
JVK
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SBS-922 Membrane Proteins

Mitochondria and respiratory chains

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Presentations
and
supplementary information

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Lectures in Membrane Proteins

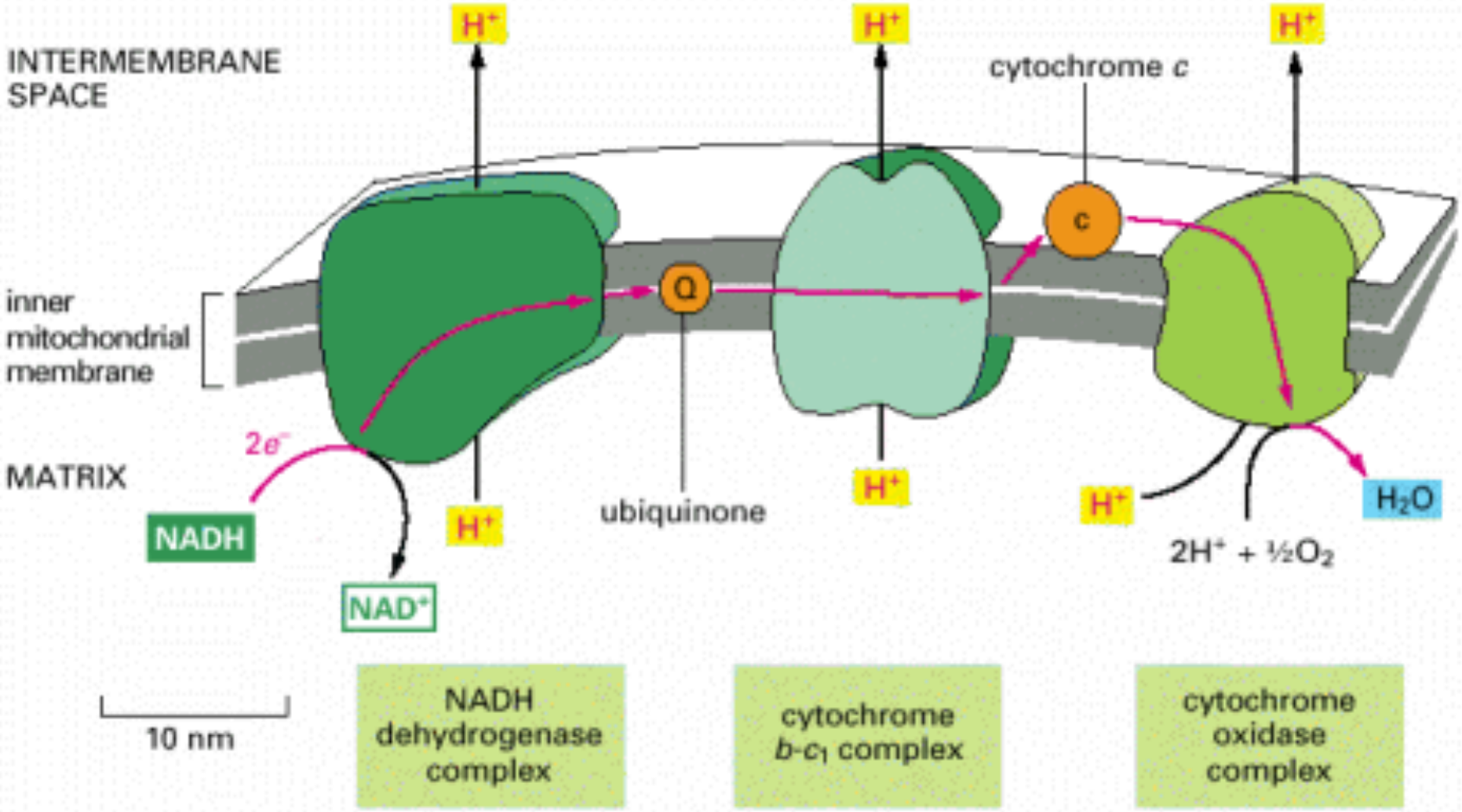
- Lecture 1. Mitochondrial membranes and chemiosmotic coupling. (Acrobat - .pdf)
- Lecture 2. Redox carriers. (Acrobat - .pdf)
- Lecture 3. Complex I. Structure and Function. Part 1 (Acrobat - .pdf)
- Lecture 4. Complex I. Part 2. Complete structure, including the hydrophobic domain. (Acrobat - .pdf)
- Lecture 5. Complex II and complex III Part 1. Structure and Function. (Acrobat - .pdf)
- Lecture 6. Complex III. The Q-cycle. (Acrobat - .pdf)
- Lecture 7. ATP Synthase. (Acrobat - .pdf)

Membrane Proteins

- Membrane Proteins course web page

References

- Nicholls DJ, Ferguson, SJ. Bioenergetics3. Academic Press/Elsevier Science 2002
- Molecular Biology of the Cell. Bruce Alberts, Alexander Johnson, Julian Lewis, Martin Raff, Keith Roberts, and Peter Walter. 5th edition. 2007. Garland Science. 4th Edition online at NCBI...

