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Superradiance and Exciton (De)localization in Light-Harvesting Complex II from Green Plants?[†]

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Fluorescence quantum yield and fluorescence lifetime measurements were performed on trimeric light-harvesting complex II (LHCII) from spinach in the temperature range 7–293 K. From the results the radiative rate was calculated, which is related to the amount of delocalization of excitations over different pigments because of intermolecular interactions. The emitting dipole strength of LHCII is very similar to that of unbound Chl *a*, and it appears to be almost independent of temperature. The apparent increase of the radiative rate upon lowering the temperature can largely be explained by the shrinking of the sample. It is concluded that at all temperatures the amount of exciton delocalization in LHCII is small.

Introduction

Photosynthesis is the process by which sunlight is converted into chemical energy in the form of organic compounds. The first important steps in this process are the absorption of light by the light-harvesting antenna and the efficient transport of the excited-state energy to the reaction center where a charge separation is initiated.¹ In green plants, more than 50% of the light absorption is accomplished by light-harvesting complex II (LHCII).² Since its three-dimensional structure at 3.4 Å resolution became available,³ intensive investigation and debate concerning the relation between the structure and spectroscopic features have led to a detailed view of the functional properties of LHCII (for a recent review see ref 4). In vivo, this complex exists in a trimeric form, the average number of trimers per PSII reaction center being four.⁵ The trimer contains between 36 and 42 chlorophyll molecules (Chl *a* to Chl *b* ratio ~ 1.4) and 9–12 xanthophyll molecules (lutein, neoxanthin, and violaxanthin in the ratio 2:1:0.07–1). The distances between neighboring Chl *a* and Chl *b* fall in the range 8.3–10.5 Å, whereas identical pigments are further apart, according to the pigment assignment of Kühlbrandt et al.³ Further progress in the elucidation of the pigment identities has been achieved by reconstitution experiments,^{6–8} showing that some pigment binding sites can bind both Chl *a* and Chl *b* (sites A3 and B3), others bind preferentially Chl *b* instead of Chl *a* (A6 and A7), and one binds only Chl *a* instead of Chl *b* (B1).⁸

Important points of discussion^{4,9–18} have been the excitonic interactions in LHCII and related to this the dipole strength of the lowest-energy electronically excited state which can be estimated with different experimental techniques and theoretical

modeling. In the presence of strong interactions between pigments, an excitation can be shared by these molecules and become delocalized (excitons), and the dipole strength of each excitonic state can adopt a value that may deviate significantly from that of monomeric pigments. In the case of weak interactions, the excitations are more or less localized on individual molecules, and the dipole strengths of the corresponding electronic transitions are close to that of an individual pigment.

Not only the intermolecular couplings but also the pigment–protein interactions play a role in the amount of delocalization of the excitation: the distinct environments of pigments bound at various sites lead to differences in their electronic transition energies (different site energies). Variations in the environment of individual pigments (static disorder) lead to the inhomogeneous broadening of absorption bands, and exciton–phonon interactions lead to additional homogeneous broadening.¹³ The extent of delocalization depends on the magnitude of the pigment–pigment coupling compared to the amount of broadening.^{19–28} The larger the spread in site energies and the amount of broadening, the less delocalized the excitations are.

In the case of LHCII, there is a lack of knowledge about the orientations of the transition dipole moments and therefore also about the sizes of the interaction strengths between the chlorophylls. However, using the crystal structure and the advances in the identification of the pigments, the interactions between them were estimated, considering dipole–dipole coupling in the point-dipole approximation.⁴ These calculations showed that the strongest coupling strength between the Chl *a* Q_y transitions is less than the inhomogeneous width of ~ 120 cm^{−1} and the homogeneous width of ~ 185 cm^{−1} at RT for the Q_y transition.²⁹ From circular dichroism measurements,^{9–11} it was concluded earlier that significant excitonic interactions occur among the chlorophylls, but the size of the coupling strength was not determined. Krawczyk et al.¹⁴ measured the Stark spectrum of LHCII and concluded that excitonic interactions are present, although the Chl *a* absorption bands, including the lowest-energy one, mostly appeared to behave like those of uncoupled Chl's.

