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The structure and function of bacterial light-harvesting complexes (Review)

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Summary

The harvesting of solar radiation by purple photosynthetic bacteria is achieved by circular, integral membrane pigment-protein complexes. There are two main types of light-harvesting complex, termed LH2 and LH1, that function to absorb light energy and to transfer that energy rapidly and efficiently to the photochemical reaction centres where it is trapped. This mini-review describes our present understanding of the structure and function of the purple bacterial light-harvesting complexes.

Keywords: LH2, RC-LH1, bacteriochlorophyll, membrane protein, photosynthesis.

Introduction

Photosynthesis is the process by which photosynthetic organisms convert light energy from the sun into a chemically useful form. In purple bacteria the initial stages of this process—the light reactions—are mediated by molecular machinery that is housed in the photosynthetic membrane. The machinery consists of two fundamentally different types of integral membrane pigment-protein complexes: the light-harvesting (LH) or antenna complexes and the photochemical reaction centres (RCs; Cogdell *et al*. 1999). The LH complexes function to absorb solar radiation and efficiently funnel that energy down a 'gradient' to the RC, where it drives a charge separation across the membrane (van Grondelle *et al*. 1994). Ultimately, this results in the production of adenosine triphosphate (ATP) that is used to fuel cellular processes.

Purple bacteria must absorb light in the region of the spectrum available to them where they live. In the wild, since they are anaerobic phototrophs, they occupy an ecological niche in aquatic environments below that occupied by oxygenic chlorophyll *a* (Chl *a*)-containing organisms. Therefore, they grow photosynthetically by absorbing the far-red and green light that has not been filtered out by the organisms that contain Chl *a*. To harvest this available light, they synthesize RC and LH complexes consisting of integral membrane proteins that bind bacteriochlorophyll *a* (which absorbs in the far-red) and carotenoids (which absorb in the green; van Gemerden and Mas 1995). These pigments are located in different chemical environments within the proteins that bind them and this gives them different spectral characteristics. This spectral heterogeneity can be illustrated by examining the absorption spectrum of photosynthetic membranes of the purple non-sulphur bacterium *Rhodpseudomonas (Rps.) acidophila* strain 10050 (Figure 1). The spectrum represents absorption by three spectrally distinct populations of bacteriochlorophyll *a* (Bchl *a*) and of the carotenoid rhodopin glucoside. Two strong near-infrared (NIR) absorption bands are located at about 800 nm and 858 nm, and these are due mainly to the Q_y transitions of Bchl *a* molecules that are located in two different binding sites within a particular type of LH protein (LH2). The shoulder at ~875 nm on the red edge of the major NIR peak results from the contribution by the Q_y transition of Bchl *a* molecules that are bound to a different type of LH protein (LH1; Cogdell *et al*. 2002). In contrast to the situation observed in the NIR region of the spectrum, the positions of the Q_x transition and Soret absorption bands of all three Bchl populations are not affected by the chemical environment of their binding sites. They all have their Soret absorbance located at ~390 nm and that of their Q_x transition located at ~590 nm. The 'three-fingered' absorption between 450 nm and 550 nm arises from the carotenoids. What types of LH complexes are responsible for such a spectrum? Sucrose density gradient centrifugation of detergent-solubilized photosynthetic membranes from most purple bacteria reveals the existence of two major types of LH complex that are termed LH1 and LH2 (Figure 2). The LH2 complexes bind Bchl *a* molecules in two different binding sites. The differences in the chemical environments provided by these binding sites give rise to the two intense NIR absorption bands at 800 nm and 850 nm seen in the absorption spectrum of the isolated complex (Figure 3). By contrast, in the LH1 complex, the Bchl *a* molecules only have a single large NIR absorption band centred at 875 nm (Figure 3). The smaller band at 800 nm is due to Bchl *a* molecules bound to the RC, which co-purifies with LH1. As can be seen from the sucrose density gradient in Figure 2, LH2 is present in much greater amounts than LH1 in photosynthetic membranes. As a result, its absorbance totally dominates the NIR region of the absorption spectrum of intact membranes (Figure 1). The RC Bchl absorbance is completely masked by the LH2 800 nm absorption band and the LH1 absorption is only visible as the ~875 nm shoulder on the red edge of the major LH2 absorption peak.

