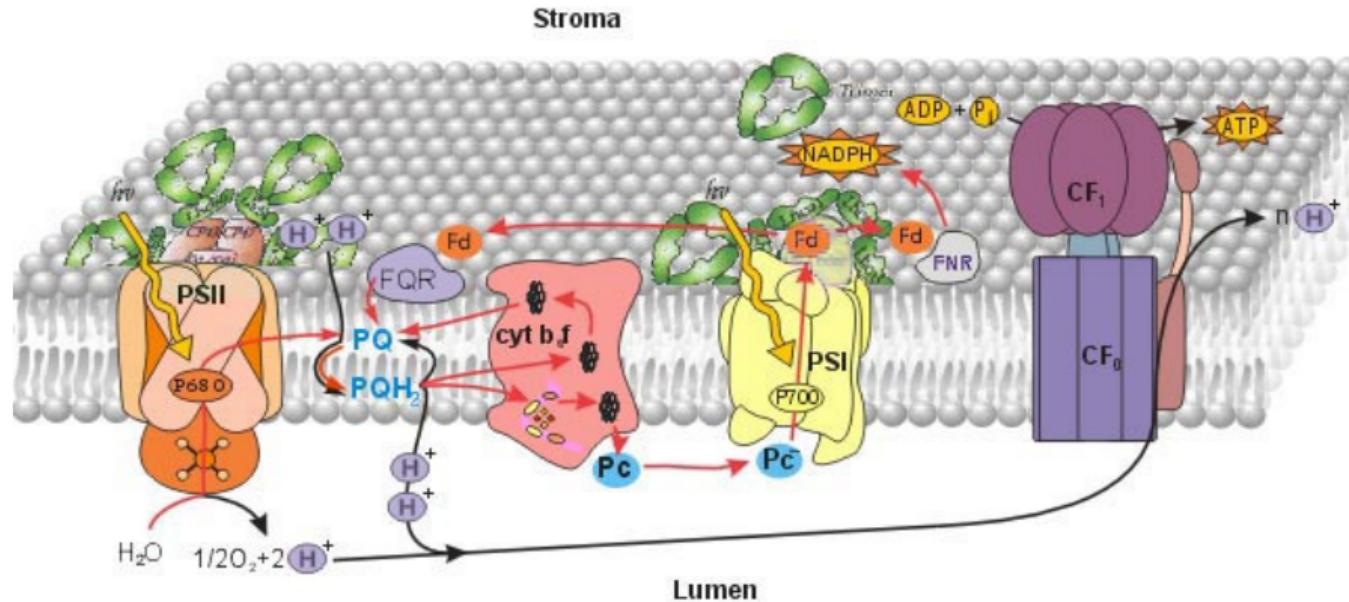


Энерго-преобразующие мембранны



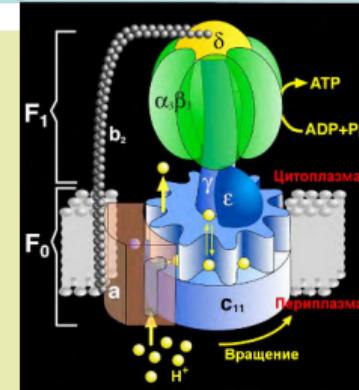
Модели первичных процессов фотосинтеза

Нано-электростанции в живой клетке

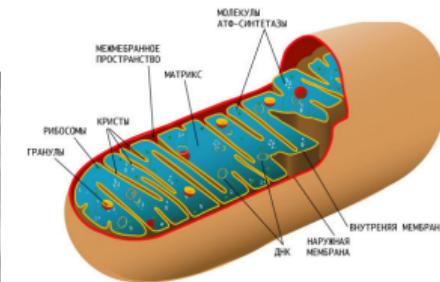
- Дыхание
- Митохондрии
- Производство энергии

АТФ из энергии солнечного света

- Хлоропласти (зеленые растения и водоросли)
и хроматофоры (бактерии)

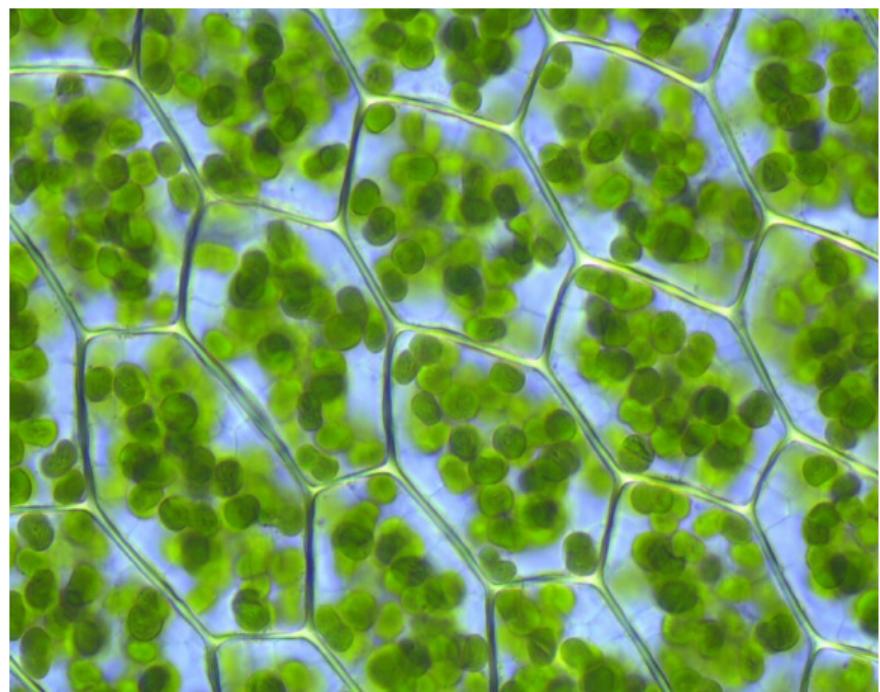
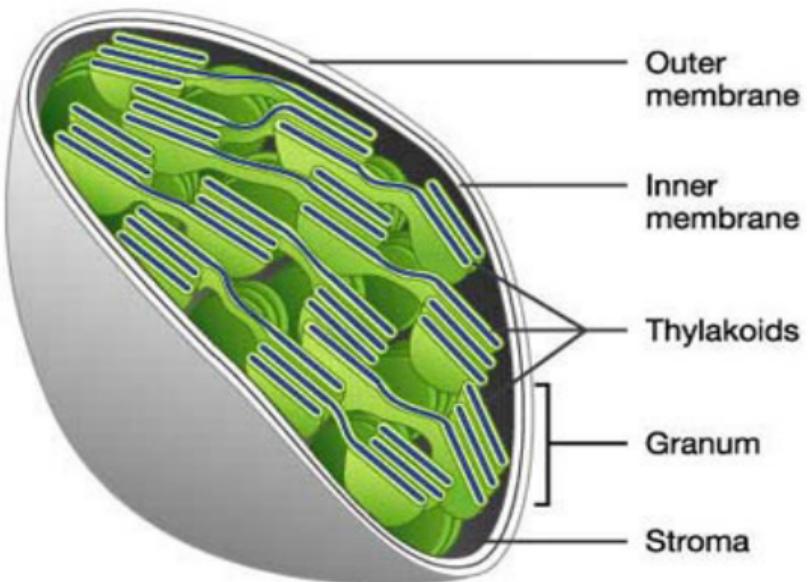


Производство энергии осуществляется в субклеточных системах



Митохондрия

Хлоропласт.

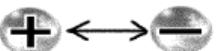


1. Поглощение света



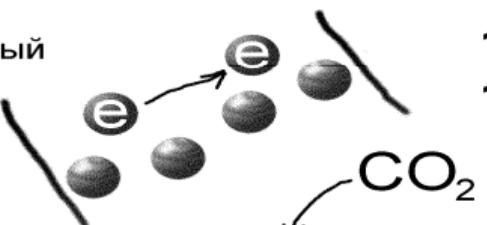
10^{-15} с

2. Разделение зарядов в реакционном центре



10^{-12} с

3. Электронный транспорт



10^{-10} –
 10^{-2} с

4. Фиксация углерода (цикл Кальвина)

секунды-
минуты

5. Транспорт веществ в растении

минуты-
часы

6. Рост растения

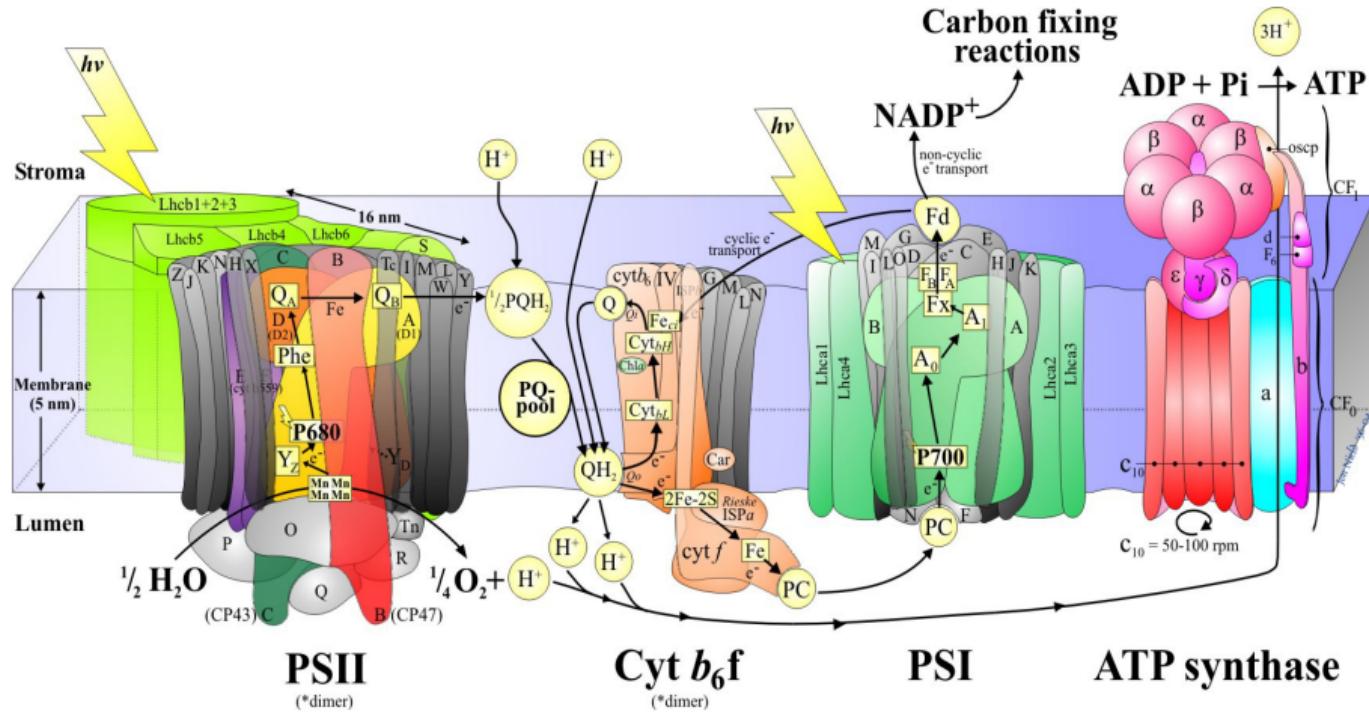
дни



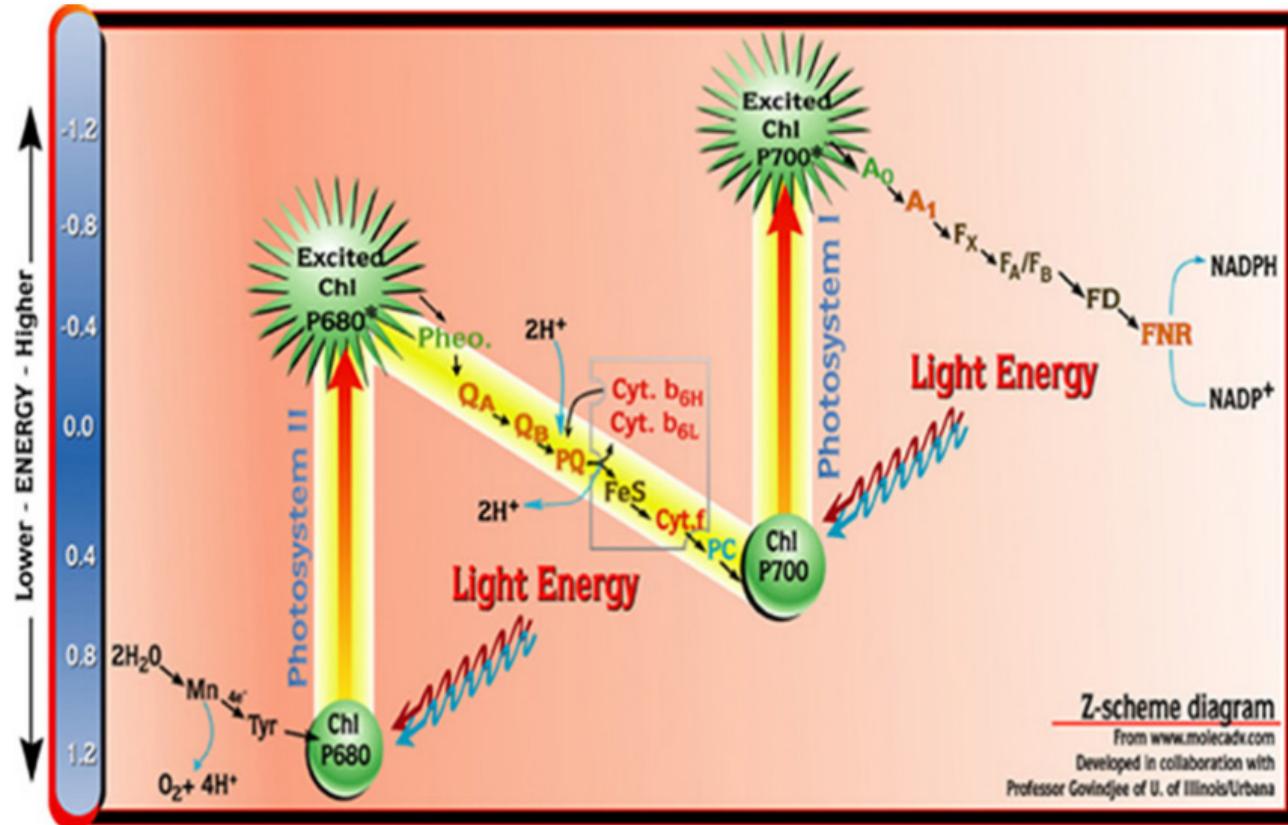
ФОТОСИНТЕЗ

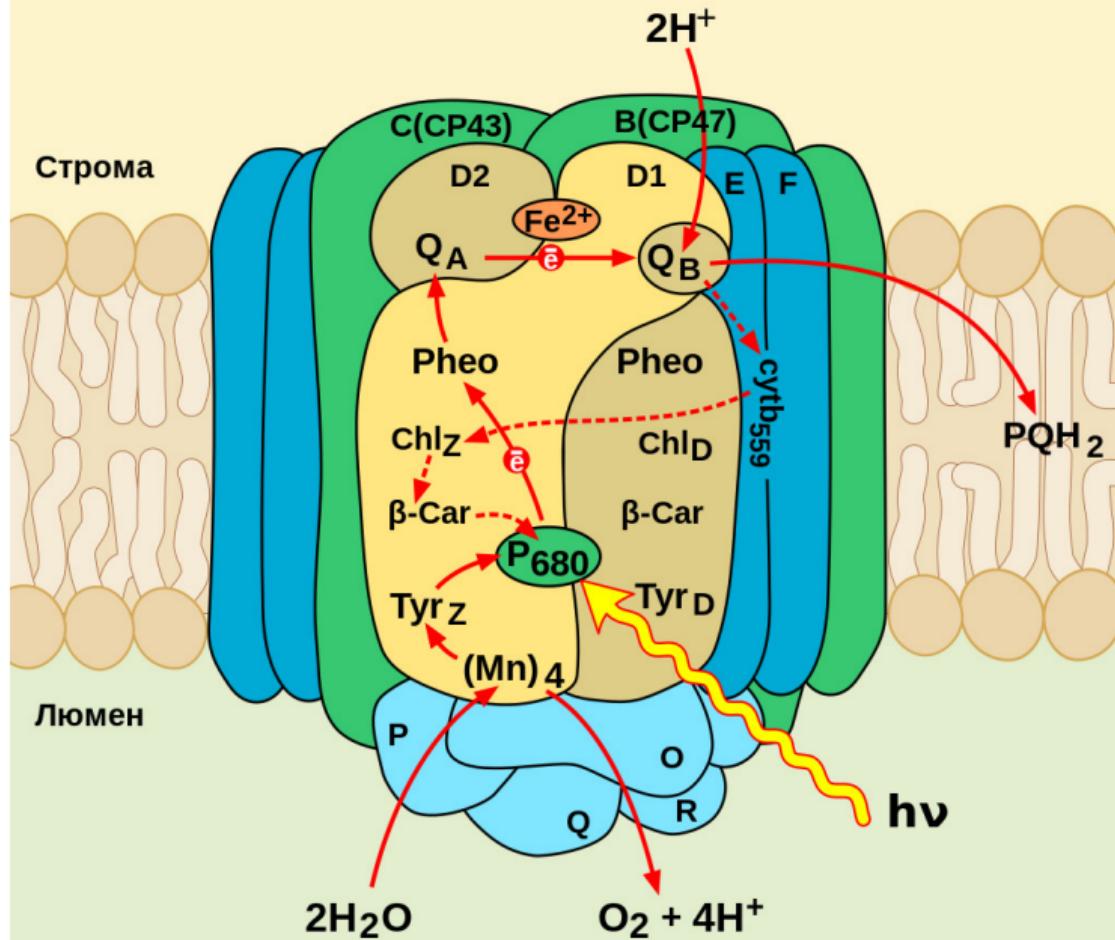
Иерархия
фотосинте-
тических
процессов

Структура мультиферментных комплексов

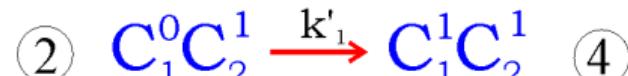
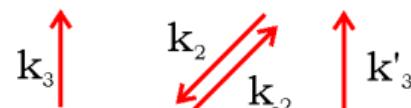
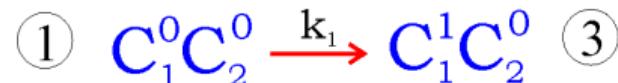


Z- схема фотосинтеза





Описание состояний комплекса из двух компонентов C_1C_2



уравнения для вероятностей состояний

$$\frac{dp_i}{dt} = \sum_{j=1}^l (p_j k_{ji} - p_i k_{ij}),$$

Начальные условия

$$p_i(0) = b_i, \quad i=1, \dots, l.$$

$$\dot{p}_1 = k_3 p_2 - k_1 p_1$$

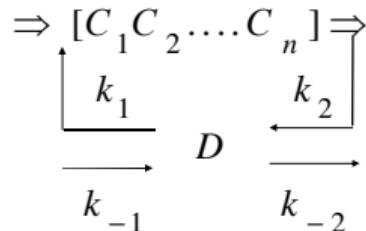
$$\dot{p}_2 = k_2 p_3 - (k'_1 + k_3 + k_{-2}) p_2$$

$$\dot{p}_3 = k_1 p_1 + k'_3 p_4 + k_{-2} p_2 - k_2 p_3$$

$$\dot{p}_4 = k'_1 p_2 - k'_3 p_4$$

$$p_1 + p_2 + p_3 + p_4 = 1$$

Взаимодействие комплекса с мобильным переносчиком D



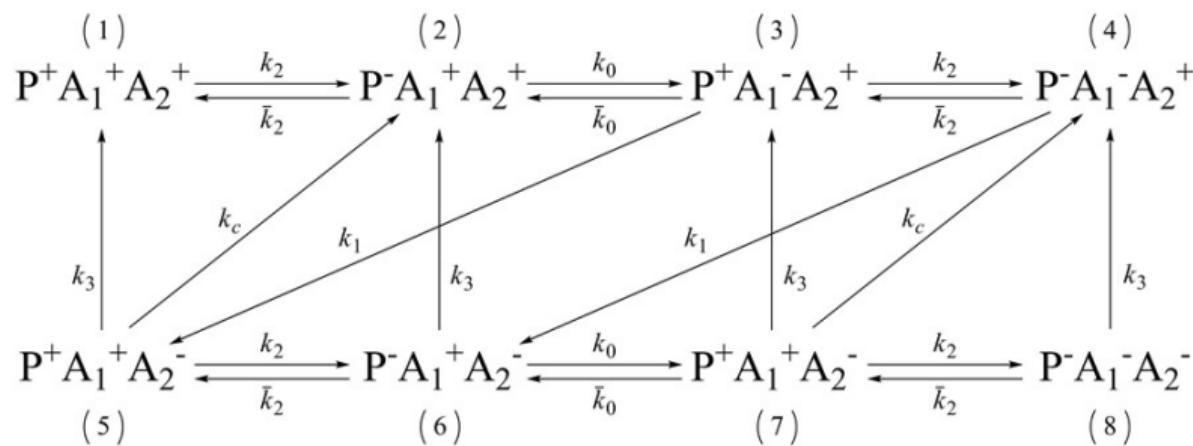
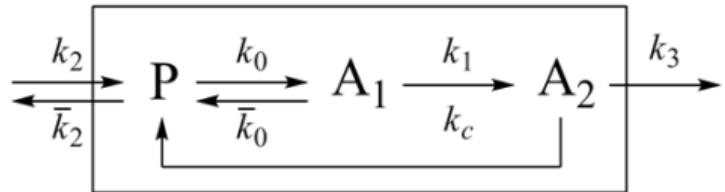
$$\frac{d[D^-]}{dt} = k_2[C_n^-][D^+] - k_{-2}[D^-][C_n^+] - k_1[D^-][C_1^+] + k_{-1}[C_1^-][D^+]$$

$[D^+]$, $[D^-]$ - концентрации мобильного переносчика в окисленной и восстановленной форме;

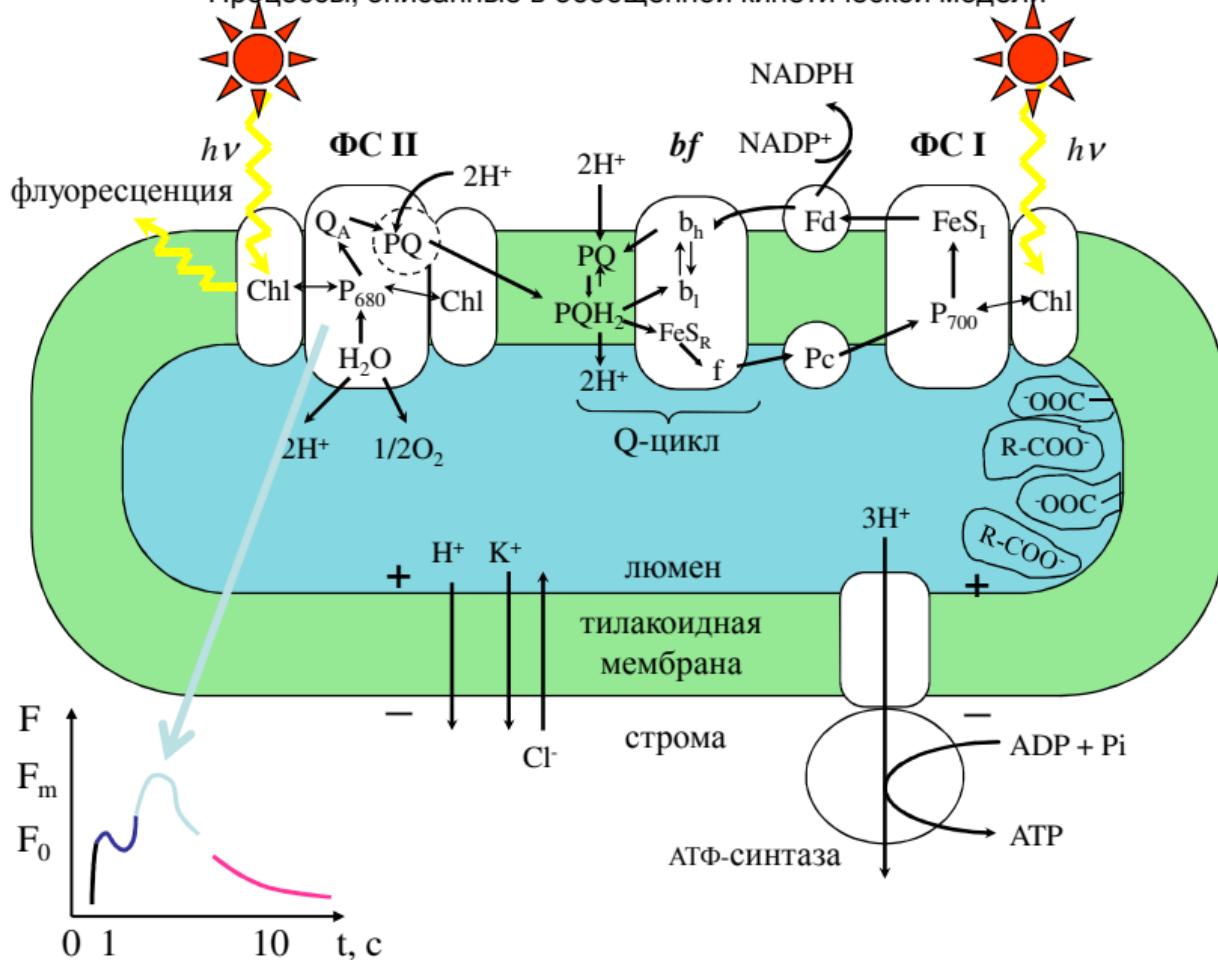
$[C_1^+]$, $[C_1^-]$, $[C_n^+]$, $[C_n^-]$ - концентрации компонентов комплекса

k_i - бимолекулярные константы скоростей.