[†] **Abbreviations.** PSII, photosystem II; Chl *a*, chlorophyll *a*; Chl *b*, chlorophyll *b*; fwhm, full width at half-maximum; LHCII, light-harvesting complex II; *n*, refractive index; OD, optical density; RT, room temperature; ACE, acetone; MeOH, methanol; k_{rad} , radiative rate.

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Several authors have addressed the issue concerning the number of Chl *a* molecules that contribute to the fluorescence at 4.2 K. Peterman et al.¹⁸ measured fluorescence line narrowing (FLN) at 4.2 K in LHCII. The FLN spectra showed vibronic features of at least two different Chl *a* molecules. This led to the conclusion that the emitting state at that temperature is delocalized over at least two different Chl *a* molecules or that emission arises from at least two independent molecules. Many spectroscopic properties of LHCII could be modeled in the same study by assuming that excitations corresponding to the lowest-energy transitions were essentially localized on individual pigments. Within the context of this model, because of the inhomogeneous spread in the site energies, 9–12 different Chl *a* molecules per trimer have approximately equal probabilities to be the lowest-energy pigment and the dipole strength of the lowest state is equal to that of monomeric Chl *a*.¹⁸ Pieper et al.¹⁵ concluded from hole-burning experiments that the lowest-energy states had a dipole strength corresponding to ~ 0.8 chlorophyll *a* molecules. Renger and May performed exciton calculations¹⁷ and concluded that significant excitonic interactions must be present, and the dipole strength of the lowest state was estimated to be ~ 0.5 . Therefore, no consensus has been reached about the degree of delocalization and the dipole strength of the lowest excited state.

The dipole strength of the lowest state(s) can also be obtained by comparing the radiative rate k_{rad} of LHCII to that of free monomeric Chl *a* molecules. This rate scales linearly with the dipole strength of the fluorescing state, and a strongly increased or decreased rate (the former corresponds to the phenomenon of superradiance) provides evidence for the formation of delocalized states.¹⁹ Superradiance has been observed in semiconductor quantum wells,³⁰ quantum wires,³¹ and J-aggregates^{32–34} and recently in a biological antenna complex.²⁵ Some concepts that account for the spectroscopic results of J-aggregates have also been applied successfully to light-harvesting complexes,^{20,21,35} but there are also some important differences. The major one is that broadening effects are much larger in pigment–protein complexes which limit the exciton delocalization length and thus the enhancement of the superradiative decay. This effect is relevant because energy transfer by incoherent hopping becomes more important at the expense of excitation delocalization.

In this paper, we present fluorescence quantum yield and lifetime measurements on trimeric LHCII, from which the dipole strength of the fluorescent state(s) is calculated. Our results show that the emitting dipole strength in this complex is 1.18 times higher at RT than that for monomeric free Chl *a*. Upon lowering the temperature, the dipole strength experiences hardly any temperature dependence, and at 7 K we found that the lowest-energy state carries an emitting dipole strength that corresponds to 1.14 Chl *a* molecules, very close to the value expected of a monomeric type of behavior. Consequences for the amount of delocalization are addressed, and existing models are tested in light of these results.

Materials and Methods

Chl *a* was purchased from Sigma and dissolved in acetone and methanol. Trimeric LHCII was purified from spinach³⁶ using anion-exchange chromatography and the detergent *n*-dodecyl- β ,D-maltoside (Sigma) for solubilization of the complexes. The samples were diluted in a medium containing 20 mM Hepes buffer at pH 7.8, 65% (v/v) glycerol to ensure a transparent sample at low temperatures, and 0.09% (w/v) *n*-dodecyl- β ,D-maltoside, except for the determination of k_{rad} at RT in which

