Unveiling the excited state energy transfer pathways in peridinin-chlorophyll α-protein by ultrafast multi-pulse transient absorption spectroscopy

Kipras Redeckas a,⁎, Vladislava Voiciuka a, Donatas Zigmantas b, Roger G. Hillerc, Mikas Vengrisa

a Department of Quantum Electronics, Faculty of Physics, Vilnius University, Saulėtekio av. 10, LT-10223 Vilnius, Lithuania
b Department of Chemical Physics, Lund University, P.O. Box 124, 22100 Lund, Sweden
c Department of Biological Sciences, Macquarie University, NSW 2109, Australia

A R T I C L E   I N F O

Article history:
Received 15 November 2016
Received in revised form 15 January 2017
Accepted 31 January 2017
Available online 2 February 2017

Keywords:
Peridinin-chlorophyll α-protein
Pump-probe
Pump-dump-probe
Intramolecular charge transfer
Excited state equilibrium
Excitation energy transfer

A B S T R A C T

Time-resolved multi-pulse methods were applied to investigate the excited state dynamics, the interstate couplings, and the excited state energy transfer pathways between the light-harvesting pigments in peridinin-chlorophyll α-protein (PCP). The utilized pump-dump-probe techniques are based on perturbation of the regular PCP energy transfer pathway. The PCP complexes were initially excited with an ultrashort pulse, resonant to the S0 → S1 transition of the carotenoid peridinin. A portion of the peridinin-based emissive intramolecular charge transfer (ICT) state was then depopulated by applying an ultrashort NIR pulse that perturbed the interaction between S1 and ICT states and the energy flow from the carotenoids to the chlorophylls. The presented data indicate that the peridinin S1 and ICT states are spectrally distinct and coexist in an excited state equilibrium in the PCP complex. Moreover, numeric analysis of the experimental data asserts ICT → Chl-α as the main energy transfer pathway in the photoexcited PCP systems.

1. Introduction

Peridinin-chlorophyll α-protein (PCP) is a light-harvesting complex, found in marine dinoflagellates Amphidinium carterae [1]. PCP attracts significant scientific interest, as it is established as an attractive model system for pigment-pigment interaction study [2–9]. A very important contribution to the progress of PCP research was made by the determination of its high-resolution crystal structure [10,11]. It revealed a tight arrangement of pigments in the protein subunits with the highest known carotenoid-to-chlorophyll ratio (4:1) among light-harvesting proteins. The close interactions between pigments are essential for effective excitation energy transfer (EET), with efficiency values as high as ca. 90% [12–16] reported for PCP. Such high EET efficiency is achieved due to excellent light harvesting abilities of peridinin [17], the primary pigment in PCP. Peridinin is a highly substituted carotenoid, containing a lactone ring and an allene moiety [18]. These substituents break its symmetry, which explains the presence of significant emission from the lowest excited state, which is usually very weak (or completely indiscernible) for carotenoids, as it is symmetry-forbidden [19,20]. Furthermore, the lowest excited state manifold of peridinin is rather unusual, as it involves an intramolecular charge transfer (ICT) state, in addition to the carotenoid-“typical” S1 [17,21–25], which is responsible for the emission observed in polar solvents. ICT state was first identified in peridinin in polar solvents, where it was proposed to account for the strong dependence of the excited state lifetime on solvent polarity [17,21,26]. For peridinin in solution, it was shown that S1 and ICT are distinct states with a dynamic equilibrium between them [27,28]. For peridinin in the PCP complex, the relation between the ICT and S1 states is not clearly understood as of yet, as the lifetime of excited peridinin is reduced dramatically due to the rapid EET to chlorophyll α (Chl-α). Several investigations [14,29–35] elucidated the general scheme of excited state dynamics in PCP, where a fraction of excitation energy is transferred to Chl-α directly from the peridinin second excited state S2, while the main channel of EET is the lowest excited state of peridinin. The latter state is usually referred to as a collective S1/ICT state. A work by Zigmantas et al. suggested that within the PCP complex, the peridinin ICT state is directly involved in energy transfer, even though it cannot be separated from S1 due to strong coupling between the states [36]. On the other hand, Stokkum et al. proposed a model of energy transfer and excited state dynamics in PCP and found no evidence of presence of the ICT state as a separate entity [37].

In this study, we focus on the nature of the S1/ICT state and its involvement in energy transfer in the PCP complex. We apply dispersed pump-dump-probe (PDP) spectroscopy, which was successfully employed to resolve peridinin S1 and ICT states in solution [27,28]. In the PDP experiment, the excited state population is selectively depleted by a dump pulse, which is tuned to interact with the emissive state [38,
In the PCP complex, the peridinin ICT state is distinguished by a stimulated emission (SE) band in the NIR, peaking at ca. 930 nm [36]. This SE band can be observed during the very early stages after the excitation, and at later times it is completely replaced by positive signal due to the increasing contribution of Chl-α excited state absorption (ESA). In our PDP experiments, the SE of the ICT state was deliberately triggered with a 950 nm pulse timed to arrive at 1.2 ps after the actinic excitation. To capture the essence of the complex experimental data, we propose a redefined model of the PCP excited state dynamics and apply global analysis to describe the EET flow. The analysis suggests the role of the ICT state as the major channel of energy transfer, while the S₁ state undergoes relaxation via a single pathway, namely, by shifting its population to the ICT state.