Комплекс из трех переносчиков



Процессы, описанные в обобщенной кинетической модели



Кинетические уравнения для вероятностей состояний ФСII имеют вид:

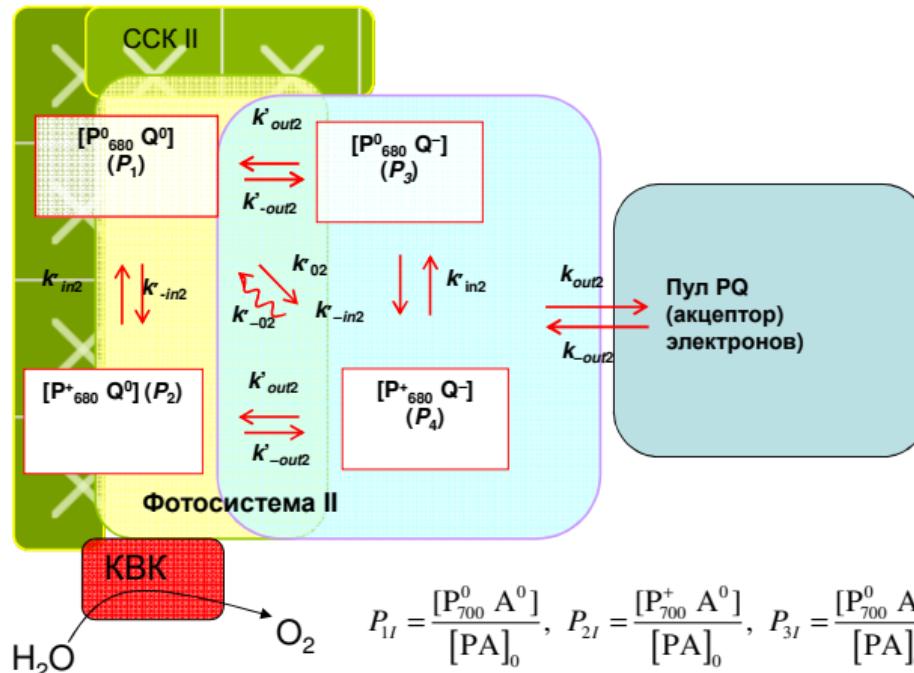
$$\frac{dP_{1II}}{dt} = -(k'_{-out2} + k'_{-in2} + k'_{02})P_{1II} + k'_{in2}P_{1II} + k'_{out2}P_{3II} + k'_{-02}P_{4II},$$

$$\frac{dP_{2II}}{dt} = k'_{-in2} \cdot P_{1II} - (k'_{in2} + k'_{-out2})P_{2II} + k'_{out2}P_{4II},$$

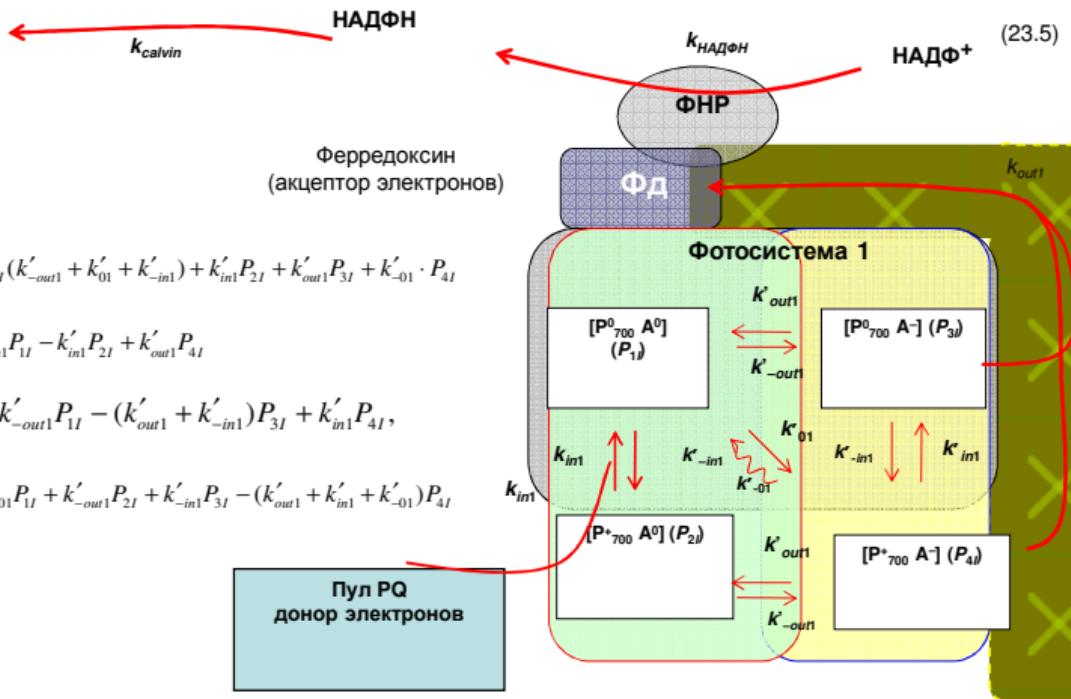
$$\frac{dP_{3II}}{dt} = k'_{out2}P_{1II} - (k'_{out2} + k'_{-in2})P_{3II} + k'_{in2}P_{4II}$$

$$\frac{dP_{4II}}{dt} = k'_{02}P_{1II} + k'_{-out2}P_{2II} + k'_{-in2} \cdot P_{3II} - (k'_{in2} + k'_{-02} + k'_{out2})P_{4II}.$$

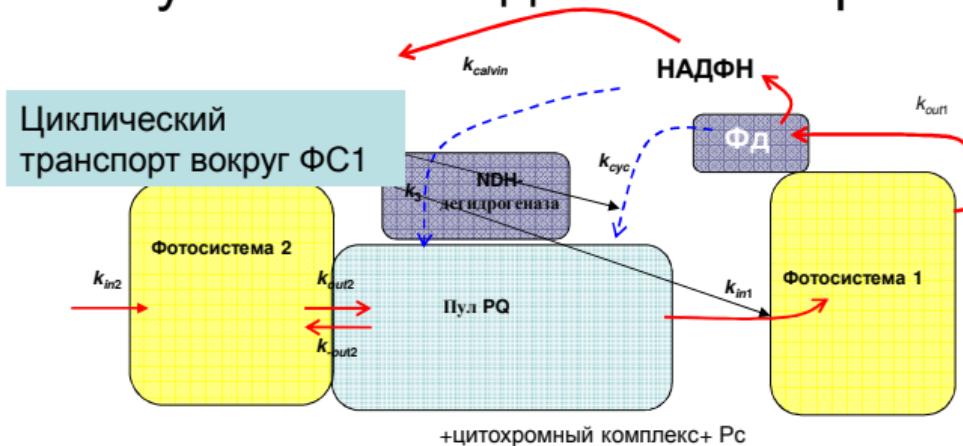
Фотосистема 2



Фотосистема 1



Взаимодействие двух фотосистем с участием подвижных переносчиков



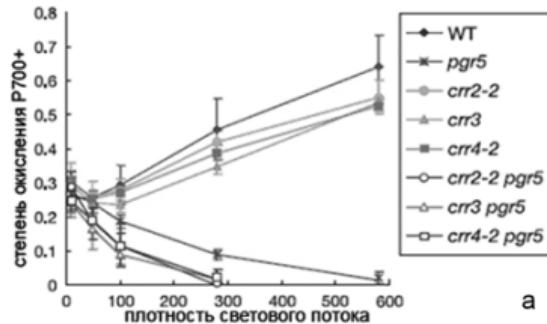
$$\frac{d[PQ^-]}{dt} = k_{out2} \cdot (P_{3II} + P_{4II}) \cdot [P_{680}] \cdot [PQ] + k_3 \cdot [\text{НАДФН}] \cdot [PQ] + k_{cyc} \cdot [\Phi_D^-] [PQ^-] - \\ - (k_{in1} \cdot (P_{1I} + P_{3I}) \cdot [P_{700}] + k_{out2} \cdot (P_{1II} + P_{2II}) \cdot [P_{680}]) [PQ^-],$$

$$\frac{d[\Phi_D^-]}{dt} = k_{out1} (P_{3I} + P_{4I}) [P_{700}] \cdot [\Phi_D] + k_{-HADFH} [\text{НАДФН}] \cdot [\Phi_D] - \\ - (k_{HADFH} [\text{НАДФ}^+] + k_{cyc} [PQ] + k_{out1} (P_{1I} + P_{2I}) [P_{700}]) [\Phi_D^-],$$

$$\frac{d[\text{НАДФН}]}{dt} = k_{HADFH} [\Phi_D^-] [\text{НАДФ}^+] - (k_{-HADFH} [\Phi_D] + k_3 [PQ] - k_{calvin}) [\text{НАДФН}].$$

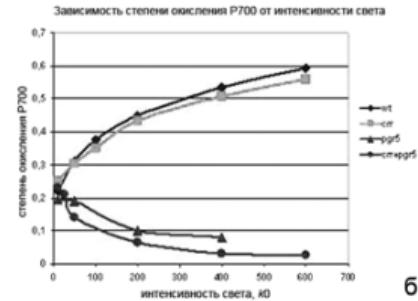
Изучение влияния мутаций

Зависимость степени окисления P700 в зависимости от интенсивности освещения у дикого типа и мутантов *Arabidopsis Thaliana*



эксперимент

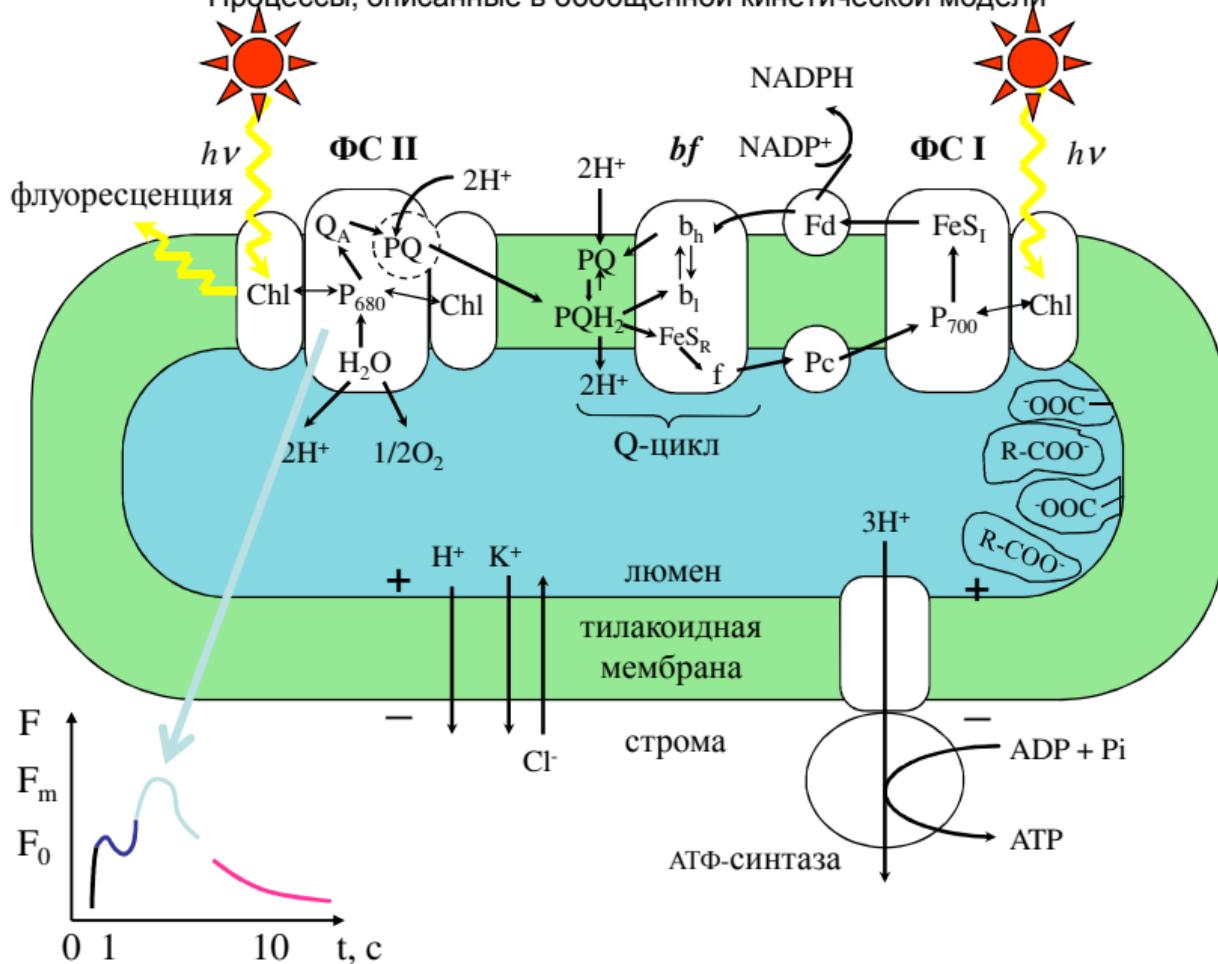
PGR proton gradient regulation



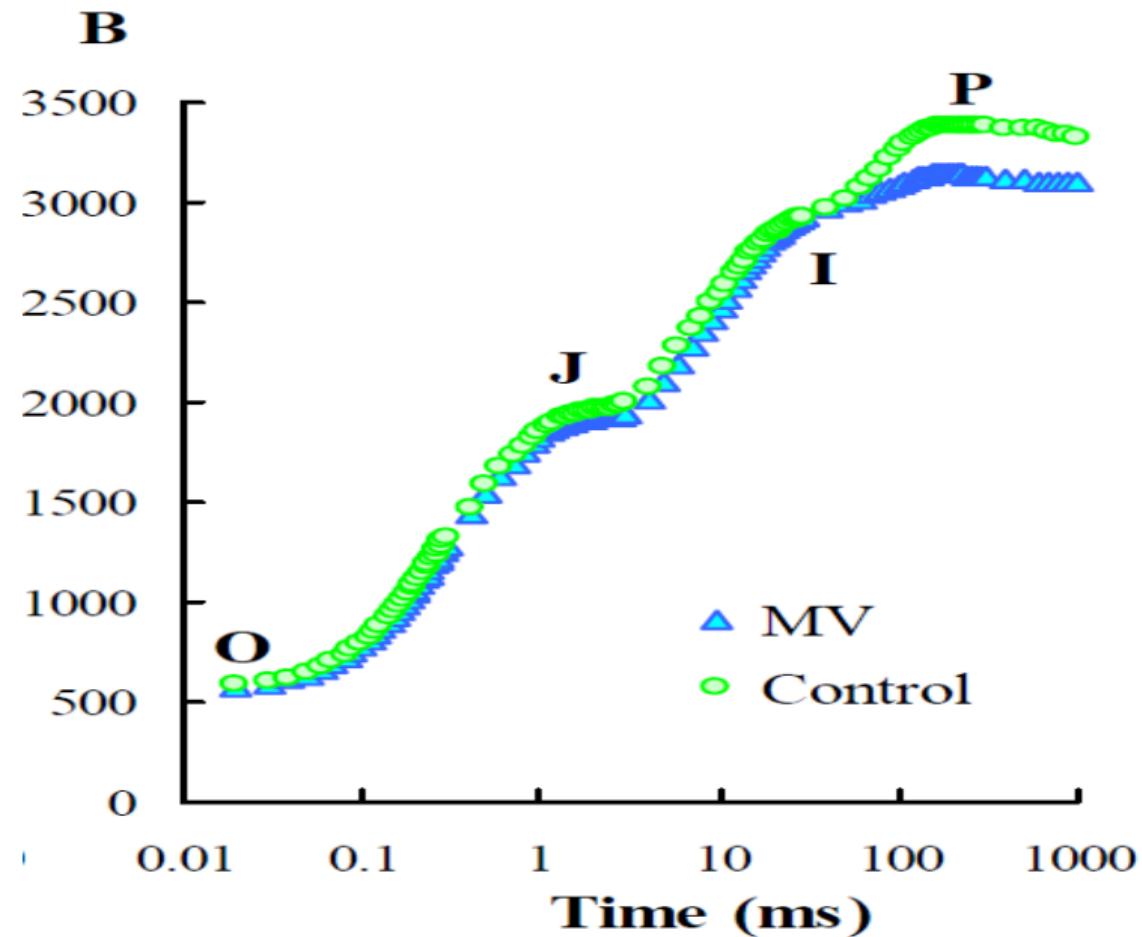
модель

у *crr*-мутантов подавлен только *NDH*-зависимый электронный транспорт, а у *pgr5*- мутантов подавлен как циклический Fd-зависимый электронный транспорт, так и электронный поток в акцепторной части ФСI.

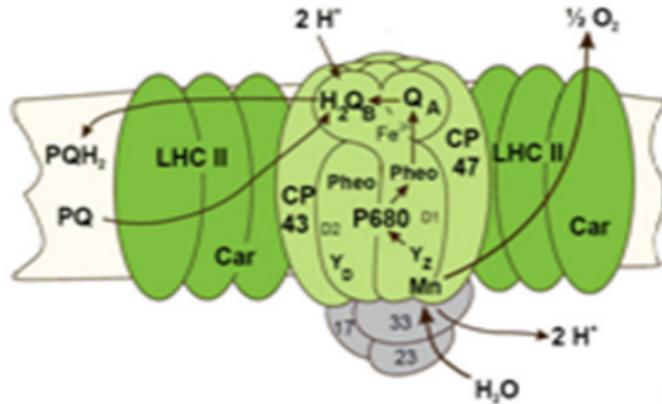
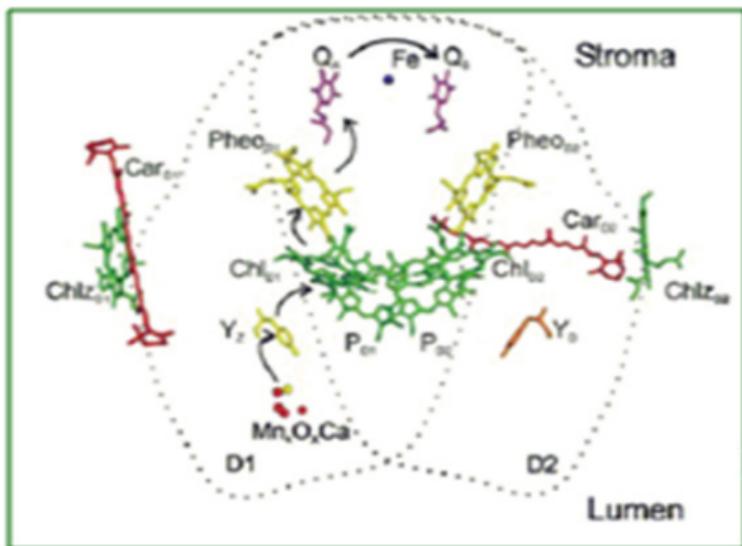
Процессы, описанные в обобщенной кинетической модели



Кинетическая
кривая
нарастающего
участка
индукции
флуоресценции

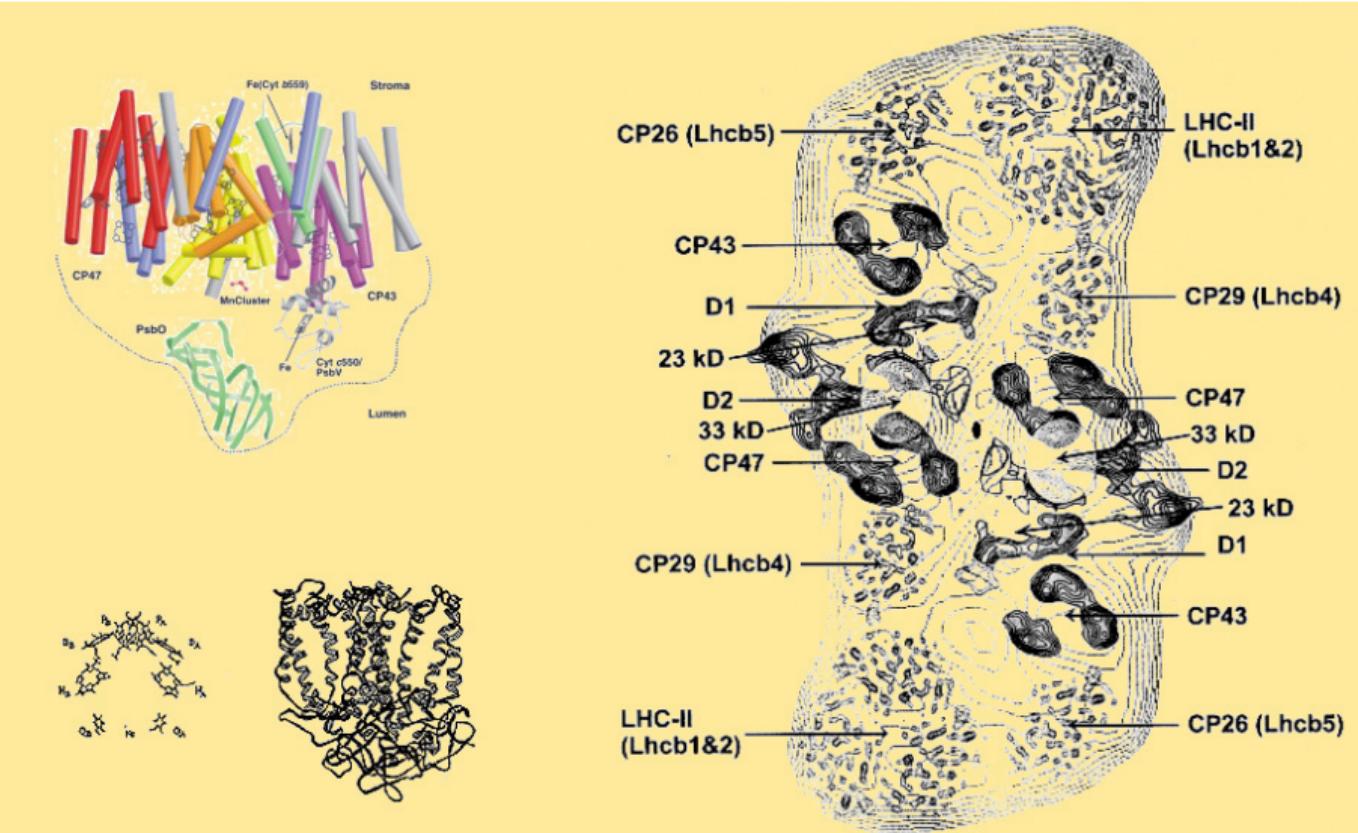


Photosystem II – the source of fluorescence

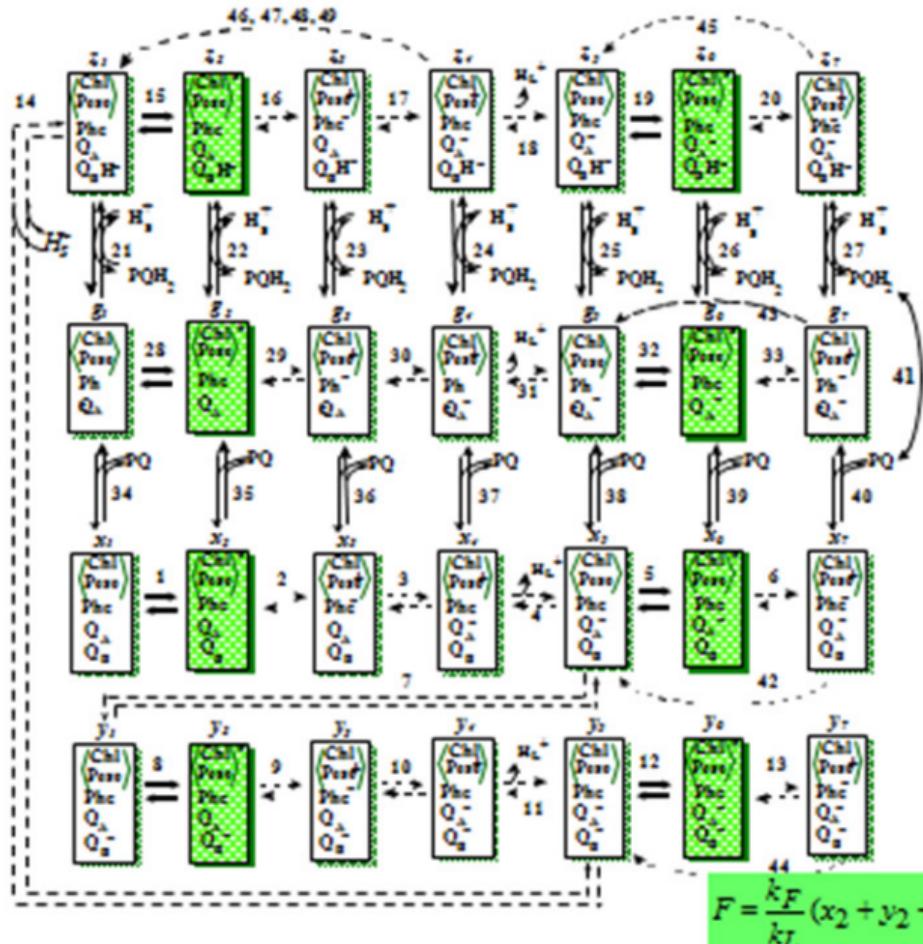


Chl, PSII chlorophyll, P680 - photoactive pigments; Phe, pheophytin; Q_A and Q_B, primary and secondary quinone acceptors; PQ, plastoquinone; PQH₂, plastoquinol; H_L⁺ and H_s⁺ protons in lumen and stroma,

Комплекс Фотосистемы 2. Подробности.

