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Purification and characterization of the polypeptides of core light-harvesting complexes from purple sulfur bacteria

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Abstract

Although the polypeptides of core light-harvesting complexes (LH1) from many purple nonsulfur bacteria have been well characterized, little information is available on the polypeptides of LH1 from purple sulfur photosynthetic organisms. We present here the results of isolation and characterization of LH1 polypeptides from two purple sulfur bacteria, *Thermochromatium* (*Tch.*) *tepidum* and *Allochromatium* (*Ach.*) *vinosum*. Native LH1 complexes were extracted and purified in a reaction center (RC)-associated form with the Q_y absorption at 914 nm and 889 nm for *Tch. tepidum* and *Ach. vinosum*, respectively. Three components were confirmed from reverse-phase HPLC for the LH1 apopolypeptides of *Tch. tepidum*. The β-polypeptide was found to be methylated at N-terminus, and two α-polypeptides were identified with one of them being modified by a formyl group at the N-terminal methionine residue. Two α- and two β-polypeptides were confirmed for the LH1 complex of *Ach. vinosum*, and their primary structures were precisely determined. Homologous and hybrid reconstitution abilities were examined using bacteriochlorophyll *a* and separated α- and β-polypeptides. The β-polypeptide from *Tch. tepidum* was capable of forming uniform structural subunit not only with the α-polypeptide of *Tch. tepidum* but also with the α-polypeptide from a nonsulfur bacterium *Rhodospirillum rubrum*. The α-polypeptide alone or β-polypeptide alone appeared only to result in incomplete subunits in the reconstitution experiments.

Abbreviations: BChl – bacteriochlorophyll; HPLC – high-performance liquid chromatography; LDAO – lauryldimethylamine *N*-oxide; LH – light-harvesting; MALDI-TOF – matrix-assisted laser desorption/ionization time-of-flight; NMR – nuclear magnetic resonance; OG – *n*-Octyl β-D-glucopyranoside; TFA – trifluoroacetic acid

Introduction

Purple photosynthetic bacteria can be classified into two groups based on their ability to use inorganic sulfur compounds as electron donors in photosynthesis. The photosynthetic complexes from purple nonsulfur organisms have been well characterized both structurally and functionally. In fact, a number of high-resolution structures are available for the reaction center (RC) and peripheral light-harvesting complex (LH2) of several purple nonsulfur bacteria (Deisenhofer et al. 1985; Allen et al. 1987; McDermott et al. 1995; Koepke et al. 1996). Two-dimensional projection maps at a resolution of 8.5 Å have been produced by electron diffraction experiments for a core light-harvesting complex (LH1) from *Rhodospirillum* (*Rsp.*) *rubrum* (Karrasch et al. 1995; Jamieson et al. 2002), a nonsulfur bacterium containing the single antenna complex. Interpretation of the ring-like structure observed from the projection maps can be made on the basis of analogies with the high-resolution structure of the LH2 antenna complexes. The LH1 complex is supposed to have 16 α-polypeptides forming the inner cylinder and 16 β-polypeptides forming the outer cylinder with a ring of bacteriochlorophylls (BChl) located between the two cylinders. In con-

trast, investigations on photosynthetic apparatus of the purple sulfur organisms are largely delayed. Only recently, a crystal structure of the RC from a purple sulfur bacterium *Thermochromatium* (*Tch*., formerly *Chromatium*) *tepidum* has been determined to atomic resolution (Nogi et al. 2000), and a number of gene sequences encoding RC and LH1 of the purple sulfur bacteria have become available (Fathir et al. 1997, 1998; Corson et al. 1999; Nagashima et al. 2002). To date, no detailed characterization of the purified core antenna polypeptides has been made for these organisms. It is of interest to compare various properties of the polypeptides with their purple nonsulfur relatives from an evolutionary point of view and also to clarify the structural organization of this ancient light-harvesting machinery.

