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Dynamic infrared band–band spectroscopy of peripheral light-harvesting complexes from *R. acidophila*

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Abstract

Infrared (IR) kinetic spectroscopy measurements have been employed to investigate and compare the electronic structure of B800 and B850 bacteriochlorophyll (BChl) pigment aggregates within peripheral (LH2) antenna complexes of photosynthetic purple bacteria. Following initial excitation of the B800 chromophores at 800 nm, a strong absorption band centred at ~1.3 μm was observed in the transient spectrum which is characteristic of monomeric BChls. Evolution of a transient spectrum in the near-IR and IR regions was then tracked while the excitation energy migrated to the B850 chromophores on a ~0.9 ps time scale. The B850 excited-state IR spectrum was found to differ substantially from that of B800, exhibiting a broader absorption band in the near-IR (centred at 1.1 μm) and new transient absorption features at 2.9 and 3.7 μm which are absent in B800. These results support a localised description of excitations for B800 but indicate the presence of considerably stronger inter-chromophore interactions between the B850 pigments. Differences between the excited-state spectra of B800 and B850 are interpreted as reflecting the formation of delocalized exciton and charge-resonance bands in B850.

Keywords: Bacteriochlorophyll; *Rhodopsuedomonas acidophila*; Band–band spectrum

1. Introduction

The conversion of light into chemical energy in plants and bacteria is essential to life on this planet [1]. Detailed understanding of the mechanism of photosynthetic light-harvesting requires knowledge of the nature of the antenna protein electronic states involved in absorption and emission processes. For recent reviews of this subject [2,3]. The symmetric arrangement and close spacing of pigments within bacterial antenna proteins [4,5], particularly, the B850 chromophores of LH2 complexes, imply the presence of strong inter-pigment interactions that must establish delocalized-electronic states. Only the average properties of these eigenstates are seen in ambient-temperature experiments but line-narrowing spectroscopies have provided convincing evidence for the presence of extended states at cryogenic temperatures [6,7]. Low-temperature measurements also point to the presence of interactions between multiple manifolds of states (such as exciton/charge-resonance couplings) and indicate a complex composition of the optically allowed levels [6,8]. Direct time-domain techniques have been extensively applied to follow the dynamics of optical excitations within LH1 and LH2 complexes [9–20]. An important goal of several time-resolved studies has been to determine whether the excitation energy equilibrates among coherently delocalized states or hops in a stochastic fashion between individual sub-units.

While the low-temperature results are highly informative regarding interactions that exist between pigments within an approximately static protein structure, elucidation of the light-harvesting mechanism requires knowledge of the electronic properties under physiological conditions. This requires femtosecond spectroscopic methods [2,3]. At ambient temperatures, thermally activated protein motions cause frequency fluctuations that can alter the absorption and emission spectra of the assemblies, potentially restrict exciton delocalization and influence the degree of inter-band coupling [21–23]. The conclusiveness of optical measurements is severely limited by the significant spectral broadening that prevails at physiological temperatures. Additional experimental methods are therefore required that can probe the band structure of antenna proteins under ambient conditions and can reveal and identify additional manifolds that interact with the Q_y exciton levels.

This article describes the application of femtosecond infrared (IR) band–band spectroscopy [24] to analyse and compare electronic properties of the B800 and B850 pigment systems within peripheral (LH2) antenna proteins.

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Band–band spectroscopy is a powerful technique for characterising the exciton level structure of interacting chromophore systems, yielding information on the manifold of the states that arises from inter-cofactor couplings that might not otherwise be obtained from conventional optical studies [24]. It is a form of two-photon absorption spectroscopy with the intermediate-resonant states being those of B800 and B850 sets of cofactors. Monitoring the evolution of the band–band spectrum by probing in the IR as excitation energy migrates from B800 to B850 provides a direct means to compare the level structures of the two pigment systems B800 and B850 and understand properties of the wave-functions of the electronic states involved in light harvesting. Furthermore, this technique enables the detection of states that interact with the Q_y exciton levels but are not be observable in the conventional ground-state optical spectrum.

