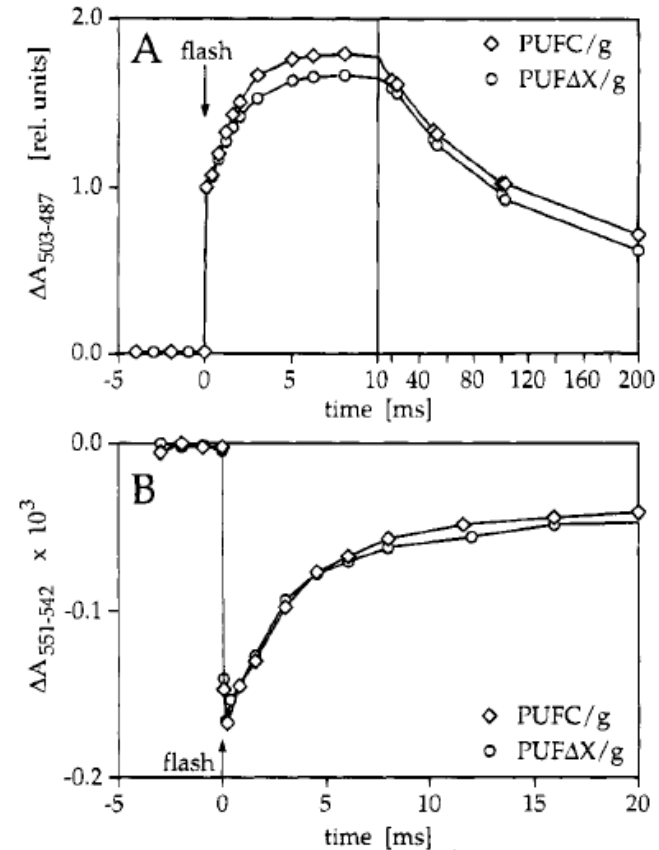
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PLoS ONE (2010)

choose 25 out of 45 system parameters for optimization.

take 7 different non-equilibrium time-resolved experiments from Dieter Oesterhelt lab (MPI Martinsried).



Biochemistry 1995, 34, 15235–15247

15235

Role of PufX Protein in Photosynthetic Growth of *Rhodospirillum rubrum*.

1. PufX Is Required for Efficient Light-Driven Electron Transfer and Photophosphorylation under Anaerobic Conditions<sup>†</sup>

Wolfgang P. Barz,<sup>‡§</sup> Francesco Francia,<sup>||</sup> Giovanni Venturoli,<sup>||</sup> B. Andrea Melandri,<sup>||</sup> André Verméglio,<sup>†</sup> and Dieter Oesterhelt<sup>\*,‡</sup>

## Parameters not optimized

Parameter	Value	Description
$bc_1::k_{on}(H^+_{out})$	$10^{10} \text{ nm}^3 \text{ s}^{-1}$	rate for proton uptake from the cytoplasm by $bc_1$
$bc_1::k_{tr}(e:Q_o \Rightarrow FeS)$	$2.3 \cdot 10^3 \text{ s}^{-1}$	rate for electron transfer from $Q_o$ to FeS
$bc_1::k_{tr}(e:c_1 \Rightarrow c_2)$	$10^5 \text{ s}^{-1}$	electron transfer rate from $c_1$ to bound cytochrome $c_2$
$bc_1::k_{tr}(e:Q_o \Rightarrow b_L)$	$10^4 \text{ s}^{-1}$	electron transfer from $Q_o$ to $b_L$ heme
$bc_1::k_{tr}(e:b_L \Rightarrow b_H)$	$10^4 \text{ s}^{-1}$	electron transfer from $b_L$ to $b_H$ heme
$\Delta\Phi::V$	$2.65 \cdot 10^4 \text{ nm}^3$	inner volume of the vesicle
$\Delta\Phi::A$	$5.28 \cdot 10^3 \text{ nm}^2$	membrane area (Q pool „volume“)
$\Delta\Phi::C_{H_{in}}$	1.0 e	effective charge of a free proton in the vesicle
$\Delta\Phi::C_{H_{m}}$	1.0 e	effective charge of a proton on the titratable groups
$\Delta\Phi::C_{prot}$	-1.0 e	effective charge of an $e^-$ translocated through an RC
$\Delta\Phi::C_{cred}$	-0.5 e	effective charge of a reduced cytochrome $c_2$
$\Delta\Phi::C_{cox}$	0.5 e	effective charge of an oxidized cytochrome $c_2$
PR:: $N_p$	80	number of titratable groups in the vesicle
PR::pK	5.0	pK of the titratable groups
$N_{core}$	10	number of dimeric core complexes (2 RC + 1 LHC)
$N_{bc_1}$	10	number of cytochrome $bc_1$ complexes
$N_{ATPase}$	1	number of ATPases
$N_{c_2}$	20	total number of cytochrome $c_2$
$N_Q$	200	total number of quinones