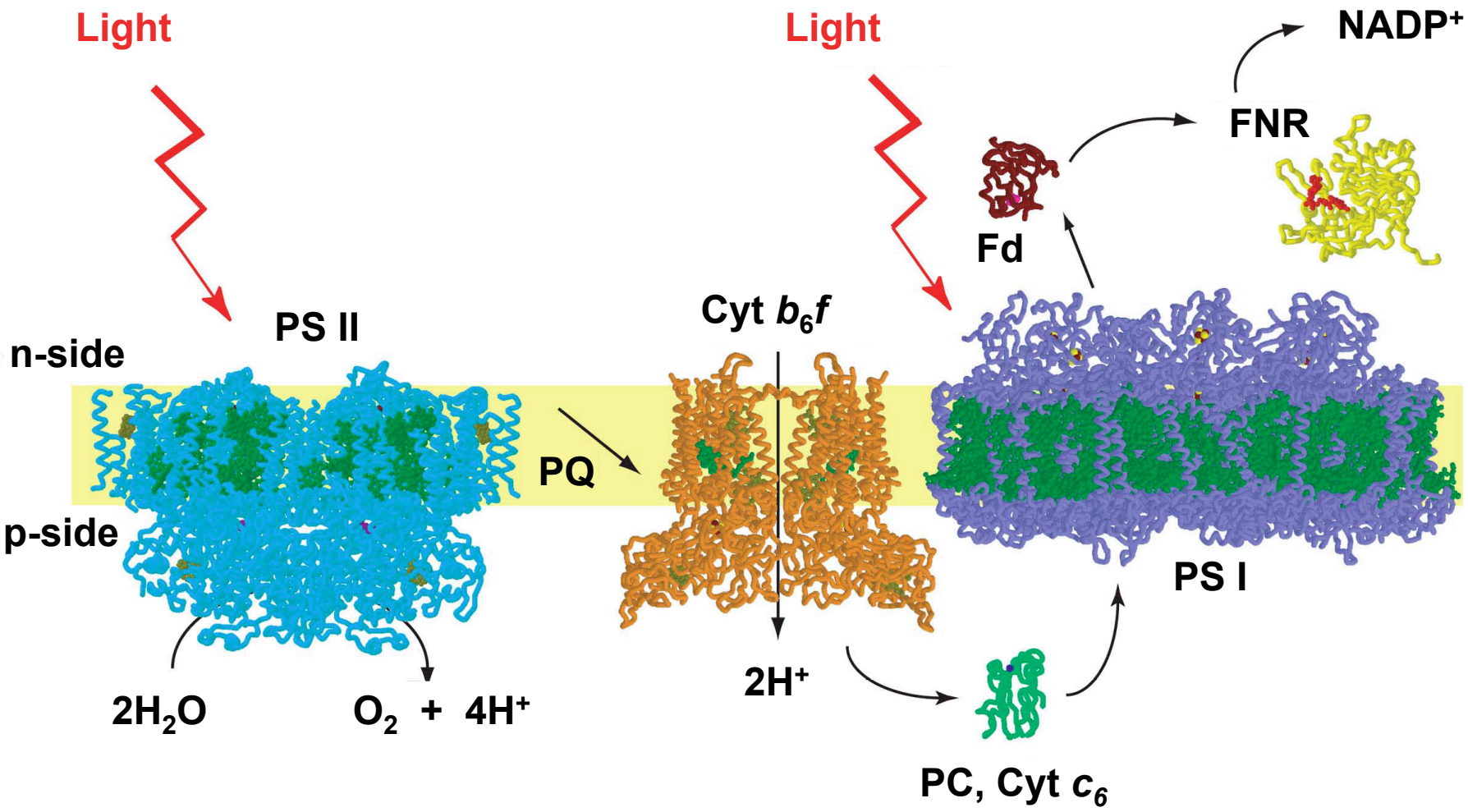


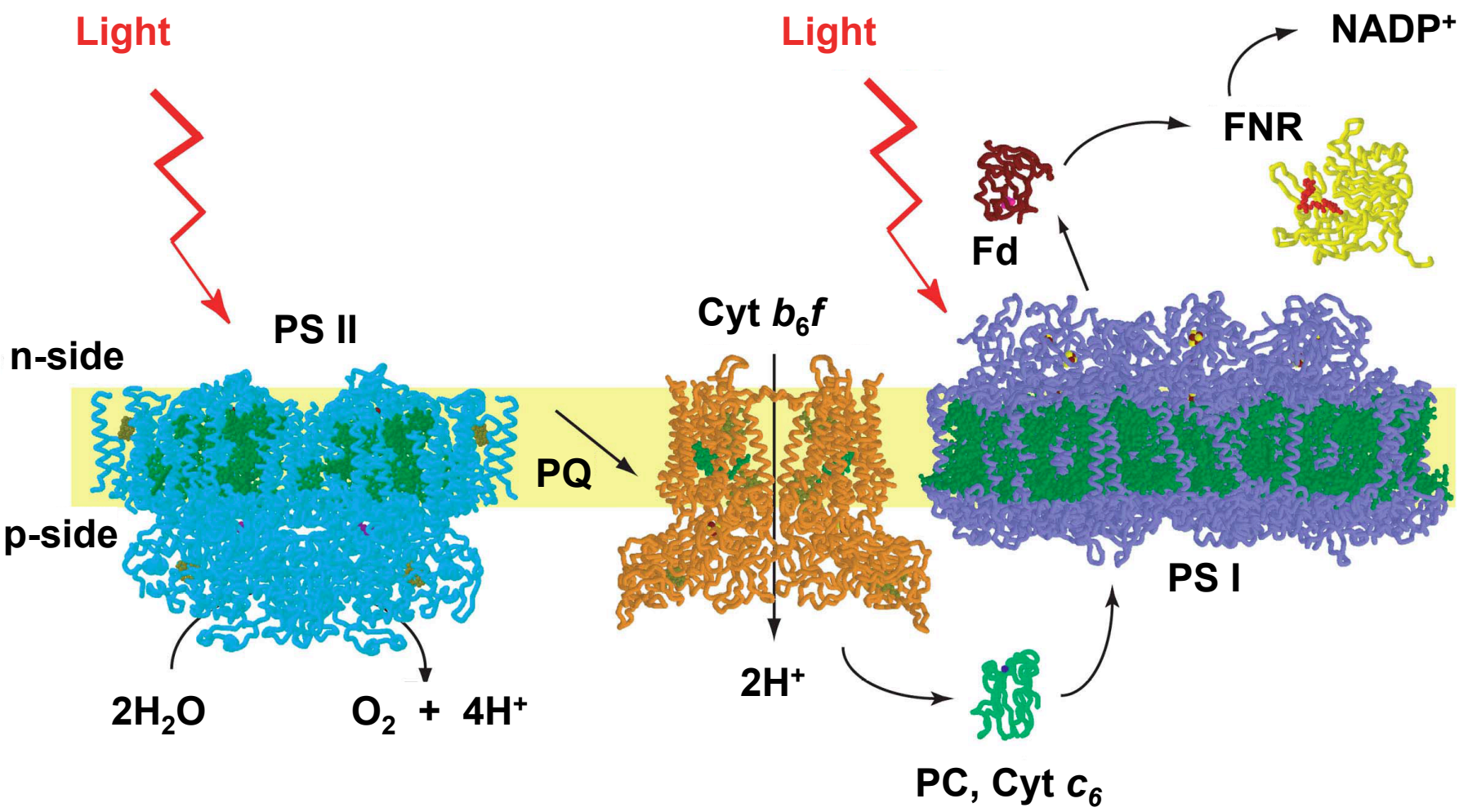
The Respiratory Chain Includes Three Large Enzyme Complexes Embedded in the Inner Membrane

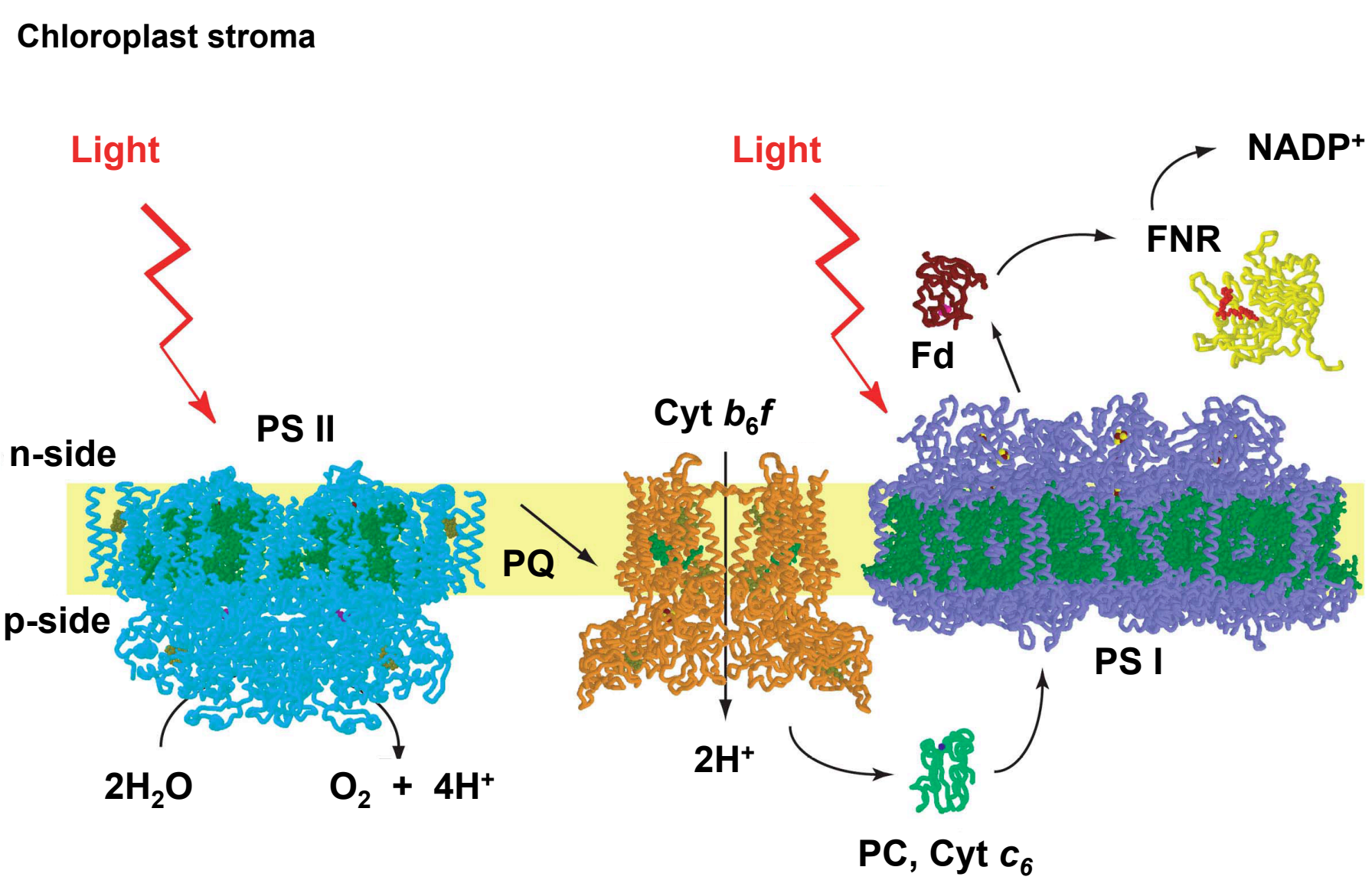
Molecular Biology of the Cell Bruce Alberts, Alexander Johnson, Julian Lewis, Martin Raff, Keith Roberts, and Peter Walter. 2007

The chloroplast homologue of respiratory complex III:

The cytochrome b_6f complex


Light
Light
NADP+
FNR
Fd
PS II
n-side
Cyt b6f
PQ
p-side
PS I
2H2O
O2 + 4H+
2H+
PC, Cyt c6


Chloroplast stroma
Light
Light
NADP+
FNR
Fd
PS II
Cyt b6f
n-side
PQ
p-side
PS I
2H2O
O2 + 4H+
2H+
PC, Cyt c6


Chloroplast stroma
Light
Light
NADP+
FNR
Fd
PS II
n-side
Cyt b_6f
PS I
PQ
p-side
2H+
2H2O
O2 + 4H+
PC, Cyt c_6
Chloroplast lumen

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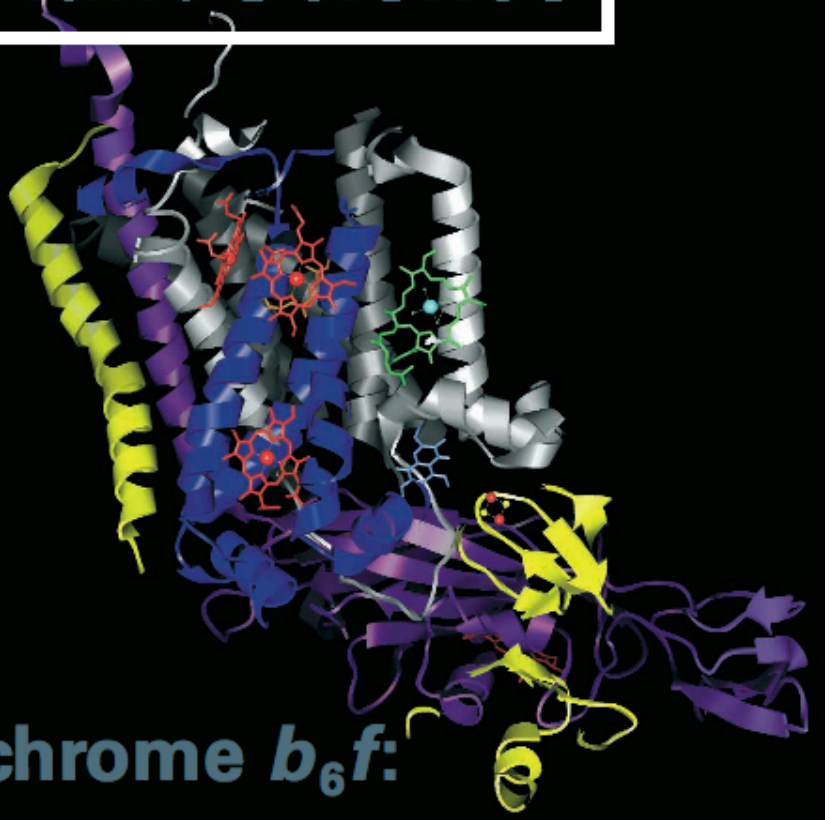
TRENDS in Plant Science



Cytochrome b_6f: bridging the divide

Pollen aperture evolution

GABA in plants: just a metabolite?