The most remarkable features reported for a limited number of LH1 polypeptides from the purple sulfur bacteria are the variability and multiplicity. Most purple nonsulfur bacteria investigated so far exhibit a simple composition in their LH1 complexes, namely α- and β-polypeptides with a fixed 1 to 1 stoichiometry (except for a BChl *b*-containing bacterium, *Rhodopseudomonas viridis*, where a third, so-called γ-polypeptide was found). According to the gene sequence information available for *Allochromatium* (*Ach*., formerly *Chromatium*) *vinosum*, a purple sulfur bacterium, there are three genes encoding each of the LH1-type α- and β-apopolypeptides (Nagashima et al. 2002). Two pairs of translated products for the α- and β-polypeptides were identified, or partially identified, by direct amino acid sequencing of the purified LH1 complexes (Nozawa et al. 1985; Bissig et al. 1990), but the translated sequences differed at several positions from the gene sequences. A marked degree of multiplicity of the core antenna polypeptides was also found in halophilic purple photosynthetic bacteria (Wagner-Huber et al. 1992). There seem to be four LH1-type antenna polypeptides, 2α and 2β, in *Ectothiorhodospira* (*Ec.*) *halochloris* and *Ec. halophila*. These bacteria belong to the family that was originally part of the Chromatiaceae, and reveal quite different spectral features from purple nonsulfur bacteria due to the variation in the types and organization of their antenna complexes. A structural subunit of LH1 complex was isolated from a marine purple sulfur bacterium, *Chromatium purpuratum*, in which only the amino acid sequence of β-polypeptide was partially determined (Kerfeld et al. 1994).

The structural subunit of LH1 is characterized by an absorption maximum around 820 nm in the infrared region (referred to as B820) and has been determined to consist of one pair α/β polypeptides and two BChl *a* molecules (Pandit et al. 2001; Arluison et al. 2002; Wang et al. 2003). In this study, we describe the isolation and the characterization of LH1 polypeptides from two purple sulfur bacteria, *Tch. tepidum* and *Ach. vinosum*, using HPLC, Edman analysis and matrix-assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI-TOF/MS). Reconstitution ability to form the structural subunit has been examined using BChl *a* and various separated α- and β-polypeptides, including those from the purple nonsulfur bacterium *Rsp. rubrum*.

Materials and methods

Isolation of the LH1-RC complexes

Tch. tepidum cells were grown anaerobically at 48 °C as described previously (Fathir et al. 1998). Chromatophores were isolated by sonication of the whole cells suspended in 20 mm Tris-HCl buffer (pH 8.5) followed by differential centrifugation. Unless otherwise stated, this buffer was used throughout the experiment. The chromatophores were first extracted with 0.35% w/v lauryldimethylamine N-oxide (LDAO) at room temperature for 60 min to remove the LH2 components, and then with 1% w/v OG under the same conditions to solubilize the LH1-containing components. The extract was further purified at 4 °C on a DEAE column (Toyopearl 650S, TOSOH Co. Ltd, Tokyo, Japan) equilibrated with the buffer containing 0.7% w/v of OG. The LH1-RC fraction was eluted by a linear gradient of NaCl from 50 mM to 250 mM. Isolation of LH1-RC complex from *Ach. vinosum* was conducted using chromatophores treated with 2% w/v Triton X-100 and 1% w/v OG at 0 °C for 60 min. The extract was loaded on the DEAE column and washed with 0.03% w/v Triton X-100, 1 M ascorbate and 100 mM NaCl. After the LH2-rich components were removed, Triton X-100 in the elution buffer was replaced by 0.8% w/v OG. The LH1-RC fraction was then eluted by a linear gradient of NaCl from 100 mM to 200 mM.

Purification of the LH1 α and β-polypeptides and Edman analysis

The LH1 α- and β-polypeptides were obtained from LH1-RC complexes by using a reverse-phase HPLC

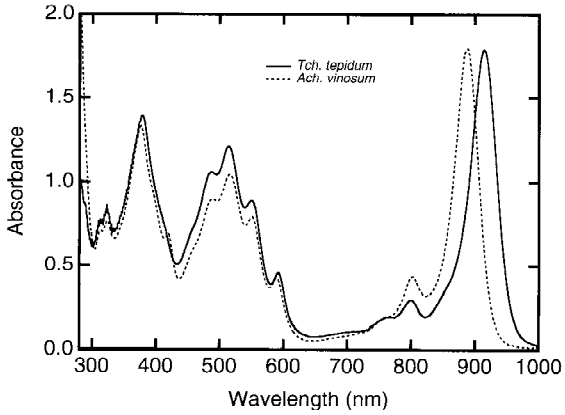

Figure 1. Absorption spectra of purified LH1-RC complexes from *Tch. tepidum* (solid curve) and *Ach. vinosum* (dotted curve).

column (TSKgel, Super ODS, 4.6 × 100 mm, TOSOH) with a mixed organic solvent system as described previously (Wang et al. 2001, 2002). Edman degradation analyses for determining the N-terminal amino acids of the polypeptides were performed by the Biochemical Analysis group of Kishida Chemical Co. Ltd, Japan.