In vivo the LH1 complex is intimately associated, in a fixed 1:1 stoichiometry, with the RC forming the so-called RC-LH1 'core' complex. All species of purple bacteria have a core complex. LH2 (or peripheral) antenna complexes are only present in some purple bacteria. The quantity of LH2 complexes per RC is variable and depends on growth conditions. When present, LH2 complexes are arranged around the core complexes where they act to effectively increase the surface area available for light absorption by the

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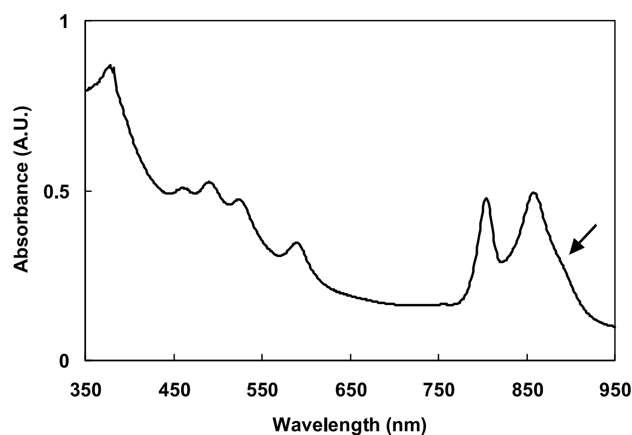


Figure 1. Absorption spectrum of photosynthetic membranes of *Rps. acidophila* strain 10050. The shoulder due to LH1 absorption at about 875 nm is arrowed.

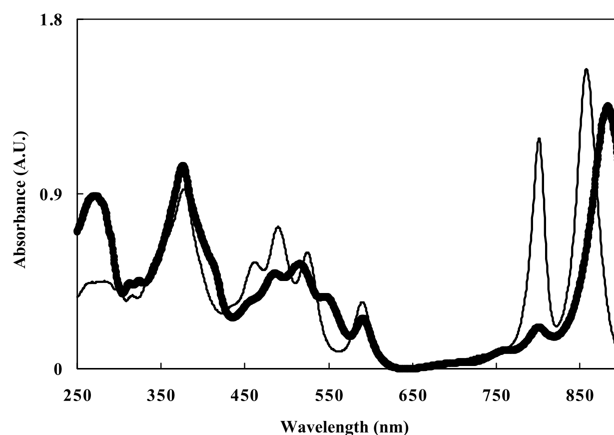


Figure 3. Absorption spectra of LH2 (fine line) and RC-LH1 'core' (heavy line) complexes of *Rps. acidophila* after partial purification. The LH2 complex possesses two major NIR peaks at 803 nm and 858 nm. The RC-LH1 complex has a single major NIR peak at 883 nm. The smaller peak at 801 nm is due to the RC accessory Bchls.

RC, over and above that provided by the fixed amount of LH1. In environments of very low light intensity, species such as *Rps. acidophila* strain 7050 can also synthesize different types of LH2 to maximize even further their light-harvesting capabilities under these extreme conditions of light stress (Gardiner *et al*. 1993).

Now is an exciting time for researchers investigating bacterial photosynthesis as the three-dimensional (3D) structures of examples of all the components involved in the photosynthetic light reactions have been elucidated. These include the structures of the cytochrome bc_1 complex (Iwata *et al*. 1998), ATP synthase (Abrahams *et al*. 1994) and the photochemical RC (Deisenhofer *et al*. 1985, Chang *et al*. 1986, Allen *et al*. 1987), as well as structures of three LH2 complexes (McDermott *et al*. 1995, Koepke *et al*. 1996, McLuskey *et al*. 2001) and an RC-LH1 core complex (Roszak *et al*. 2003). Their roles in the light reactions are summarized in schematic form in Figure 4. The LH structures illustrate how elegantly the antenna system has been adapted to optimize the absorption of solar energy and its efficient transfer to the RC (Pullerits and Sundström 1996, Sundström *et al*. 1999). In the next section of this review we describe the structures of both the LH2 and RC-LH1 complexes, before embarking on a discussion of the energy transfer reactions that take place both within and between these complexes and the RC.

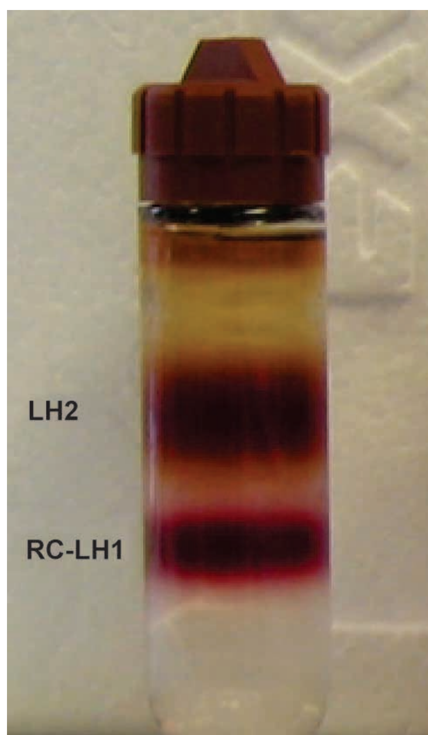


Figure 2. Discontinuous sucrose density gradients of detergent-solubilised photosynthetic membranes of *Rps. acidophila* reveal the existence of two major types of antenna complex. The upper band is LH2 and the lower band is RC-LH1 core complex. The minor band at the very top of the gradient is due to the presence of free carotenoid, in this case rhodopin glucoside (RG).