**Table S1: Model Parameters Not Included in the Optimization Process**

# Parameter optimization through evolutionary algorithm

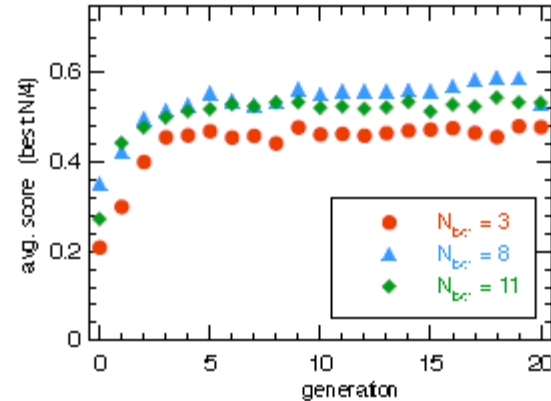
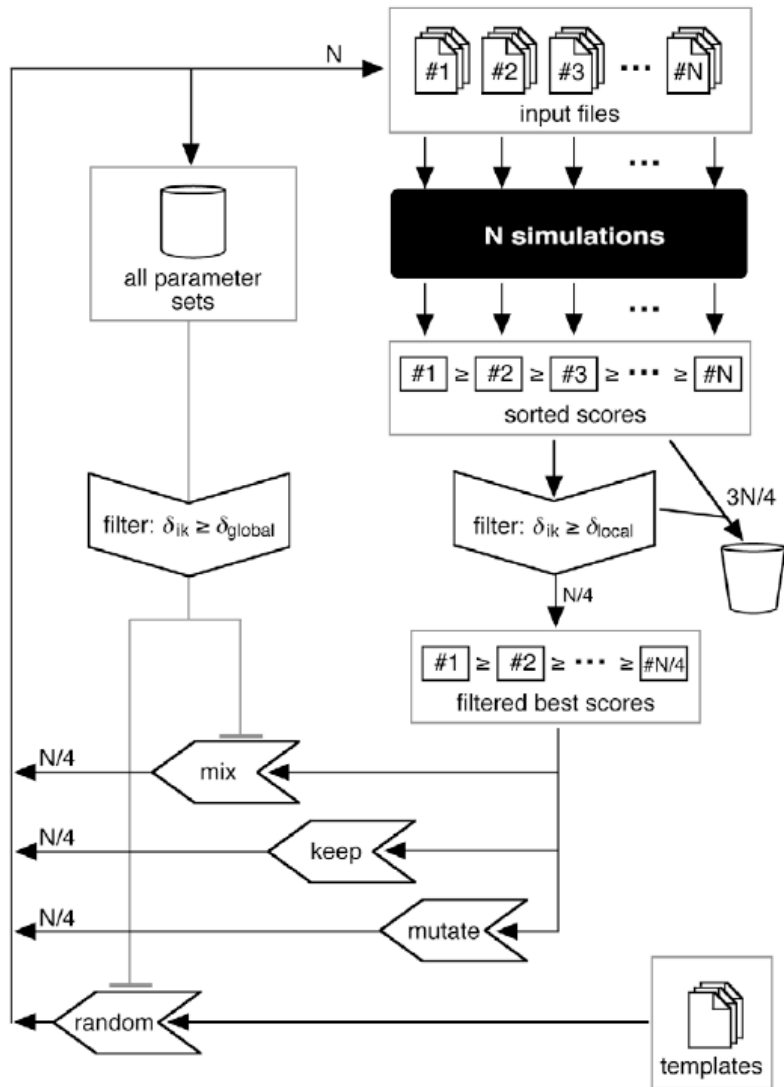