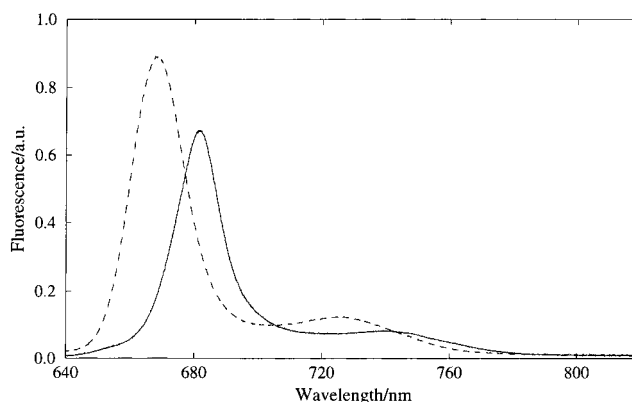


Figure 1. Room-temperature fluorescence emission spectra of LHCII (solid line) and Chl *a* (dashed line) in acetone upon excitation at 609 nm. The spectra were corrected for the wavelength-dependent sensitivity of the detection and for the number of absorbed photons.

case no glycerol was used. Measurements were performed in 1.0 cm path length acryl cuvettes at OD = 0.05 at the Q_y absorption peak to minimize the effects of self-absorption, except for the case of time-resolved measurements at RT, in which the OD was 0.15 in a rotating cell with a path length of 3 mm. A cryosystem Utreks-LSO which consists of an Utreks helium bath cryostat in conjunction with a temperature controller unit was used to vary the temperature. During the measurements, the alignment of the setup was kept constant. Fluorescence emission spectra were detected at 90° using a $\frac{1}{2}$ m spectrograph (Chromex) and a cooled CCD camera (Texas Instruments) and corrected for the wavelength-dependence sensitivity of the detection system. The sample was excited at 609 nm using a 150 W tungsten halogen lamp and an interference filter with a fwhm of 20 nm. The excitation intensity was ~ 1.5 mW/cm². Room-temperature fluorescence quantum yields were calculated by dividing the ratio of integrated fluorescence intensities on a wavenumber scale of Chl *a* and LHCII by the ratio of their total absorption factor $(1 - T)$ for the spectral region where the samples were excited, that is, around 609 nm. At lower temperatures, they were corrected for the shrinking of the sample diluted in a buffer with glycerol (see Results). Fluorescence lifetimes were measured with a time resolution of 5 ps at RT and 20 ps at lower temperatures using a streak-camera system (Hamamatsu) coupled to a CCD camera. The sample was excited with a repetition rate of 125 kHz by means of a frequency-doubled output of an Optical Parametric Amplifier which was pumped by a Ti–sapphire laser. The excitation was approximately 300 μ W/cm², and the excitation wavelength was 400 nm.

Results

Room Temperature. Figure 1 shows the emission spectra at RT of LHCII (solid line) and Chl *a* (dashed line) in acetone normalized on the number of absorbed photons. The Chl *a* spectrum exhibits a peak at 668.3 nm with a fwhm of 20 nm, whereas the LHCII emission spectrum peaks at 681.4 nm and has a fwhm of 18 nm. At room temperature, the calculated fluorescence quantum yield of LHCII was 0.22 ± 0.1 , taking a quantum yield for Chl *a* in acetone of 0.30.³⁷

The fluorescence lifetime measured for Chl *a* in acetone was 6.1 ± 0.2 ns, close to the value given by Vladkova (6.15 ± 0.2 ns).³⁸ In the case of LHCII, the decay was essentially monophasic with a decay time of 4.0 ± 0.1 ns. A minor component (1%) with a lifetime of 0.30 ± 0.01 ns was also present. Similar results were recently obtained by Barzda on trimeric LHCII from peas

TABLE 1. Room-Temperature Fluorescence Quantum Yields and Lifetimes of LHCII and Chl *a*

sample	quantum yield ($\pm 3\%$)	lifetime (ns) ($\pm 3\%$)	rad rate ^a (ns ⁻¹) ($\pm 6\%$)	refractive index <i>n</i>	emit dipole strength (D ²) ($\pm 6\%$)
Chl <i>a</i>	0.30 ^b	6.13	0.049	1.36	1.00
LHCII	0.22	4.00	0.054	1.33	1.18