The minimal structural unit of all bacterial LH complexes is a heterodimer of very hydrophobic, low molecular weight (4–7 kDa) α and β polypeptides, each consisting of between 40 and 70 amino acid residues (Figure 5; Zuber and Brunisholz 1991, Zuber and Cogdell 1995). Both polypeptides have a tripartite structure consisting of polar N- and C-terminal domains and a central, single transmembrane α-helical domain. The N-terminus is located on the cytoplasmic side of the membrane and the C-terminus on the periplasmic side (Zuber 1985). Native antenna complexes are ring-like structures composed of oligomers of this minimal subunit and its non-covalently bound Bchl *a* and carotenoid pigment molecules.

The structure of LH2

Our understanding of photosynthetic light harvesting took a dramatic leap forward when, in 1995, the 3D crystal structure of the LH2 complex from *Rps. acidophila* strain 10050 was elucidated (McDermott *et al*. 1995). This was quickly followed by the structure of LH2 from *Rps. molischianum*

(Koepke *et al*. 1996) and an LH2 complex from low-light-grown *Rps. acidophila* strain 7050 (McLuskey *et al*. 2001). LH2 from *Rps. acidophila* strain 10050 is a nonamer of dimers formed from two apoproteins (an α-subunit of 53 amino acid residues and a β-subunit of 41 amino acids) and their associated Bchl *a* and carotenoid molecules (Figure 6). The transmembrane helices of the 9 αβ-apoprotein dimers form two concentric rings with radii of 18 Å and 34 Å, respectively, that surround a lipid-filled central cavity (Prince *et al*. 2003). The helices of the α-apoproteins lie perpendicular to the plane of the membrane whereas those of the β-apoproteins are tilted by about 15° with respect to it. Although there are some strong helix–helix interactions between the α-polypeptides, it is the extensive pigment–pigment interactions that dominate the structure. No αβ-apoprotein helix–helix interactions occur within the transmembrane domain of the complex (Freer *et al*. 1996). The N- and C-termini of both apoproteins fold over and interact with one another to enclose the cytoplasmic and periplasmic surfaces of the ring. The whole structure is interlocked by hydrogen bonds formed between large aromatic residues located at the C-termini of both apoproteins and Bchl *a* molecules. It is this protein structure that acts as a scaffold for the non-covalent attachment of the photosynthetic pigments (McDermott *et al*. 1995; Freer *et al*. 1996).

Each minimal αβ-subunit of the complex binds a total of three Bchl *a* and two rhodopin glucoside carotenoid molecules (Kramer *et al*. 1984, Cogdell *et al*. 1999, Papiz *et al*. 2003) giving a total of 27 Bchl *a* and 18 rhodopin glucoside molecules in the holocomplex. In each dimer unit, two of the three Bchl molecules are arranged near the periplasmic surface of the complex as a closely coupled dimer, with the plane of their bacteriochlorin rings approximately perpendicular to the plane of the membrane. These dimers form a ring of 18 overlapping pigments that are sandwiched between the α- and β-apoproteins of the fully assembled LH2 complex. Each pair of Bchls are coordinated, via their central Mg^{2+}, to two highly conserved histidine residues, one located in each of the α- and β-polypeptides. It is a combination of excitonic and protein–pigment interactions that causes the absorption maximum of these Bchls to red-shift dramatically, from about 770 nm for monomeric Bchl *a* in organic solvents to about 850 nm in LH2. Hence, these pigments are known as B850 Bchls. The remaining nine Bchl *a* molecules of the LH2 complex are located a further 16 Å into the membrane. In contrast to the B850 Bchls, these molecules absorb at 800 nm and are therefore termed B800 Bchls. The B800 molecules are located in a rather polar binding pocket between the α-helices of the outer β-polypeptides, and their bacteriochlorin rings lie in a plane parallel to the plane of the membrane. Each B800 is essentially monomeric, being separated from its neighbour by a distance of ~21 Å. Unlike the B850s, the central Mg^{2+} ions of the B800 molecules are not liganded to a histidine residue but rather to a carboxylate moiety on the N-terminal amino group of α-Met1 (the Met1 residue of the α polypeptide; Papiz *et al*. 2003). The B800 is further held in place by hydrogen bonds formed between an acetyl group on the bacteriochlorin ring and the Arg20 residue of the β-polypeptide. The whole B800 binding site is stabilized by hydrogen bonding between the α-Met1 residue and the α-Asn2, α-Gln3 and β-His12 residues (Papiz *et al*. 2003). The polar environment of the binding pocket and the weak coupling between B800 Bchls account for the absorption characteristics of this particular Bchl *a* population.