Mass measurements

Masses of the polypeptides were determined by MALDI-TOF/MS (REFLEX III, Bruker Analytische, Germany) as described previously (Wang et al. 2001). Improvement was made on the solvent system for dissolving the α-polypeptides. Mixed solvents of chloroform and methanol (1:1, v/v) were used to first dissolve the α-polypeptides and then mixed with the matrix solution α-cyano-4-hydroxycinnamic acid.

Reconstitution of the structural subunits

Detailed procedure of reconstitution of the B820 subunits from pigment molecules and separated α- and β-polypeptides has been given elsewhere (Wang et al. 2001). Absorption spectra were recorded with a Beckman DU-640 spectrophotometer (Beckman Instruments, Inc., Fullerton, California). All operations were conducted at room temperature.

Results

Purification of the LH1 polypeptides

The LH1 complexes from both *Tch. tepidum* and *Ach. vinosum* were obtained in a RC-associated form after

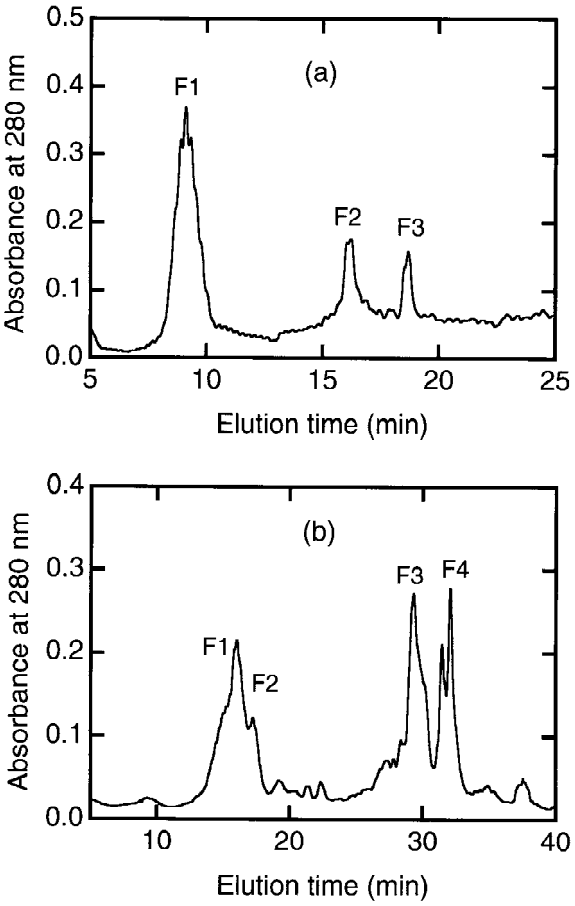

Figure 2. Reverse-phase HPLC chromatograms of LH1 apopolypeptides from *Tch. tepidum* (a) and *Ach. vinosum* (b).

elution from the DEAE column, as can be confirmed from their absorption spectra in Figure 1. Both the spectra showed similar shapes except that the LH1 from *Tch. tepidum* had a Q_y maximum at 914 nm, 25 nm red-shift compared with that (889 nm) from *Ach. vinosum*. Lyophilized LH1-RC complexes were first treated with benzene and methanol to remove all pigments and then were subjected to separation by a reverse-phase HPLC column. Figure 2 shows chromatograms for the LH1 apopolypeptides from the two bacteria eluted by linear gradients. Three fractions were eluted for the LH1 polypeptides of *Tch. tepidum* within a range of 60% $\sim$ 80% of the organic solvent and were separated into distinct peaks. Edman degradation analyses of the N-terminal ten amino acid residues revealed that the first fraction (F1) corresponded to the β-polypeptide and both the second (F2) and third (F3) fractions corresponded to the α-polypeptides. A quite different elution profile was

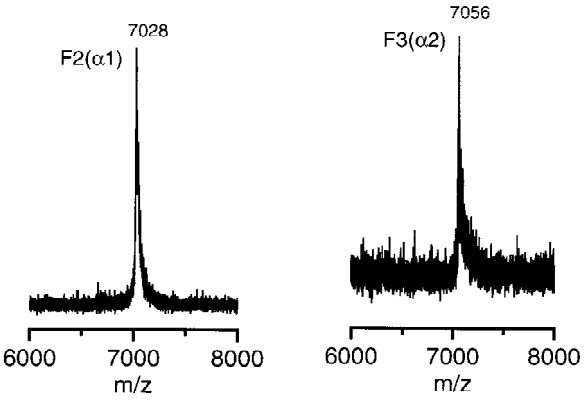

Figure 3. MALDI-TOF mass spectra of individual α1- and α2-polypeptides from *Tch. tepidum*.