^a Radiative rates were calculated according to $k_{\text{rad}} = \phi_{\text{fl}}/\tau_{\text{fl}}$. ^b Taken from ref 37. ^c Emitting dipole strengths were calculated using eq 1. The value of the dipole strength is an average of the calculated ones for LHCII when compared to Chl *a* in acetone and methanol, which are each less than 4% different from the one shown. ^d Note that the difference in dipole strengths has been corrected for the difference in refractive indexes according to eq 2.

(single exponential with the lifetime 3.5 ns).³⁹ Upon lowering the detergent concentration, aggregation did occur which led to the appearance of an additional faster component. Some aggregation possibly also explains the observation of multiexponential decays in earlier studies: Ide et al.⁹ reported lifetimes of 3.53 ± 0.4 ns with the amplitude 0.78 and 1.12 ± 0.1 ns with the amplitude 0.22; Vasil'ev et al.⁴⁰ obtained two lifetimes, 4.3 and 1.9 ns, with amplitudes 0.94 and 0.06, respectively; and Seydack et al.⁴¹ even found four exponential decays with lifetimes 4.0, 1.1, 0.16, and 0.008 ns with amplitudes 0.49, 0.18, 0.33, and 0.3, respectively.

In Table 1, we have listed the fluorescence quantum yields ϕ_{fl} and lifetimes τ_{fl} found at RT for LHCII and Chl *a* with the radiative rates calculated according to the relation $k_{\text{rad}} = \phi_{\text{fl}}/\tau_{\text{fl}}$.

From the radiative rate, the emitting dipole strength $|\vec{\mu}|^2$ can be obtained using the equation for the rate of spontaneous emission for molecules present in a medium with refractive index *n*:

$$k_{\text{rad}} = A_{10}(n) = n \frac{16\pi^3 \nu^3}{3\epsilon_0 h c^3} |\vec{\mu}|^2 \quad (1)$$

where ϵ_0 is the vacuum dielectric constant, *h* is Planck's constant, *c* is the speed of light, and ν is the emission frequency. We have used $|\vec{\mu}|^2$ instead of $f^2 |\vec{\mu}|^2$ which was given in ref 42. Note that $|\vec{\mu}|^2$ is dependent on *n* and a phenomenological relation was recently proposed by Knox⁴³ to account for the effect of the medium on the dipole strength of Chl *a*:

$$|\vec{\mu}|^2 = 20.07 + 24.31(n - 1) D^2 \quad (2)$$

To compare the dipole strengths of LHCII and Chl *a*, we correct for the effect of the difference in the medium surrounding Chl *a* in acetone or methanol and in LHCII by applying this equation. The refractive indexes for acetone and methanol are 1.36 and 1.33, respectively. For LHCII, we estimated the refractive index of the solvent at all temperatures (see below). The choice of the solvent and not the value of the direct protein environment seems justified because the wavelength of the light that interacts with the pigments is much larger than the size of LHCII, although the effect of the protein can probably not be ignored completely. At room temperature in the case of LHCII, the refractive index of water (1.33) was used. We approximated ν^3 by

$$\langle \bar{\nu}^3 \rangle = \frac{\int \bar{\nu}^3 I(\bar{\nu}) d\bar{\nu}}{\int I(\bar{\nu}) d\bar{\nu}} \quad (3)$$

In the last expression, $\langle \rangle$ means average, $\bar{\nu}$ the frequency in wavenumbers, and $I(\bar{\nu})$ the emission spectrum on a wavenumber