Often overlooked, but important structural moieties of the LH2 complex are the Bchl *a* phytyl tails. These play a central role in ensuring that the transition dipoles of the Bchl *a* macrocycles are aligned in such a manner as to maximize the energy transfer between them. The tails perform this function by providing the protein with a ‘handle’ to ‘grab’ onto and use to hold the planes of the bacteriochlorin rings in the correct orientation with respect to each other. Once the Bchls are in the correct position, they are fixed into place by a combination of ligation of the central Mg^{2+} to the protein and of H-bonding between the bacteriochlorin rings and the protein (Freer *et al*. 1996). The Bchl phytyl chains also play a role in maintaining the structural integrity of the LH2 complex. On examination of the structure, one can see that the phytyl tails of the B800 and β-B850 Bchls (the B850 molecules liganded to the β-polypeptides) wrap around each other. These interactions effectively lock the more exposed B800 Bchls into place within the complex (Prince *et al*. 1997).

The carotenoid pigments of LH2 have a number of important functions. These include accessory light harvesting, photoprotection and structure stabilization (Frank and Cogdell 1995). The original electron density map of LH2 at 2.5 Å showed only a single carotenoid molecule per αβ-subunit. This carotenoid has a typical *all trans* conformation and spans the whole depth of the complex. Its glucoside ring interacts with polar residues on the N-terminus of the α-apoprotein, and its hydrocarbon, conjugated tail passes across the face of the β-B850 macrocycle of an adjacent αβ-subunit. In doing so, this carotenoid acts as a cross-strut to lock adjacent αβ-dimers in place within the LH2 structure. The stabilization role played by the carotenoids has been illustrated in studies with carotenoid deletion mutants of purple bacteria (Zurdo *et al*. 1993, Lang and Hunter 1994). In the absence of coloured carotenoids LH2 fails to assemble.

Recently, a higher resolution structure of LH2 at 2.0 Å has revealed the existence of a second carotenoid molecule per αβ-dimer (Figure 6; Papiz *et al*. 2003). This carotenoid is located on the periphery of the complex between the β-polypeptides and it is severely bent owing to the presence of two *cis* bonds in the hydrocarbon tail. The distortion of this molecule may, however, not reflect its conformation *in vivo*. Its location on the periphery of the LH2 ring makes it vulnerable to attack by detergents. Therefore, it is possible that during the detergent solubilization step of sample preparation, some loss of carotenoid has occurred. The resulting partial occupancy may offer an explanation for the severely distorted conformation of the second carotenoid observed in the 2.0 Å crystal structure. It is more probable, therefore, that the actual *in vivo* conformation of the second carotenoid is more like that of the first. It is interesting to note that the second carotenoid molecule is oriented in the opposite direction to the first. However, both are in van der Waal’s contact with all the Bchl pigments of each LH2 dimer unit. This allows them to fulfill their accessory light-harvesting role by transferring absorbed solar energy to the Bchl