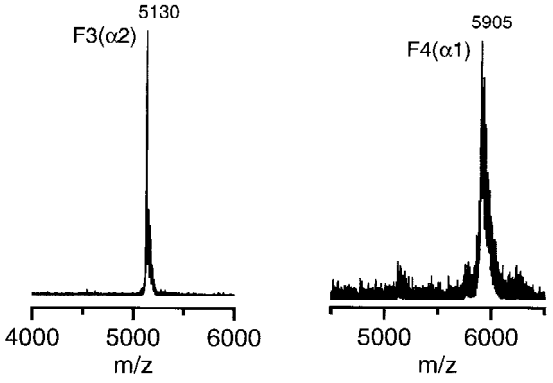

Figure 4. MALDI-TOF mass spectra of individual α1- and α2-polypeptides from *Ach. vinosum*.

obtained for the LH1 polypeptides of *Ach. vinosum* as shown in Figure 2b. By carefully collecting each of the fractions and from the result of Edman degradation, it was confirmed that the relatively broad peak eluted at about 16 min contained 2 components (F1, F2) corresponding to 2 β-polypeptides and fractions at about 29 min (F3) and 32 min (F4) corresponded to two α-polypeptides. Another pair of LH1 α- and β-polypeptides as predicted from the gene sequence of *Ach. vinosum* was not identified in our experiment.

Characterization of the LH1-apopolypetides

Molecular masses of the LH1 α- and β-apopolypeptides calculated from the *Tch. tepidum* gene sequences (*pufA* and *pufB*) are 7028.4 Da and 5529.4 Da, respectively (Fathir et al. 1997). The β-polypeptide has been identified by nuclear magnetic resonance (NMR) and MALDI/TOF-MS to have a *N*-methylated alanine as the N-terminal residue with a measured molecular weight of 5410 (Wang et al. 2002). The F2 and F3 fractions were measured by MALDI/TOF-MS, yielding molecular masses of 7028 Da and 7056 Da, respectively (Figure 3). Clearly, the F2 fraction has a primary structure exactly same as that coded by the *pufA* gene sequence without any modification. The F3 fraction corresponds to a modified α-polypeptide whose N-terminal methionine residue is formylated. This was confirmed by mass measurement and NMR spectroscopies using the N-terminal tryptic fragment (data not shown). The primary structures determined for the *Tch. tepidum* LH1 polypeptides are given in Table 1.

Ach. vinosum LH1 polypeptides yielded four fractions from the reverse-phase HPLC (Figure 2b). The

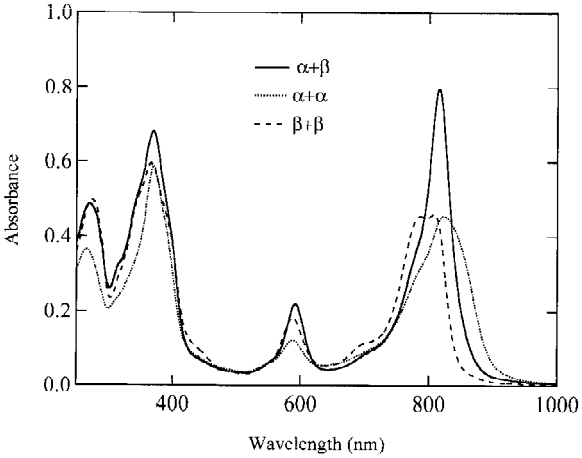

Figure 5. Absorption spectra of the reconstituted B820 subunits at room temperature using the LH1 polypeptides from *Tch. tepidum* and BChl *a*. The α-polypeptide used was a mixture of α1- and α2-polypeptides.

F1 fraction has a measured molecular mass of 5396 Da and is assigned to β3-polypeptide as its amino acid sequence corresponds to *pufB3* gene (Nagashima et al. 2002). The primary sequence is also in agreement with that reported from Edman degradation (β-B890-2) (Bissig et al. 1990) except that it has a *N*-methylated alanine as the N-terminal residue (Wang et al. 2002). The F2 fraction with a molecular mass of 5133 Da is assigned to β1-polypeptide because its amino acid sequence corresponds to *pufB1* gene except for a removal of the N- terminal methionine. A previous study (Bissig et al. 1990) on amino acid sequence of the purified polypeptide (β-B890-1) wrongly assigned the Phe at position 16 to Ala. Figure 4 shows the TOF/MS results for F3 and F4 fractions. The F3 fraction has a measured molecular mass of 5130 Da which is