Figure S1: Determining the Number of bc1 Complexes: Evolution of the Master Score

# 25 optimization parameters

Analyze 1000 best  
parameter sets among  
32.800 simulations:

$$\langle P \rangle = \exp[\langle \log P \rangle]$$

$$\sigma^2 = \langle (\log P - \langle \log P \rangle)^2 \rangle$$

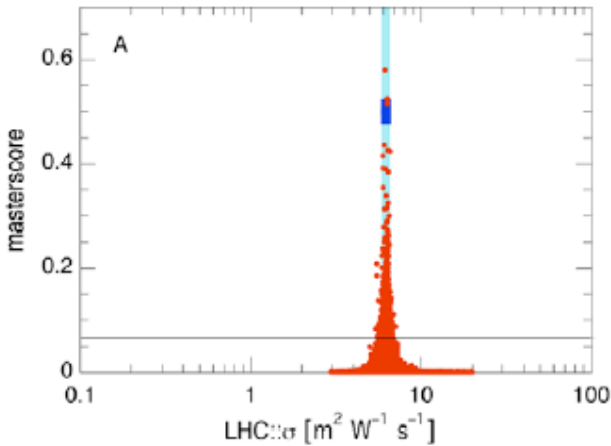
$$P_{\min} = \exp[\langle \log P \rangle - \sigma]$$