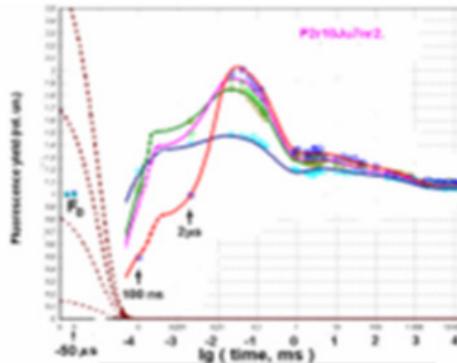
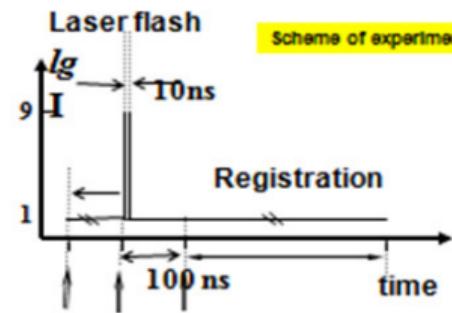
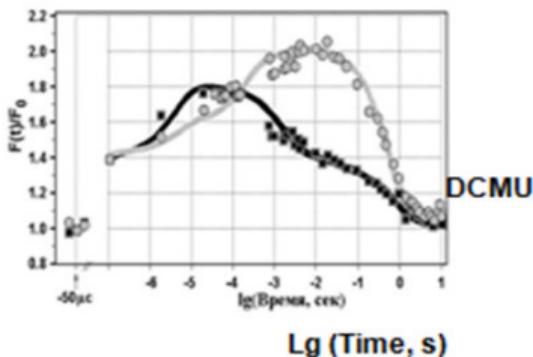


Scheme of PSII states



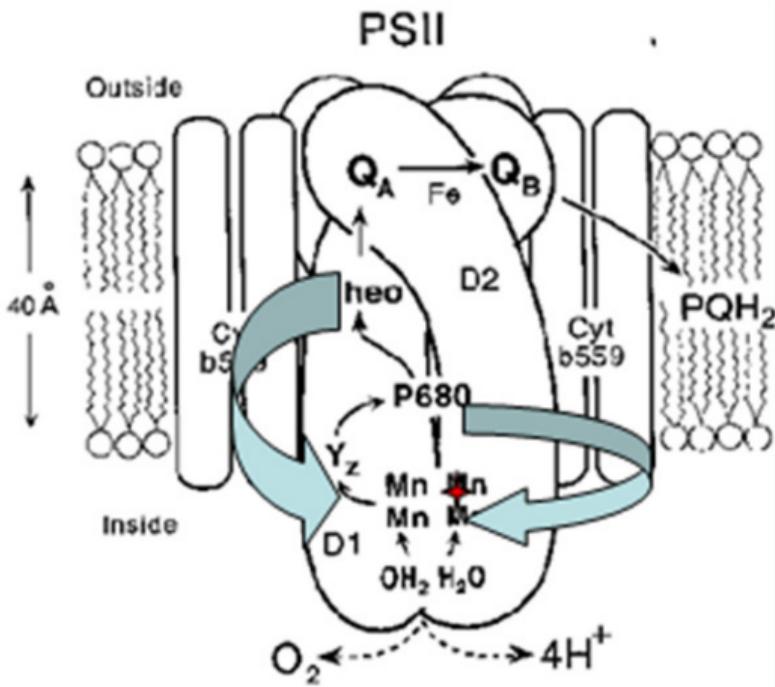
Моделирование отклика системы на короткую вспышку

Experiment (dots) and simulation (solid lines). Fluorescence induction curves after the saturating 10 ns laser flash, cells of thermophilic *Chlorella pyrenoidosa* Chick.
lab. Prof. G.Renger (Berlin)



Belyaeva, Renger et al., Phot.Res.
2008-2016

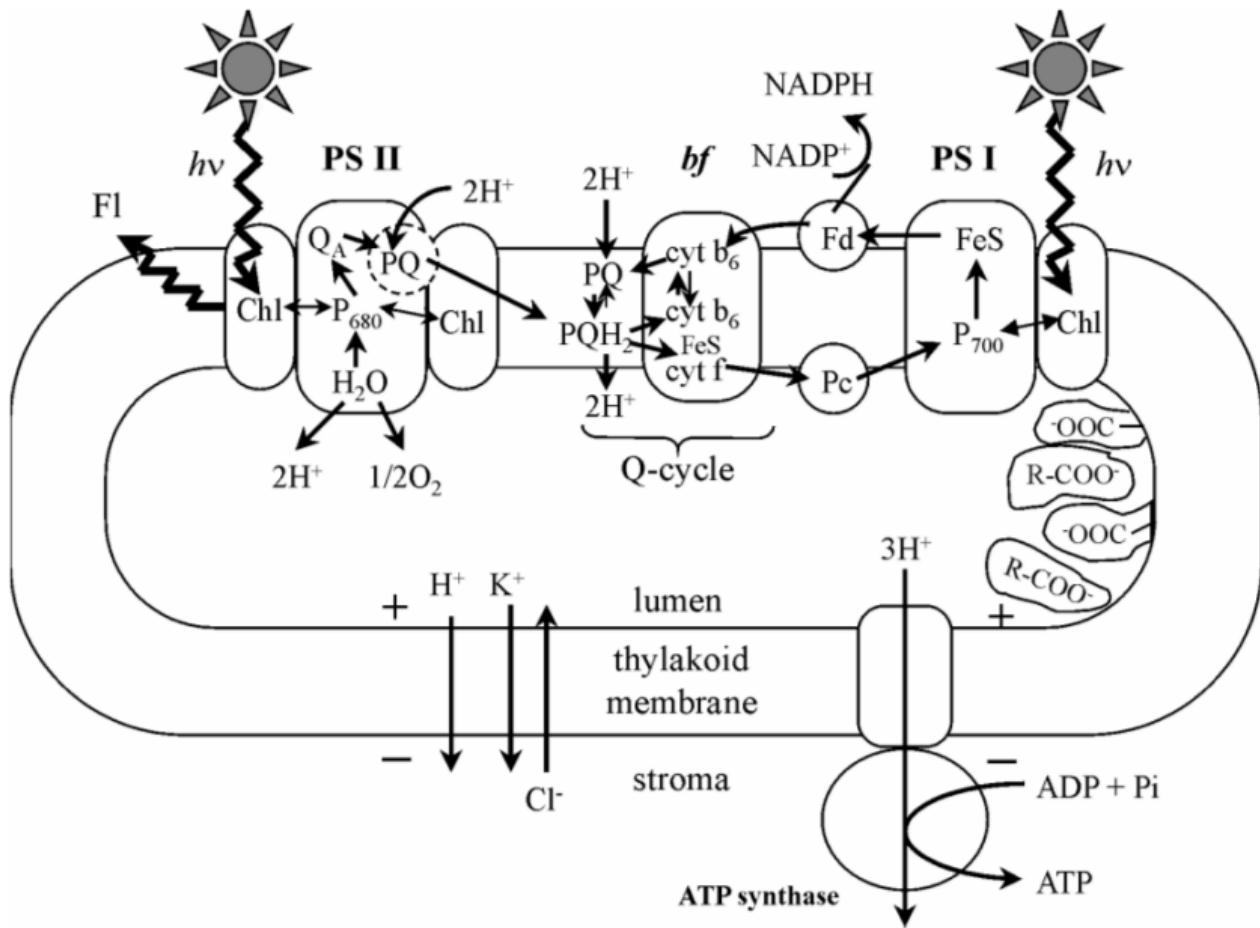
laser energies: $7.5 \cdot 10^{15} \text{ ph/cm}^2$ flash (dark blue), $6.2 \cdot 10^{15} \text{ ph/cm}^2$ flash (magenta), $3.0 \cdot 10^{15} \text{ ph/cm}^2$ flash (beige);
 $5.4 \cdot 10^{14} \text{ ph/cm}^2$ flash (light-green).



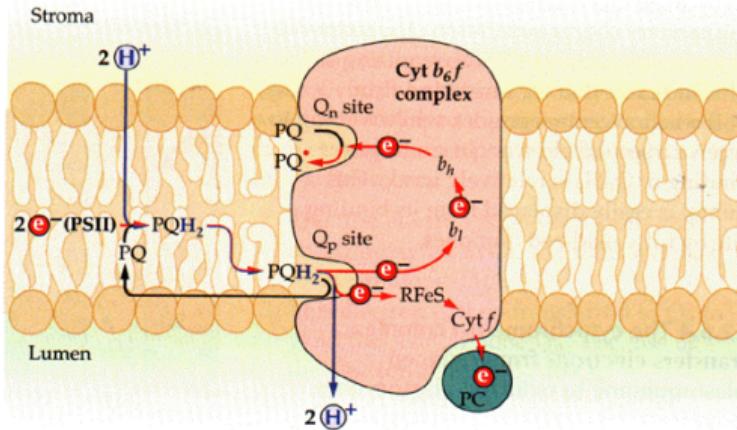
Electron transport in PSII

Arrows – the processes of non-radiation relaxation

Rate constants of this processes can be evaluated only by simulation (not directly in experiment)



(A) First turnover



(B) Second turnover

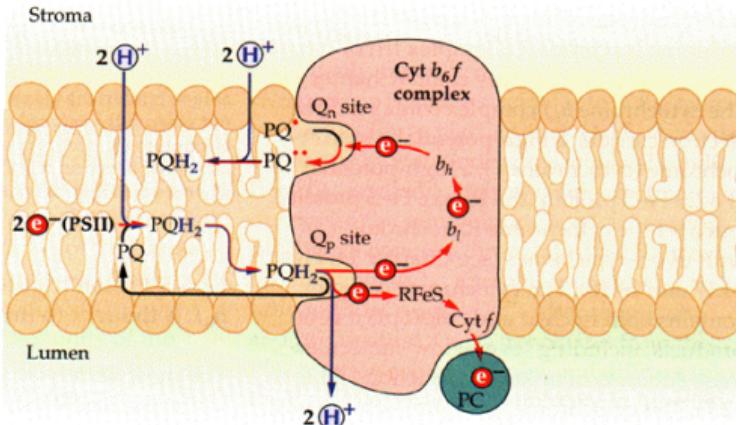
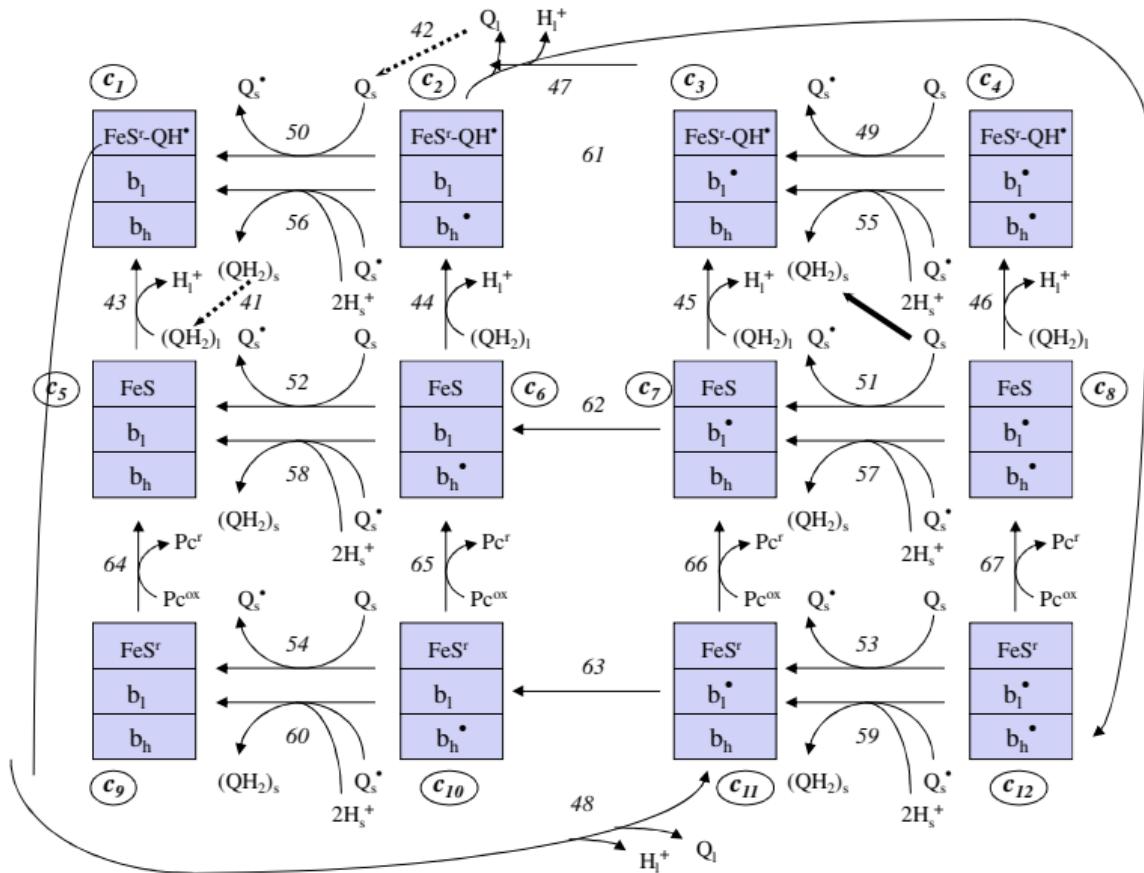
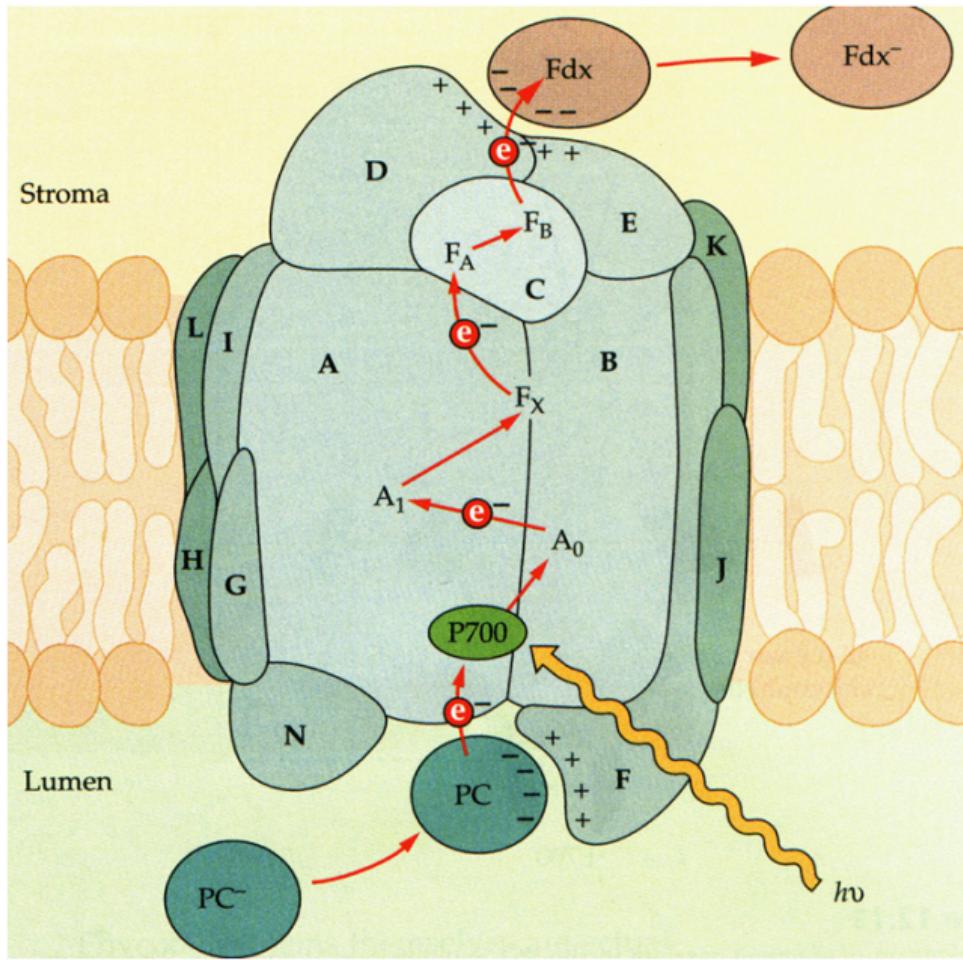


Схема Митчела
функционирования
цитохромного
комплекса.

Сопряжение
электронного
транспорта и
трансмембранного
переноса протонов

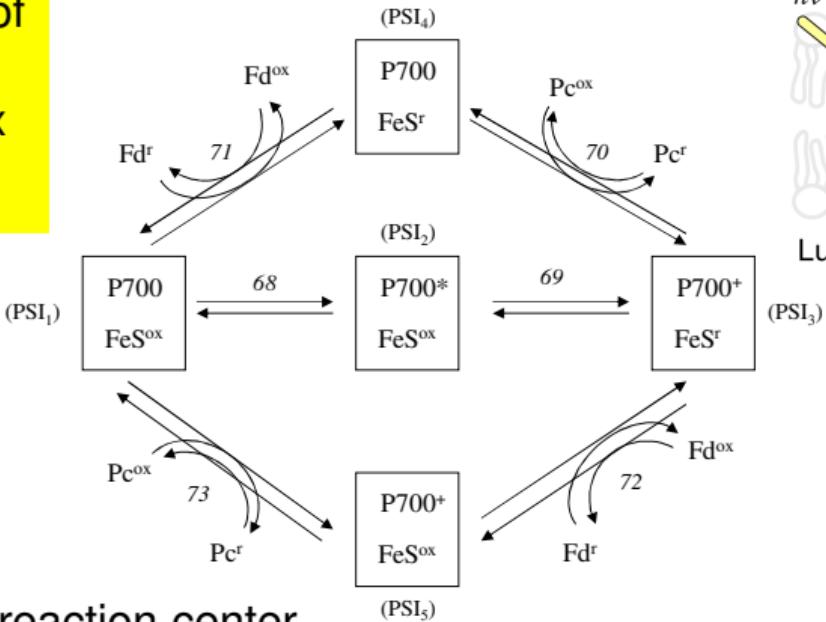
States of the Cytochrome complex





Комплекс
Фото-
реакционного
центра
Фотосистемы I

Scheme of PS1 Complex States



P700 - the reaction center chlorophyll,

FeS - the entire acceptor complex;

Fd, ferredoxin;

Pc, plastocyanin;

superscripts mark the reduced (r) and oxidized (ox) states.

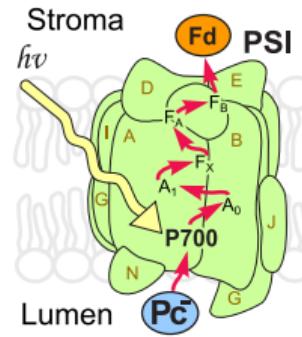
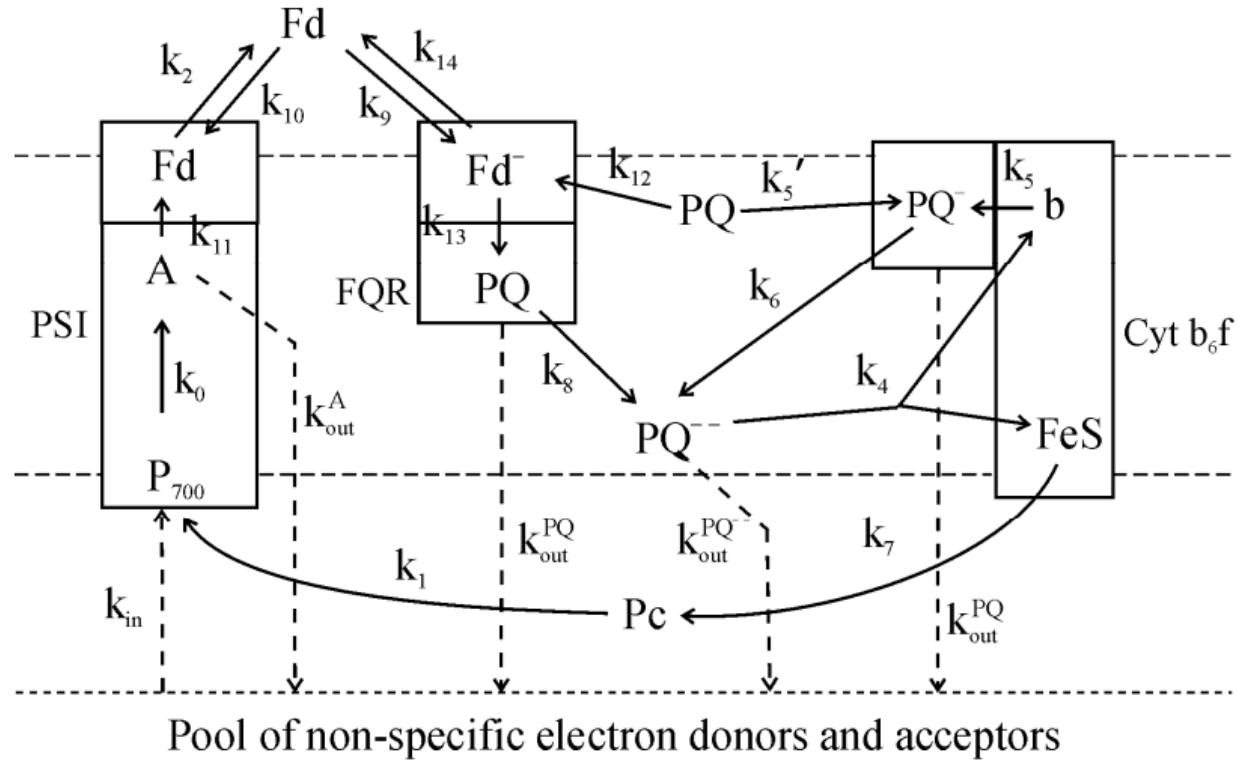


Схема процессов в ФС1 и циклических потоков вокруг ФС1



General kinetic model. Fluorescence induction curves simulation

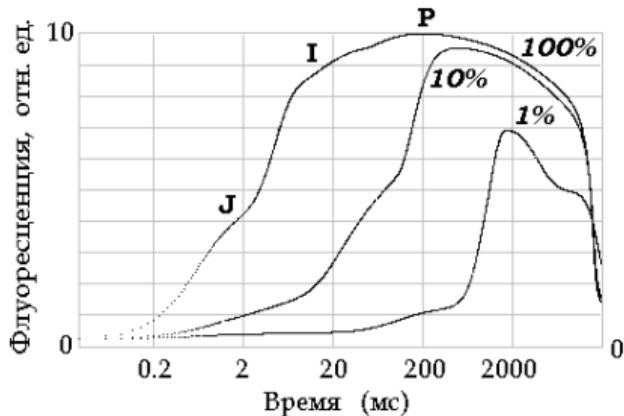
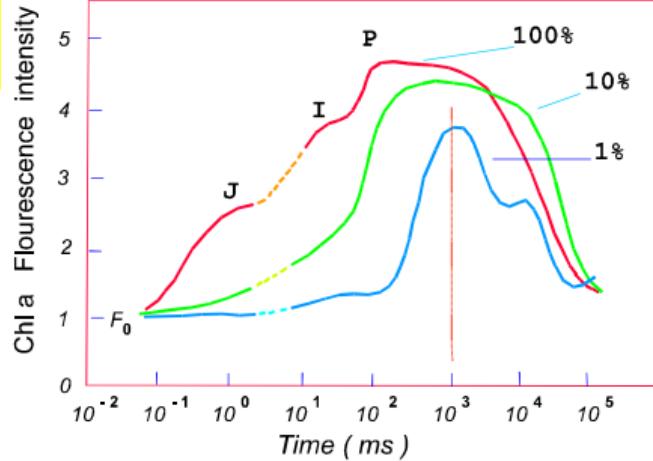
Experiment

Red light (650 нм) Intensity
600 (100%), 60 (10%) and 6
(1%) $W \cdot m^{-2}$.