Figure 7

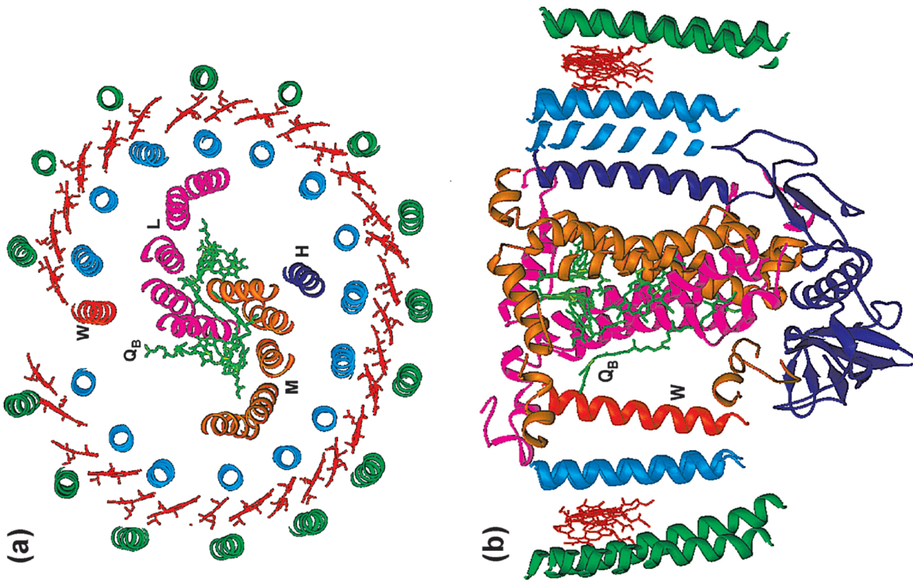


Figure 4

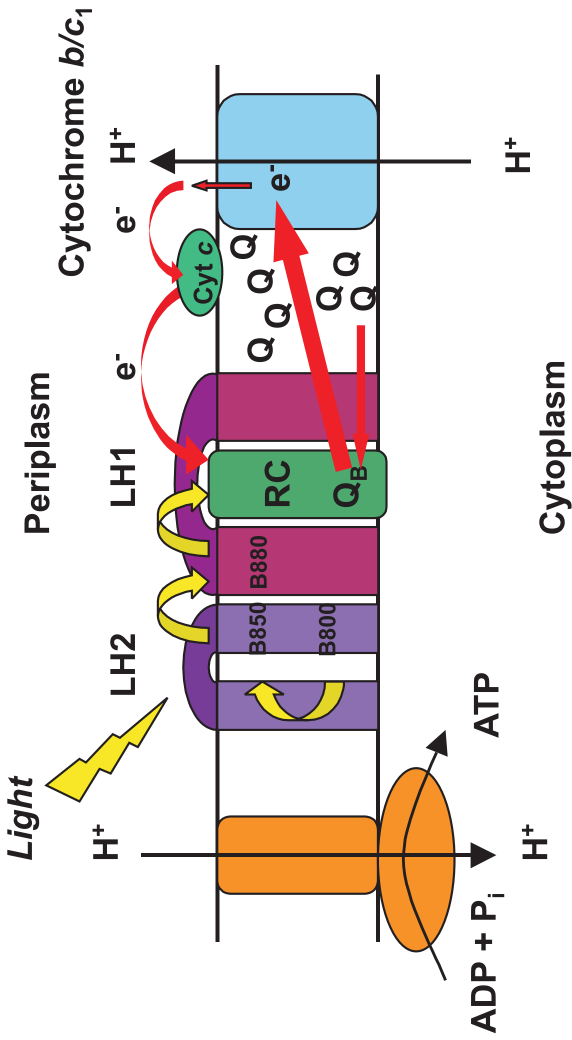
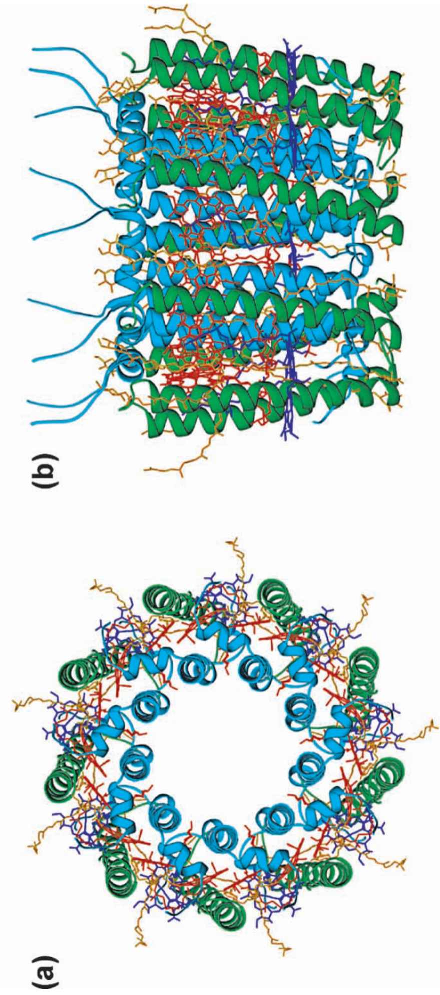


Figure 6



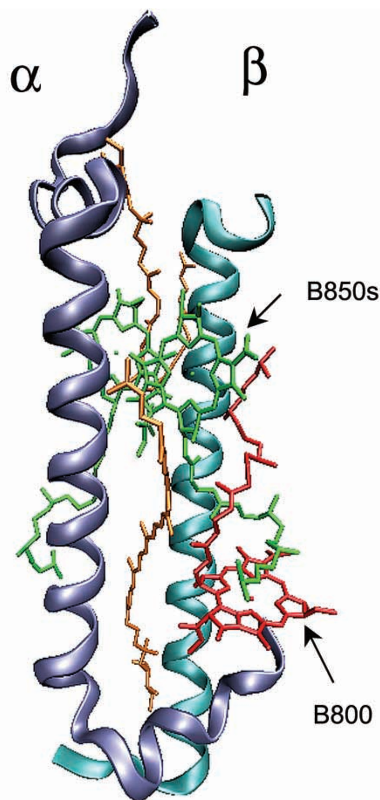


Figure 5. The minimal heterodimer subunit of the LH2 B800-850 complex of *Rps. acidophila* (Papiz *et al*. 2003). A colour version of this figure in which the carotenoid molecules are coloured orange is available in the online issue of the journal. The α and β polypeptides are depicted as dark grey ribbons (or dark and light blue ribbons respectively online). The dimeric B850 Bchl *a* molecules and monomeric B800 Bchl *a* are depicted as stick models in light grey (green and red respectively online).

molecules. The efficiency of the carotenoid to Bchl energy transfer depends on both the type of carotenoid and type of LH complex. In *Rps. acidophila* LH2 this energy transfer is about 55% efficient (Angerhofer *et al*. 1986) while in *Rhodobacter (Rb.) sphaeroides* 2.4.1 LH2 it occurs with about 95% efficiency (Cogdell *et al*., 1981).