2. Materials and methods

2.1. Sample preparation

PCP complexes (isolated as described earlier in refs. [10,40] and stored in the dark at −40 °C) were dissolved in buffer solution (25 mM TRIS, 2 mM KCl, pH 7.5). Solution concentration was adjusted to ca. 0.3 OD at the excitation wavelength (520 nm) in a 1 mm quartz cuvette. All the experiments were performed at room temperature. The sample cuvette was constantly translated with respect to irradiation beams during the measurements to distribute the energy equally within the solution. In addition, sample stability was controlled by measuring the steady-state absorption spectrum before and after the experiment, and the differences were below 5% (the degradation was only very minor, most likely due to the fact that dump pulse is not absorbed by the PCP molecules in the steady state). In addition, the difference absorption traces measured in the different stages of experiment (which consisted of a large number of kinetic scans) were identical within the signal-to-noise (see Fig. SM-1 in the Supplementary Material for more detail).

2.2. Time-resolved spectroscopy

Pump-probe (PP) and pump-dump probe (PDP) spectra were measured with a home-built multi-pulse setup described in detail earlier [41,42]. Briefly, two tunable femtosecond actinic pulses, used for exciting the sample and dumping the excited state population back to the ground state, were produced using two travelling-wave optical parametric amplifiers (Topas-800, Light Conversion) pumped by a commercial Ti:Sapphire laser system (Libra, Coherent; 3.5 W at 1 kHz, 50 fs). Both pulses were individually timed using separate optical delay lines. The actinic excitation (pump) wave- and PDP spectra could be measured. The temporal resolution of the setup was approximately 120 fs. The actinic excitation (pump) wavelength was set at 520 nm (see Fig. 1), whereas dump wavelength was 950 nm. The polarizations of pump and dump beams were parallel to each other and were set at 54.7° with respect to the polarization of the probe. The energy of the pump pulse was in the vicinity of 100 nJ, whereas that of the dump pulse was in the order of 600 nJ. To cover a broad spectral range, transient data were separately recorded in two overlapping spectral windows (430–740 nm and 700–1000 nm) in two successive PDP experiments and were later merged into a single dataset for data analysis.

3. Results

3.1. Pump-probe dynamics

PCP complexes were excited at 520 nm, i.e., a wavelength corresponding to the low-energy wing of peridinin S₀ → S₂ absorption (see Fig. 1) [36], thus reducing to a minimum the contributions arising from vibrational relaxation within the S₂ state. PCP PP data have already been presented and discussed in previous studies (for visible region see refs. [14,37], results in the NIR are given in ref. [36]), and are thus not addressed in greater detail in this work. To summarize, the 520 nm excitation initially populates the optically-allowed S₂ state of peridinin, resulting in the appearance of SE at 540–560 nm and ESA at 820–880 nm in the sub-100-femtosecond transient absorption spectra (see Fig. 2). Within a sub-100-femtosecond period after the actinic excitation, S₂ is populated and the resulting sub-5-pico-second transient absorption spectra are mostly comprised of contributions from peridinin—namely, ground state bleaching (GSB), peaking at ca. 485 nm, and ESA, spanning the broad spectral range from 550 to 800 nm, with a maximum located at ca. 650 nm. This early spectral outline decays concurrently with the increasing contribution of Chl-α signal, observed at early times most distinctly as a sharp dip at 672 nm. The transformation of peridinin-to-chlorophyll signal continues until a plateau is reached at ca. 10 ps. Besides the GSB/SE band at 672 nm, corresponding to Chl-α Qₐ transition, Chl-α signals also contain a less intense GSB band at the blue-most edge of the experimental window (seen both in the early and the late transient absorption spectra), peaking at around 430 nm. In addition, a weak and featureless Chl-α ESA is displayed throughout the entire VIS-to-NIR spectrum. Such evolution of transient absorption directly reflects the process of EET from peridinin to Chl-α.

The region of special importance to this work is located in the NIR, around 800–1000 nm (see the enlargement in Fig. 2(b)), where a low-intensity negative band appears at early probe delay times (<3 ps). This particular band has been ascribed to SE originating from the peridinin ICT state [36]. Subsequently, the sign of the NIR-based signal is reversed to positive due to the eventual emergence of broadband, mostly featureless, long-lived Chl-α ESA (see Fig. 2(a)). In order to identify the contributions of the ICT state to the excited state dynamics of the entire PCP complex, we introduced a dump pulse, resonant with this emissive band. If the ICT state can be separated from the other concurrent states at this delay time, the dump pulse should allow us to single out its contribution to the PP signal by depopulating it, without affecting other excited-state species. Even though possible in principle, the repumping effect on the Chl-α species (i.e., population transfer to higher electronic states) is expected to be negligible, as at the time of dump transient absorption signal is dominated by the contributions from peridinin and most excitations are not yet transferred to Chl-α, which is supported by the fact that the overall PP signal is negative and the
دریافت فوری متن کامل مقاله

امکان دانلود نسخه تمام متن مقالات انگلیسی
امکان دانلود نسخه ترجمه شده مقالات
پذیرش سفارش ترجمه تخصصی
امکان جستجو در آرشیو جامعی از صدها موضوع و هزاران مقاله
امکان دانلود رایگان ۲ صفحه اول هر مقاله
امکان پرداخت اینترنتی با کلیه کارت های عضو شتاب
دانلود فوری مقاله پس از پرداخت آنلاین
پشتیبانی کامل خرید با بهره مندی از سیستم هوشمند رهگیری سفارشات