Owing to the rigorous selection rules (such as $\Delta k = 0$ for a linear chain, or $\Delta k = 0 \pm 1$ for a circular aggregate) which are typical for optical transitions in perfect molecular aggregates at sufficiently low temperatures, transitions from the non-degenerate ground state can usually access only a small subset (usually one or two, but this depends on the amount of disorder) of the N levels within the one-exciton manifold, where N is the total number of pigments. An elegant experimental technique for observing *all* states within the manifold, first proposed by Rashba and co-workers [25,26], involves the measurements of transitions between the optical one-exciton band and a second band also comprised of N states: the selection rules can be satisfied for each and every level in this fashion. A powerful application of this approach corresponds to the case where the width of the second-exciton band is negligible in comparison with that of the first. In such a case, the spectra of band–band transitions directly yield the density of states (DOS) function for the first band after appropriate correction for the Boltzman distribution. Early examples of experimental determination of band-level structure by this method are the vibrational exciton (vibron)–electronic exciton absorption measurements [27] and the exciton luminescence studies [28–30] performed on crystalline forms of aromatic compounds. The ability to follow evolution of band–band spectra on ultrafast time scales provides a means to study population dynamics within portions of the exciton band that cannot otherwise be observed directly from the ground state. Furthermore, consideration of the properties of band–band transitions reveals *a* significant sensitivity of the transition anisotropy to coherent exciton delocalization.

Results are reported in this paper for IR measurements in the 1–4 μm spectral region. The findings from this study provide new insight into the differences in electronic structure of the B800 and B850 pigments through information on previously undetected manifolds of B850 states that will help to understand the factors that influence energies of Q_y states participating in photosynthetic energy transfer.

2. Experimental

2.1. Sample preparation

The LH2 (B800–B850) antenna complex was prepared from cells of *Rhodopsuedomonas acidophila* strain 10050 as previously described [31]. The purified LH2 complex was suspended in 20 mM Tris–HCl pH 8.0, 0.1% LDAO and stored frozen until required. The absorption spectrum of the complex was recorded before and after the experiments to check that it was not damaged during the experimental procedures.

2.2. Femtosecond laser apparatus

Femtosecond IR measurements were conducted with 140 fs IR probe pulses generated from a KTP-based optical parametric amplifier that was pumped by 80 fs, 810 nm pulses from a regeneratively amplified Ti sapphire system. Photo excitation pulses at 800 nm were obtained by separating a small fraction of the 810 nm beam prior to the OPA system and selecting the appropriate spectral profile using an 800 nm interference filter (Corion, 10 nm full-width at half-maximum (FWHM)). The use of 10 nm FWHM pump pulses at 800 nm was necessary to selectively excite the B800 pigments and avoid direct excitation of the B850 chromophores. Photo excitation energies of 50 nJ were used for these experiments, and the pump beam was focused to a 400 μm diameter (FWHM) spot at the sample position. LH2 samples were contained in a 200 μm spinning cell with CaF_2 windows.

3. Results and discussion

3.1. Infrared inter-band transitions of LH2 antenna proteins

Femtosecond IR kinetic signals of the LH2 proteins in the 1–2 μm spectral range are presented in Fig. 1. Following photo excitation at 800 nm, absorption signals were seen to appear on the time scale of the instrument response throughout the 1–1.5 μm range. The maximum intensity of the rapidly rising initial signal (see Fig. 1) occurs at 1.3 μm (7700 cm^{-1}). Transient absorption in this region is typical for bacteriochlorophyll (BChl) *monomers* [32,33]. With increasing delay, absorption strength is seen to intensify in the 1–2 μm region and the transition peak shifts to shorter wavelength (1.1 μm or 9100 cm^{-1}). The kinetics of the near-IR spectral evolution are wavelength dependent. At shorter wavelengths, the difference signal rises with a time constant of 0.5–0.7 ps (0.56 ps at 1.1 μm). No initial signal is present in the 1.5–2.0 μm region, and absorption intensity grows in on a 0.85 ps time scale across this range. Similarly, the 2–4.0 μm spectral region is initially devoid of transient absorption after 800 nm photo excitation and a strong