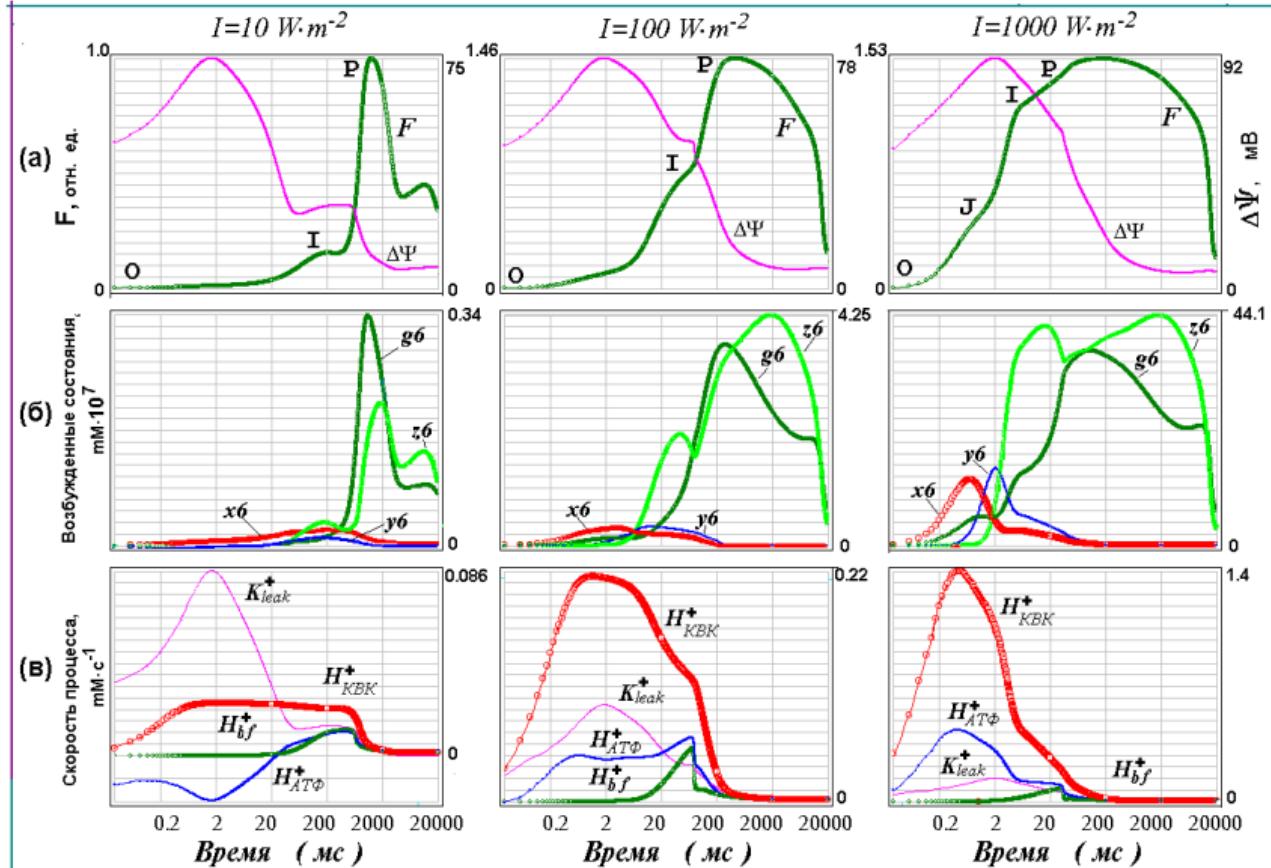
Strasser R.J., Srivastava A.,
Govindjee // Photochemistry
and Photobiology. 1995. V.61.
P.32-42 44.

Model

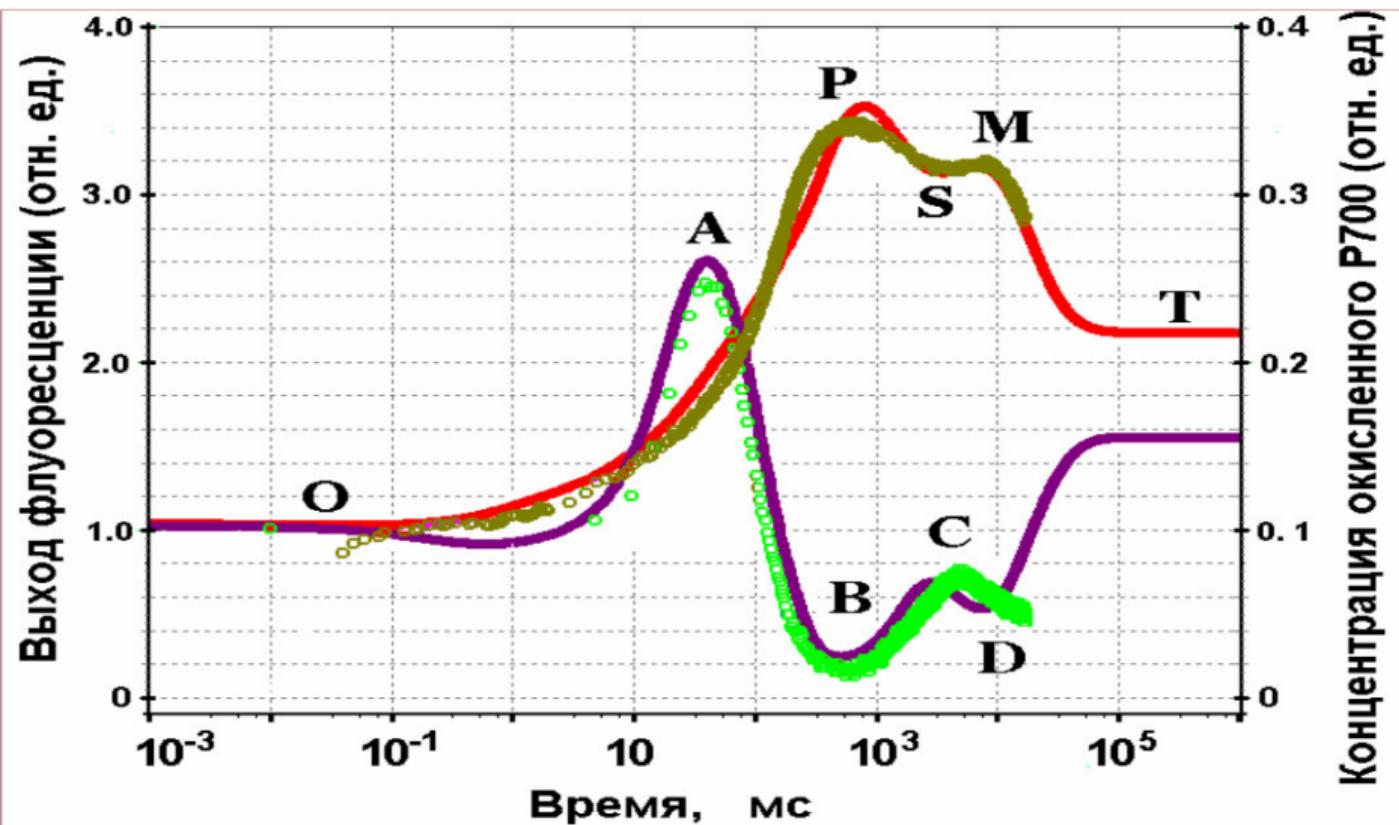
Light constants:
 $1500, 150 \text{ и } 15 \text{ } c^{-1}$.



Kinetic curves of variables of the model



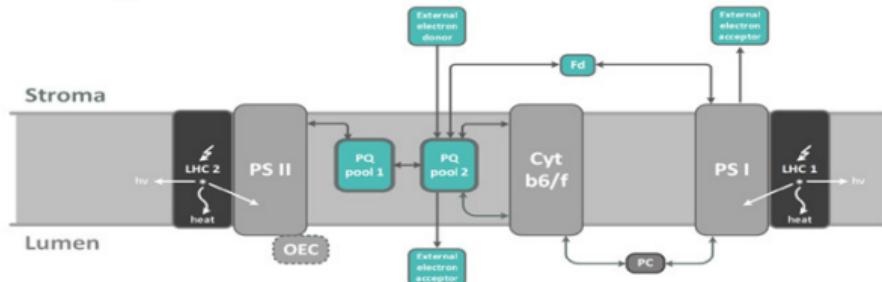
Одновременное фитирование данных флуоресценции и кинетики редокс превращений P700 - фотоактивного пигмента ФС1



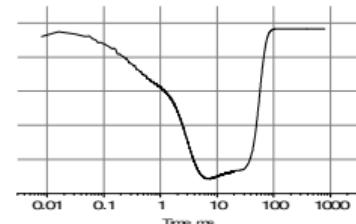
Belyaeva,
Bulychev,
Riznichenko,
Rubin. Phot. Res.
2016

Кинетические Монте Карло модели

A

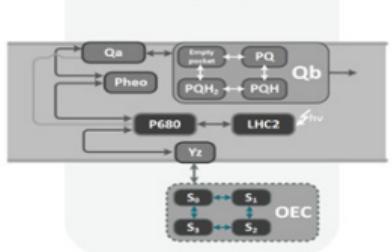


P700 redox transformations



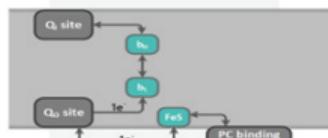
B

PS II



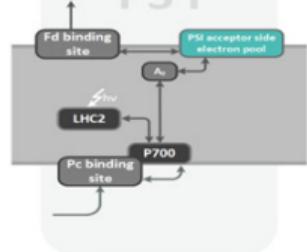
C

Cyt b6/f complex



D

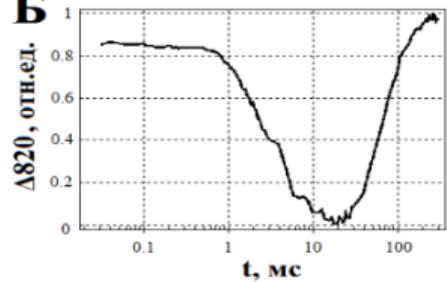
PS I



3 millions of Photosynthetic Chains
As in a real micro algae cell

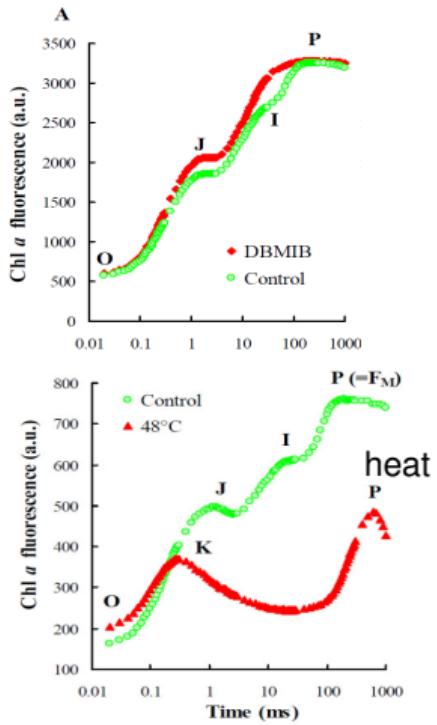
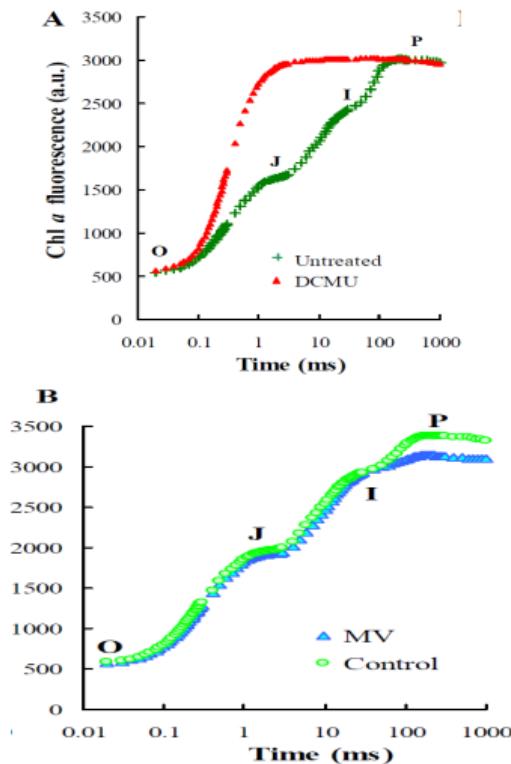
МОДЕЛЬ

Б



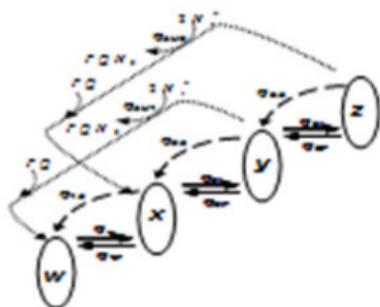
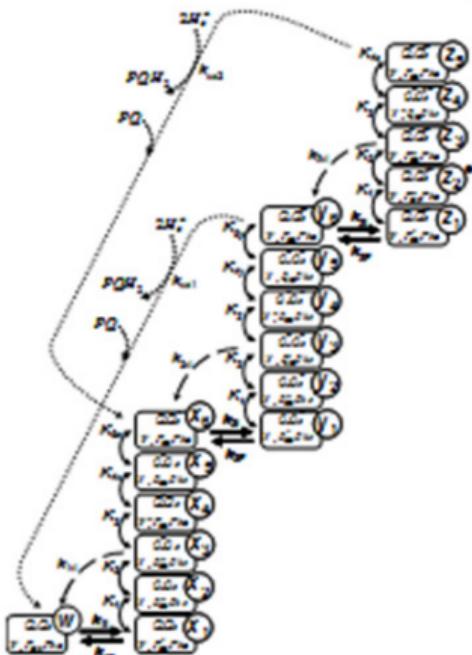
эксперимент

Симуляция воздействия ингибиторов



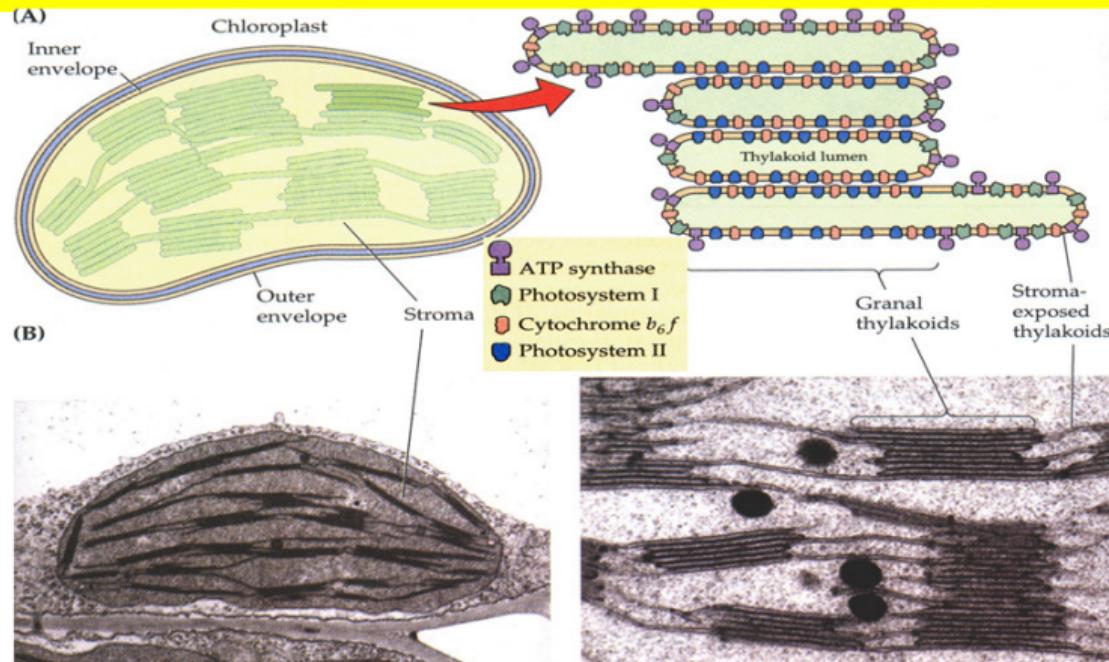
Как использовать кинетические модели

- Фитирование экспериментальных кривых и оценка параметров. Не определяемых экспериментально (параметры безызлучательной релаксации в ФРЦ)
Оценка параметров фотосинтетической цепи в разных условиях : для разных видов, в ходе роста культуры, при разных режимах культивирования и режимах освещения, при стрессе

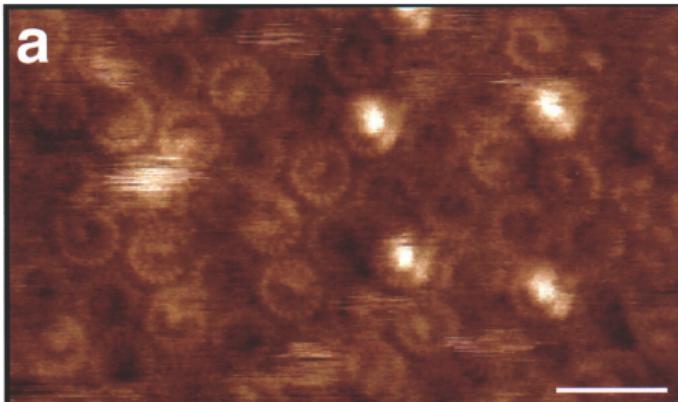
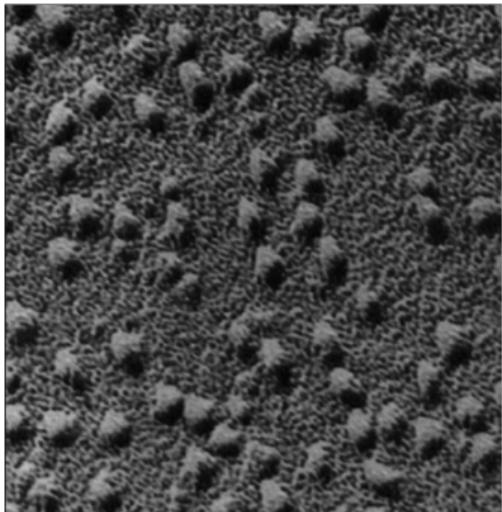


Недостатки кинетических моделей

- Трудности в описании пространственной гетерогенности
- Несвободная диффузия подвижных переносчиков
- Невозможность проследить судьбу отдельного участника процесса



Изображения мембраны тилакоида

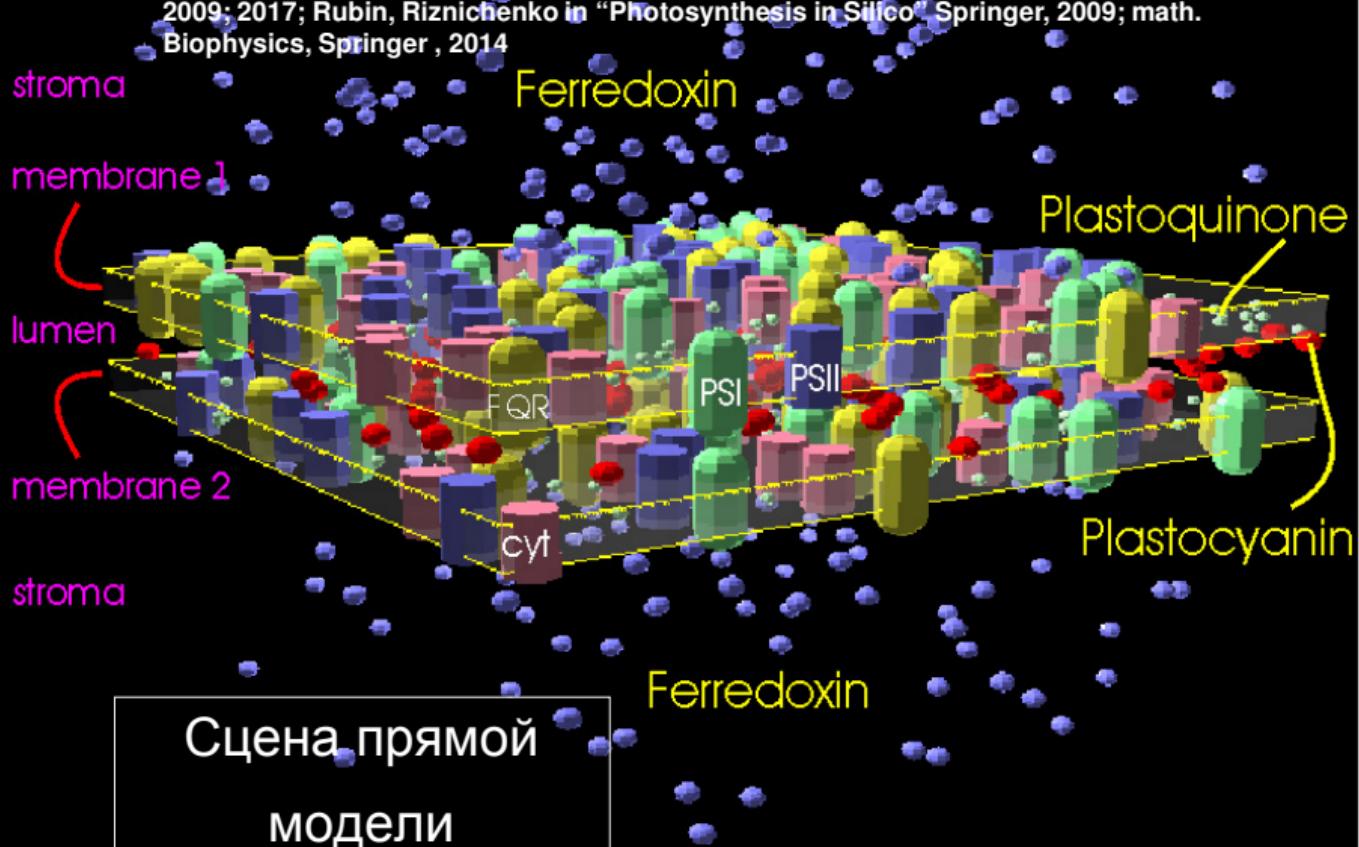


Атомная силовая микроскопия

Вид участка тилакоидной мембраны в электронный микроскоп. Размер изображения 4 мкм. Грана – структурная единица тилакоида, имеет форму диска диаметром 500 нм и толщиной 15-20 нм

Метод прямого многочастичного моделирования

Коваленко и др., 2003, 2007, 2008, 2009; Kovalenko et al., 2006; Абатурова и др., 2008;
Дьяконова и др., 2008; 2016; Устинин и др., 2013; Хрущев и др. 2015; Ризниченко и др.,
2009; 2017; Rubin, Riznichenko in "Photosynthesis in Silico" Springer, 2009; math.
Biophysics, Springer , 2014



Броуновская динамика (Brownian dynamics)

Для каждой частицы решается уравнение:

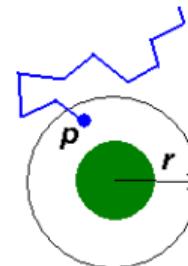
- $\dot{\xi} \frac{dx}{dt} = f(t)$

- Здесь $f(t)$ – случайная сила, распределенная по Гауссу с нулевым средним и дисперсией, равной $2kT\xi$, k – постоянная Больцмана, T – температура, ξ – коэффициент трения в среде, вычисляемый (в предположении о сферичности частицы) по формуле ,