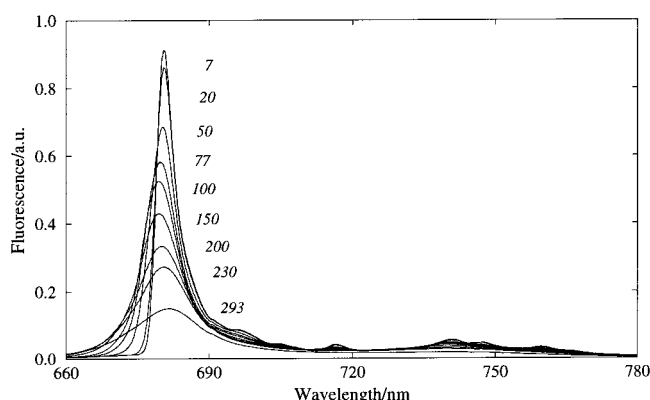


Figure 2. Fluorescence emission spectra of LHCII at different temperatures. The spectra were corrected for the wavelength dependence of the detection. The excitation wavelength was 609 nm.

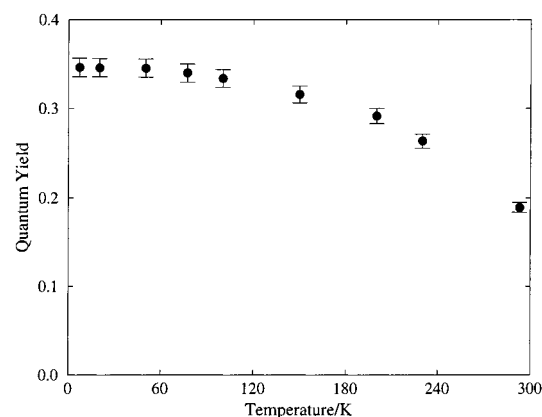


Figure 3. Fluorescence quantum yields in LHCII derived from the emission spectra of Figure 2. Note that the quantum yield determined with glycerol is 0.19 (1.15 times lower than in the case without glycerol).

scale. It should be noted that eq 1 is only used to compare the radiative rates of LHCII and Chl *a* and, since their spectra are rather similar, the approximation made for ν^3 is sufficiently accurate for our present purposes.

Looking at Table 1, we can conclude that the radiative rate at RT for LHCII is 1.1 times the radiative rate of free Chl *a* but the calculated emitting dipole strength is 1.18 times higher when corrected for the difference in the refractive indexes. If instead of the refractive index of the solvent that of the protein (values close to 1.55 have been estimated by several groups^{44–46}) is taken, a value of 0.85 is obtained. This value can be considered as an absolute lower limit, but most likely the value should be much closer to 1.18.

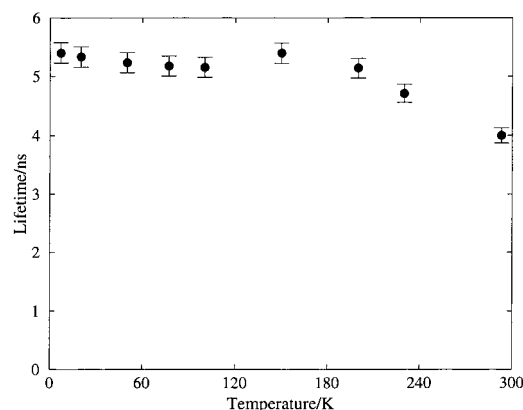
Temperature Effect. The effect of the temperature on the emission spectrum of LHCII is given in Figure 2. The emission spectrum at 7 K has a fwhm of 4.7 nm and peaks at 680.4 ± 0.1 nm, close to the values at 4.2 K reported by Pieper et al.¹⁵ (680.3 ± 0.2 nm) and Peterman et al.¹⁸ (680.7 nm). Pieper et al. argued that most of the fluorescence at this temperature arises from the lowest state in LHCII located at 679.8 nm. Upon raising the temperature, we observed that the maximum shifts first to the blue (679.4 nm at 100 K). Above 100 K, the maximum gradually shifts to the red. This peculiar behavior was previously observed,^{10,11,18} and it is possibly related to an apparent shift of some of the absorption subbands and a (small) conformational change.