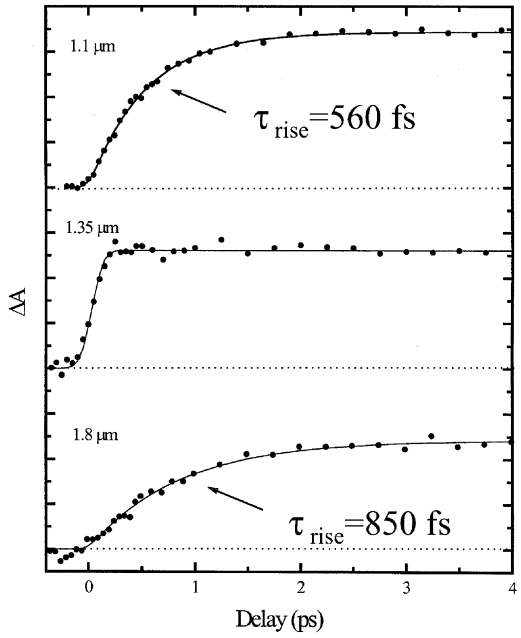

Fig. 1. LH2 near-infrared transient absorption signals across the 1–2 μm spectral region. These signals were fit (solid line in Fig. 2) to functions comprised of an instantaneous component with an instrument-response-limited rise time, and a rising exponential component. The instantaneous component was found to be absent at 1.8 μm, whereas the slower component was absent at 1.3 μm. Time constants, found from fitting the rising component to an exponential were in the range 0.5–0.9 ps.

transient absorption is seen to develop with increasing delay (see Fig. 2). The time scale of the appearance of transient absorption is ∼0.8–0.85 throughout this range. The correspondence of this ca. 0.8 ps time scale with that measured for

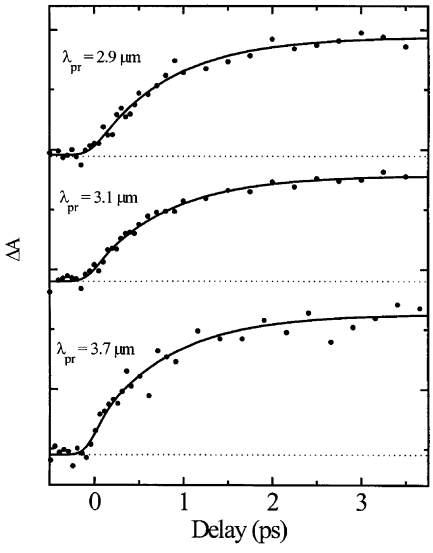

Fig. 2. LH2 infrared transient absorption signals across the 2.5–4.0 μm spectral region. The signals were fit to a single rising exponential function at each wavelength (solid lines); no instantaneous component was present. Time constants for the exponential rising function were in the range of ∼0.8–0.85 ps at all wavelengths in this interval.

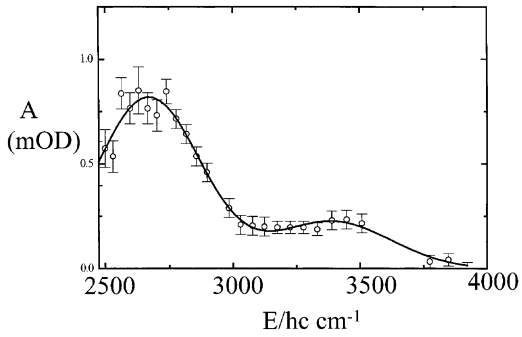

Fig. 3. LH2 infrared transient absorption spectrum in the 2.5–4.0 μm spectral region. The solid line represents a two-component Gaussian curvefit to the spectrum with maxima at 2700 and 3400 cm^{-1}.