Table 1. Amino acid sequences of LH1 polypeptides from *Tch. tepidum* and *Ach. vinosum*

Tch. tepidum	
α1	MFTMNANLYKIWLILDPRRVLVSIVAFQIVLGLLIHMIVLSTD LNWLDDNIPVSYQALGKK
α2	f-MFTMNANLYKIWLILDPRRVLVSIVAFQIVLGLLIHMIVLSTDLNWLDDNIPVSYQALGKK
β	N-(CH_3)AEQKSLTGLTDDEAKEFHAIFMQSMYAWFGLVVIAH LLAWLYRPWL
Ach. vinosum	
α1	f-MSPDLWKIWLLVDPRRILIAVFAFLTVLGLAIHMILLSTAE FNWLEDGVPAA
α2	f-MHKIWQIFDPRRTLVALFGFLFVLGLLIHFILLSSPAFNWL SGS
β1	ANSSMTGLTEQEAQEFHGIFVQSMTAFFGIVVIAHILAWLWRPWL
β3	N-(CH_3)ADQKSMTGLTEEEAKEFHGIFTQSMTMFFGIVIIAHILAWLWRPWL

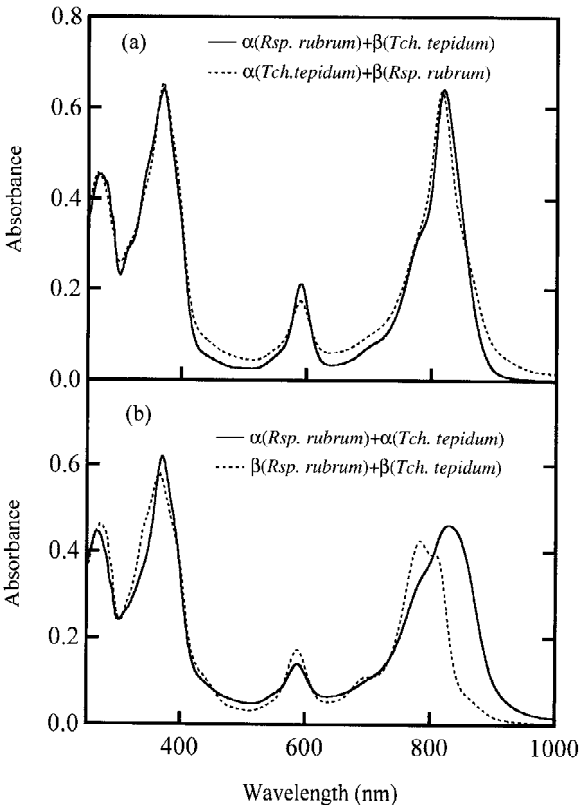

Figure 6. Absorption spectra of reconstituted B820 subunits using the LH1 polypeptides from *Tch. tepidum* and *Rsp. rubrum*. (a) Cross reconstitutions using the α-polypeptide from *Rsp. rubrum* and β-polypeptide from *Tch. tepidum* (solid curve), and the α-polypeptide from *Tch. tepidum* and β-polypeptide from *Rsp. rubrum* (dotted curve). (b) Hybrid reconstitutions using the α-polypeptide from *Rsp. rubrum* and α-polypeptide from *Tch. tepidum* (solid curve), and the β-polypeptide from *Tch. tepidum* and β-polypeptide from *Rsp. rubrum* (dotted curve).

27 Da larger than that calculated from its gene sequence (*pufA2*) (Nagashima et al. 2002) and therefore is assigned to α2-polypeptide. The result, along with that obtained from the N-terminal tryptic fragment (data not shown), indicates that the α2-polypeptide has a formylated N-terminal methionine, and its C-terminus is a Ser residue rather than Gly as previously reported from the amino acid sequencing (Bissig et al. 1990). The F4 fraction was measured to have a molecular mass of 5905 Da which is much smaller than that (7211 Da) calculated from gene sequence (*pufA1*) (Nagashima et al. 2002) and is assigned to α1-polypeptide. The α1-polypeptide was also identified before and its amino acid sequence of N-terminal domain was partially determined (Nozawa et al. 1985). Further analyses of the tryptic fragments revealed that the N-terminal methionine was formylated and twelve amino acid residues in the C-terminal domain coded by the gene sequence were removed after expression. The primary structures determined for the *Ach. vinosum* LH1 polypeptides are summarized in Table 1. The polypeptides corresponding to *pufA3* and *pufB2* genes were not detected in this work.