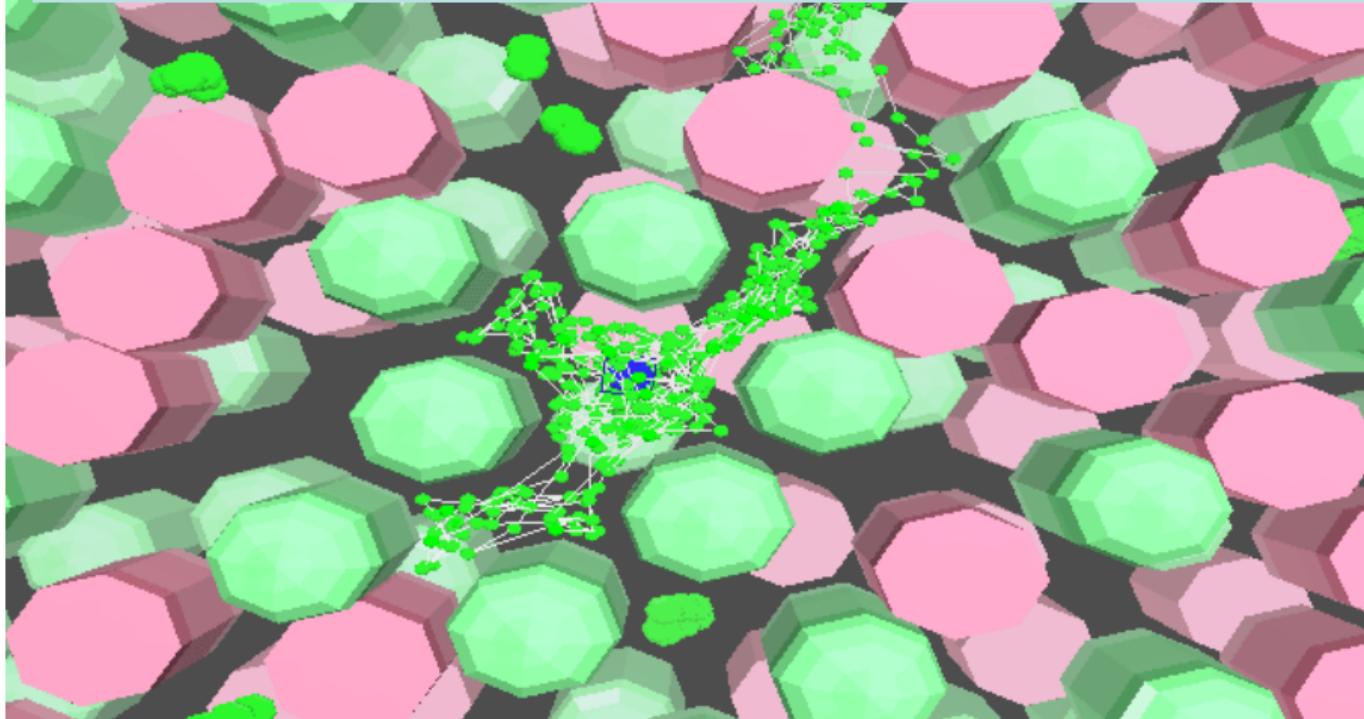
$$\xi = 6\pi\eta a$$

- где η – вязкость среды, a – радиус частицы

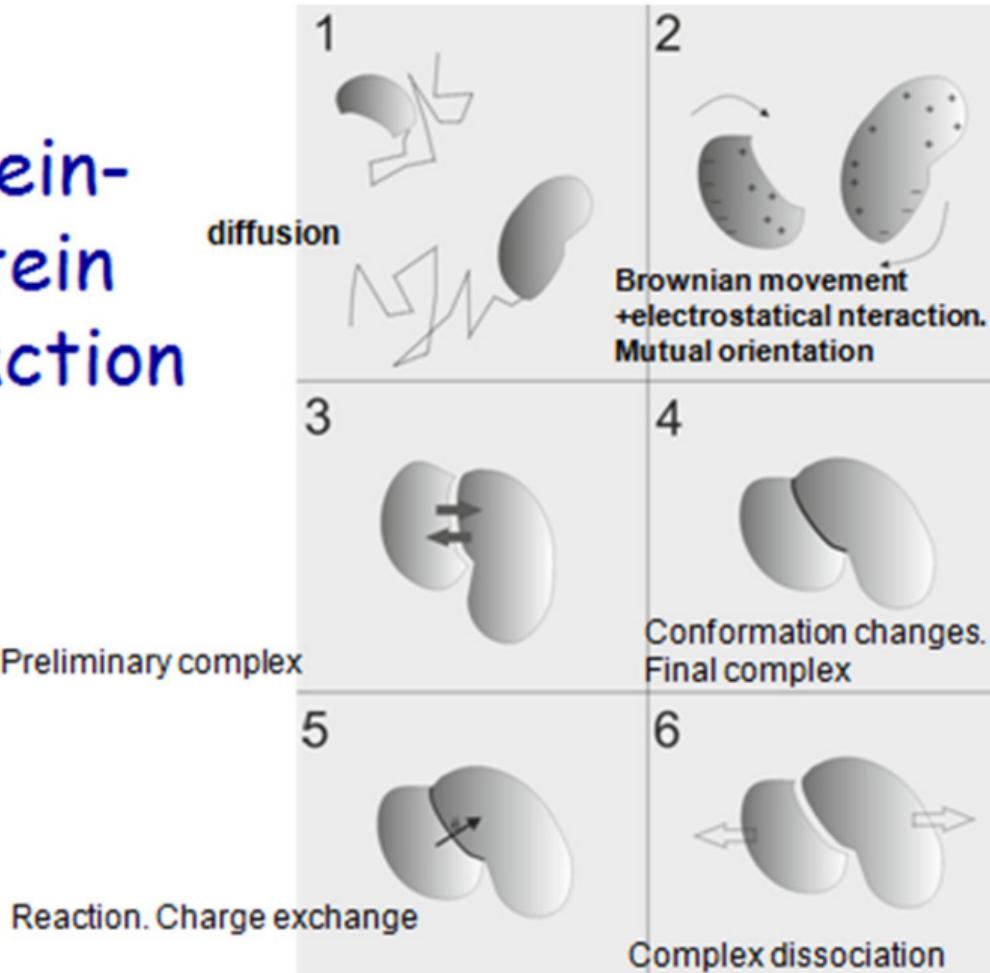
Параметры прямой модели:
Эффективный радиус взаимодействия
Вероятность докинга



Модельная траектория молекулы PQ в мембране с встроенными ФС1, ФС 2 и цитохромными комплексами



Protein-protein interaction



Description of protein movement by Langevin Equations

Transition

$$\xi_t^x \frac{dx}{dt} = f_x(t) + F_x \quad \langle f_x(t) \rangle = 0 \quad \langle f_x(t)^2 \rangle = \frac{2kT\xi_t^x}{\Delta t}$$

x -coordinate,

ξ_t^x -vicious friction coefficient along x ,

$f_x(t)$ and F_x -projections of casual and electrostatic forces on the axes x , respectively

k -Boltzmann factor,

T -temperature

$F_x = -q \cdot \frac{d\varphi}{dx}$ φ -potential

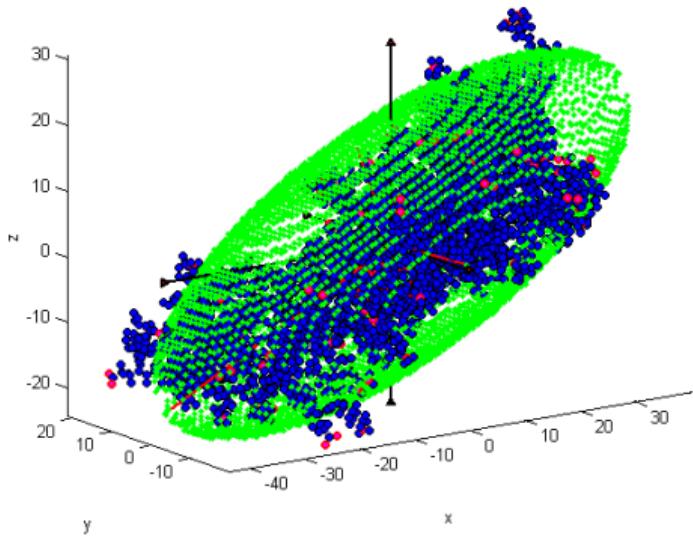
Rotation

$$\xi_r^x \frac{d\varphi}{dt} = m_x(t) + M_x \quad \langle m_x(t) \rangle = 0 \quad \langle m_x(t)^2 \rangle = \frac{2kT\xi_r^x}{\Delta t}$$

φ -the angle of rotation around the axes x , ξ_r^x -vicious friction coefficient for rotation around the axes x , $m_x(t)$ and M_x -moments of casual and electrostatic forces relative to the axes x , respectively

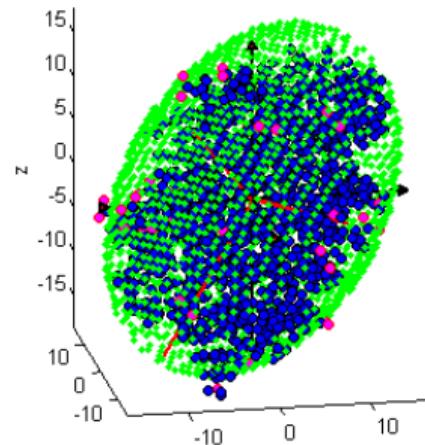
To simulate the diffusion at the distance more than 35 Å

Approximation of cyt f and Pc by ellipsoids of rotation

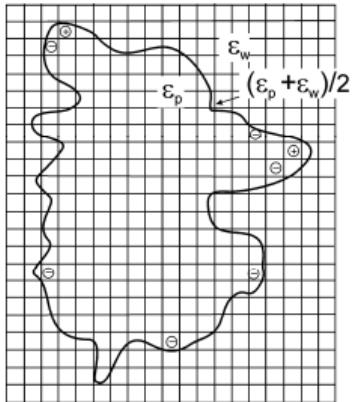


Molecular mass
Axes of ellipsoids of rotation

Cyt f
 $M = 27.9 \text{ КДа}$
 $a=47 \text{ \AA}, b=17 \text{ \AA}$



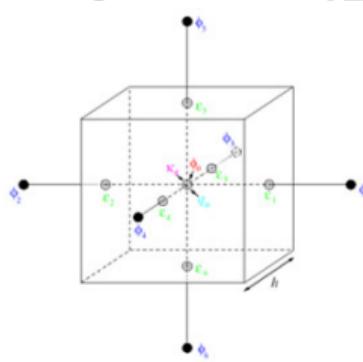
Pc
 $M = 10.5 \text{ КДа}$
 $a=21 \text{ \AA}, b=14 \text{ \AA}$



The grid to calculate electrical potential around the protein

$$\varphi_0 = \frac{\left(\sum_{i=1}^6 h \epsilon_i \varphi_i \right) + 4\pi q_0}{\left(\sum_{i=1}^6 h \epsilon_i \right) + h^3 K_0^2}$$

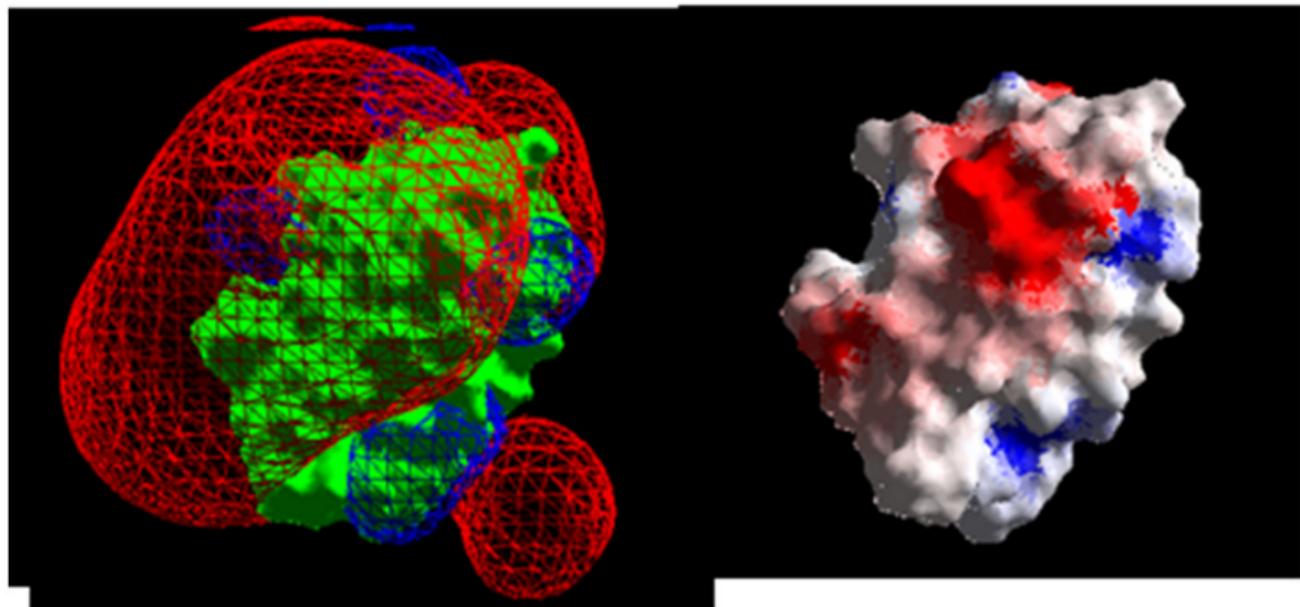
Separate cell of the greed



$$K^2 = \frac{8\pi N_A e^2 I}{k_B T} \quad I = \frac{1}{2} \sum_{i=1}^K c_i^{\text{bulk}} Z_i^2$$

$$\nabla \epsilon \nabla \varphi = -4\pi \rho + \kappa^2 \varphi$$

Equipotential surfaces (left) (-10mB, +10mB) and surface electrostatic potential (right) of plastocyanin,
 $pH=7$, $I=100\text{ M/M}^3$

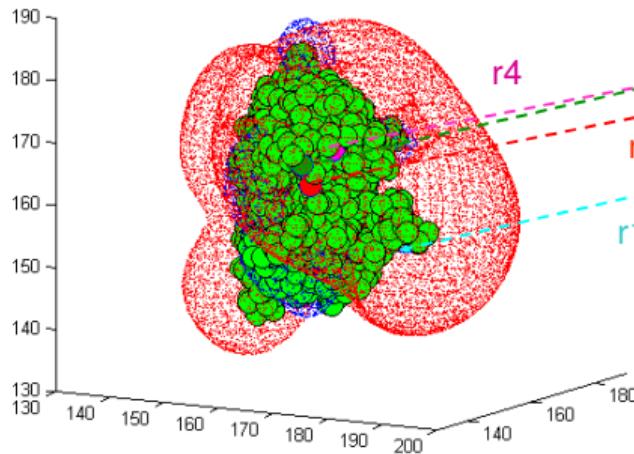


To calculate interactions at the distance less than 35Å

Equipotential surfaces calculated according to Poisson-Boltzmann equations

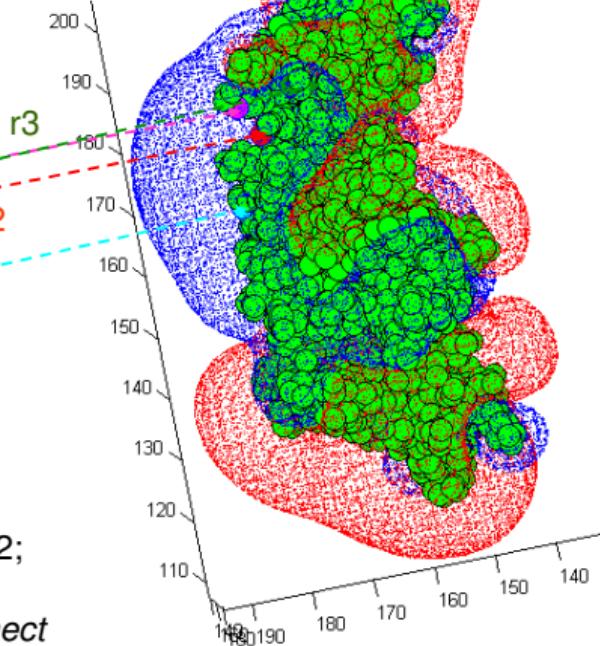
Oxidesed Pc

WT



Reduced cyt f

WT



Ion strength - 100 mM, pH=7, $\epsilon_{p-pa}=80$; $\epsilon_{белка} = 2$;
red -6.5 mV, blue + 6.5 mV;

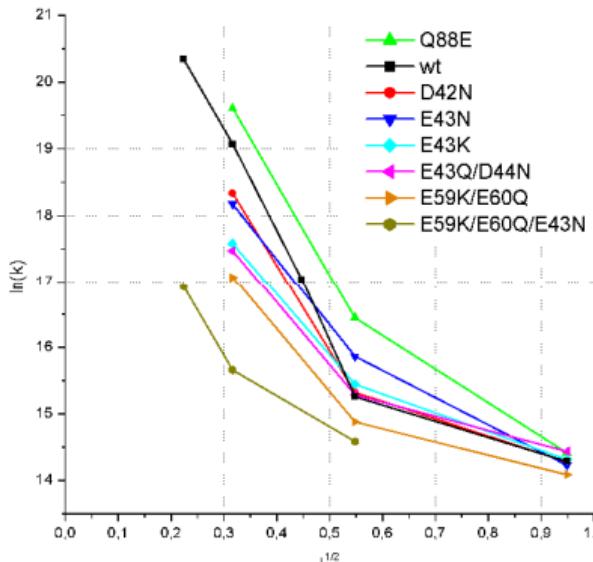
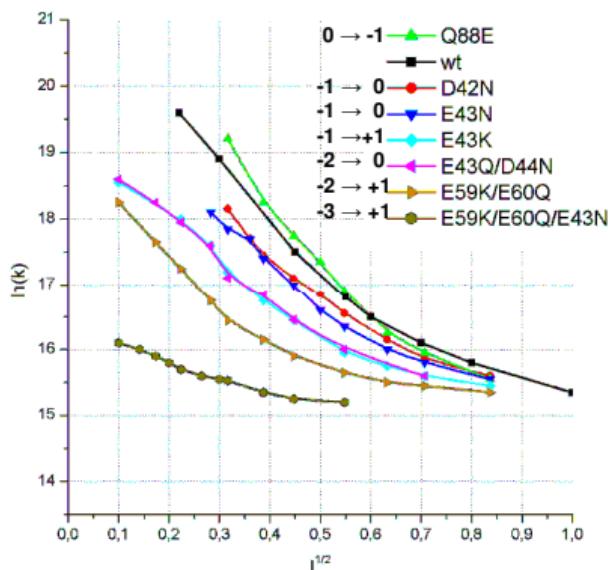
green – atoms of molecules. Dotted lines connect residues on Pс and Cytf that were used by simulation for calculation the distance between proteins

Reaction between cyt f and different Pc mutants in solution

Dependence of Log k from Ion strength

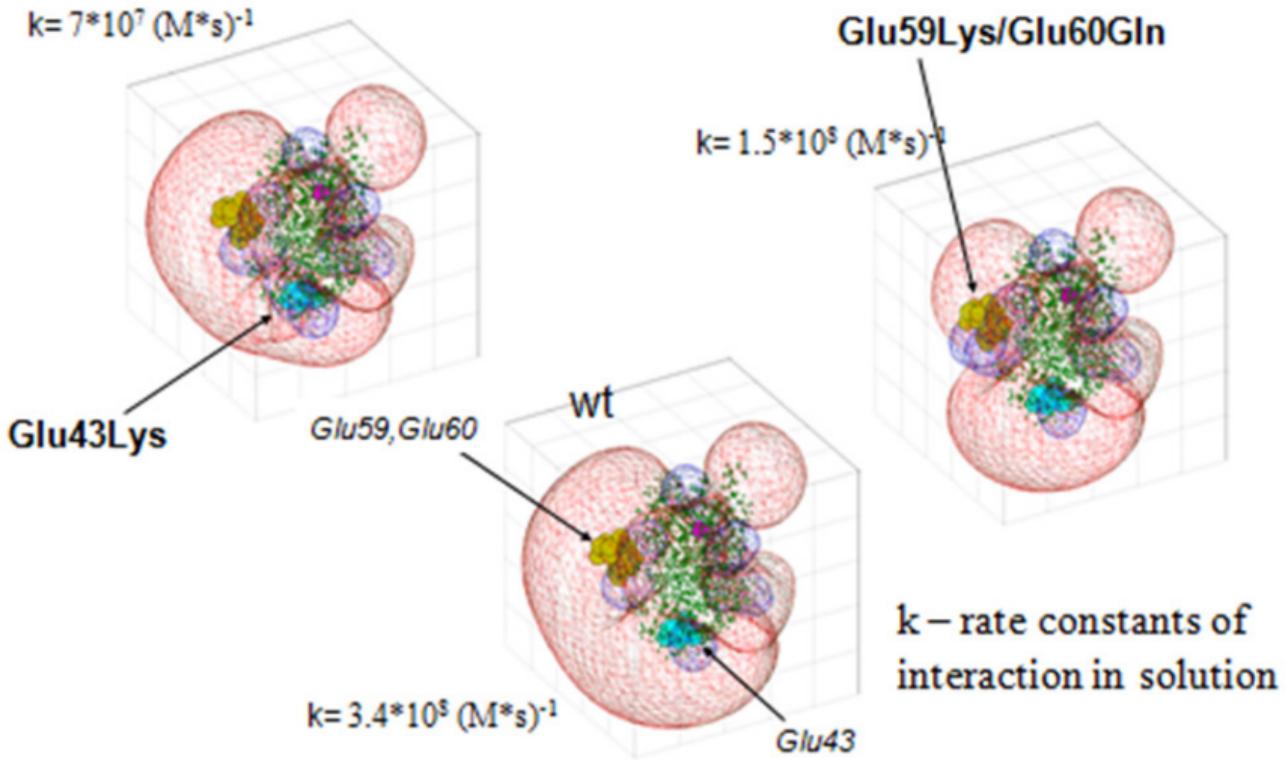
experiment A. Kannt et al.(1996)

modeling



$k - (M \cdot c)^{-1}$, $I - M$; pH=7 ; r D42-R209 -18 A, r E43-K187 -18 A, r D44-K187 -18 A, r E60-K58 -25 A, r Cu-Fe – 40A; P=0.01; dt=100 ps

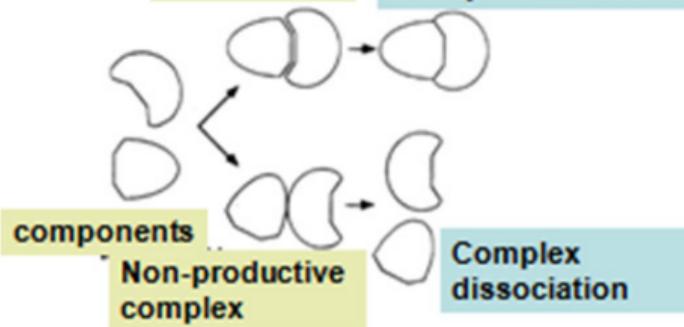
Equipotential surfaces of Pc and its mutants



Preliminary complex

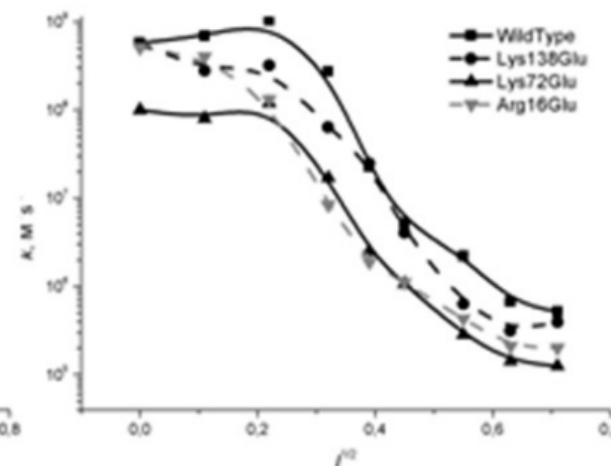
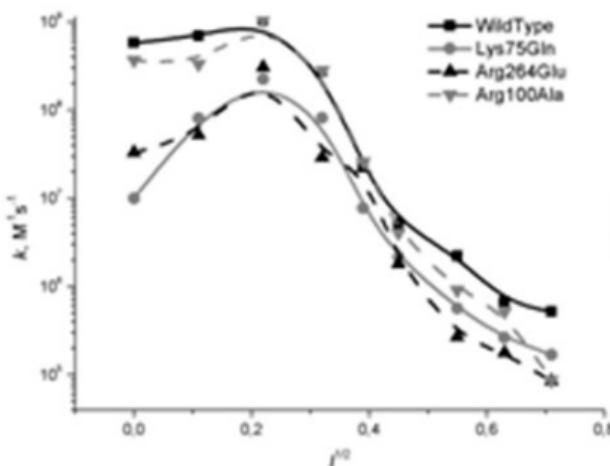
Final Complex → reaction

Kovalenko, Djakonova, et al., 2011

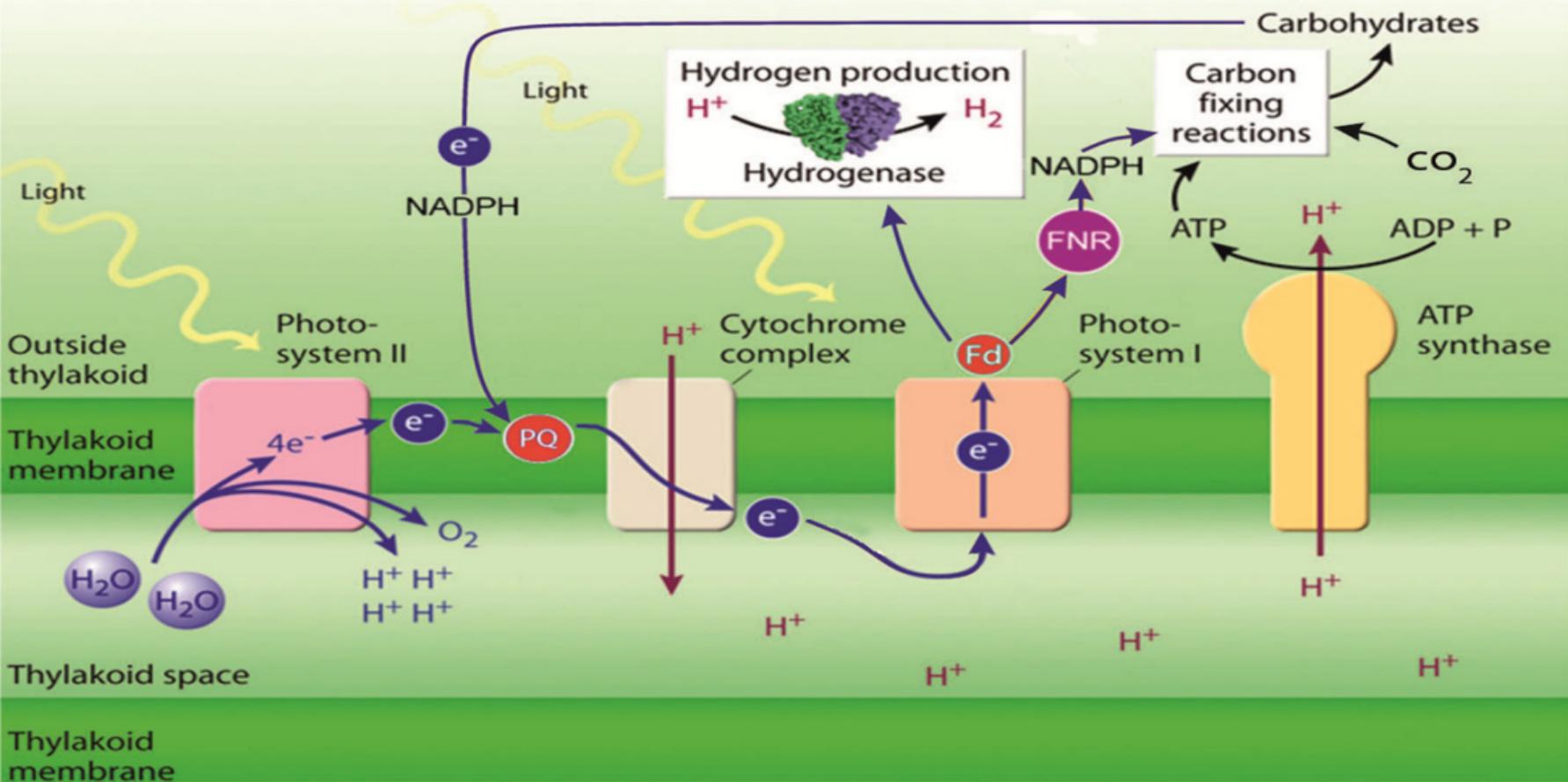


Non-monotonous dependence of the Rate constant of complexes interaction on the ion strength