B800–B850 energy transfer [3] establishes that the evolving signal is associated with the B850 pigments. The transient IR spectrum measured at a delay of 4 ps after 800 nm excitation, when there are no further changes occurring, is shown in Fig. 3 for the 2.5–4.0 μm region. An absorption peak occurs at 3.7 μm (2700 cm^{-1}) and a distinct second peak is seen at ca. 2.9 μm (3450 cm^{-1}).

3.2. Level structure of the B800 and B850 pigment systems

Fig. 4 depicts the situation following excitation at 800 nm. B800 excitations that are about 700 cm^{-1} higher in energy than those of B850 are first created. The B800 spectra are

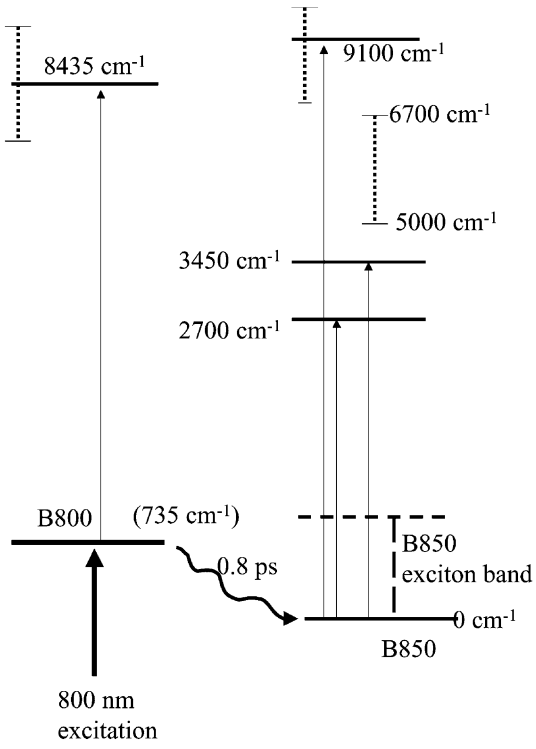

Fig. 4. Energy level scheme for the B800 and B850 pigment systems within LH2 proteins.

monomer-like in that they have a transient absorption at ca. 1.3 μm exciting the level at 8435 cm^{-1} in Fig. 4 [32,33] but none in the 3.7–1.5 μm region. The B850 ring becomes excited with a time constant of ca. 0.8 ps at which point the electronic excitations in the IR are significantly altered. A monomer-like transition is still observed but it is shifted to 9100 cm^{-1}, from the 7700 cm^{-1} transition seen with B800. Broad absorption, shown as a shaded region in Fig. 4, occurs in the 5000–6700 cm^{-1} region that is not present for B800. Finally, two new B850 transitions appear at 2700 and 3450 cm^{-1}. The 2700 cm^{-1} transition has a FWHM of ca. 450 cm^{-1} and is ca. four times more intense than the one at 3450 cm^{-1}. The energy zero for B850 transitions in this discussion and Fig. 4 is chosen as the bottom of the exciton band of eighteen B850 states which extend up to ca. 1000 cm^{-1} [3]. In reality, the B850 transitions will originate from a thermal distribution of these exciton states and hence will be band–band transitions [24], i.e. transitions between two bands of excitonic states. At 300 K the average energy of a B850 exciton is 380 cm^{-1} if a 250 cm^{-1} exciton coupling is assumed [3], so only a few levels at the bottom of the excitation band have significant populations in the equilibrium distribution.