Mitochondrial carriers in *Arabidopsis*

Trends Plant Sci. March 2004 Vol. 9 No. 3, pp. xxx–xxx ISSN 1360-1385

Chemiosmosis and the Q-cycle

At about the same time as the Z-scheme, a common mechanism for photosynthetic and oxidative phosphorylation was proposed. Peter Mitchell's chemiosmotic theory [6] made several radical assumptions. One was that electron and hydrogen transfers are arranged vectorially across bioenergetic membranes, thus moving protons (hydrogen ions) across the membrane to establish an electrochemical gradient. Another cornerstone was the idea that this gradient, or 'proton motive force', supplies energy for ATP synthesis, because this, too, is coupled to vectorial movement of protons between aqueous phases on each side of the membrane [7,8].

The Q-cycle was a later addition [9,10] designed initially to explain higher observed proton-to-electron ratios than the original chemiosmotic theory seemed to predict [11]. Simple inspection of quinone redox chemistry suggested two electrons move only two protons (Equation 1).

$$Q + 2e^- + 2H^+ \rightarrow QH_2 \rightarrow Q + 2H^+ + 2e^- \qquad \text{(Eqn 1)}$$

At the Q_i site:

$$Q + \underline{1e^-} \rightarrow Q\cdot^- \qquad \text{(Eqn 2)}$$

$$Q\cdot^- + 2H^+_{[in]} + 1e^- \rightarrow QH_2 \qquad \text{(Eqn 3)}$$

At the Q_i site:

$$Q + \underline{1e^-} \rightarrow Q\cdot^- \qquad \text{(Eqn 2)}$$

$$Q\cdot^- + 2H^+_{[in]} + 1e^- \rightarrow QH_2 \qquad \text{(Eqn 3)}$$

At the Q_o site:

$$QH_2 \rightarrow Q\cdot^- + 1e^- + 2H^+_{[out]} \qquad \text{(Eqn 4)}$$

$$Q\cdot^- \rightarrow Q + \underline{1e^-} \qquad \text{(Eqn 5)}$$

At the Q_i site:

$$Q + \underline{1e^-} \rightarrow Q\cdot^- \qquad \text{(Eqn 2)}$$

$$Q\cdot^- + 2H^+_{[in]} + 1e^- \rightarrow QH_2 \qquad \text{(Eqn 3)}$$

At the Q_o site:

$$QH_2 \rightarrow Q\cdot^- + 1e^- + 2H^+_{[out]} \qquad \text{(Eqn 4)}$$

$$Q\cdot^- \rightarrow Q + \underline{1e^-} \qquad \text{(Eqn 5)}$$

Sum, for one electron transferred (Equation 6):

$$2H^+_{[in]} \rightarrow 2H^+_{[out]} \qquad \text{(Eqn 6)}$$

At the Q_i site:

$$Q + \underline{1e^-} \rightarrow Q\cdot^- \qquad \text{(Eqn 2)}$$

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$$Q\cdot^- \rightarrow Q + \underline{1e^-} \qquad \text{(Eqn 5)}$$

Sum, for one electron transferred (Equation 6):

$$2H^+_{[in]} \rightarrow 2H^+_{[out]} \qquad \text{(Eqn 6)}$$

The effect is to re-cycle one of the electrons from plastoquinol (PQH_2, also called 'plastohydroquinone'). This electron (underlined in Equations 2 and 5) is supplied by the plastosemiquinone anion intermediate ($PQ\cdot^-$) at the Q_o site, to a short chain of two *b*-haems, and given back to plastosemiquinone at the Q_i site. The whole process gives two protons translocated for each single electron transferred through the quinone pool, that is, a total of four protons, not two, for each pair of electrons passing through the quinone pool and the cytochrome complex (Equation 7).

At the Q_i site:

$$Q + \underline{1e^-} \rightarrow Q\cdot^- \qquad \text{(Eqn 2)}$$

$$Q\cdot^- + 2H^+_{[in]} + 1e^- \rightarrow QH_2 \qquad \text{(Eqn 3)}$$

At the Q_o site:

$$QH_2 \rightarrow Q\cdot^- + 1e^- + 2H^+_{[out]} \qquad \text{(Eqn 4)}$$

$$Q\cdot^- \rightarrow Q + \underline{1e^-} \qquad \text{(Eqn 5)}$$

Sum, for one electron transferred (Equation 6):

$$2H^+_{[in]} \rightarrow 2H^+_{[out]} \qquad \text{(Eqn 6)}$$

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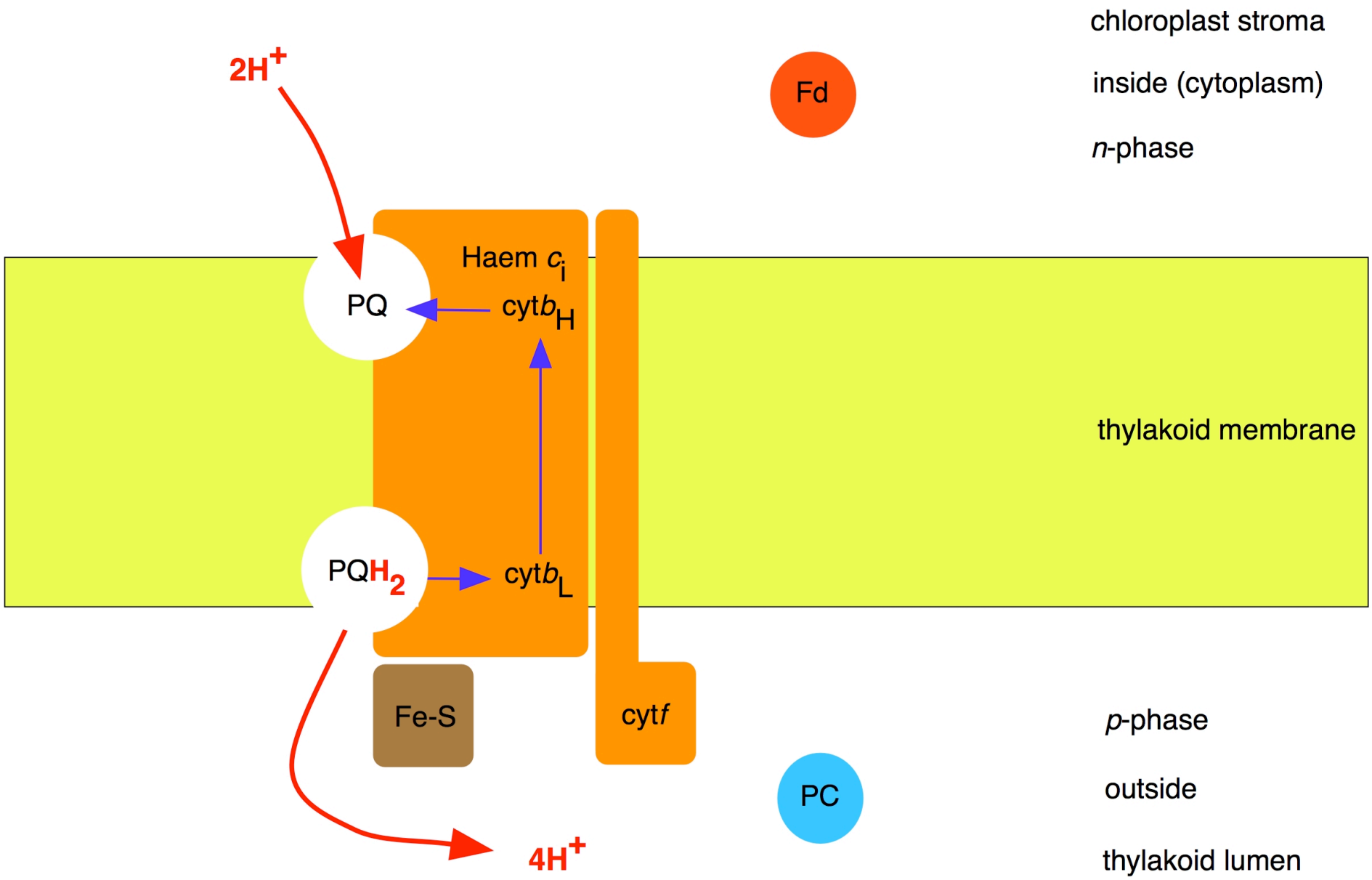
$$Q + 2e^- + 4H^+_{[in]} \rightarrow QH_2 + [2H^+] \rightarrow Q + 4H^+_{[out]} + 2e^- \qquad \text{(Eqn 7)}$$