The structure of the RC-LH1 core complex

The LH1 antenna is built using the same modular principle as the LH2 complex. The αβ-dimer unit of LH1 binds two Bchl *a* molecules and a single carotenoid (Cogdell *et al*. 1982). Even though the sequence identity between the α- and β-polypeptides of the heterodimer subunit of the two complexes is relatively low, the His residues that coordinate the B850 Bchls in LH2 are totally conserved in LH1 (Zuber and Brunisholz 1991). The latter, therefore, ligates a 'ring' of Bchl *a* molecules that are located at the same depth in the membrane as the LH2 B850s. This has important implications for energy transfer. Slight differences in the chemical environments of the LH2 and LH1 Bchl *a* binding sites result in the absorption maximum of the LH1 Bchls being further red-shifted to ~875 nm. There is no equivalent to the LH2 B800 binding site in LH1.

Until very recently, the best information we had about the structure of the LH1 antenna came from an 8.5 Å resolution projection map of the reconstituted LH1 complex from *Rhodospirillum (Rs.) rubrum* that was derived by electron crystallography of two-dimensional (2D) crystals (Karrasch *et al*. 1995). This appeared to show LH1 as a closed ring consisting of 16 αβ-dimer units and their associated pigments. The ring had a hole in the centre large enough to house an RC *in vivo*. A model of the core complex in which the LH1 ring completely surrounds the RC was supported by image analysis of 2D crystals of the native RC-LH1 complex from a carotenoidless strain of *Rs. rubrum* (Walz and Ghosh 1997). Further confirmation of such an arrangement came from projection structures of 2D crystals of RC-LH1 from *Rb. sphaeroides* (Walz *et al*. 1998).

The number of subunits that constitute the LH1 complex has long been a contentious matter. Although the projection maps of LH1 from *Rs. rubrum* and *Rb. sphaeroides* indicated it consisted of 16 αβ-subunits, other studies of

Figure 4. Cartoon representation of the major protein components involved in the photosynthetic light reactions. Light energy (yellow arrow) absorbed by the LH2 antenna (purple) B800 and B850 Bchl *a* molecules is transferred to the LH1 (maroon) Bchl *a*s, and is then trapped by the RC (green). In the RC, the special pair Bchl *a* molecules (not shown) are oxidized and electron flow occurs across the photosynthetic membrane to a primary electron acceptor, ubiquinone (UQ_A, not shown), which is reduced. Subsequently, the electron is transferred to the secondary electron acceptor, UQ_B (depicted here as Q_B). A second RC turnover results in the full reduction of UQ_B^- to UQ_BH_2. This fully reduced ubiquinone leaves the RC, passes its electron to the cytochrome b/c_1 complex (blue), and is replaced by an oxidized ubiquinone (Q). Electrons are passed from cytochrome b/c_1 to cytochrome *c* (green) and back to the RC. Concomitantly, cytochrome b/c_1 pumps protons (H^+) across the membrane to produce a proton gradient that drives ATP synthesis via the ATP synthase (orange). Reproduced from Roszak, *et al.* (2003) with permission.

Figure 6. Structure of the LH2 complex of *Rps. acidophila* at a resolution of 2.0 Å. (*a*) Top view of the complex looking down from the periplasmic side of the membrane. The α and β polypeptides are drawn as ribbons and are coloured turquoise and green, respectively. The pigments are all drawn as stick models. The B850 Bchl *a* molecules are red and the B800 Bchls are purple. The rhodopin glucoside carotenoids are orange. (*b*) The LH2 complex viewed parallel to the plane of the membrane with the periplasmic side on the top. The colour coding of the polypeptides and pigments is the same as in (*a*) (Papiz *et al*. 2003).

Figure 7. 4.8 Å resolution structure of the RC-LH1 core complex from *Rps. palustris*. (*a*) View perpendicular to the plane of the membrane. The protein components of the complex are drawn as ribbons. The RC H, L and M subunits are coloured blue, mauve and orange, respectively. The LH1 α and β polypeptides are shown in turquoise and green, respectively. Sandwiched between these polypeptides are the Bchl *a* molecules (red). Protein W is labelled and depicted in red. The RC pigments and ubiquinones are represented as stick models and shown in lime green. (*b*) Cross sectional view of the core complex viewed parallel to the plane of the membrane. The components are colour coded as described in (a) above. Reproduced from Roszak *et al.* (2003) with permission.

core complexes have suggested the LH1 ring is composed of 12 αβ-subunits (Boonstra *et al*. 1994, Meckenstock *et al*. 1994). However, the latter would be too small to encircle the RC completely. Biochemical methods of measuring the ring size have also given apparently conflicting results (Gall 1994, Francke and Amesz 1995). These differences have led some to question whether LH1 actually forms a complete ring *in vivo*. Electron microscopy analysis of tubular membranes from an LH2-null mutant of *Rb. sphaeroides* has shown that, in this particular strain, the LH1 rings are incomplete and form arcs around the RC (Jungas *et al*. 1999). However, another way to enable an LH1 antenna of less than 16 αβ-subunits to encircle an RC completely would be to include an additional component in the ring. A strong body of evidence suggests that a protein, termed PufX, may be such a component in the LH1 antenna of *Rb. capsulatus* and *Rb. sphaeroides* (Farchaus and Oesterhelt 1989, Lilburn and Beatty 1992, Lilburn *et al*. 1992, Barz and Oesterhelt 1994, Barz *et al.* 1995a, Pugh *et al*. 1998).