The two peaks at 2700 and 3450 cm^{-1} are very unlikely to be vibrational transitions. Vibrational difference spectra could arise if the IR spectrum of the excited B850 state was quite different from that of the ground and B800 states. However, the C–H modes are the only fundamentals near to the observed frequencies, and they lie in between these two peaks. There are no other strong IR active modes in the observed spectral region. A more complete study of the full IR region down to 10 μm is expected to confirm this hypothesis.

The spectral changes that occur and the new bands that appear dramatise the very different electronic structures of B800 and B850. In the case of the excited special pair (P^*) of the reaction centres from photosynthetic bacteria, which is a strongly coupled dimer of BChl molecules, there are also electronic transitions in the IR between 1.25 and 5 μm [33]. These were attributed to charge resonance-transitions and double-triplet excitations [34]. The present results imply that, the B850 ring of cofactors is a much more strongly coupled set than B800 since the IR electronic transitions not present in the monomer or B800 must be related to the inter-cofactor coupling in B850.

The IR spectral dynamics observed in this study are consistent with the Q_y optical excitations being coupled so weakly that the charge resonance and the resulting spectral changes accompanying the passage of energy from this weakly interacting set of BChl pigments (B800) to a strongly coupled aggregate (B850) can be understood within the framework of the set of states shown in Fig. 4. The 800 nm excitation produces B800 excitations with distinctive IR absorption. Transitions of the B800 pigments are characteristic of monomeric BChls, by having Q_y excitations in the ~800 region and transitions from these to higher states at ~1.3 μm (8435 cm^{-1} region in Fig. 4). These near-IR transitions are broadened by the disordering of the individual transition energies. Within the B850 system, the cofactors interact to establish an exciton band of collective states. The inter-pigment couplings reduce the apparent inhomogeneity.

The spectral behaviour observed across the 1–4 μm region in this study is fully consistent with the scheme shown in Fig. 4. As excitation energy is transferred from B800 to B850 within 0.8 ps, the near-IR absorption at 1.3 μm blue-shifts to 1.1 μm and broadens significantly. These features at 1.1 μm probably also correspond to transitions from the thermally populated low-energy states of the Q_y manifold of B850 into the higher band that derives from BChl monomer two-photon states. The spectral broadening reflects a combination of inhomogeneity and the dispersion of exciton state energies within the lower B850 band. As a result of the phases of the excitations of the excitonic B850 states not all transitions to them from the ground are equally allowed. It is generally believed that only the first few levels at the bottom of the band of 18 states are involved in transitions to the ground state [3]. However, all transitions from B850 excitations to higher states are allowed with equal probability, thus, the absorption profile should track the room-temperature thermal populations of the exciton levels. The appearance of the new absorption features at 3.7, 2.7 μm and in the 1.5–2.0 μm range that are not seen for either BChl or B800 requires the presence of strong interactions between the BChls within B850 compared with B800. These are the band–band transitions [24]. We have not yet studied the B800/B850 spectral *evolution* in the 4–10 μm range. This will be necessary to determine whether some of these newly discovered excited states derive from monomer levels that are shifted to higher energy due to the inter-pigment coupling. More likely, they correspond to charge resonance [33] or double-triplet [34] levels that are stabilised and activated by the interactions within B850.

4. Summary and conclusions

The near-IR and IR transient spectral evolution reported in this article highlight the substantial differences in the electronic structure of the B800 and B850 pigments at room temperature. Following Q_y photo excitation of the B800 chromophores, transient signals in the near-IR region are seen to evolve from an initial monomer-like spectrum into a broad, asymmetric feature suggestive of band–band transitions within a weakly disordered system. Concomitant with the near-IR spectral changes, new absorption bands appear in the 2.5–4.0 μm region on the time scale characteristic of B800–B850 energy transfer. These findings emphasise the presence of strong inter-pigment interactions in the B850 aggregate that are absent in the B800 chromophores.