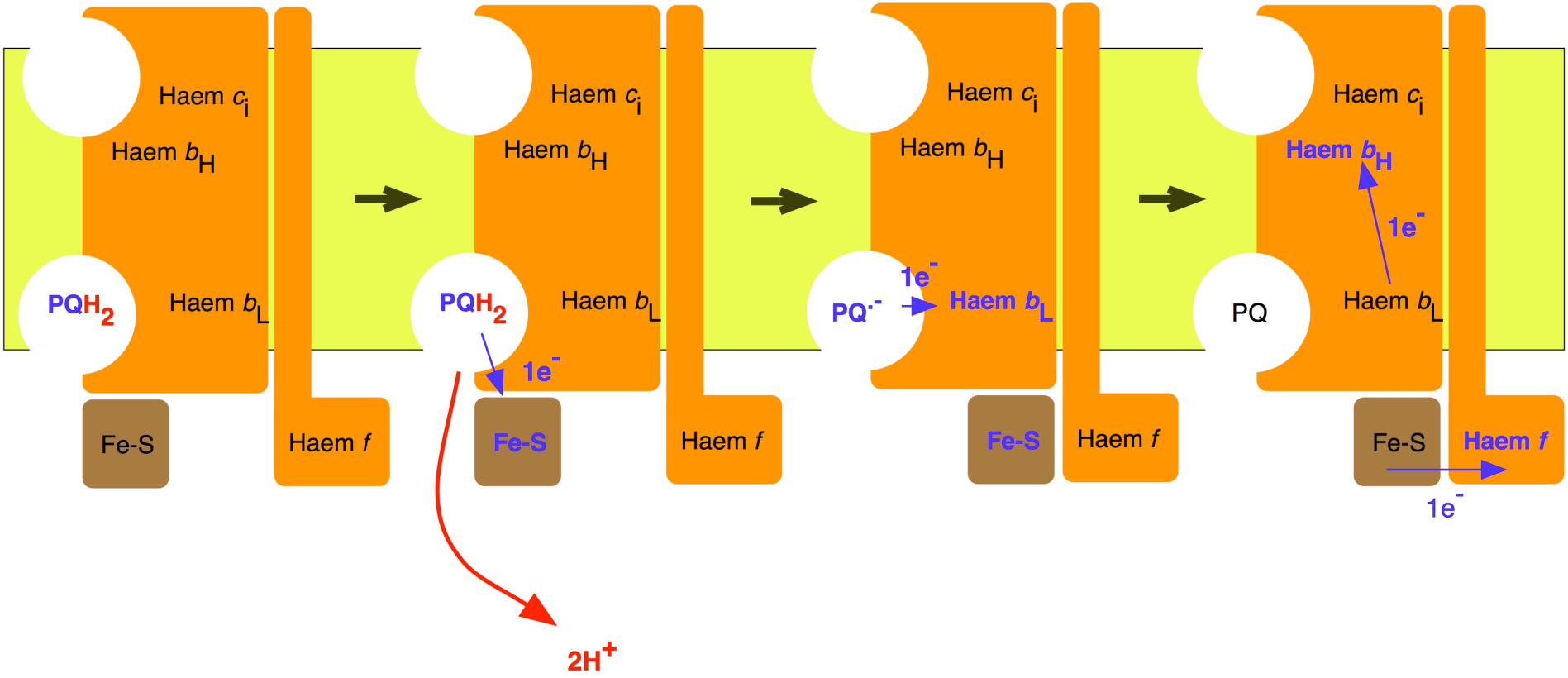

chloroplast stroma
inside (cytoplasm)
n-phase
2H+
Fd
Haem ci
PQ
cytbH
thylakoid membrane
PQH2
cytbL
Fe-S
cytf
p-phase
outside
PC
4H+
thylakoid lumen

At the Q_o site

Each plastoquinol (PQH_2) oxidation releases two protons (H^+) into the lumen.

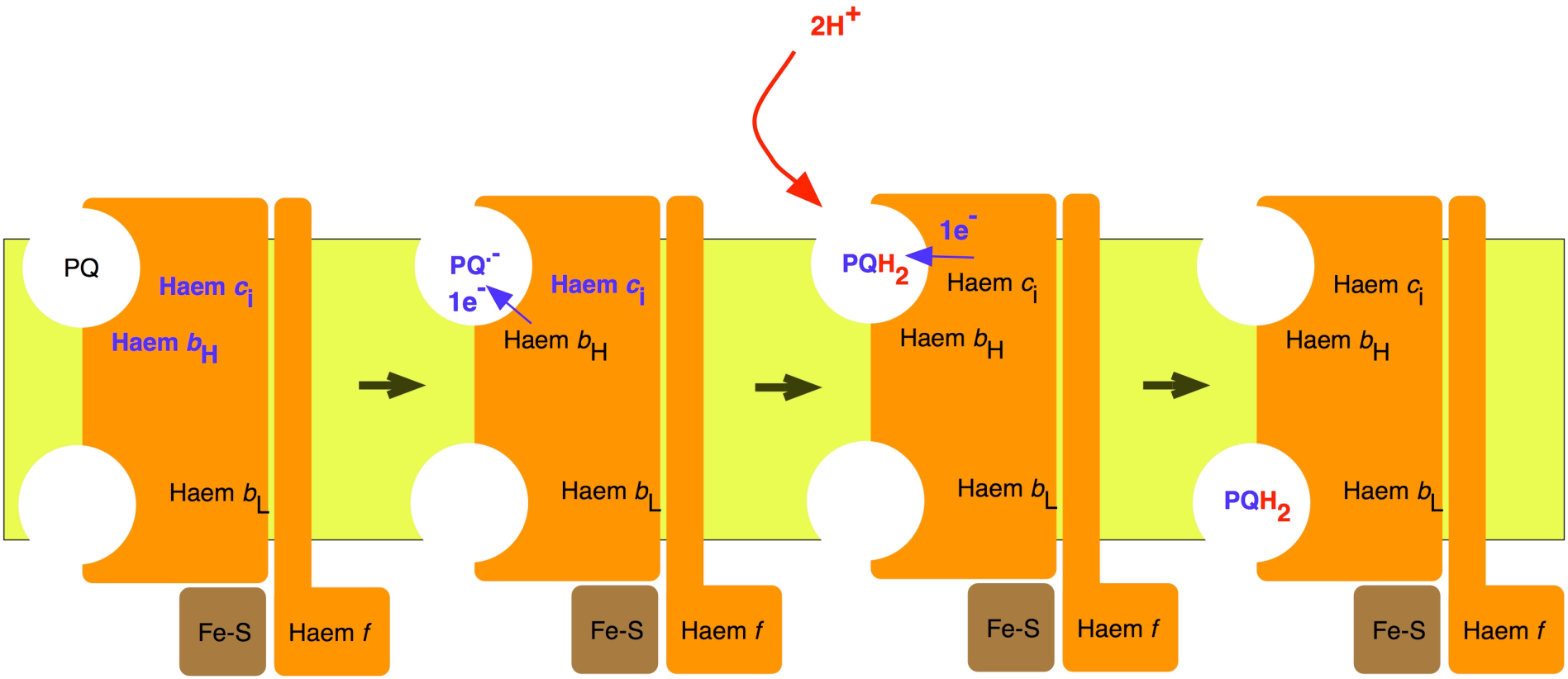
One electron (e^-) passes to cytochrome *f*. The other electron passes towards the Q_i site, via the cytochrome *b* haems.

One PQH_2 arrives from the Q_i site; another PQH_2 arrives from photosystem II.



At the Q_i site

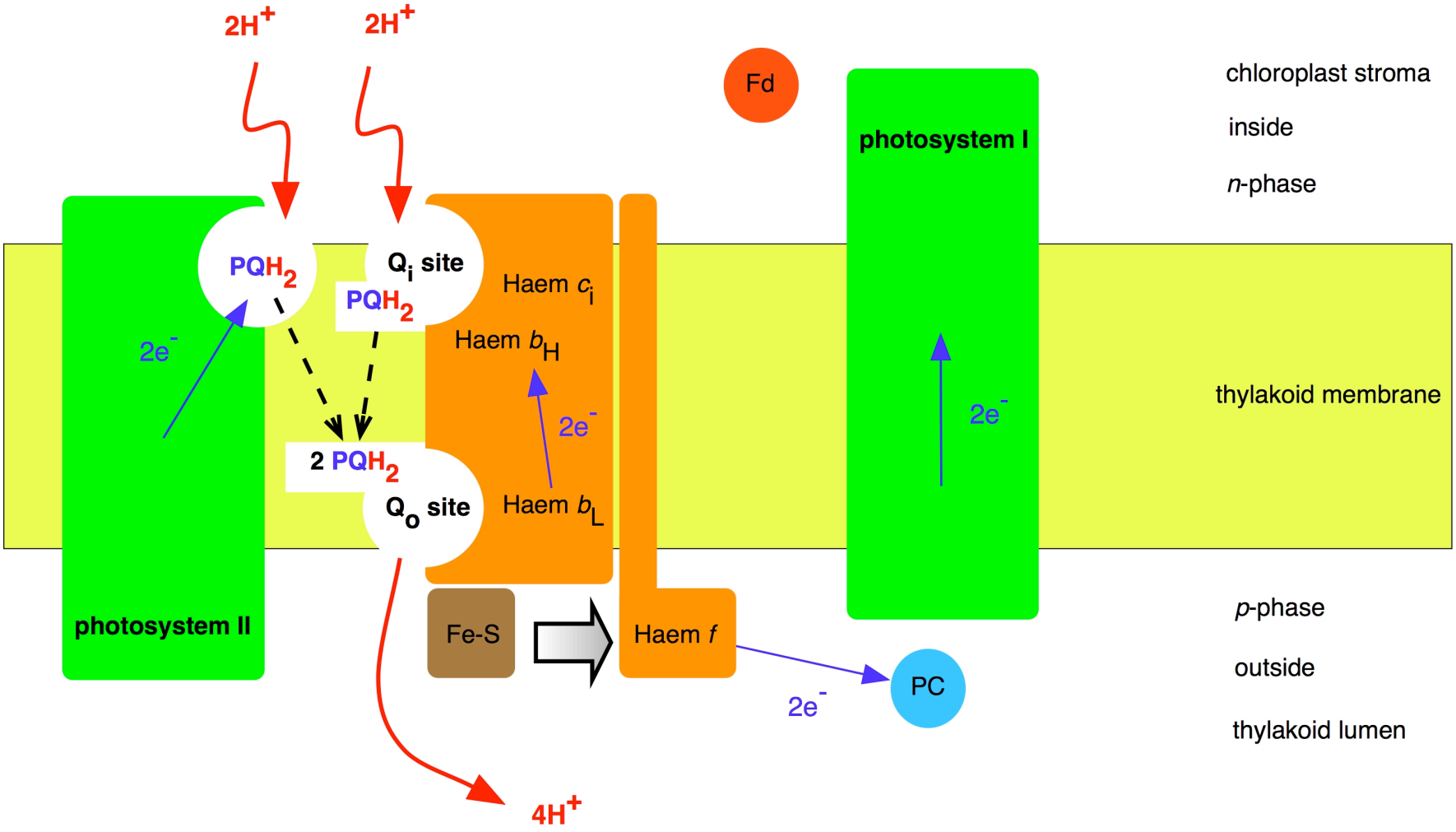
Each plastoquinone (PQ) reduction gives one plastoquinol (PQH_2), and two protons (H^+) are taken up from the stroma.