The quantum yields obtained from the emission spectra as a function of temperature are plotted in Figure 3 and listed in Table 2. The results were reproducible within the indicated error

TABLE 2. Temperature Dependence of Fluorescence Quantum Yields and Lifetimes of LHCII Together with the Radiative Rates and Relative Dipole Strengths Corrected for the Difference in Refractive Indexes

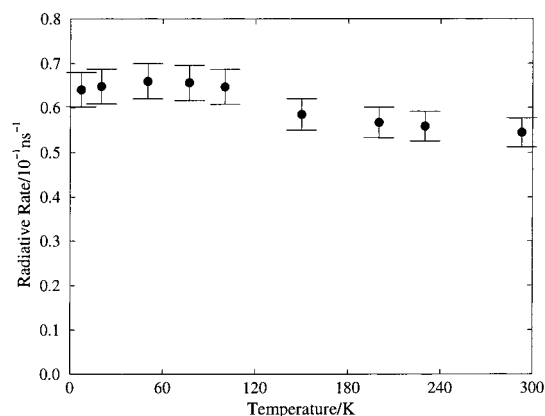
<i>T</i> (K)	quantum yield (±3%)	lifetime (ns) (±3%)	rad rate ^a (ns ⁻¹) (±6%)	refractive index ^b <i>n</i>	emit dip ^c strength (D ²) (±6%)
230	0.26	4.71	0.056	1.45	1.01
200	0.29	5.14	0.057	1.46	1.01
150	0.32	5.40	0.059	1.47	1.03
100	0.33	5.16	0.065	1.47	1.12
77	0.34	5.18	0.066	1.47	1.16
50	0.35	5.24	0.066	1.47	1.17
20	0.35	5.34	0.065	1.47	1.15
7	0.35	5.41	0.064	1.47	1.14

^a Radiative rates were calculated according to $k_{\text{rad}} = \phi_{\text{f}}/\tau_{\text{f}}$. ^b The refractive index was estimated from the refractive index of a glycerol–water mixture (1.43) at RT, taking into account the decrease in volume of the sample upon lowering the temperature. ^c Emitting dipole strengths were calculated using eq 1, and their values were divided by the dipole strength of the individual Chl *a* molecules. They are an average of the calculated ones for LHCII when compared to Chl *a* in acetone and methanol, which are each less than 4% different from the ones shown. They were also corrected for the difference in refractive indexes.

**Figure 4.** Fluorescence lifetimes of LHCII resulting from a global analysis fit of the fluorescence decay. The excitation wavelength was 400 nm.

margins. The values of the quantum yield were corrected for the shrinking of the sample upon decreasing the temperature which leads to an increase of the concentration and therefore to a concomitant increase of the number of LHCII complexes that can be “seen” by the detection system. A decrease in volume of ~12% at temperatures below 150 K estimated from the increase in the integrated area under the absorption spectrum of LHCII leads to an increase of 8% of the detected fluorescence solely because of this effect (note that the number of observed molecules along the viewing direction does not change, and therefore only the effect of shrinking in two directions has to be considered). The temperature dependence of the fluorescence quantum yield in LHCII is moderate and similar to the behavior observed for the CP47 complex.⁴⁷ At 7 K, the quantum yield is 1.8 times the value at RT. The shape of the curve differs from the one reported by Ruban et al.⁴⁸ The relative quantum yields in the latter study were slightly lower and probably some aggregates instead of trimers were present, as indicated by a pronounced shoulder near 700 nm, which correlates with quenching of the fluorescence.^{9,40,49–51}

In Figure 4, the fluorescence lifetimes are plotted as obtained from a global analysis fit of the decay of the fluorescence in LHCII at the temperatures studied. The temperature dependence of the lifetime in LHCII shows also two different regimes in

**Figure 5.** Radiative rates in LHCII calculated according to $k_{\text{rad}} = \phi_{\text{f}}/\tau_{\text{f}}$.

the studied temperature range. A similar behavior was previously reported.^{40,41}