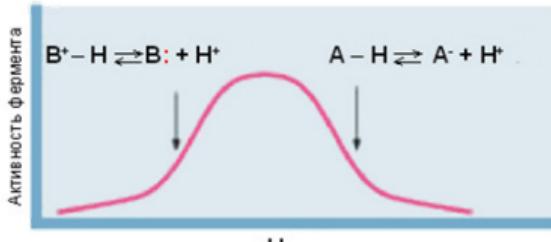
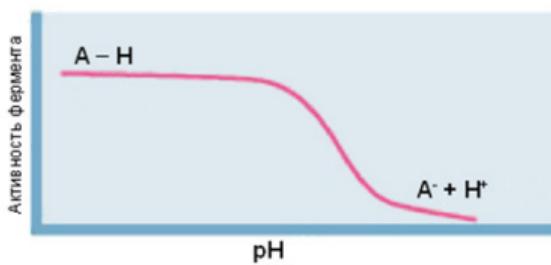
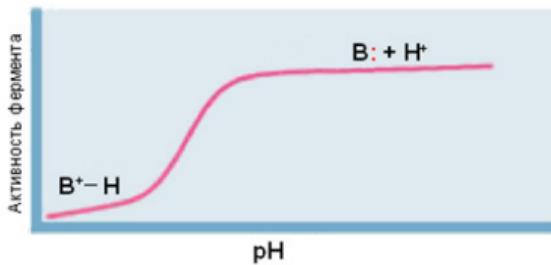
Fd-FNR interaction



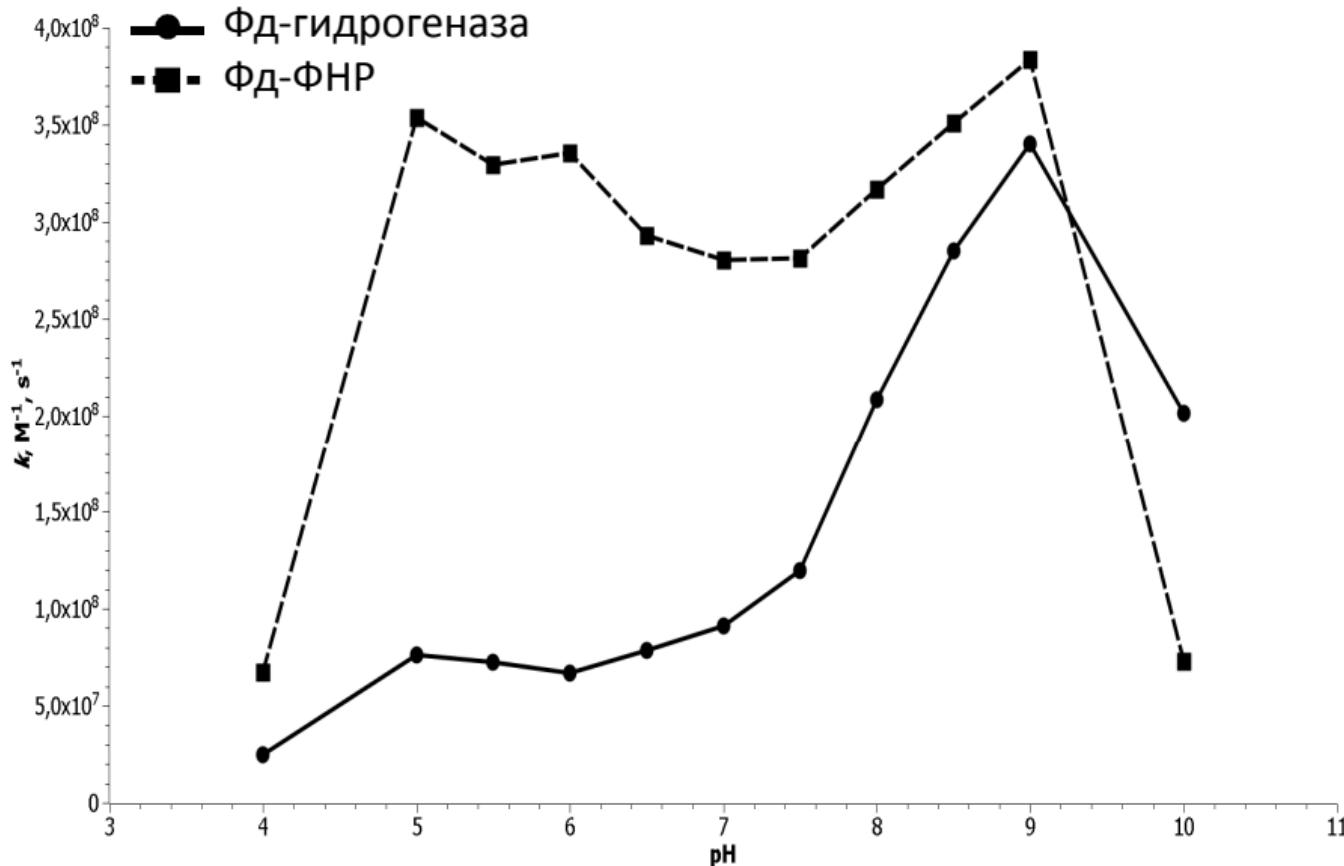
Пути переноса электрона в условиях стресса¹



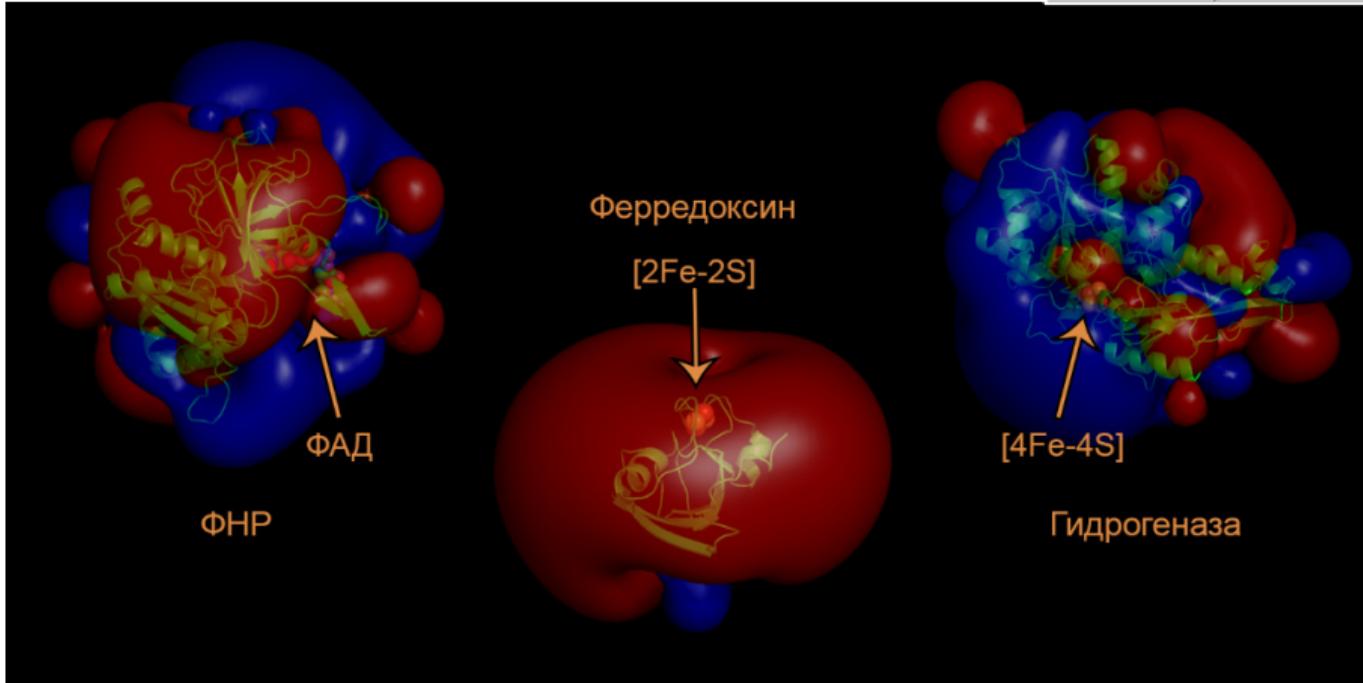
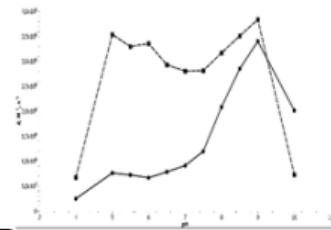
Влияние pH на белковые молекулы



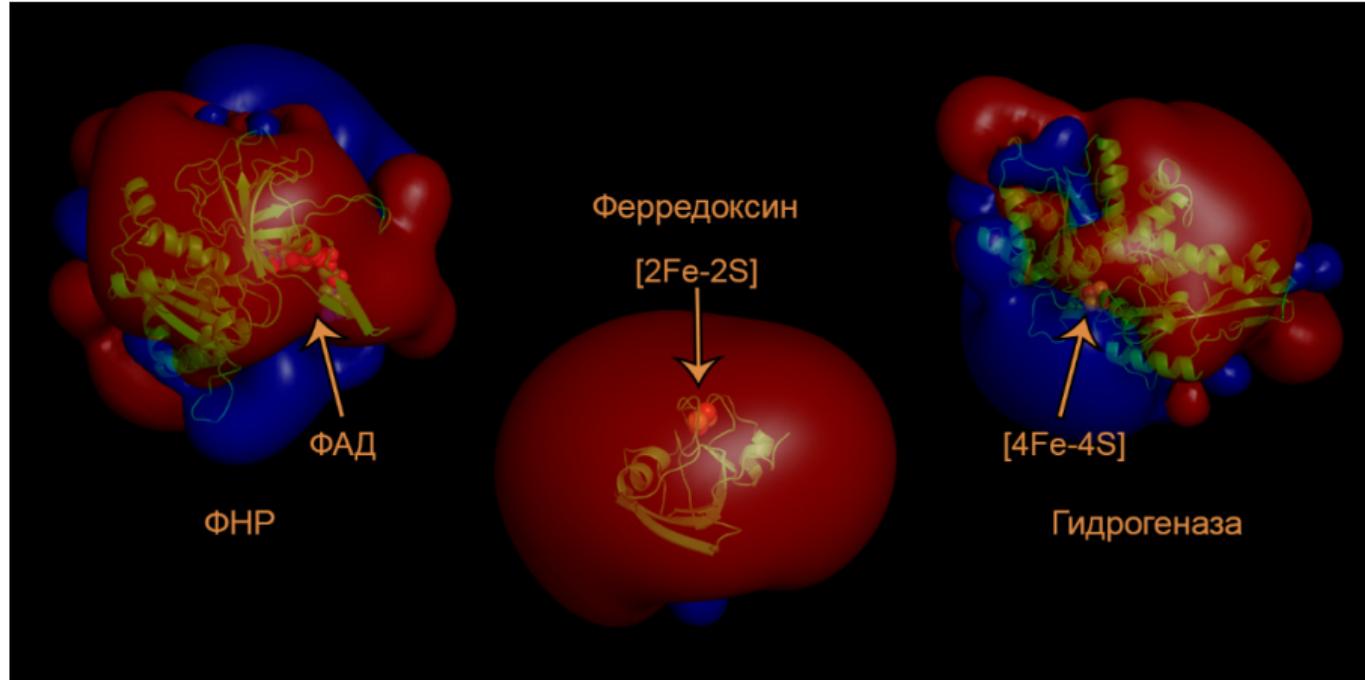
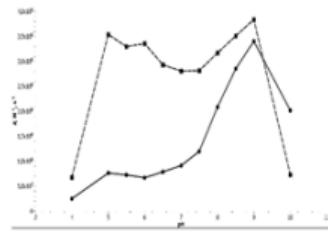
Зависимость константы скорости образования комплекса Фд-ФНР и Фд-гидрогеназы от pH



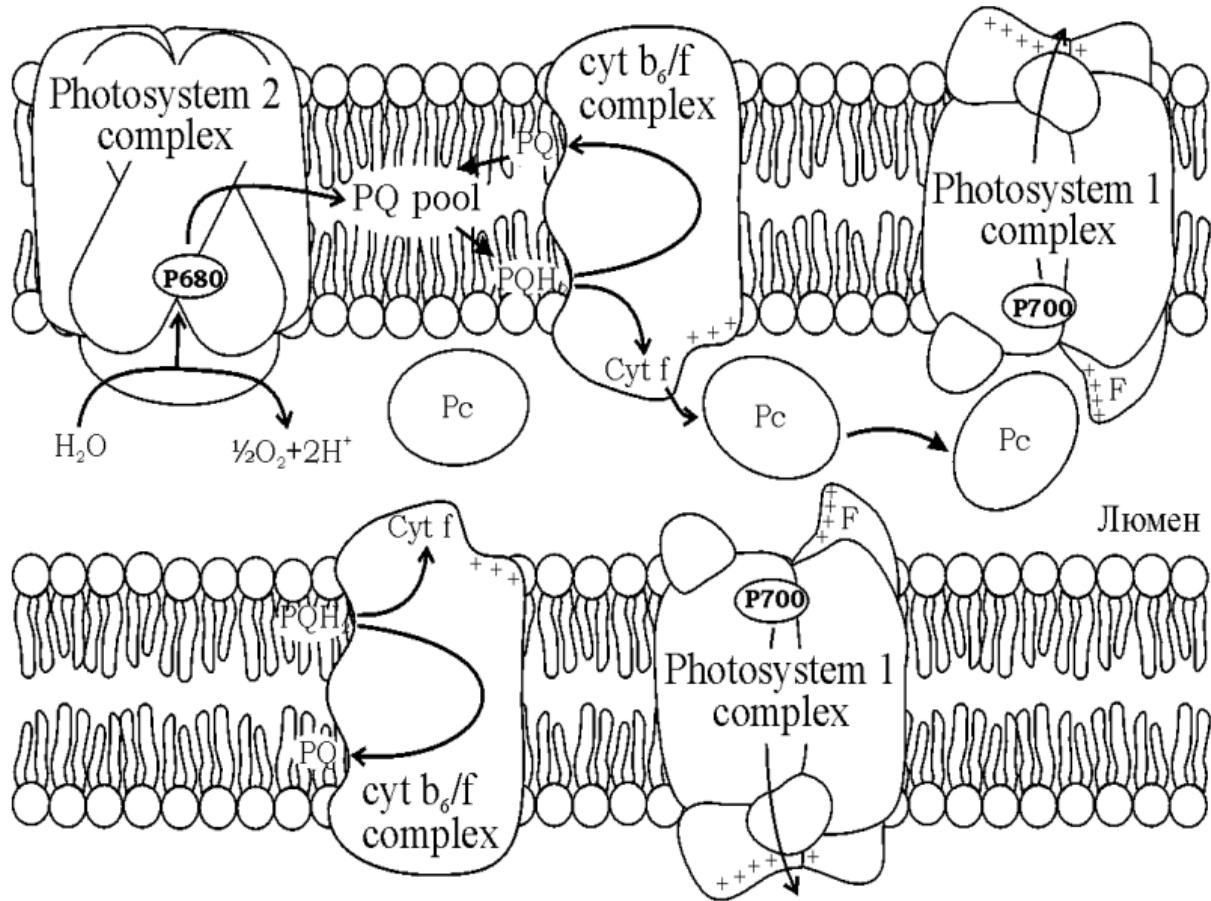
Эквипотенциальные поверхности Фд, ФНР и гидрогеназы при рН 6



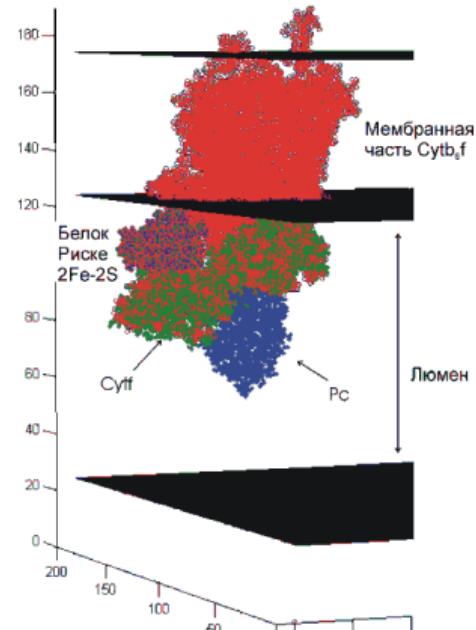
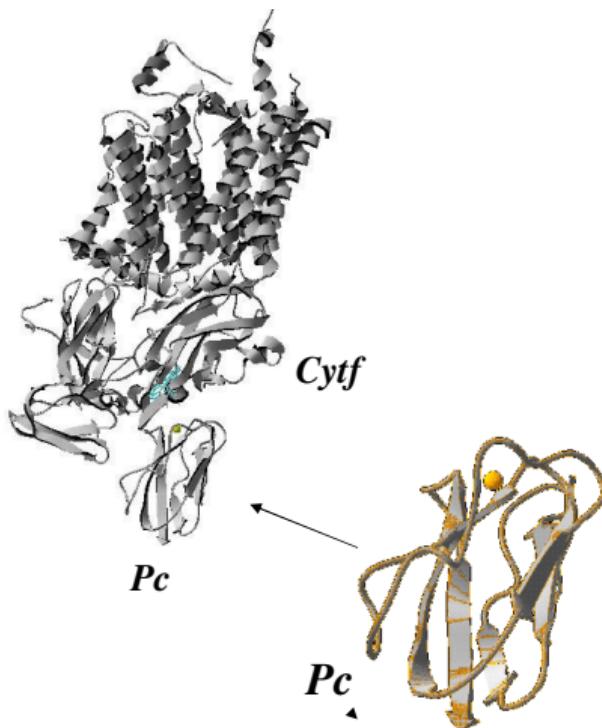
Эквипотенциальные поверхности Фд, ФНР и гидрогеназы при pH 8



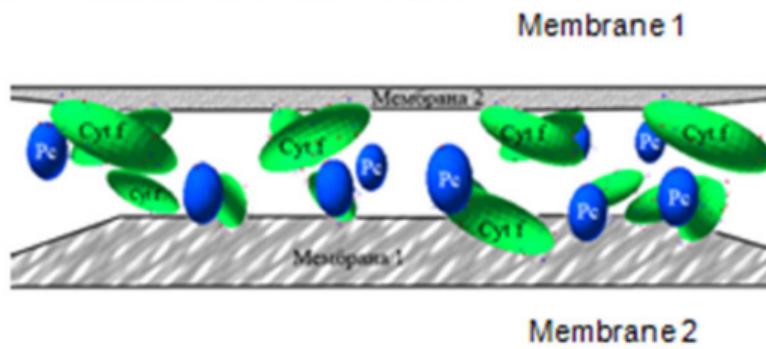
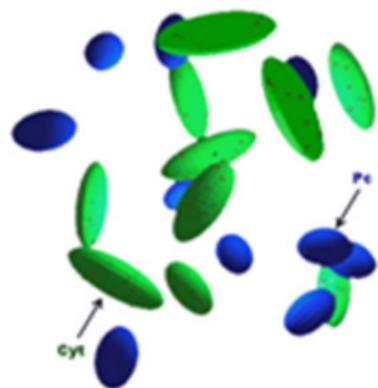
Моделирование процессов в люмене тилакоида



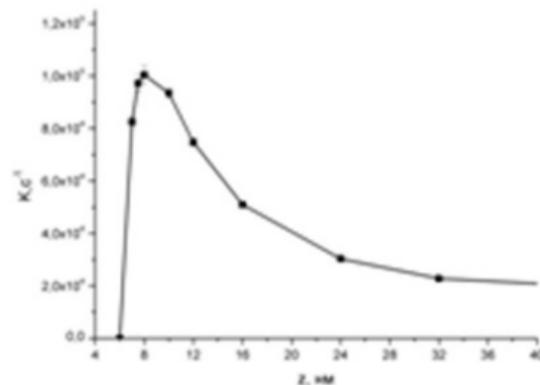
Взаимодействие между Рс и сутф



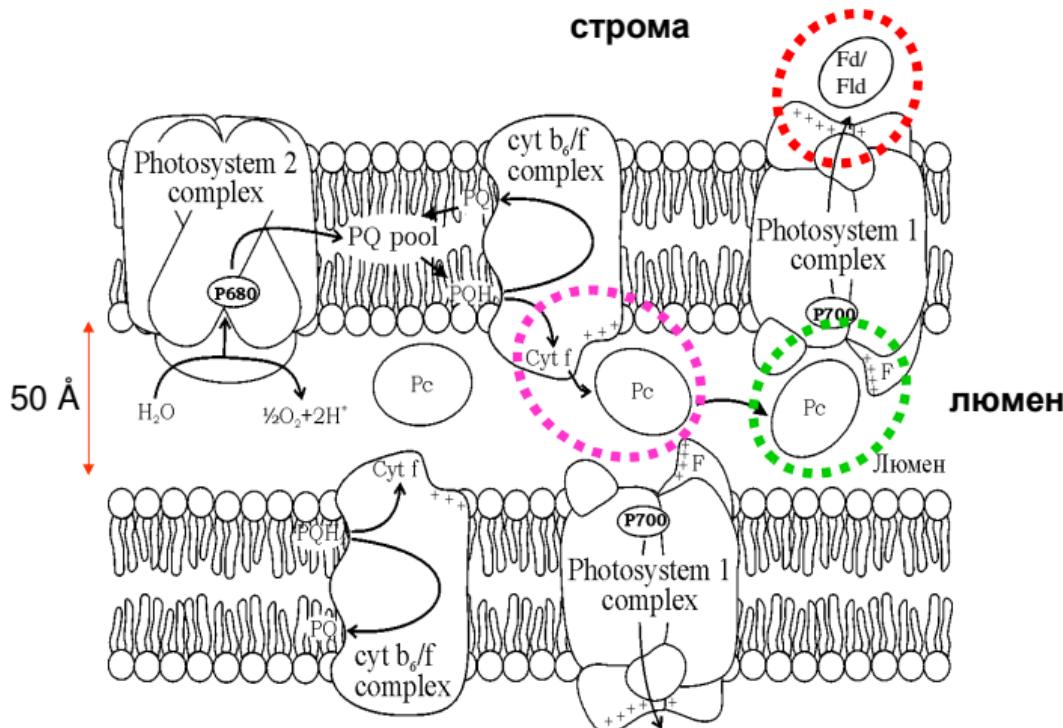
Simulation of Pc-cyt f interactions in solution and in lumen



Rate constant of the reaction of complex Pc-Cyt f formation in thylakoid lumen as a function on the distance z between the membranes