A number of recent studies have revealed significant differences in the optical properties of B800 and B850 pigments at cryogenic temperatures. Hole-burning and

single-molecule fluorescence experiments have demonstrated that the B850 band behaves as a superposition of non-interacting monomeric absorptions, whereas the B850 transitions are characteristic of collective excitations [6,8,20]. The separations between B850 eigenstates, however, could be satisfactorily explained within the framework of a simple exciton model based on crystallographic structures of LH2 proteins. Interactions of the exciton levels with charge-resonance states and structural distortions have been recognised as possible causes of the observed level splittings.

While the line-narrowing measurements confirm the presence of delocalized B850 exciton states at cryogenic temperatures, elucidation of the light-harvesting mechanism requires understanding of pigment electronic properties under physiological conditions. The results from this study confirm that room-temperature protein fluctuations certainly do not cause complete energy localisation within B850. Furthermore, the IR measurements have revealed low-energy inter-band transitions that provide important information on states that interact with the optical exciton levels but are not detected via one-photon measurements from the ground state. The nature of these levels need to be examined by polarised femtosecond IR measurements in order to establish a detailed understanding of them and the interactions that drive photosynthetic energy transfer.

It is of some interest to compare the transitions seen here with those reported in the two-photon spectra of LH2 [35]. These single beam two-photon spectra show a peak at a two-photon wavenumber of $13{,}900 \pm 150\,\mathrm{cm}^{-1}$. Therefore, they correspond to transitions to a state that is about $2235\,\mathrm{cm}^{-1}$ above the energy corresponding to 850 nm. The lowest energy peak we see is $2700\,\mathrm{cm}^{-1}$ above this energy (see Fig. 4). However, the two results should not coincide, because these two-photon spectra are ascribed to the carotenoid spheroidene [35]. However, it is interesting that the excited states reported here do not appear in these two photon experiments, indicating that they have very small two photon cross-sections for light in the $7000\,\mathrm{cm}^{-1}$ region and are not yet detectable by two photon induced fluorescence. Because the new IR transitions have vanishingly small absorption cross-sections in transitions from the ground state they must have very small exciton bandwidths. The contribution to the exciton bandwidth from the multipole interactions (through-space electrostatic interactions) must be vanishingly small leaving only the electron exchange interactions. The latter are dependent on the overlap of wave functions on different molecules and must also be extremely small [23], in the range of a few cm^{-1}. It follows that the shape of these IR transitions of B850 should signal the distribution of exciton states in B850, since the distribution in the final states is so narrow. These band–band spectra are also expected to be sensitive to disorder. Therefore, we anticipate that as more accurate transient IR spectra become available they will provide essential information regarding both the coupling and the energy transport in the B850 assembly.