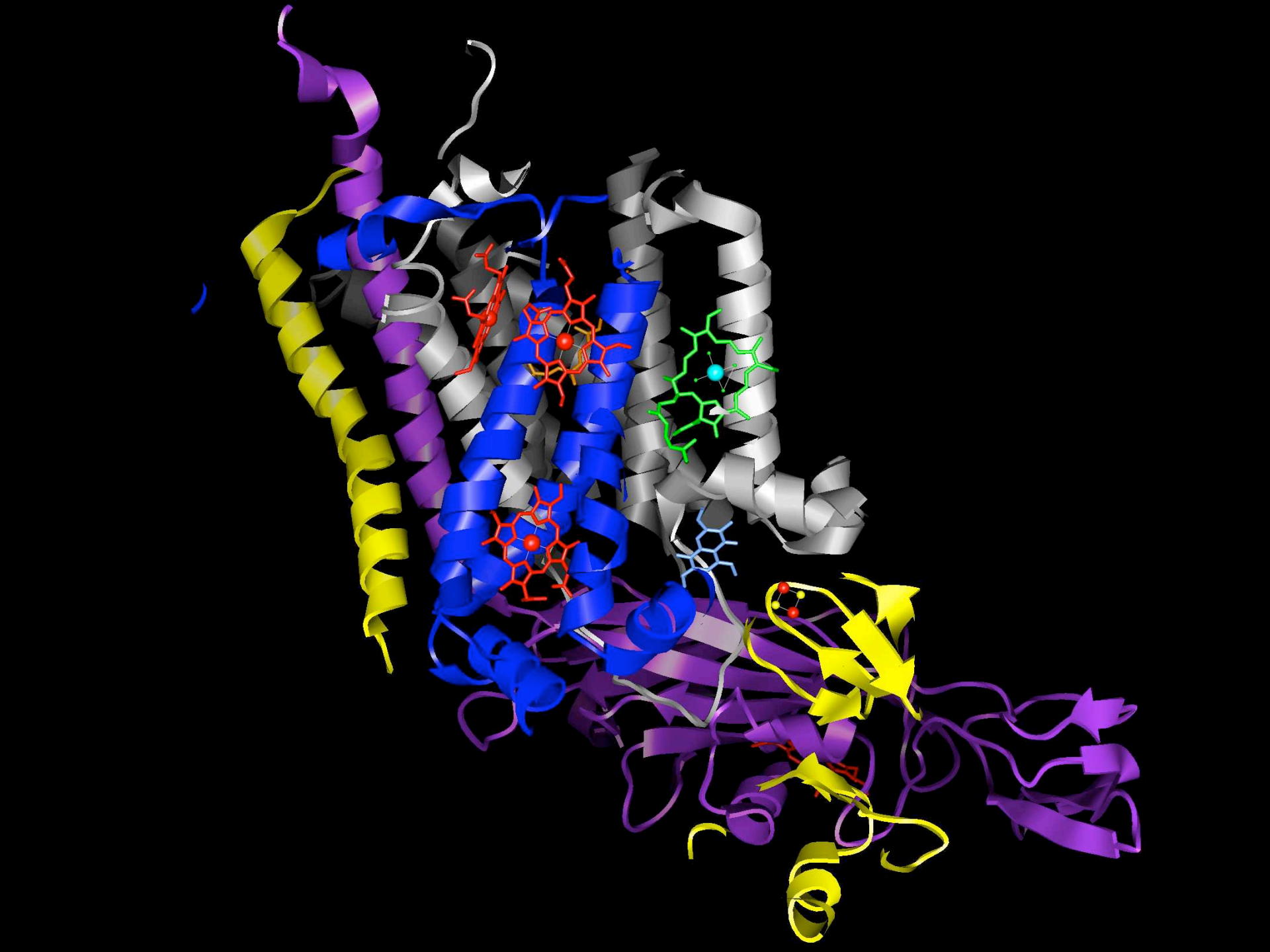


Cytochrome b_6f connects photosystems I with photosystem II and carries out the Q-cycle

Plastoquinone (PQ) reduction by two electrons (e^-) from photosystem II gives one molecule of plastoquinol (PQH_2).
There are two PQH_2 oxidations at the Q_o site for each PQ reduction at the Q_i site, and for each PQ reduction in photosystem II .

Four protons (H^+) are therefore translocated from the stroma to the lumen for each pair of electrons passing through cytochrome b_6f.





An Electronic Bus Bar Lies in the Core of Cytochrome bc_1

Monika Świerczek,[1] Ewelina Cieluch,[1] Marcin Sarewicz,[1] Arkadiusz Borek,[1] Christopher C. Moser,[2] P. Leslie Dutton,[2] Artur Osyczka[1]*

The ubiquinol–cytochrome c oxidoreductases, central to cellular respiration and photosynthesis, are homodimers. High symmetry has frustrated resolution of whether cross-dimer interactions are functionally important. This has resulted in a proliferation of contradictory models. Here, we duplicated and fused cytochrome b subunits, and then broke symmetry by introducing independent mutations into each monomer. Electrons moved freely within and between monomers, crossing an electron-transfer bridge between two hemes in the core of the dimer. This revealed an H-shaped electron-transfer system that distributes electrons between four quinone oxidation-reduction terminals at the corners of the dimer within the millisecond time scale of enzymatic turnover. Free and unregulated distribution of electrons acts like a molecular-scale bus bar, a design often exploited in electronics.

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[2]The Johnson Research Foundation, Department of Biochemistry and Biophysics, University of Pennsylvania, PA 19104, USA.

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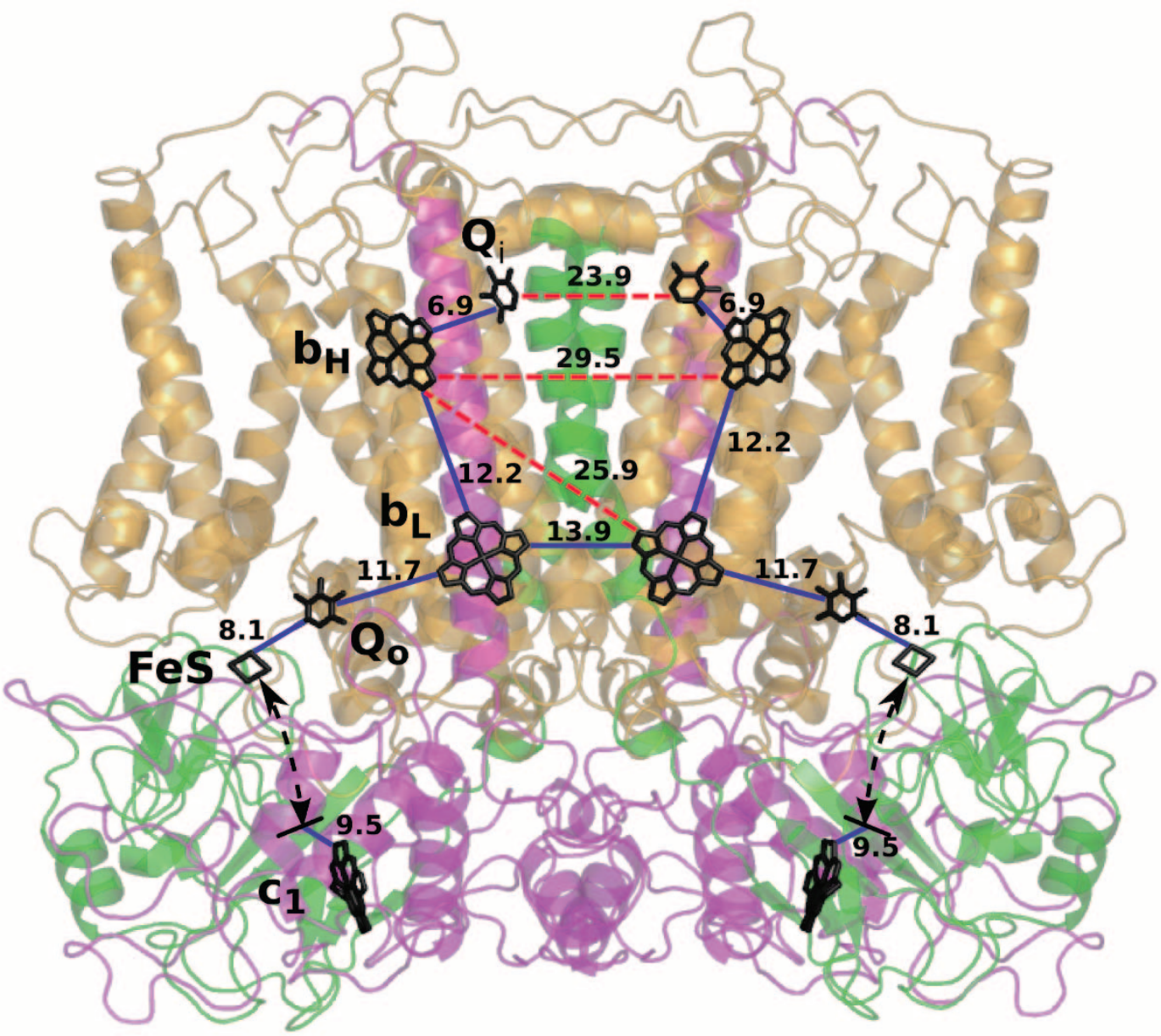