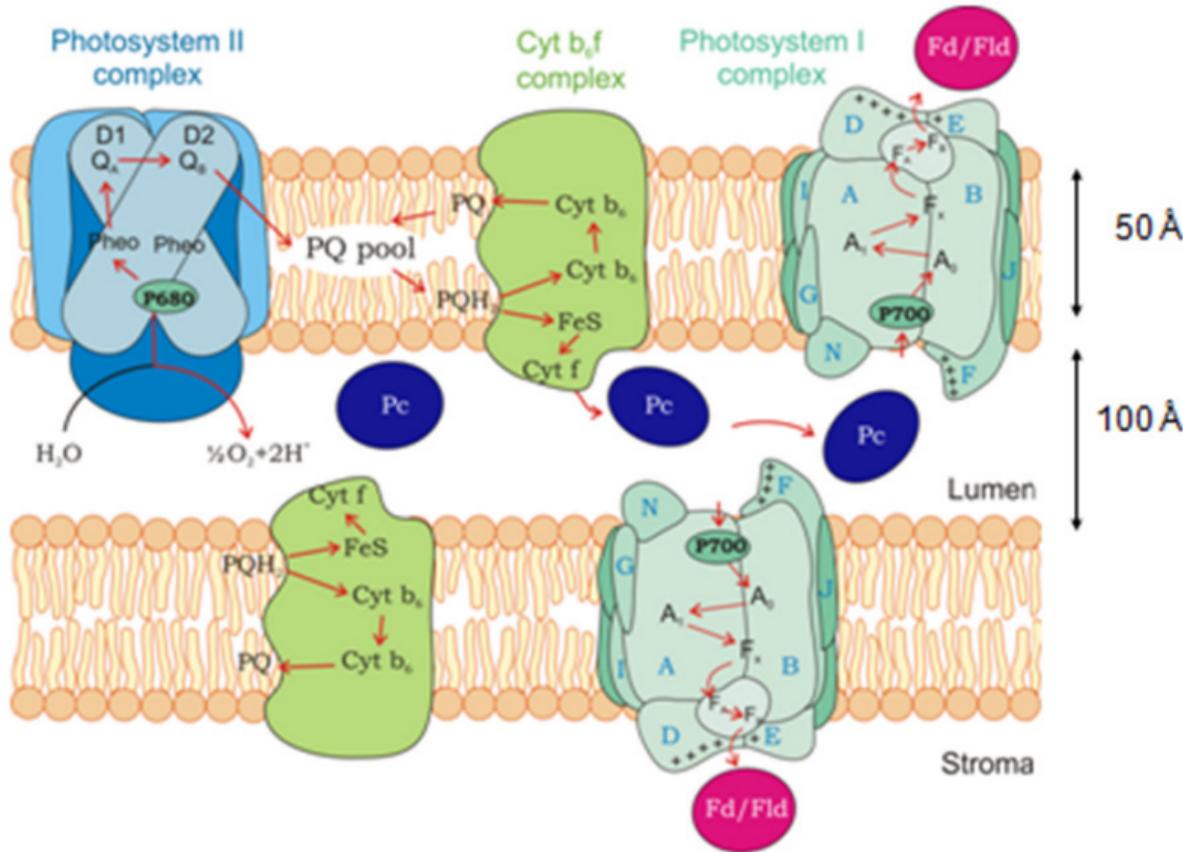


Фотосинтетический электронный транспорт. Комплексы и подвижные переносчики

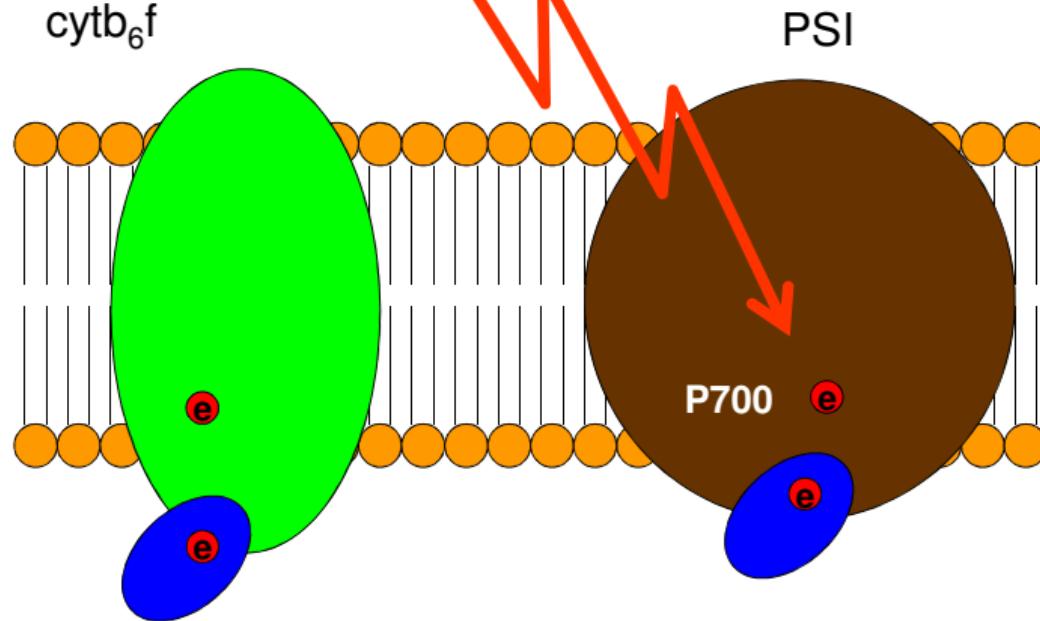


Фрагмент Тилакоида

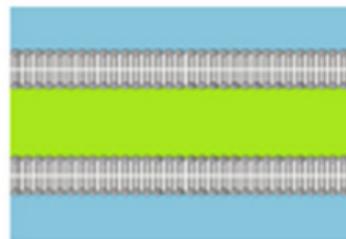
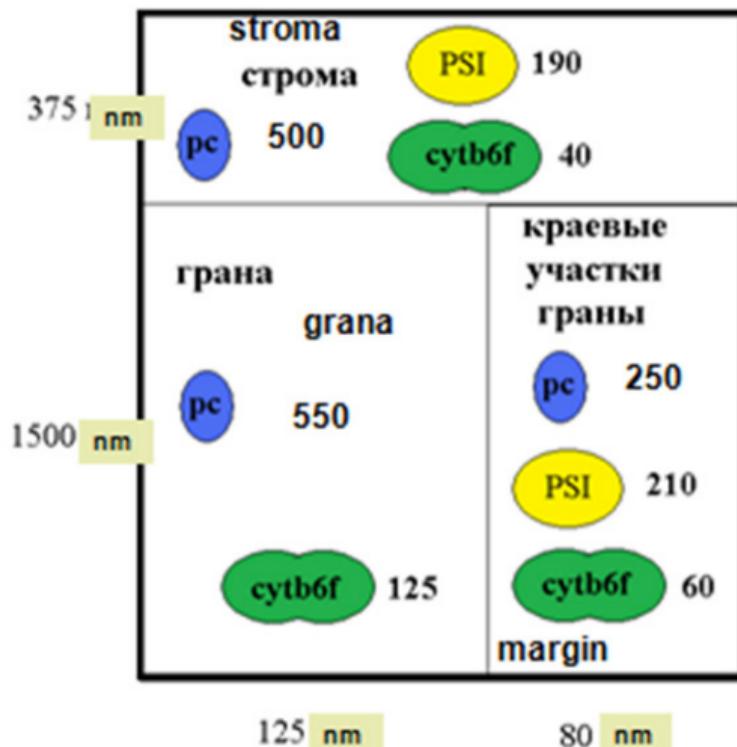
PC transfers electrons from cytochrome complex to PSI



Electron transition by P_c molecule from cytochrome complex to PSI



Model scene: The number of protein and multienzyme complexes



- margin – take about 40% of the grana surface;

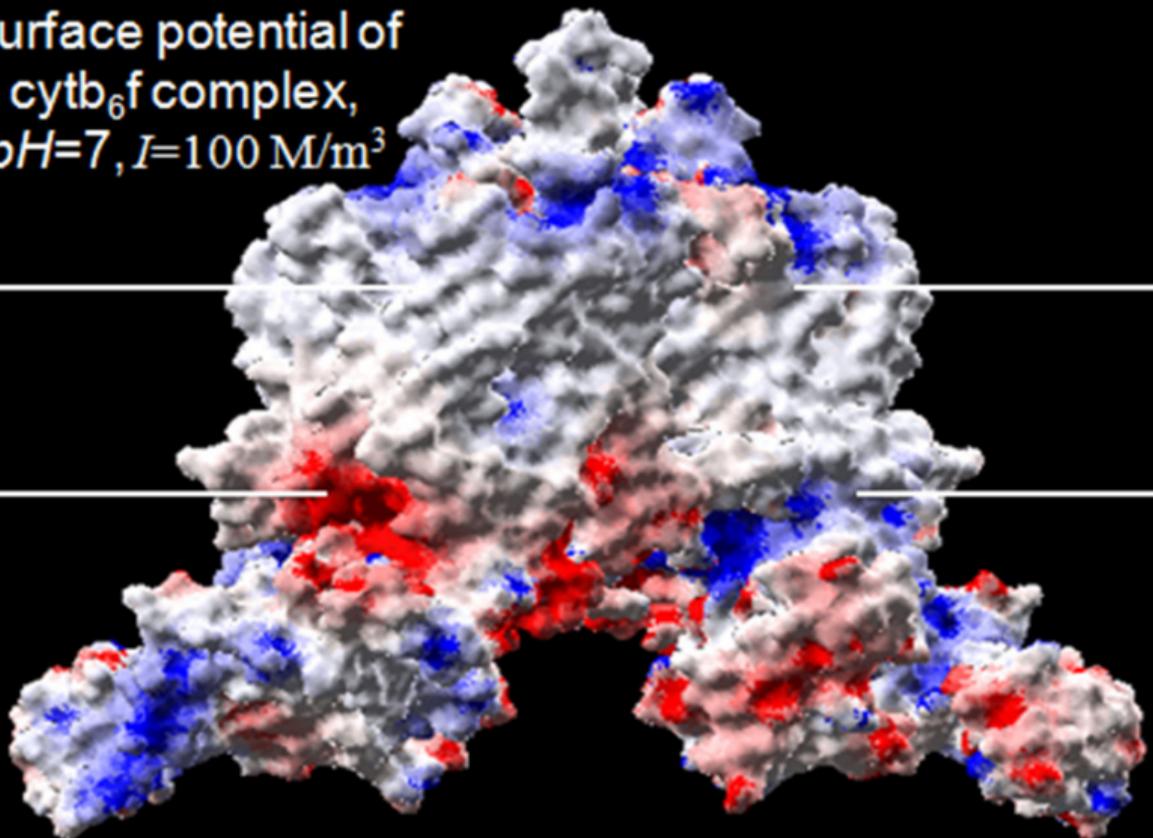
- stroma takes 20% of the tylakoid membrane surface

PSI – Photosystem

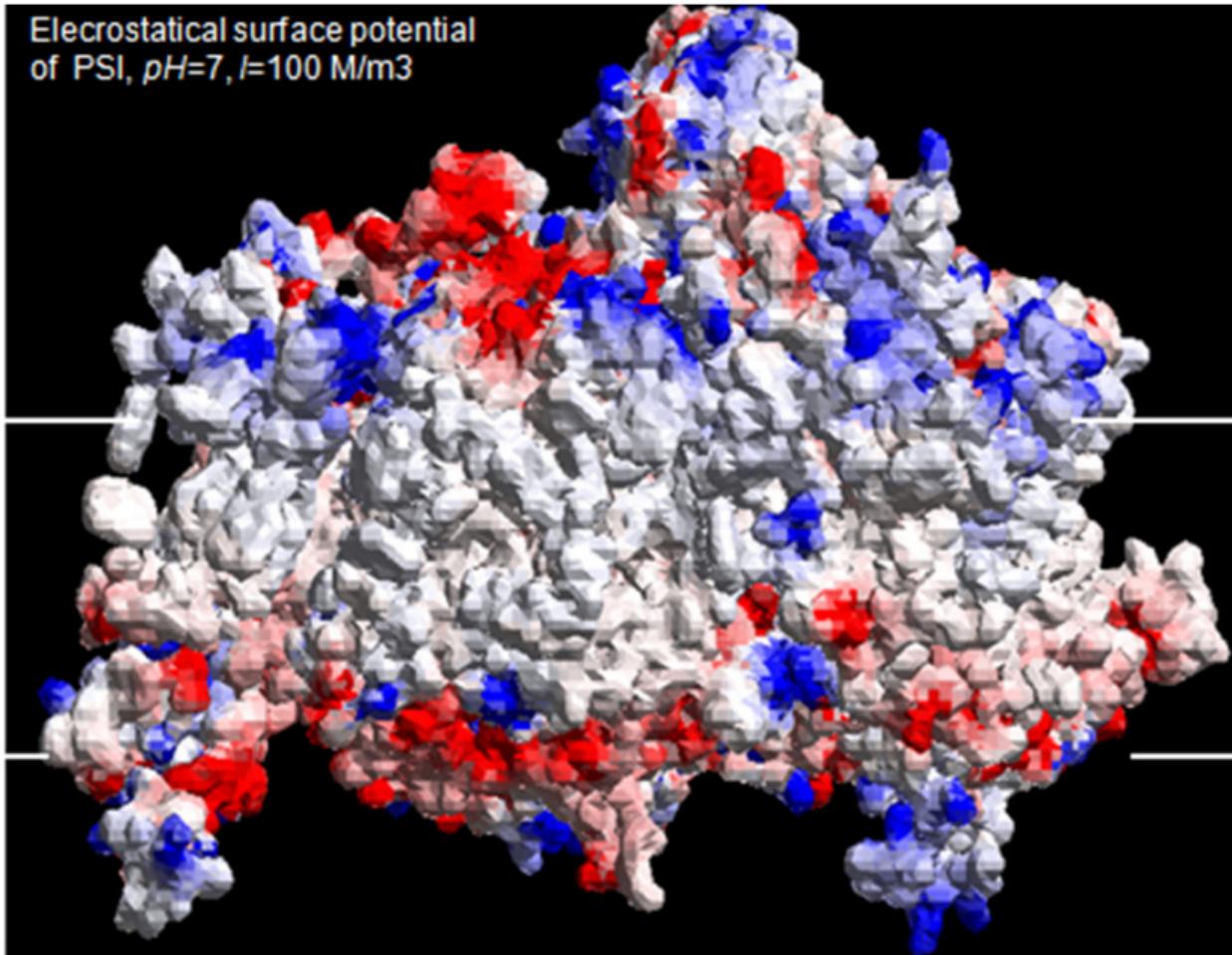
cytb6f – cytochrome b6f complex (dimer)

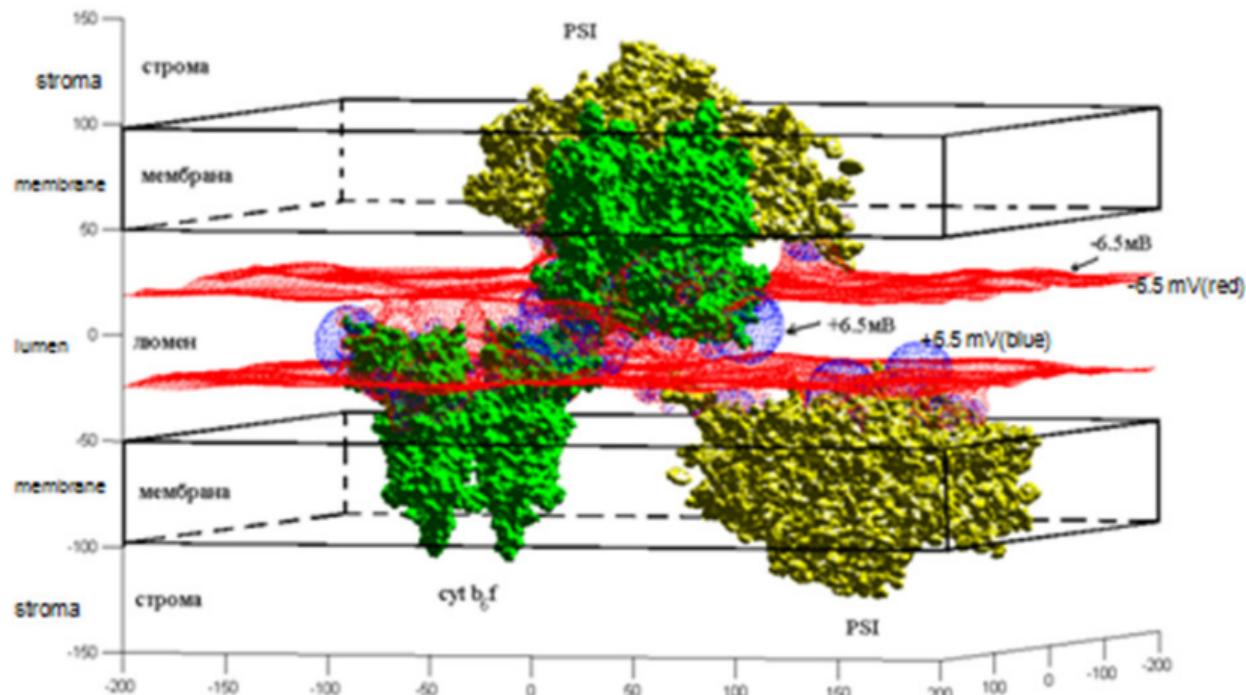
pc - plastocyanin

Electrostatic
surface potential of
cytb₆f complex,
 $pH=7, I=100 \text{ M/m}^3$



Electrostatic surface potential
of PSI, $pH=7$, $I=100$ M/m³





Equipotential surfaces (6.5 mV) in lumen of
chloroplast thylakoid, $pH=7$, $I=100 \text{ mM}$,
 $\sigma=-47.5 \text{ mQ/m}^2$

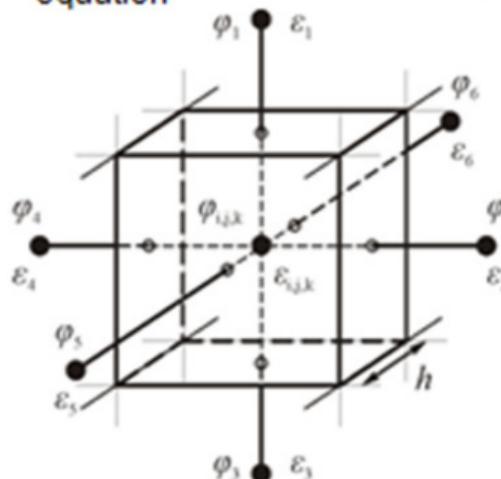
Electrostatic potential calculation

Poisson-Bolzmann equation:

$$\nabla(\varepsilon(\vec{r})\nabla\varphi(\vec{r})) = -\frac{1}{\varepsilon_0} \sum_i c_i^{\text{bulk}} e_0 z_i e^{\frac{-z_i e_0 \varphi(\vec{r})}{kT}} - \frac{1}{\varepsilon_0} (\rho_{\text{prot}} + \rho_{\text{memb}})$$

φ – potential, ε – dielectrical constant, ρ_{prot} – protein charge density, ρ_{memb} – membrane charge density, e_0 – electron charge, I – ion strength of solution, z_i – charge number, c_i^{bulk} – volume charge concentration

Linear Poisson-Bolzmann
equation



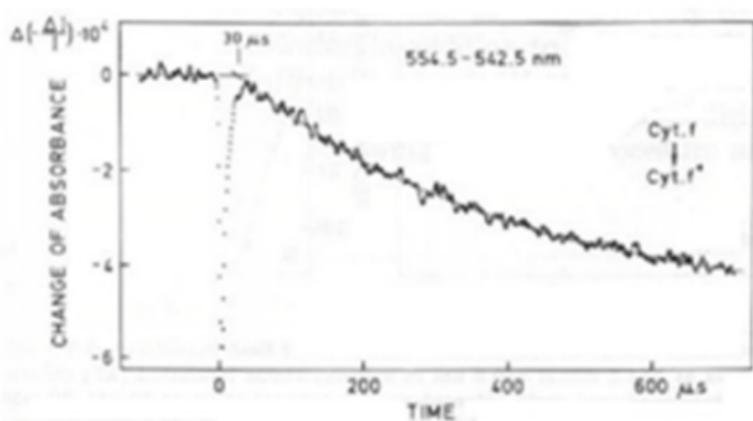
$$\nabla(\varepsilon(\vec{r})\nabla\varphi(\vec{r})) = -\frac{1}{\varepsilon_0} \left(-2Ie_0^2 \frac{\varphi(\vec{r})}{kT} + \rho_{\text{prot}} + \rho_{\text{memb}} \right)$$

$$\varphi^{n+1}_{i,j,k} = \frac{\sum_{m=1}^6 \frac{(\varepsilon_m + \varepsilon_{i,j,k})}{2} \varphi^{n-1}_m + \frac{1}{\varepsilon_0 h} (q^{\text{prot+memb}}_{i,j,k})}{\sum_{m=1}^6 \frac{(\varepsilon_m + \varepsilon_{i,j,k})}{2} + \kappa h^2}$$

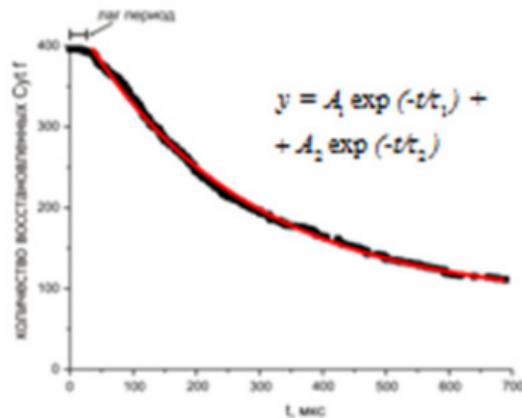
$$\kappa = \frac{2Ie_0^2}{\varepsilon_0 kT}, h \quad \text{- space step of calculation}$$

G. M. Ullmann (2004)

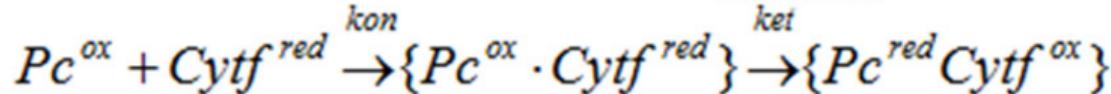
Cytochrome f oxidation after the shot light pulse



$\tau_1 \sim 101\text{--}190$ mks, $\tau_2 \sim 635\text{--}1240$ mks,
Lag-period 30–50 mks (Haehnel 1980)



$\tau_1 \sim 241$ mks, $\tau_2 \sim 1030$ mks,
Lag-period 25–30 mks
simulation



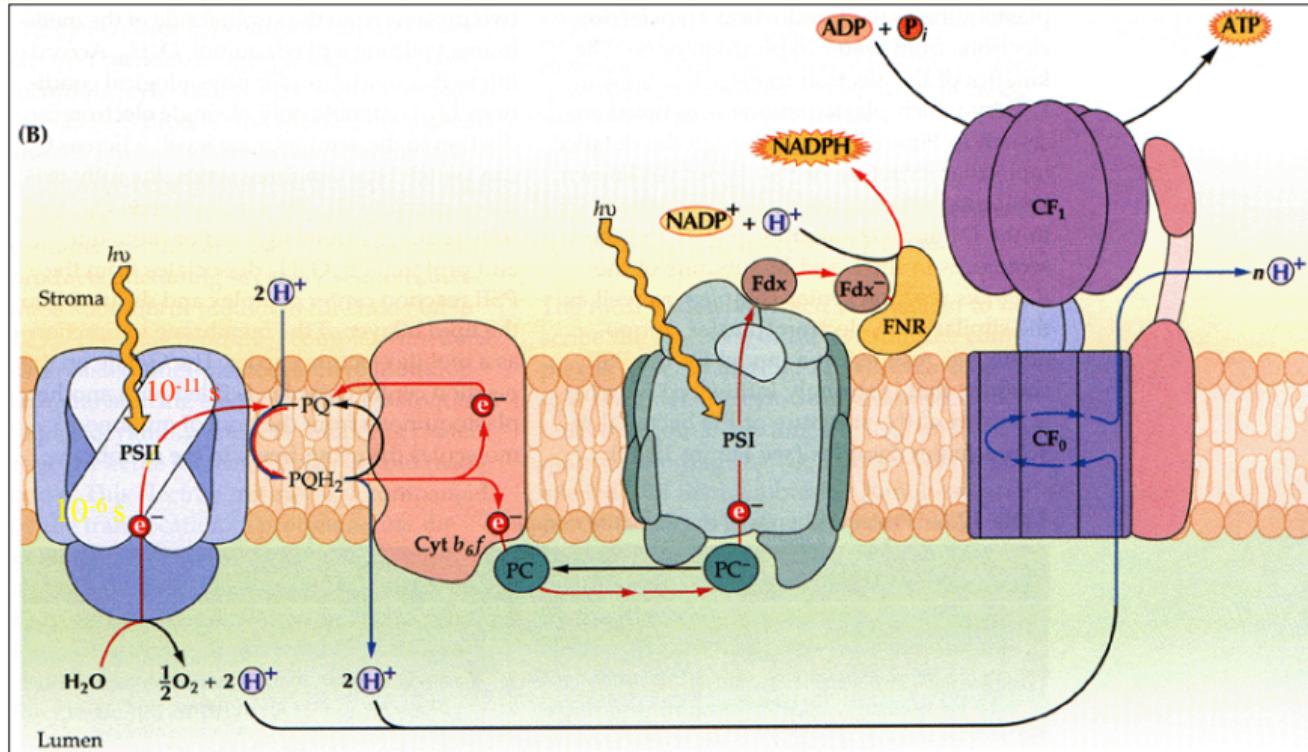
$$k_{et} = 26 \cdot 10^3 [1/c] \text{ (Hope 2000)}$$

Kovalenko, Knyazeva et al.