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References

[1] G. Porter, Femtosecond processes in photosynthesis, in: J. Mantz, L. Woeste (Eds.), Femtosecond Chemistry, VCH Publishers, Weinheim, Germany, 1995, pp. 625–632.
[2] G.R. Fleming, R. van Grondelle, Femtosecond spectroscopy of photosynthetic light harvesting systems, Curr. Opin. Struct. Biol. 7 (1997) 738–748.
[3] W. Sundstrom, T. Pulletits, R. van Grondelle, Photosynthetic light-harvesting: reconciling dynamics and structure of purple bacterial LH2 reveals function of photosynthetic unit, J. Phys. Chem. B 103 (1999) 2327–2346.
[4] G. McDermott, S.M. Prince, A.A. Freer, A.M. Hawthornthwaite-Lawless, M.Z. Papiz, R.J. Cogdell, N.W. Isaacs, Crystal structure of an integral membrane light-harvesting complex from photosynthetic bacteria, Nature (London) 374 (1995) 517.
[5] J. Koepke, X. Hu, C. Muenke, K. Schulten, H. Michel, The crystal structure of the light-harvesting complex II (B800-850) from *Rhodospirillum molischianum*, Structure 4 (1996) 581–597.
[6] N.R.S. Reddy, G.J. Small, M. Seibert, R. Picorel, Energy transfer dynamics of the B800-B850 antenna complex of *Rhodobacter sphaeroides*: a hole burning study, Chem. Phys. Lett. 181 (1991) 391–399.
[7] H.-M. Wu, M. Ratsep, R. Jankowiak, R.J. Cogdell, G.J. Small, Comparison of the LH2 antenna complexes of *Rhodopseudomonas acidophila* (Strain 10050) and *Rhodobacter sphaeroides* by high-pressure absorption, high-pressure hole burning, and temperature-dependent absorption spectroscopies, J. Phys. Chem. B 101 (1997) 7641–7653.
[8] A.M. van Oijen, M. Ketelaars, J. Kohler, T.J. Aartsma, J. Schmidt, Unraveling the electronic structure of individual photosynthetic pigment-protein complexes, Science 285 (1999) 400–402.
[9] S.E. Bradforth, R. Jiminez, F. van Mourik, R. van Grondelle, G.R. Fleming, Excitation transfer in the core light-harvesting complex (LH-1) of *Rhodobacter sphaeroides* — an ultrafast fluorescence depolarisation and annihilation study, J. Phys. Chem. 99 (1995) 16179.
[10] R. Jiminez, S.N. Dikshit, S.E. Bradforth, G.R. Fleming, Electronic excitation transfer in the LH2 complex of *Rhodobacter sphaeroides*, J. Phys. Chem. 100 (1996) 6825.
[11] V. Nagarajan, R.G. Alden, J.C. Williams, W.W. Parson, Ultrafast Exciton relaxation in the B850 antenna complex of *Rhodobacter sphaeroides*, Proc. Natl. Acad. Sci. U.S.A. 93 (1996) 13774.
[12] M. Chachisvilis, O. Kuhn, T. Pullerits, V. Sundstrom, Excitons in photosynthetic purple bacteria: wavelike motion or incoherent hopping? J. Phys. Chem. B 101 (1997) 7275.
[13] T. Pullerits, M. Chachisvilis, V. Sundstrom, Exciton delocalization length in the B850 antenna of *Rhodobacter sphaeroides*, J. Phys. Chem. 100 (1996) 10787.
[14] R. Kumble, S. Palese, R.W. Visschers, P.L. Dutton, R.M. Hochstrasser, Ultrafast dynamics within the B820 sub-unit from the core (LH-1) antenna complex of *Rhodospirillum rubrum*, Chem. Phys. Lett. 261 (1996) 396.
[15] H.M. Visser, O.J.G. Somsen, F. van Mourik, S. Lin, I.H.M. van Stokkum, R. van Grondelle, Direct observation of sub-picosecond equilibration of excitation energy in the light-harvesting antenna of *Rhodospirillum rubrum*, Biophys. J. 69 (1995) 1083.