$$P_{\max} = \exp[\langle \log P \rangle + \sigma]$$

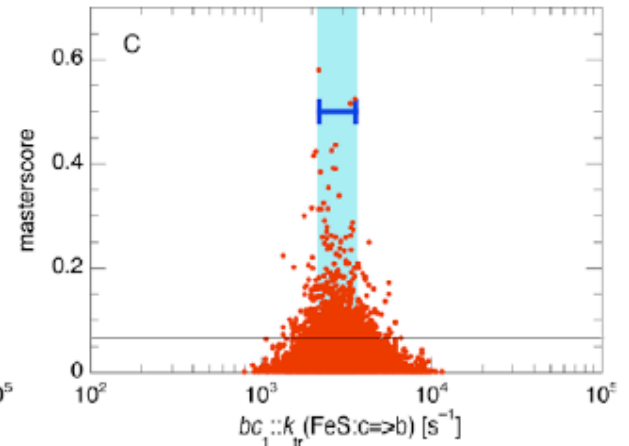
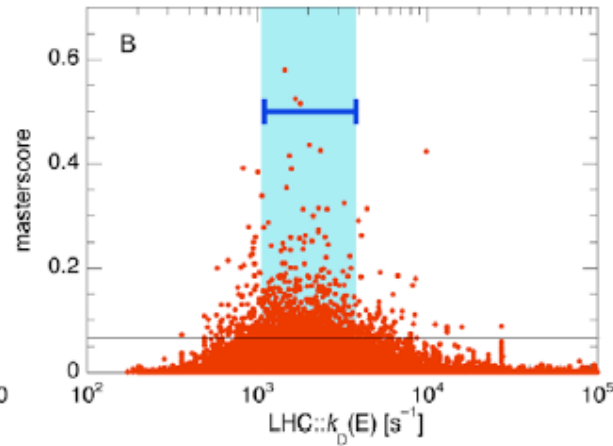
parameter	units	$\langle P \rangle$	$P_{\min} \dots P_{\max}$	$\frac{P_{\min}}{P_{\max}}$
LHC:: $\sigma$	$\text{m}^2 \text{W}^{-1} \text{s}^{-1}$	6.22	6.02...6.42	0.94
LHC:: $N_0$	1	1.31	0.81 ... 2.13	0.38
LHC:: $k_D(E)$	$\text{s}^{-1}$	$1.9 \times 10^3$	$(1.1 \dots 3.8) \times 10^3$	0.29
RC:: $k_{on}(E)$	$\text{s}^{-1}$	$24 \times 10^6$	$(1.2 \dots 4.5) \times 10^6$	0.27
RC:: $k_{on}(H^+)$	$\text{nm}^3 \text{s}^{-1}$	$1.4 \times 10^8$	$(1.3 \dots 1.6) \times 10^8$	0.81
RC:: $k_{on}(Q)$	$\text{nm}^2 \text{s}^{-1}$	$6.0 \times 10^4$	$(4.4 \dots 8.1) \times 10^4$	0.54
RC:: $k_{off}(QH2)$	$\text{s}^{-1}$	87	70...108	0.65
RC:: $k_{on}(c2red)$	$\text{nm}^3 \text{s}^{-1}$	$9.2 \times 10^5$	$(7.3 \dots 11.5) \times 10^5$	0.63
RC:: $k_{off}(c2ox)$	$\text{s}^{-1}$	$2.2 \times 10^3$	$(1.6 \dots 3.0) \times 10^3$	0.53
bc1:: $k_{on}(QH2@Q_0)$	$\text{nm}^2 \text{s}^{-1}$	$1.2 \times 10^4$	$(0.79 \dots 1.7) \times 10^4$	0.46
bc1:: $k_{off}(Q@Q_0)$	$\text{s}^{-1}$	28.3	26.3...30.4	0.86
bc1:: $k_{r1}(Q:Q_0 - > Q)$	$\text{s}^{-1}$	$4.9 \times 10^3$	$(3.6 \dots 6.7) \times 10^3$	0.54
bc1:: $k_{on}(Q@Q_i)$	$\text{mm}^2 \text{s}^{-1}$	$6.7 \times 10^5$	$(4.5 \dots 10) \times 10^5$	0.45
bc1:: $k_{off}(QH2@Q_i)$	$\text{s}^{-1}$	86	68...110	0.62
bc1:: $k_{r1}(QH2:Q_i - > Q_0)$	$\text{s}^{-1}$	$3.8 \times 10^3$	$(2.6 \dots 5.5) \times 10^3$	0.47
bc1:: $k_{on}(c2ox)$	$\text{nm}^3 \text{s}^{-1}$	$94 \times 10^6$	$(6.3 \dots 14) \times 10^6$	0.47
bc1:: $k_{off}(c2red)$	$\text{s}^{-1}$	$6.0 \times 10^3$	$(3.3 \dots 11) \times 10^3$	0.30
bc1:: $k_{off}(H+@Q_0)$	$\text{s}^{-1}$	$24 \times 10^4$	$(1.3 \dots 4.3) \times 10^4$	0.30
bc1:: $k_{r1}(FeS:b - > c)$	$\text{s}^{-1}$	$3.9 \times 10^3$	$(3.1 \dots 5.1) \times 10^3$	0.61
bc1:: $k_{r1}(FeS:c - > b)$	$\text{s}^{-1}$	$2.8 \times 10^3$	$(2.2 \dots 3.6) \times 10^3$	0.61
bc1:: $k_{r1}(e:b_H - > Q_i)$	$\text{s}^{-1}$	$7.7 \times 10^3$	$(5.0 \dots 12) \times 10^3$	0.42
bc1:: $\Phi_0$	mV	102	83...114	0.73
$\Delta\Phi::U_0$	mV/e	10.3	9.5...11	0.85
$\Delta\Phi::\Delta\Phi_0$	mV/pH	10	7.6...13.7	0.55
PR::pK	1	4.84	3.9...5.9	0.66

# Sensitivity of master score

Decay rate of excitons  
in LHC



Absorption cross section  
light harvesting complex



Kinetic rate for hinge  
motion of FeS domain in  
bc1 complex

Some parameters are very sensitive, others not.