Мезоскопический подход

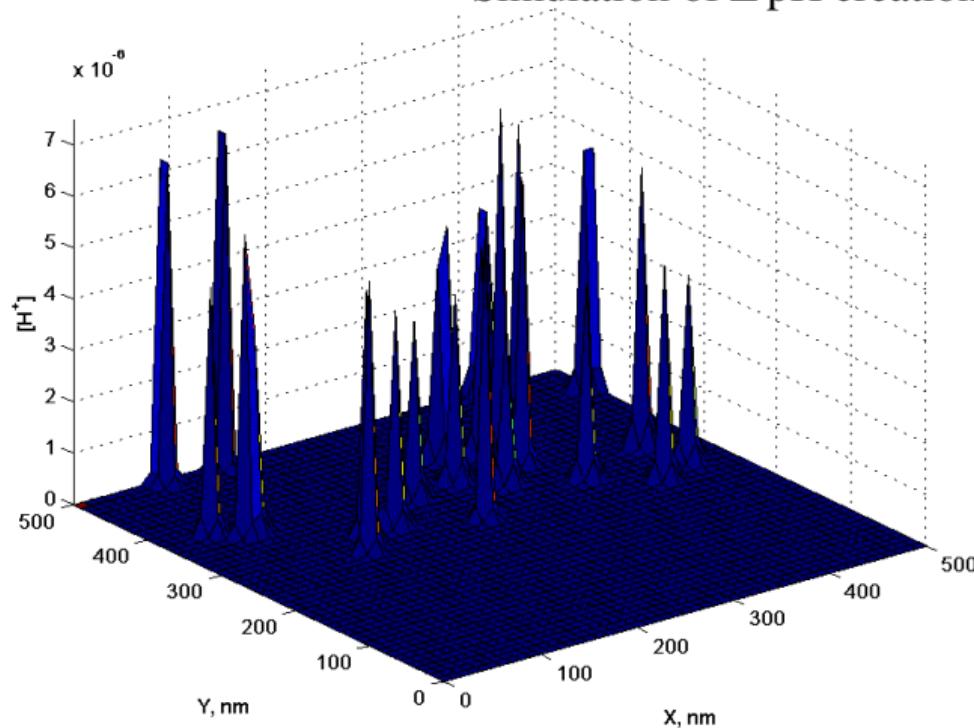
- Описание процессов внутри комплексов с помощью уравнений для вероятностей состояний
- Многочастичная Броуновская Динамика для подвижных переносчиков
- Уравнения в частных производных для распространения электрохимического потенциала в люмене.

Фотосинтетическая мембрана зеленых растений и водорослей

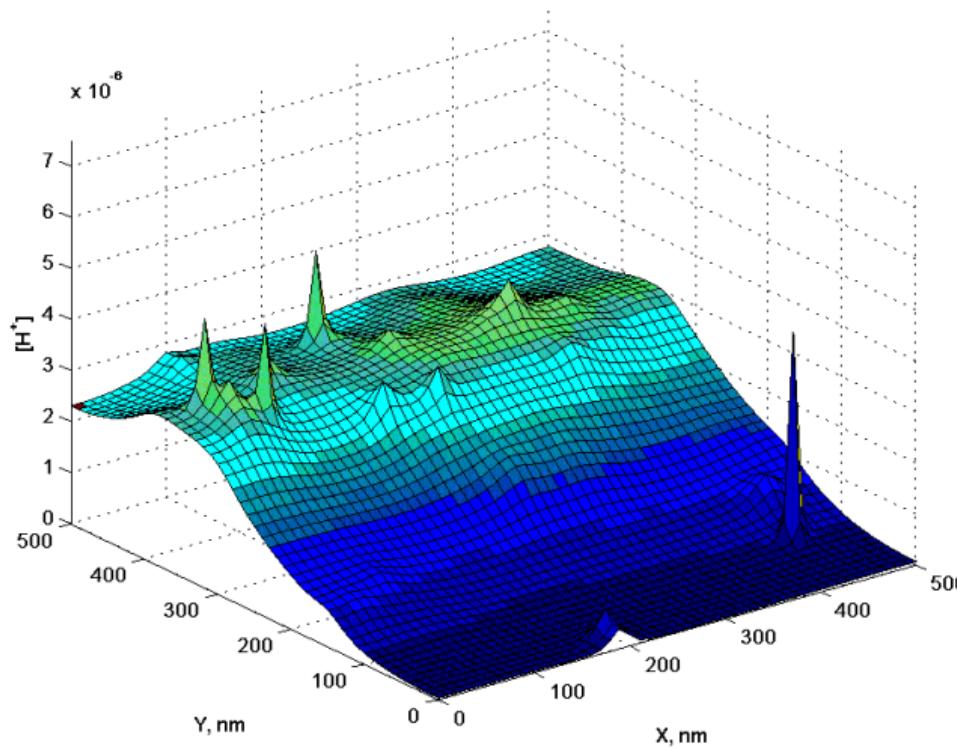


Initial profile of proton concentration on membrane surface

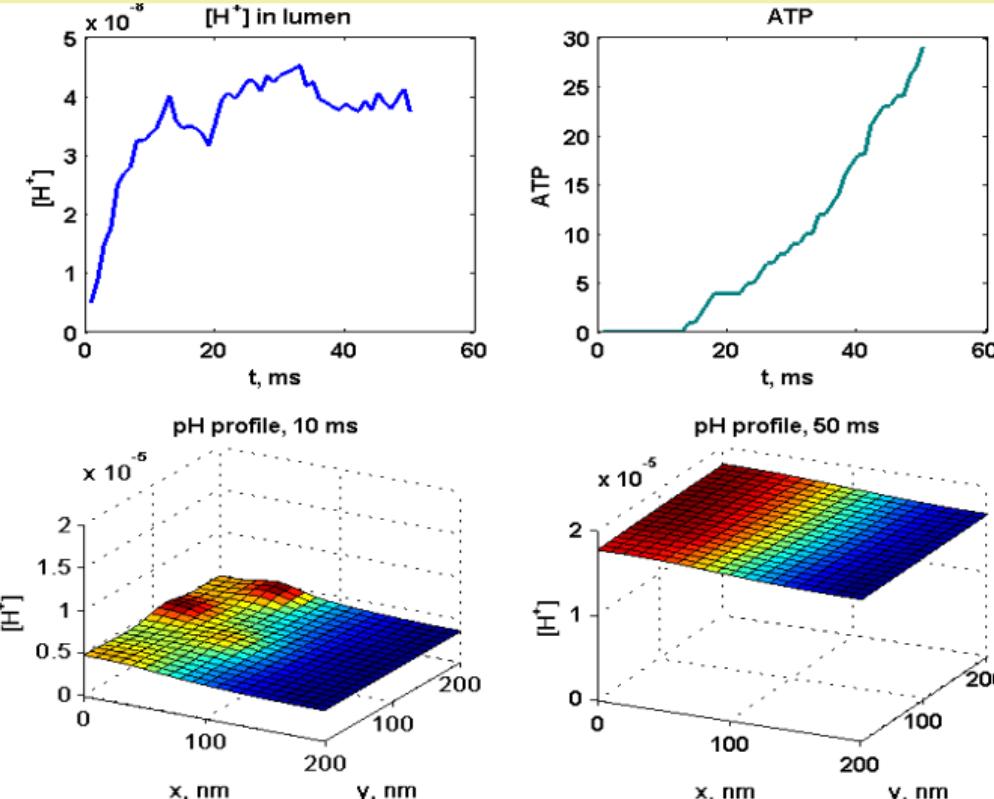
Simulation of ΔpH creation

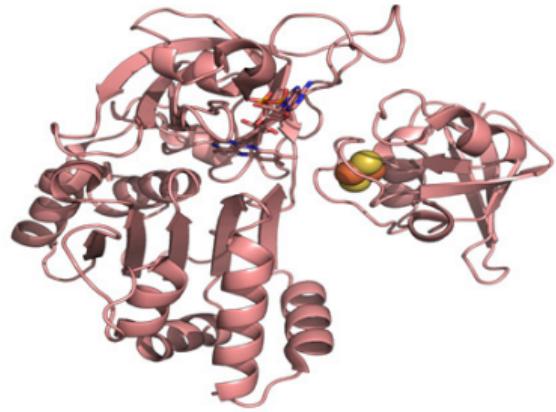
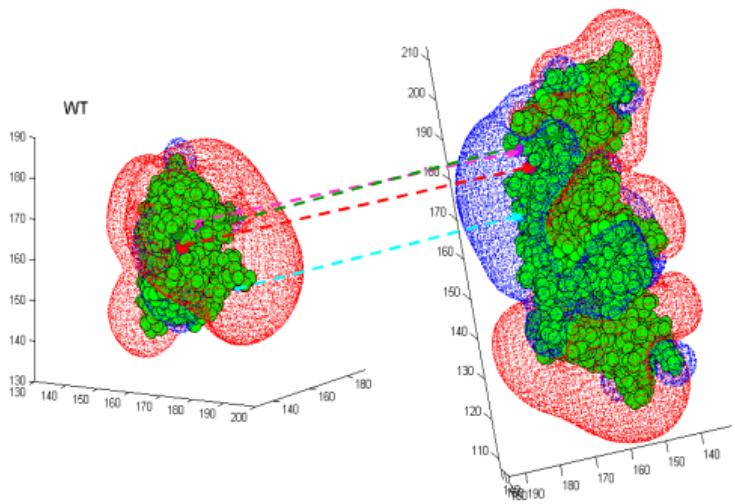


Proton concentration in lumen



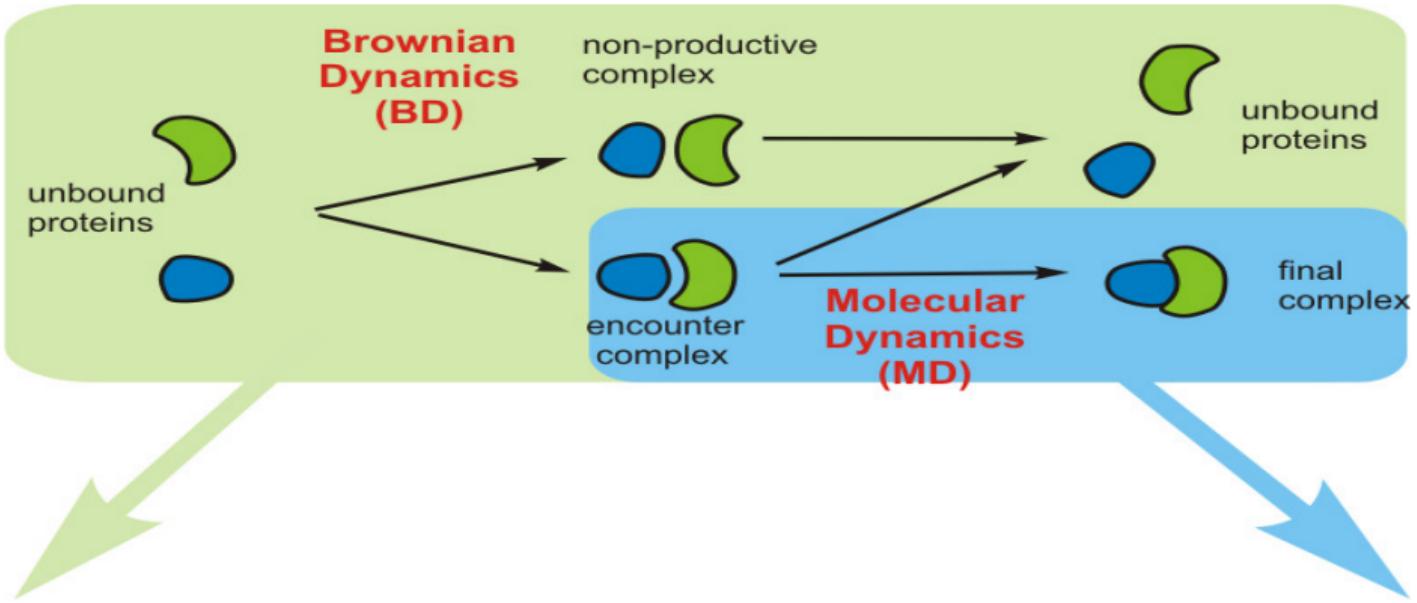
Proton concentration in lumen, ATP-formation, and pH profile





Гибридная модель Броуновская +
молекулярная динамика

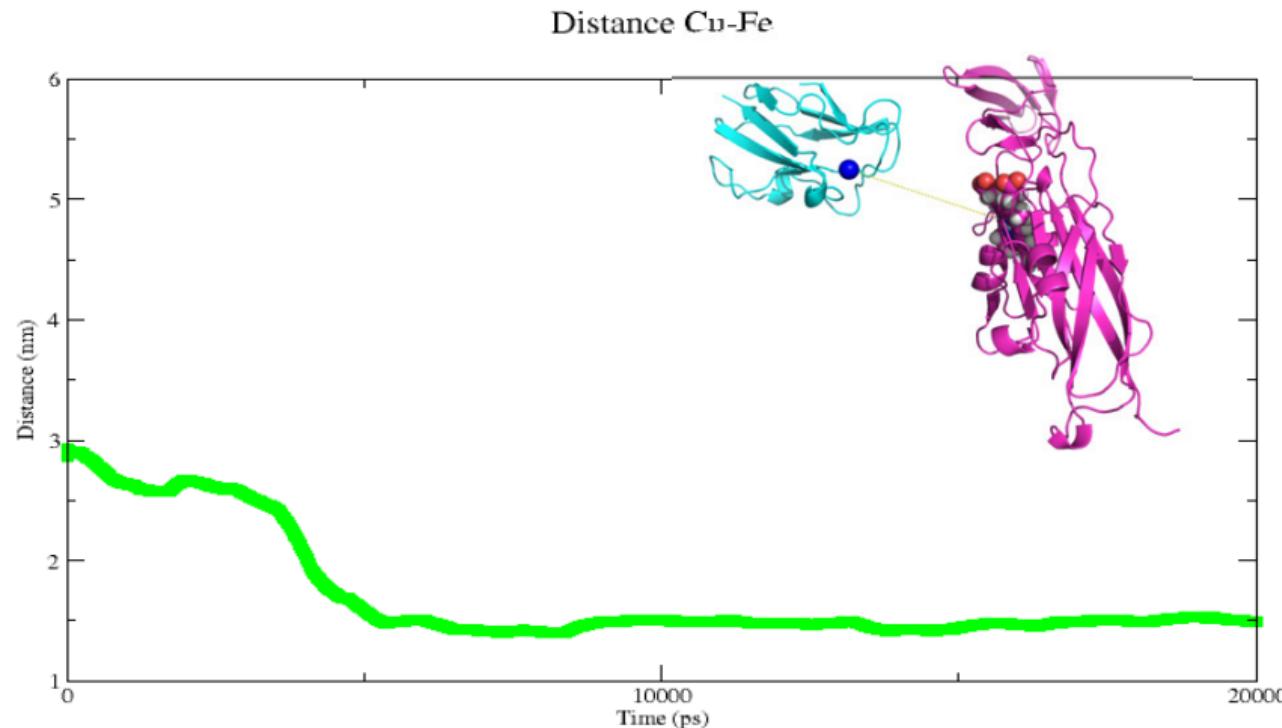
Protein-protein complex formation



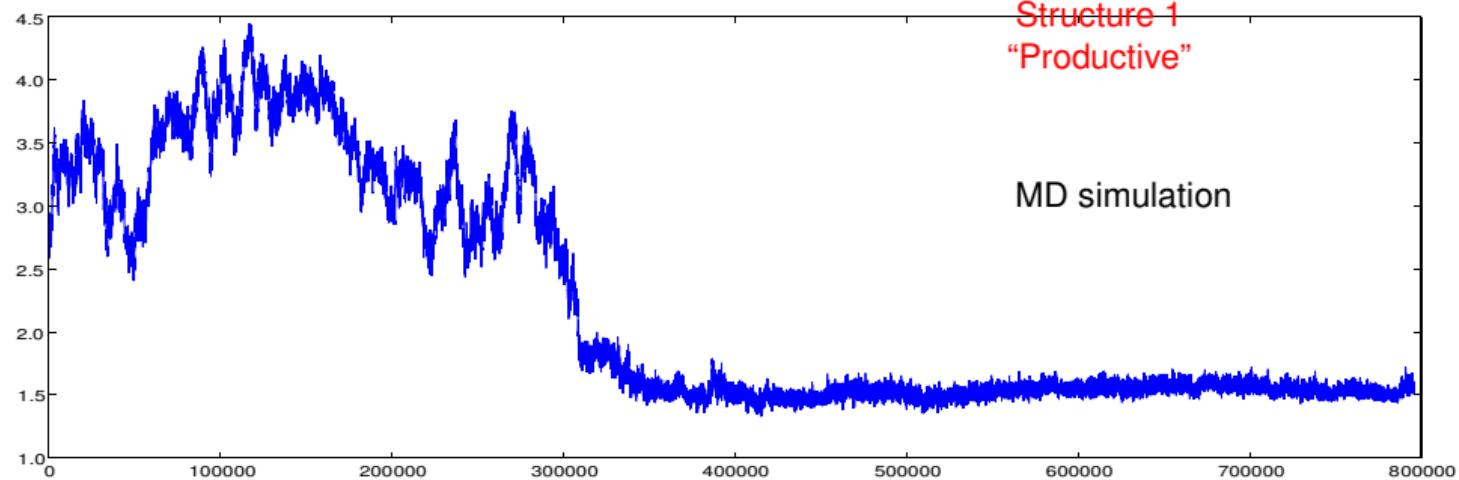
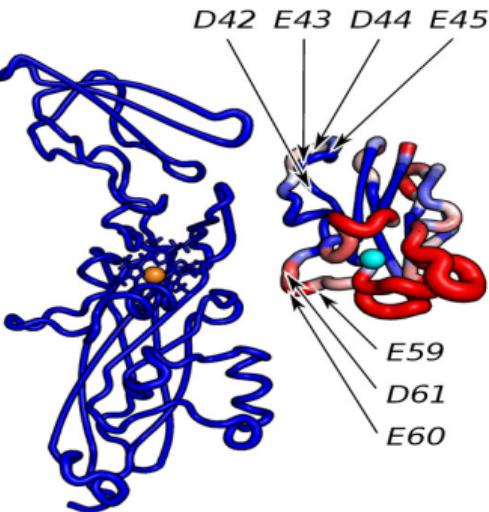
Encounter complex simulation by
Brownian Dynamics

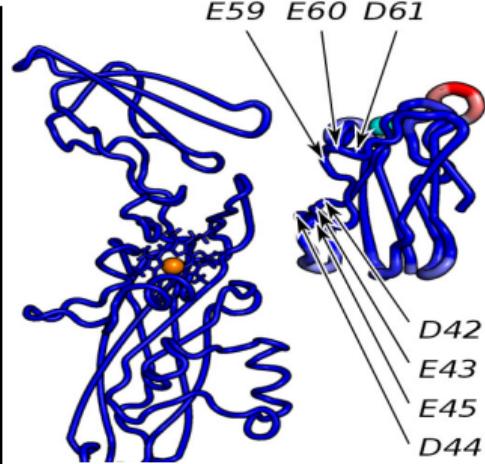
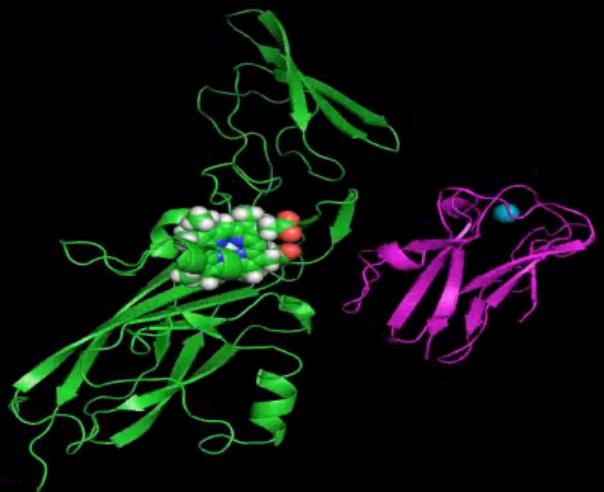
Final complex simulation by
Molecular Dynamics

Образование комплекса пластроцианина и цитохрома f



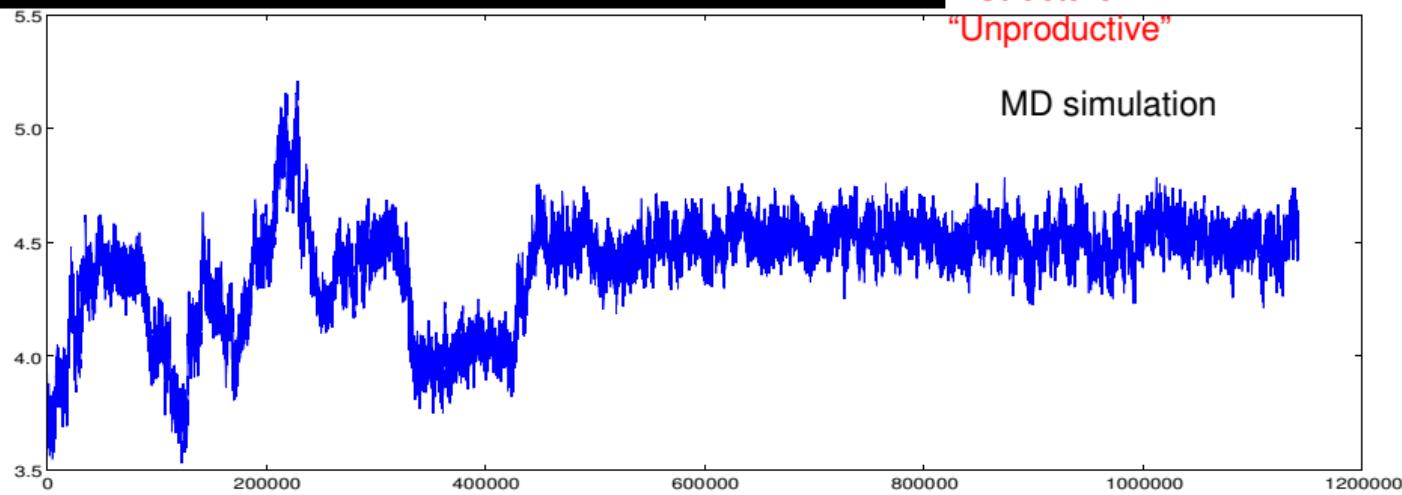
Зависимость расстояния (нм) между Cu в молекуле Pc и Fe Cyt f от времени

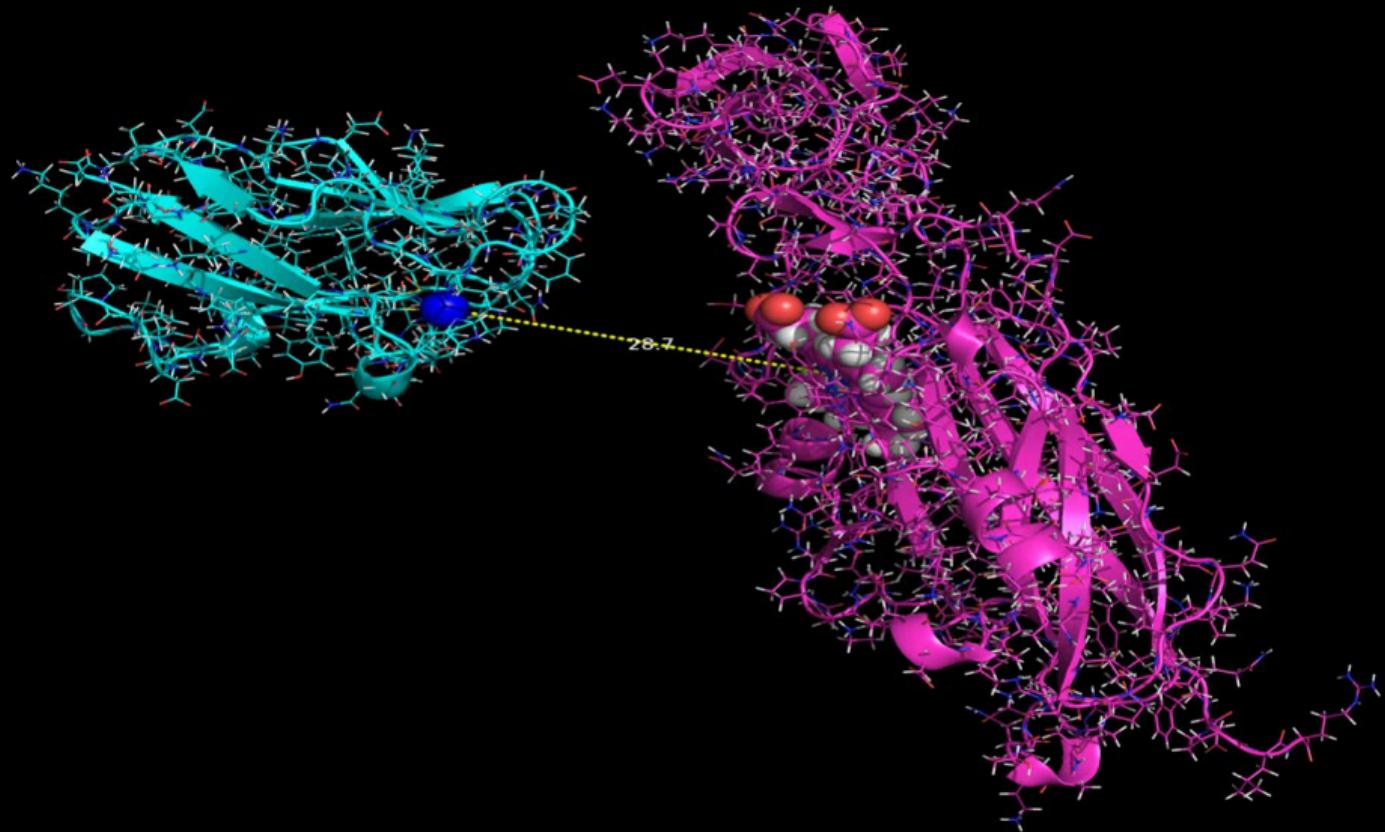




Structure 2
"Unproductive"

MD simulation





Supercomputer «Lomonosov» Moscow State University



Общая схема процессов