[16] S. Hess, E. Akesson, R.J. Cogdell, T. Pullerits, V. Sundstrom, Energy transfer in spectrally inhomogeneous light-harvesting pigment-protein complexes of purple bacteria, Biophys. J. 69 (1995) 2211.
[17] J.T.M. Kennis, A.M. Streltsov, T.J. Aartsma, T. Nozawa, J. Amesz, Energy transfer and exciton coupling in isolated B800–850 complexes of the photosynthetic purple sulphur bacterium *chromatium tepidum*. the effect of structural symmetry on bacteriochlorophyll excited states, J. Phys. Chem. 100 (1996) 2438.
[18] J.T.M. Kennis, A.M. Streltsov, H. Permentier, T.J. Aartsma, J. Amesz, Exciton coherence and energy transfer in the lh2 antenna complex of Rhodopseudomonas acidophila at low temperature, J. Phys. Chem. B 101 (1997) 8369.
[19] A.P. Shreve, J.K. Trautman, H.A. Frank, T.G. Owens, A.C. Albrecht, Femtosecond energy-transfer processes in the B800–850 light-harvesting complex of *Rhodobacter sphaeroides* 2.4.1, Biochem. Biophys. Acta 1058 (1991) 280.
[20] L.D. Book, A.E. Ostafin, N. Ponomarenko, J.R. Norris, N.F. Scherer, Exciton delocalization and initial dephasing dynamics of purple bacterial LH2, J. Phys. Chem. B 104 (2000) 8295–8307.
[21] T. Meier, V. Chernyak, S. Mukamel, Multiple exciton coherence sizes in photosynthetic antenna complexes viewed by pump–probe spectroscopy, J. Phys. Chem. B 101 (1997) 7332–7342.
[22] T. Meier, Y. Zhao, V. Chernyak, S. Mukamel, Polarons, localisation, and excitonic coherence in superradiance of biological antenna complexes, J. Chem. Phys. 107 (1997) 3876–3893.
[23] S. Gnanakaran, G. Haran, R. Kumble, R.M Hochstrasser, Energy transfer and localisation: applications to photosynthetic systems, in: D.L. Andrews, A.A. Demidov (Eds.), Resonance Energy Transfer, Wiley, West Sussex, UK, 1999.
[24] R. Kumble, R.M. Hochstrasser, Disorder-induced exciton scattering in the light-harvesting systems of purple bacteria: influence on the anisotropy of emission and band–band transitions, J. Chem. Phys. 109 (1998) 855–865.
[25] V.L. Broude, E.I. Rashba, E.F. Sheka, Spectroscopy of Molecular Excitons. Springer Series in Chemical Physics, Vol. 16, Springer, Berlin, 1985, p. 271.
[26] E.I. Rashba, Fiz. Tverd. Tela (Leningrad) 5 (1963) 1040.
[27] S.D. Colson, D.M. Hanson, R. Kopelman, G.W. Robinson, Direct observation of the entire exciton band of the first excited singlet states of crystalline benzene and naphthalene, J. Chem. Phys. 48 (1968) 2215.
[28] R.M. Hochstrasser, T.-Y. Li, H.-N. Sung, J. Wessel, A.H. Zewail, Experimental studies of triplet exciton bands of molecular crystals, Pure Appl. Chem. 37 (1974) 85.
[29] T.-Y. Li, Ph.D. Dissertation, University of Pennsylvania, Philadelphia, 1974.
[30] R.M. Hochstrasser, J.D. Whiteman, Exciton band structure and properties of a real linear chain in a molecular crystal, J. Chem. Phys. 56 (1972) 5945.
[31] R.J. Cogdell, A.M. Hawthornthwaite, Preparation, purification and crystallisation of purple bacterial antenna complexes, in: J.R. Norris, J. Deisenhofer (Eds.), The Photosynthetic Reaction Centre, Academic Press, New York, 1993, pp. 23–42.
[32] G. Haran, K. Wynne, C.C. Moser, P.L. Dutton, R.M. Hochstrasser, Femtosecond infrared studies of photosynthetic reaction centres: new charge transfer bands and ultrafast energy redistribution, Ultrafast Phenomena X, Springer, Berlin, 1996, pp. 326–327.
[33] K. Wynne, G. Haran, G.D. Reid, C.C. Moser, P.L. Dutton, R.M. Hochstrasser, Femtosecond infrared spectroscopy of low-lying excited states in reaction centres of *Rhodobacter sphaeroides*, J. Phys. Chem. 100 (1996) 5140–5148.
[34] P.O.G. Scherer, S. Fischer, Interpretation of a low lying excited state of the reaction centre of *Rhodobacter sphaeroides* as a double triplet, Chem. Phys. Lett. 268 (1997) 133–142.
[35] B.P. Krueger, J. Yom, P.J. Walla, G.R. Fleming, Observation of the S1 state of spheroidene in LH2 by two-photon fluorescence excitation, Chem. Phys. Lett. 310 (1999) 57–64.