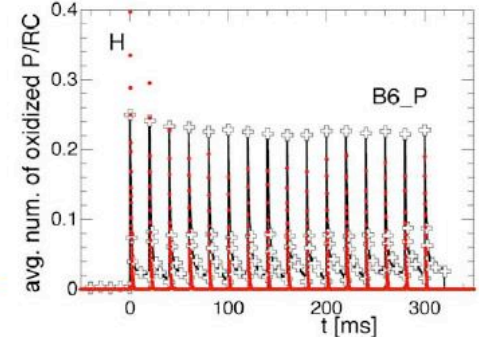
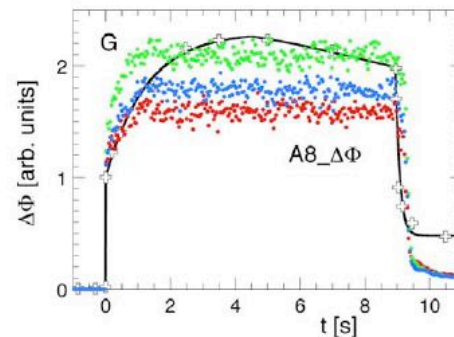
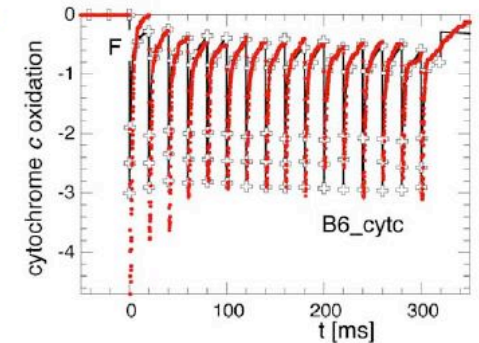
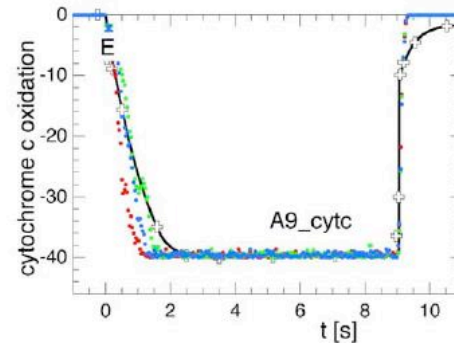
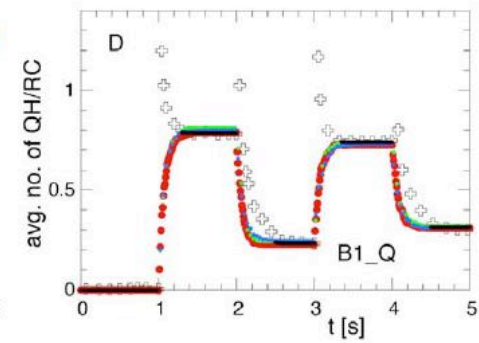
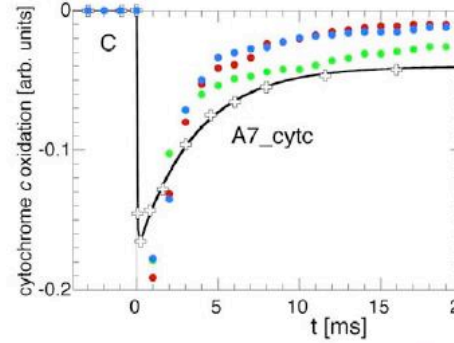
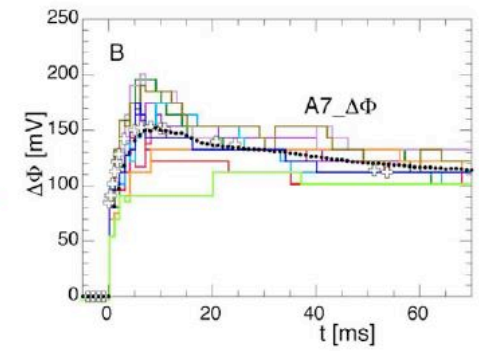
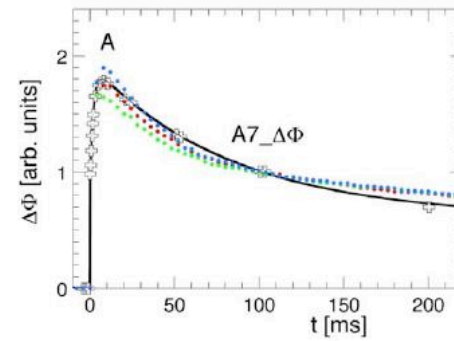
# Three best-scored parameter sets