Reconstitution of the structural subunits

Reconstitution ability to form the structural subunits was examined using separated α-and β-polypeptides with pigment molecules. Figure 5 shows the absorption spectra of reconstitution of α- and β-polypeptides from *Tch. tepidum* with BChl *a*. A mixture of the α1- and α2-polypeptides with the same molar ratio as that found in LH1 complex was used in the experiment as no essential difference was found between the two polypeptides in their reconstitution behaviour. The ratio of absorbance at 817 nm to that at 280 nm was used as an indicator of reconstitution quality. Complexes which gave ratios of greater than 1.6 were considered to be high-quality and to have a complete subunit structure. By this definition, only

the combination of α- and β-polypeptides with BChl *a* was shown to form a complete structural subunit with the Q_y transition at 817 nm, whereas neither the α-polypeptide only nor β-polypeptide only with pigment molecules were capable of reconstituting a high-quality B820 complexes. Attempts for reconstitution of LH1-type complex (B870) from the subunits following a procedure (Parkes-Loach et al. 1988) by gradually reducing the OG concentration to 0.6% w/v at 4 °C were not successful. Although high homologies were confirmed in amino acid sequence of LH1 polypeptides between sulfur and nonsulfur bacteria, it is not clear whether structural elements required for the formation of functional B820 subunit would be the same as those in the nonsulfur bacteria. For this purpose, hybrid reconstitutions were conducted using the LH1 polypeptides from *Tch. tepidum* with those from a purple nonsulfur bacterium *Rsp. rubrum*, and the result is shown in Figure 6. Any combination of α- and β-polypeptides with BChl *a* resulted in B820 subunits stable for several hours at room temperature, whereas mixed α-polypeptides, or β-polypeptides, from different bacteria were unable to form a complete structural subunit.

Two pairs of the LH1 α- and β-polypeptides isolated from *Ach. vinosum* were employed for examination of their reconstitution ability. Because the two α-polypeptides and also the two β-polypeptides are different slightly in their amino acid sequences, these polypeptides were used separately in the reconstitution experiments. Figure 7 shows the absorption spectra obtained at two different OG concentrations. Each pair of the α- and β-polypeptides was capable of forming the structural subunit with BChl *a* at OG concentration of 0.9%. The Q_y transitions observed for all combinations were in a range of 821 nm to 823 nm. With decreasing the OG concentration to 0.6%, the Q_y transitions were red-shifted to 854 nm $\sim$ 863 nm corresponding to the incomplete LH1-type complexes, probably an oligomeric form of the structural subunits. Further efforts by decreasing the OG concentration and adjusting other reconstitution conditions did not yield the LH1-type complexes.

Discussion

LH1 complexes from both *Tch. tepidum* and *Ach. vinosum* were highly purified in a RC associated form. Attempts for isolation of pure LH1 complexes under various conditions, such as at higher OG concentrations and for longer treatment time, were not successful. This may reflect a strong interaction between the LH1 and RC complexes. The isolation method for the LH1-RC complex from *Tch. tepidum* has been improved by employing two-step solubilization by LDAO and OG instead of Triton X-100 (Fathir et al. 1998), followed by DEAE chromatography at 4 °C. The resultant LH1-RC complex has a Q_y absorption band at 914 nm for the antenna BChl *a*, more than 20 nm red-shift compared with that from most other species. The 914 band was found to shift to 880 nm by addition of various salts at room temperature, and such process was reversible (Fathir et al. 1998). Recently, a LH1-RC complex isolated from a purple sulfur bacterium, strain 970, was reported to exhibit a Q_y absorption band at 963 nm for the antenna BChl *a* (Permentier et al. 2001). The extremely large red-shift was explained in terms of enhanced exciton interaction between the BChl *a* molecules and specific interactions with the polypeptides. However, no information is available for the primary structure of the LH1 polypeptides of this strain.

Like the LH1 complexes from other nonsulfur bacteria (Parkes-Loach et al. 1988), B820 structural subunits can be obtained by addition of detergent OG to the LH1-RC complexes of the two sulfur bacteria of this study. We have also observed a stable intermediate species with Q_y around 850 nm (Figure 8) for the LH1 complex from *Tch. tepidum* by addition of cetyl trimethylammonium bromide (CTAB). Because the CTAB is a cationic detergent and considering that the Q_y transition can be reversibly converted between 914 nm and 885 nm by addition and removal of various salts (Fathir et al. 1998), a strong ionic interaction may be involved in the LH1 polypeptides of *Tch. tepidum*.