PufX is vital for photosynthetic growth in *Rb. sphaeroides* and *Rb. capsulatus* strains that contain native LH1 antenna (Lilburn *et al*. 1992, Barz and Oesterhelt 1994). Its presence is a requirement for both efficient ubiquinone/ubiquinol exchange between the RC and the cytochrome bc_1 complex as well as light-driven electron-transfer and photophosphorylation (Barz *et al*. 1995a,b). Photosynthetic growth is not compromised by deletion of the *pufX* gene in RC-only mutants or those in which the LH1 antenna is reduced in size (McGlynn *et al*. 1994). The PufX protein from *Rb. capsulatus* and *Rb. sphaeroides* has been isolated and used in reconstitution experiments to examine its effect on LH1 antenna formation *in vitro* (Recchia *et al*. 1998). The isolated protein was shown to have a specific, high affinity for the LH1 α-polypeptide and that it was inhibitory to LH1 formation at low concentrations. Further studies showed that the transmembrane region of PufX was responsible for this inhibition (Parkes-Loach *et al*. 2001). To explain these results it was suggested that PufX interrupts the molecular architecture of the LH1 ring (presumably at a position adjacent to the Q_B site of the RC) by binding to the LH1 α-polypeptide in the presence of Bchl *a*, thus preventing LH1 from impeding the free passage of ubiquinol from the RC to cytochrome bc_1. PufX not only has been implicated in organizing a specific orientation of the RC within the LH1 ring but, in LH2-minus mutants of *Rb. sphaeroides* that were grown under dark, partially aerobic conditions, it has also been shown to be responsible for the formation of long-range regular arrays of core complexes in the photosynthetic membrane (Frese *et al*. 2000). The exact function of PufX, especially relating to its postulated role in disrupting the LH1 ring, will have to await the arrival of a high resolution, 3D structure of a PufX-containing core complex.

Very recently, our laboratory has published a 4.8 Å resolution 3D structure of the RC-LH1 core complex from *Rps. palustris* (Roszak *et al.* 2003). This structure shows the RC surrounded by an oval, rather than a circular LH1 complex consisting of 15 αβ-dimer units and their associated pigment molecules (Figure 7). The outer ring of the elliptical LH1 complex has approximate dimensions of 110 Å by 95 Å. The longest dimension of the inner LH1 ellipsoid is approximately 78 Å. This allows the RC (whose longest in-membrane dimension is about 70 Å) to be accommodated. The orientation of the long axis of the LH1 ellipse coincides with the long axis of the RC to allow LH1 to wrap tightly around the RC. The LH1 oval is prevented from completely encircling the RC by a single transmembrane helix (termed protein W) that is out of register with the array of inner α-polypeptides. While protein W has not been fully characterized, mass spectrometry has shown it to have a mass of 10.7 kDa. The presence of protein W raises the question as to whether it is a 16th α-polypeptide of the LH1 inner ring or a PufX-like protein that acts to facilitate ubiquinone exchange. An exact equivalent gene for the PufX protein has not been found in the *Rps. palustris* genome. This is not entirely surprising because the PufX protein sequences, even for two such closely related species as *Rb. sphaeroides* and *Rb. capsulatus*, show little identity (Parkes-Loach *et al*. 2001). However, an analysis of the *Rps. palustris* genome has revealed the existence of at least 10 genes that could encode putative membrane spanning proteins with a mass about 10.7 kDa (J. T. Beatty, personal communication).

The structure reveals a second interesting and important feature at the location of protein W. The elliptical LH1-structure has a unique orientation with respect to the RC. Both helix W and the break in the outer ring of β-polypeptide helices of the LH1 complex are positioned on the opposite side of RC with respect to the single transmembrane helix of the RC H subunit. The latter itself breaks the overall 2-fold pseudo-symmetry of the RC. Helix W is therefore located adjacent to the groove in the RC through which the tail of the secondary UQ_B projects. The hydrophobic tail of UQ_B points towards the gap in the LH1 complex next to the W helix, strongly suggesting that protein W forms a 'portal' through which fully reduced UQ_B can communicate with the UQ pool located in the membrane lipid phase outside the LH1. It further suggests that the location of helix W imparts a significant role in the unique positioning of the RC within the RC-LH1 complex.

Although this structure explains much of the biophysical and biochemical data obtained for RC-LH1 core complexes, does it actually reflect the *in vivo* structure? 10 Å resolution AFM images of core complexes in native membranes from *Rps. viridis* have shown that LH1 consists of a closed ellipsoid of 16 subunits (Scheuring *et al*. 2003). However, the low resolution of the AFM images prevents visualization of the break in the LH1 structure. Interestingly, these experiments have also shown that the LH1 subunits rearrange into a circular ring structure after removal of the RC from the core complex. These AFM studies clearly indicate that the oval structure for the RC-LH1 complex from *Rps. palustris* observed in the crystalline state is representative of its *in vivo* condition.