Fig. 1. Cofactors and distances in homodimer of cytochrome bc_1 [Protein Data Bank ID: 1ZRT (*1*)]. Each monomer comprises cytochrome b (yellow), cytochrome c_1 (magenta), and FeS subunit (green). Functional distances (blue lines) and nonfunctional distances (red dashed lines) between cofactors (black) are in angstroms. Q_o site quinone is approximated from the crystallographic position of stigmatellin (*1*), and Q_i site quinone position is adopted from (*28*). FeS head domain movement (*29*) is indicated by the dashed arrow.

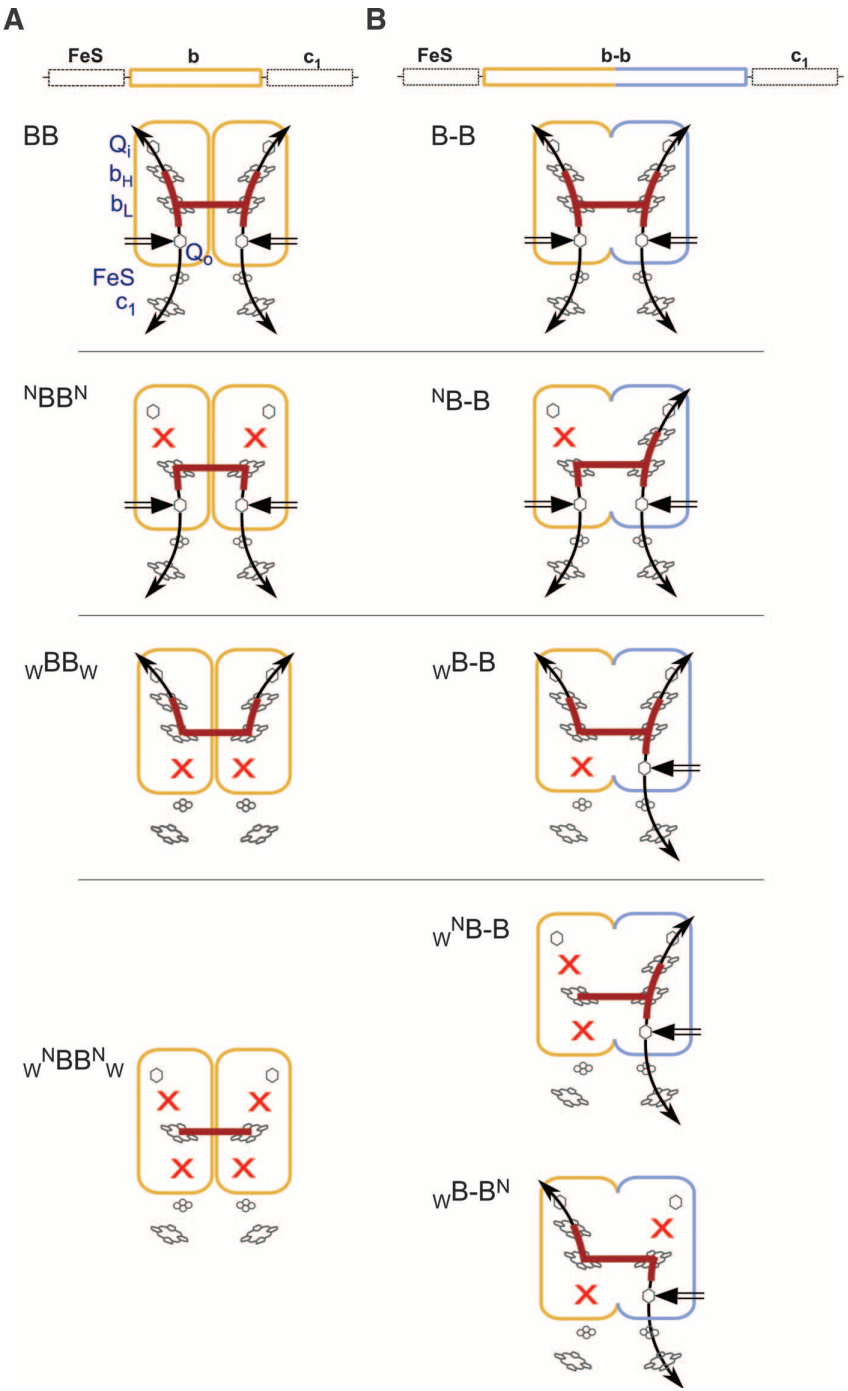
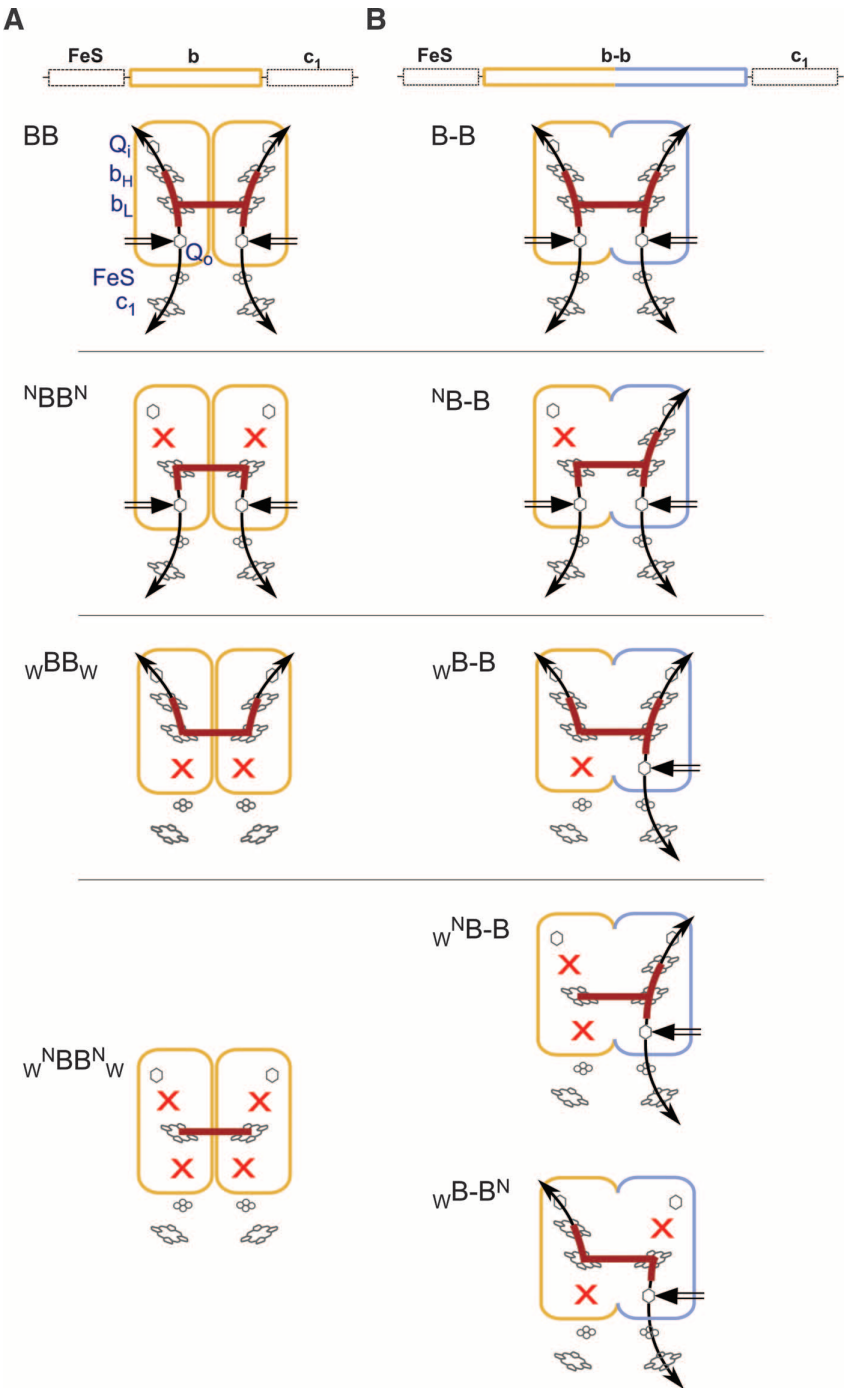


Fig. 2. Symmetric and asymmetric knockout patterns. Distribution of the knockouts (red crosses) constructed with unfused native operon coding (**A**) and fused gene coding (**B**). BB, native dimer: $^{N}BB^{N}$, both upper branches removed; $_{W}BB_{W}$, both lower branches removed; $_{W}{}^{N}BB^{N}{}_{W}$, all four branches removed. B-B, fused protein: ^{N}B-B, one upper branch removed; $_{W}$B-B, one lower branch removed; $_{W}{}^{N}$B-B, two branches on the same side removed; $_{W}$B-B^{N}, two branches across removed. N and W refer to H212N and G158W point mutations in cytochrome b (G, Gly; H, His; N, Asn; W, Trp). Black arrows, functional branches. Black double arrow, electron entry point at the Q_o site. Brown overlay: intraprotein electronic bus bar.