One β-polypeptide and two α-polypeptides were identified for the *Tch. tepidum* LH1 complex. Although it is difficult to precisely evaluate the ratio of α- and β-polypeptides solely from the HPLC chromatogram, the amount of the β-polypeptide is apparently larger than the total amount of the two α-polypeptides even by taking into account of one more tryptophan and phenylalanine residues in the β-polypeptide. The ratio of the three polypeptides as calculated by integrating peak areas of the HPLC chromatogram (Figure 2a) was approximately $\alpha 1{:}\alpha 2{:}\beta$ = 2:1:10. This feature is of particular interest because most LH1 polypeptides of the photosynthetic bacteria investigated so far are characterized by a simple composition, each of these antenna complexes being composed of an α/β heterodimer with a fixed one to one stoichiometry.

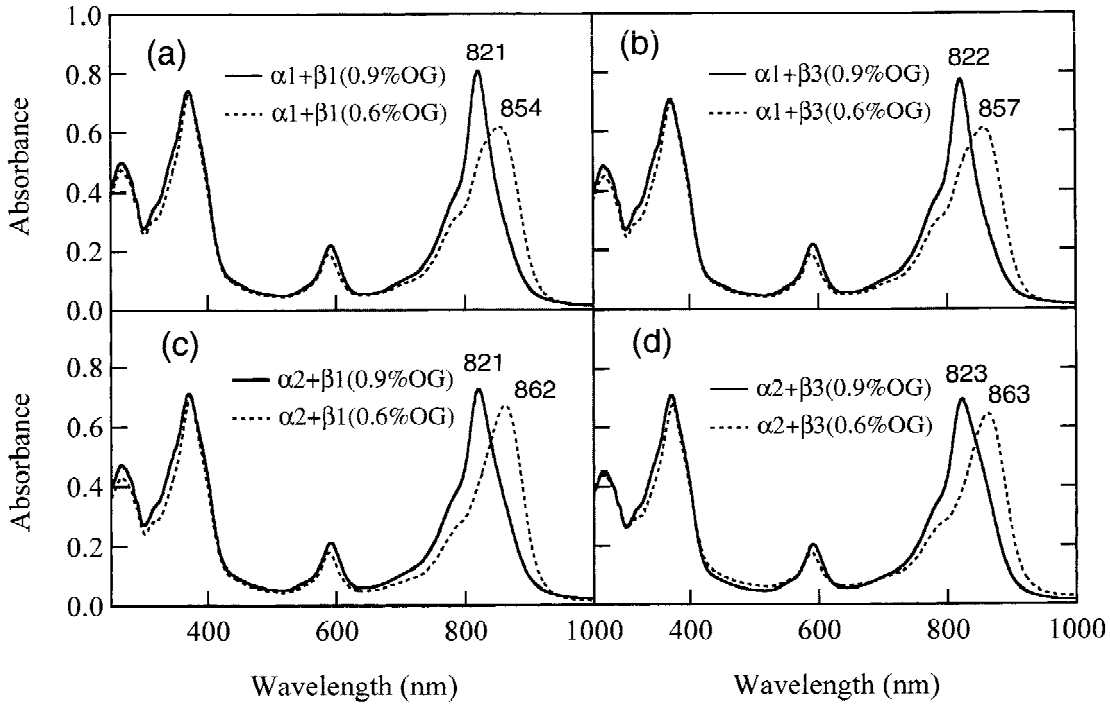

Figure 7. Absorption spectra of species formed on reconstitution of LH1 α- and β-polypeptides from *Ach. vinosum* with BChl *a* in 0.9% OG (solid curve) and in 0.6% OG (dotted curve). (a) Reconstitution using α1- and β1-polypeptides. (b) Reconstitution using α1- and β3-polypeptides. (c) Reconstitution using α2- and β1-polypeptides. (d) Reconstitution using α2- and β3-polypeptides.

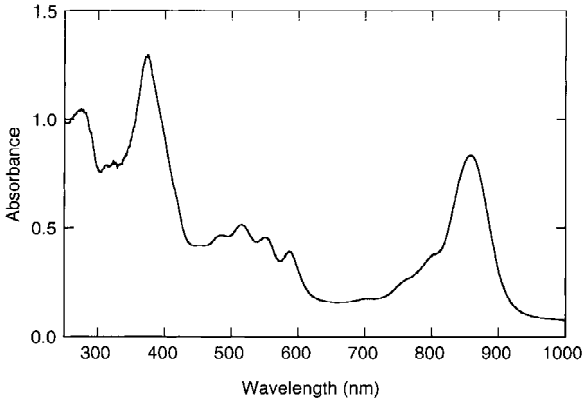

Figure 8. Absorption spectrum of *Tch. tepidum* LH1-RC core complex treated with 0.05% CTAB for 30 min at room temperature.