Energy transfer in the photosynthetic unit

Energy transfer within and between LH complexes and the RC is generally recognized to occur via a Förster mechanism (Förster 1948) and is governed by parameters such as the distance between pigments, the spectral overlap between

the fluorescence emission of the donor pigment and the absorption of the acceptor pigment, and the excited state lifetimes and the relative geometric arrangement of the transition dipole moments of the pigments involved. The wealth of detailed structural information available for the components of the bacterial photosynthetic unit (PSU, defined as the combination of a RC and the LH pigments that contribute excitation energy to that RC; Aagard and Siström 1972) along with sophisticated spectroscopic techniques has enabled a detailed kinetic analysis of the energy transfer events that occur upon absorption of a photon (Fleming and van Grondelle 1997, van Grondelle and Novoderezhkin, 2001). When a Bchl *a* molecule absorbs light, the lifetime of the excited electronic singlet state is about 1 ns. Transfer of this energy to the RC must occur within the same timeframe. In the purple bacterial PSU, the direction of energy transfer is driven by an energy gradient going from LH2 to LH1 (B800 → B850 → B875) and then to the RC 'special pair' Bchls (Pullerits and Sundström 1996, Fleming and van Grondelle 1997, Yang *et al*. 2001). In this way the photosynthetic light-harvesting system acts as a 'funnel' to direct excitation energy to the RC. It is this directionality that is the key to its efficiency.

The structure of LH2 reveals an organization that is beautifully adapted to optimize the orientation of the Bchl *a*'s for a rapid and efficient energy transfer. Measurements of the decay of the anisotropy of the excited B800 population at room temperature indicated a rapid, but limited B800-to-B800 energy transfer step. The time constant for this decay was 0.3 ps, which translated into a B800-to-B800 transfer time of about 0.5 ps (Sundström *et al*. 1999). Energy transfer from B800 to B850 takes place with a rate constant of 0.9 ps at room temperature (Kennis *et al*. 1996). This energy transfer reaction is remarkably temperature insensitive and decreases to only 1.8 ps at 77 K and 2.4 ps at 1.4 K (Reddy *et al*. 1993, Vulto *et al*. 1999). When the excitation energy arrives at the B850 ring it can remain there for >1 ns provided no other LH complex is nearby (van Grondelle *et al*. 1994). The B850 molecules are strongly interacting and the excited state is rapidly delocalized, with transfers between Bchls occurring on the 50 fs–150 fs timescale (De Caro *et al*. 1994). The exact extent of this delocalization is not known, but it is probably delocalized over just a part of the ring (Alden *et al*. 1996, Koolhaas *et al*. 1996, Sauer *et al*. 1996). The rapid delocalization of excitation energy around the circumference of the LH2 ring has important implications for the function of the PSU. Within the excited state lifetime of LH2, every B850 molecule has the possibility of being visited by the excited state many times and so the rings of B850 molecules can be thought of as 'storage rings'. Therefore, the probability of energy transfer out of the ring is equal from each and every B850 molecule. This means that energy is available for transfer from any part of the ring to any part of a neighbouring ring, provided they are close enough. As such, a precise arrangement of LH2 and LH1 antenna complexes is not a prerequisite for the efficient function of the PSU. LH1 complexes simply need to lie close enough to excited LH2 complexes for energy transfer to occur with high efficiency.

The next energy transfer step, from B850 of LH2 to B880 of LH1, has a rate constant of 2–4 ps (Sundström *et al*. 1999, Hess *et al*. 1995). Once again, when the energy reaches the circular array of B880 molecules it is rapidly delocalized, with B880-to-B880 transfers occurring in about 80 fs (Visser *et al*. 1995). The final and slowest energy transfer step, from B880 to the RC, occurs in about 30–50 ps (Otte *et al*. 1993, Kennis *et al*. 1994, van Grondelle *et al*. 1994). The relatively slow rate of this transfer is a consequence of the distance between the RC special pair Bchls (P870) and the antenna Bchls. Why is this transfer rate so slow, and why are the LH1 Bchl *a* molecules not positioned nearer to the RC special pair Bchls? Oxidation of just a single Bchl *a* molecule in LH1 results in a strong quenching of the fluorescence yield (which is equivalent to the singlet excited state lifetime) thereby preventing LH1 from acting as an effective antenna for the RC (Law and Cogdell 1998). As oxidized P870 is strong enough to oxidize antenna Bchl *a* molecules, it makes sense for the LH1 antenna Bchls to be located sufficiently far away to prevent this oxidation from occurring. Therefore, the actual positioning of the LH1 antenna Bchls relative to P870 is clearly a compromise: they are sufficiently close to allow efficient energy transfer but not close enough as to allow the possibility of electron transfer.

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