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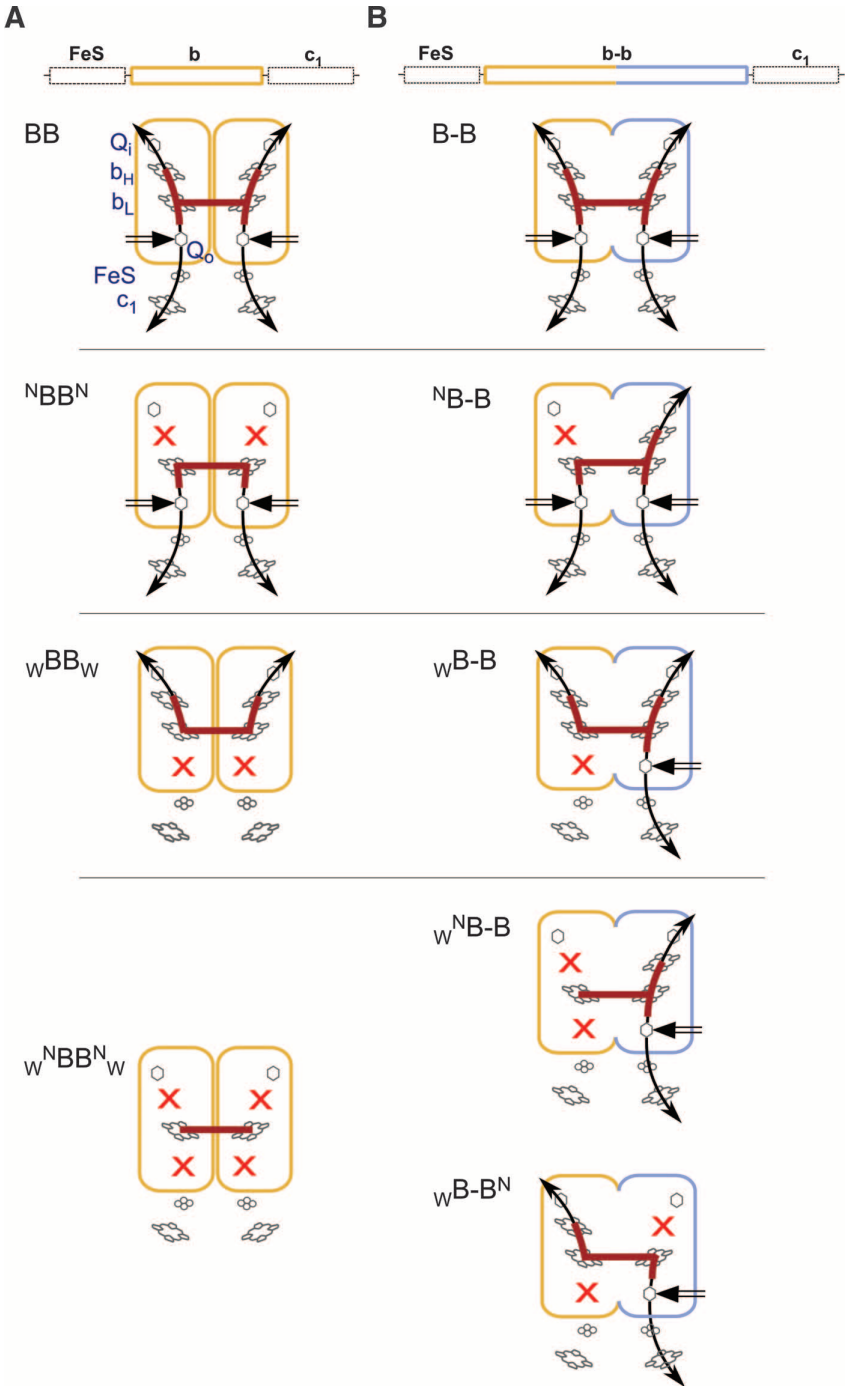

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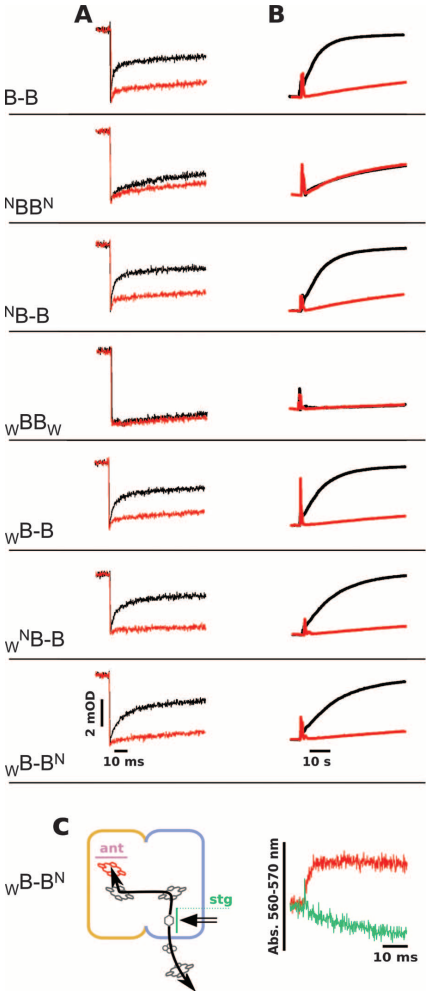

Fig. 4. Testing functional branch connection in the H-shaped electron transfer system. (**A**) Light-induced oxidation and re-reduction of cytochrome c at 550 minus 540 nm in membranes containing complete knockout variations described in Fig. 2. Black, uninhibited; red, inhibited with antimycin. B-B$_W$ and B-B$^N{}_W$ displayed kinetics similar to that of $_{W}$B-B and $_{W}{}^{N}$B-B, respectively (not shown). (**B**) Corresponding steady-state enzymatic reduction of cytochrome c at 550 nm. Rates are listed in Table 1. (**C**) Light-induced heme b_H kinetics in $_{W}$B-B^{N} in the presence of antimycin abolishing Q_i action (red) or stigmatellin abolishing Q_o action (green). In $_{W}$B-B^{N} blocked with antimycin (ant), the only route to reduce heme b_H (red) must involve the heme b_L to b_L electron transfer. stg, stigmatellin.

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Review

The Q cycle of cytochrome *bc* complexes: A structure perspective

William A. Cramer [a,*,1], S. Saif Hasan [a], Eiki Yamashita [b]

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[b] *Institute for Protein Research, Osaka University, Suita, Osaka 565-0871, Japan*