Further investigations are needed to clarify the *in vivo* structural arrangement of these polypeptides and its relationship with physiological functions.

The LH1 β-polypeptide of *Tch. tepidum* was shown to have a methylated N-terminus (Wang et al. 2002). It is for the first time to have confirmed formylated and unmodified α-polypeptides coexisting in the LH1 complex, as only formylated α-polypeptides have been reported for most purple bacteria. The α-polypeptide with a formylated N-terminus eluted later, with a slightly smaller amount, from the reverse-phase HPLC column than the unmodified α-polypeptide. Differences of the two α-polypeptides in their physiological functions and how the removal of the formyl group is regulated *in vivo* are not known.

We have identified two polypeptides for each of the α- and β-polypeptides of LH1 from *Ach. vinosum*. The α3- and β2-polypeptides as reported from gene sequences remain undiscovered. The result is in agreement with those reported previously by organic solvent extraction and subsequent reverse-phase chromatography (Nozawa et al. 1985; Bissig et al. 1990). The two β-polypeptides show a high homology (80%) to each other, and one of the β-polypeptide (β3) has a methylated N-terminus (Wang et al. 2002). N-terminal methionine residues of both the α-polypeptides are formylated. One of the α-polypeptides (α1) experienced a carboxyl-terminal processing, in which twelve residues were truncated from the C- terminal domain. The multiplicity and variability of the LH1 complex in *Ach. vinosum* are considered as a consequence of adjustment to the growth environment such as ambient light intensity and quality, because this bacterium

is also known to have multiple peripheral antenna complexes (Zuber and Cogdell 1995).

Reconstitution experiments were conducted to ensure that the high homologies in amino acid sequence of LH1 polypeptides from the two sulfur bacteria and between sulfur and nonsulfur bacteria bring about ability to form the structural subunit. Although the reconstitution procedure used in this study has been well established and similar behavior were observed, the result provides new information demonstrating that structural elements required for the formation of functional B820 subunit are also contained in the LH1 polypeptides of the sulfur bacteria and they are essentially the same as those of the nonsulfur bacteria. This is important from the evolutionary point of view because these structural elements are shown to be conserved among the purple bacteria, however LH1 polypeptides of the sulfur bacteria reveal more diversity compared with their nonsulfur counterparts. The multiplicity of LH1 polypeptides implies that different structural organization may be adopted for the core light-harvesting complex in sulfur bacteria. Reconstitution of the B820 structural subunits can be achieved by using separated α- and β-polypeptides of the two sulfur bacteria with BChl *a*. In the homologous reconstitution experiments, any α/β polypeptide pair with BChl *a* formed complete B820 complexes, whereas α-polypeptide alone or β-polypeptide alone with BChl *a* failed to yield the structural subunits. We have not been able to reconstitute a LH1-type complex from its components. It has been shown for the nonsulfur bacteria that β-polypeptides can form the subunit complexes with BChl *a* in the absence of an α-polypeptide, and both α- and β-polypeptides are required for the formation of a LH1-type complex (Parkes-Loach et al. 1988; Loach et al. 1994). The structural role of α-polypeptide on the formation of B820 subunit and possible requirement of other factors for the formation of LH1-type complex in the sulfur bacteria might indicate somewhat different arrangement of the two polypeptides and pigment molecules in the LH1 complex. In the hybrid experiments where α- and β-polypeptides of *Tch. tepidum* were examined for reconstitution with β- and α-polypeptides of *Rsp. rubrum*, each pair with BChl *a* resulted in B820 complexes. The amino acid homologies between the two α- and two β-polypeptides are about 40% and 50%, respectively, in which sufficient information is contained to allow specific interactions for the formation of B820 subunit. Hybrid reconstitutions using the LH1 polypeptides from four nonsulfur bacteria demonstrated that a central core region of amino acids is important for formation of a functional complex and the N-terminal hydrophilic region of each polypeptide contains a specific portion to stabilize complex formation through ionic interactions between oppositely charged groups on the α- and β-polypeptides (Loach et al. 1994). The results of this study show that the LH1 polypeptides of sulfur bacteria are functionally similar to those of nonsulfur bacteria, but the structural organization might be different due to the multiplicity and variability. Further detailed analysis on the structural basis of LH1 complexes will be useful in the study of the evolution of the light-harvesting apparatus in photosynthetic bacteria.

Acknowledgements

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