Score of individual parameter set  $i$  for matching one experiment:

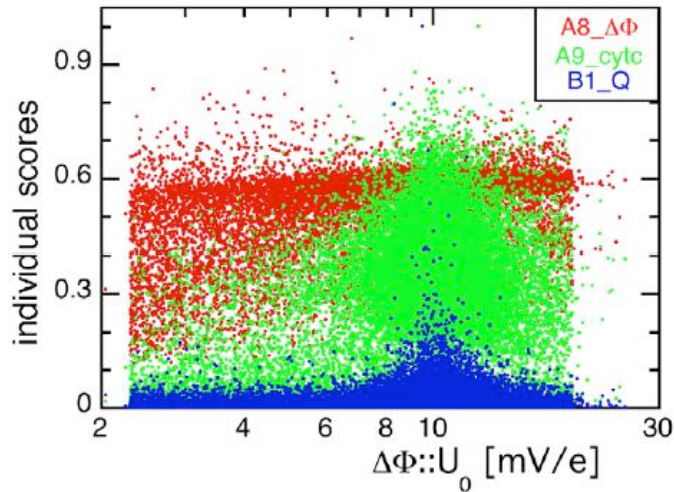
$$s_i = \frac{C_i}{\sum (x(t_i) - f(t_i))^2}$$

$x(t_i)$ : simulation result  
 $f(t_i)$ : smooth fit of exp. data

**Master score** for one parameter set: defined as product of the individual scores  $s_i$



# Different experiments yield different sensitivity



“importance score”:  
Sum of the sensitivities  
 $P_{min} / P_{max}$  of all relevant  
parameters

**Table 2.** Importance scores and correlation coefficients between the master score and the respective individual scores of the experimental scenarios denoting the relative importance of each of the experiments for the parameter value optimization.

experiment	A7 cytc	A7 ΔΦ	A8 ΔΦ	A9 cytc	B1 Q	B6 P	B6 cytc	BC1
importance score	4.4	7.7	5.8	9.7	3.8	5.2	8.9	5.5
correlation	0.09	0.44	0.22	0.38	0.83	0.17	0.31	0.41

The importance scores are determined as the sums of the sensitivities of all relevant parameters against the individual scores (see table S2 for all the individual values). The correlation coefficients are obtained from a linear fit of the master score against the respective individual score.

Analysis could suggest new  
experiments that would be  
most informative!

# Summary

Only 1/3 of the kinetic parameters previously known.

Stochastic parameter optimization converges robustly into the same parameter basin as known from experiment.

Two large-scale runs (15 + 17 parameters) yielded practically the same results.

If implemented as grid search, less than 2 points per dimension.

It appears enough to know 1/3 – 1/2 of kinetic rates about a system to be able to describe it quantitatively (IF connectivities are known).