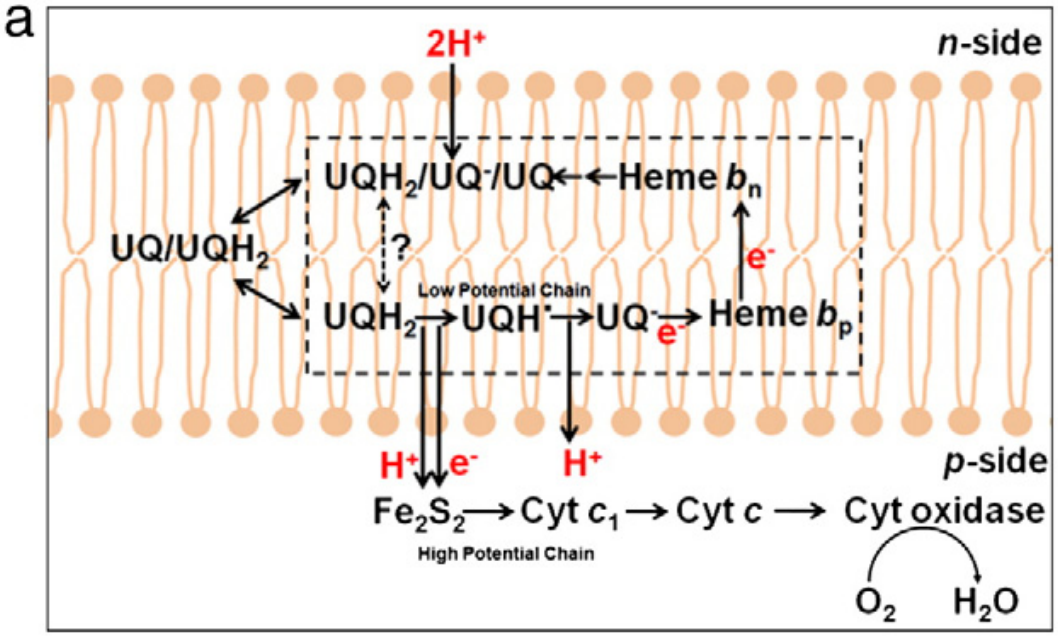
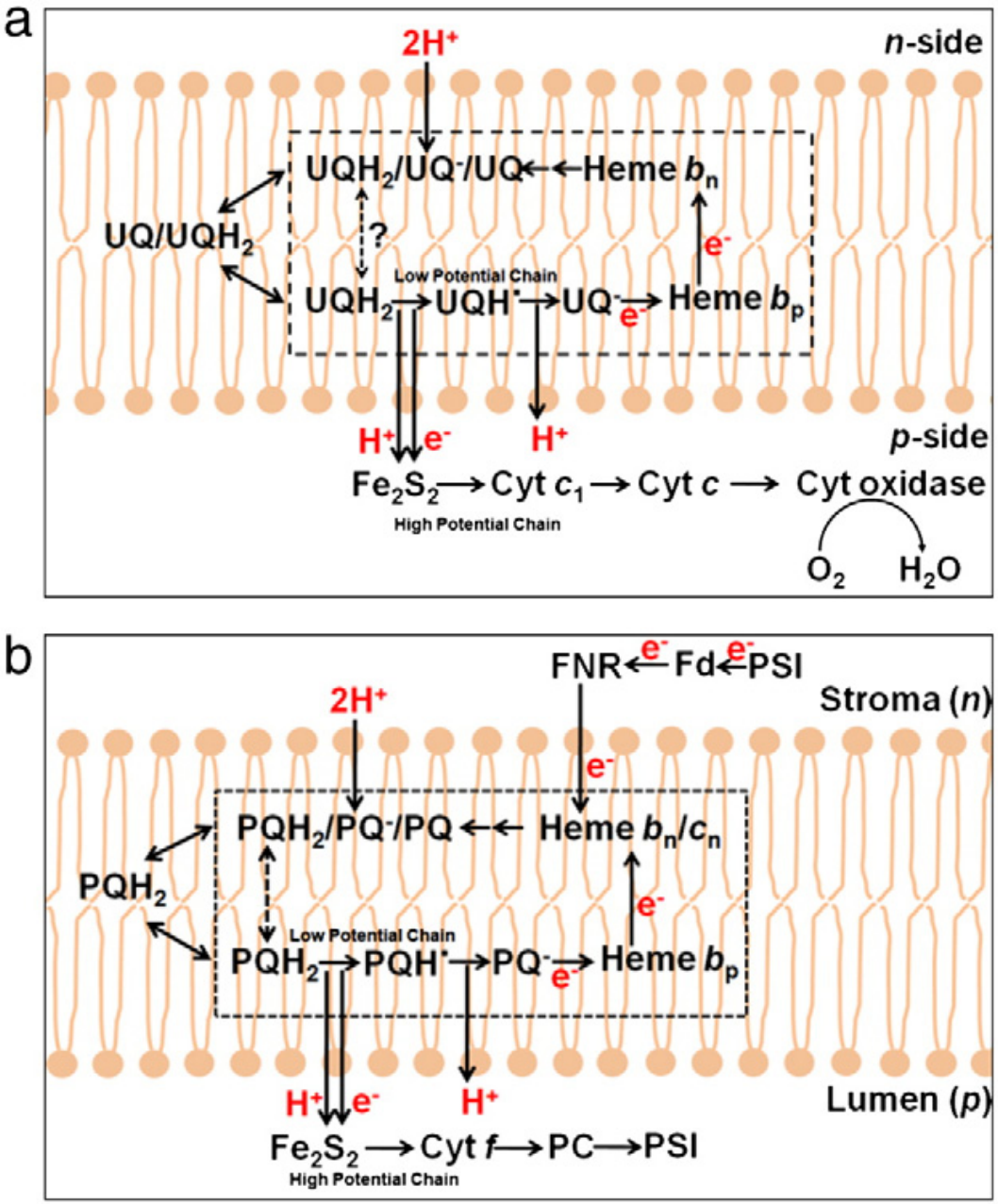


Fig. 3. Q cycle models for electron transfer and proton translocation through (A) the bc_1 complex in the respiratory chain [176] and the purple photosynthetic bacteria [48], reaction sequence (Table 3A1–3) and (B) the b_6f complex that functions in oxygenic photosynthesis (Table 3B). The original "Q cycle" model [172,174] for proton translocation, formulated in the aftermath of the experiment of the discovery of oxidant-induced reduction of heme *b* [171], focused on the mitochondrial bc_1 complex. Fundamental features of the classical Q cycle are: (i) the [2Fe–2S] complex on the p-side of the complex that functions as the one electron oxidant of the lipophilic quinol electron and proton donor, resulting in a bifurcated pathway into high and low potential chains; (ii) the high potential segment of the bifurcated pathway, initiated by electron transfer to cytochrome c_1 or *f*, which transfers one electron to the high potential electron terminal acceptor, (a) cytochrome oxidase or (b) photosystem I, while generating the semiquinone; (iii) the semiquinone donates the second electron to the two trans-membrane hemes *b*, b_p and b_n, in the low potential segment of the bifurcated chain that reduces a quinone or semiquinone [53] bound at the Q_n site.



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ATP synthase. Structure and Function

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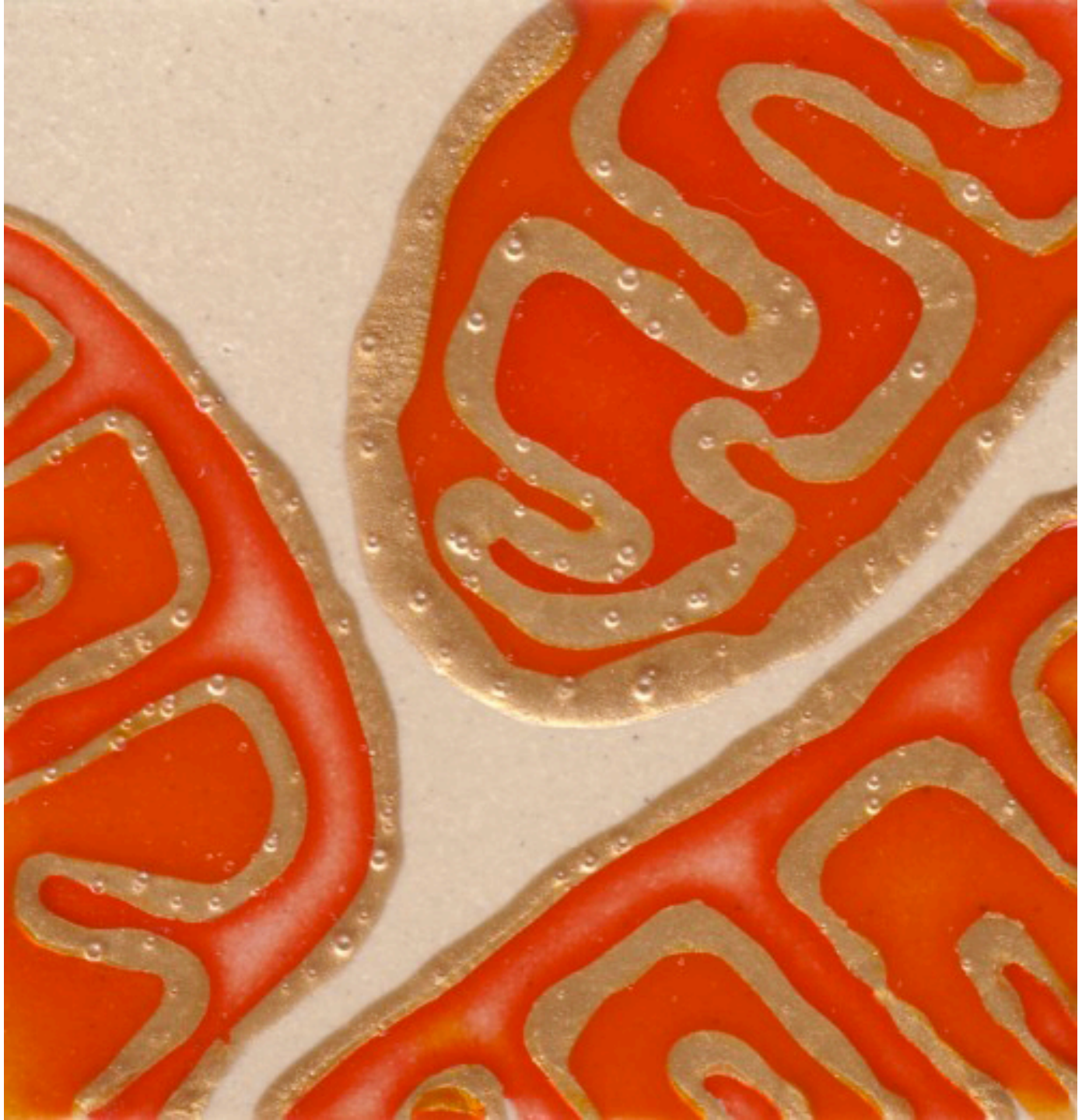


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SBCS-922 Membrane Proteins

Mitochondria and respiratory chains

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Queen Mary, University of London