From the fluorescence quantum yields and lifetimes, the radiative rates were calculated. They are plotted in Figure 5 and listed in Table 2 with the dipole strengths obtained according to eq 1. At every temperature, the refractive index *n* was estimated from the refractive index of the glycerol–water mixture at room temperature (1.43)⁵² according to the relation⁵³

$$\epsilon_r - 1 \propto \frac{1}{V}$$

where *V* is the volume of the sample. Its temperature dependence due to shrinking was again calculated from the increase in the integrated area under the absorption spectrum. The calculated radiative rate decreases 18% when raising the temperature from 7 K to RT.

Discussion

We have presented values of the emitting dipole strength of trimeric LHCII in the temperature range from 7 K up to RT. To calculate these, we used the slowest fluorescence decay time, which varies between 4 ns (RT) and 5.3 ns (7 K). A fast component of ~300 ps had an amplitude of only 1% or less in these samples and is probably due to a tiny amount of aggregation. The contribution of this component to the fluorescence yield can easily be calculated to be less than 0.1%, and it can safely be neglected.

The emitting dipole strength at RT is 1.18 times as large as that of free Chl *a* after correction for the difference in refractive indexes. This factor is found to be 1.14 at 7 K. Thus, in LHCII there is hardly any temperature dependence for the emitting dipole strength, and the value obtained for the lowest-energy emitting states(s) is close to the one expected from a monomeric-like type behavior, that is, 1. The fact that it is slightly higher can be because of close interactions between the Chl *a* and Chl *b* molecules.^{3,54} It is also useful to give an estimate of the absolute value for the dipole strength in LHCII and compare it to the absolute value for Chl *a*. To do so, we take a ratio of 0.8 (R. S. Knox, personal communication) for the 0–0 area of the emission from *S*₁ to *S*₀ to the area of the entire emission band and calculate a dipole strength of 34.6 D² (*n* = 1.33) for LHCII at RT. For Chl *a* in acetone, we calculate 29.1 D² (*n* = 1.36), close to the value 28.8 D² that is obtained from eq 2 using *n* = 1.36.

The value of the emitting dipole strength per se does not provide information about the amount of delocalization of the

excitations, although in the presence of strong delocalization the dipole strength may differ substantially from that of free Chl *a*, depending on how the summed dipole strength of the individual pigments is distributed over the exciton transitions. For instance, Voigt et al.¹⁶ performed exciton calculations for LHCII monomers and concluded that the lowest-energy state is located at 679.6 nm and carries a dipole strength of 1.16 Chl molecules, in agreement with the values obtained above. However, in their simulation the exciton transition just above the lowest-energy one is twice as strong. Therefore, from this one might expect an increase of the emitting dipole strength upon raising the temperature because this next-highest exciton state would also become significantly populated. However, the observed temperature dependence for the dipole strength is very weak, indicating that all states that contribute to the emission and consequently to the main absorption band around 676 nm have similar dipole strengths which are close to that of free Chl *a*. Peterman et al. arrived at the same conclusion.¹⁸ It should be noted that Voigt et al. used the Chl identities as proposed by Kühlbrandt et al. Similar calculations were performed by van Amerongen and van Grondelle⁴ where the latest information on the pigment identities was used. The calculated dipole strength of the total Chl *a* absorption band was 8% higher when compared to that of free Chl *a* because of interactions between the Chl *a* and Chl *b* molecules. Therefore, the predicted emitting dipole strength from this study for the lowest state is 1.08 as high as that for free Chl *a*, close to the observed values. According to van Amerongen and van Grondelle,⁴ an exciton interaction leads to some redistribution of dipole strength over the different transitions but, because of inhomogeneous and homogeneous broadening and differences in the site energies, this effect is relatively small; that is, there is no strong delocalization.

Another theoretical study was performed by Renger and May¹⁷ who calculated a dipole strength for the lowest-excited state in LHCII which is only 50% of that of free Chl *a*, implying significant delocalization; the present study is in disagreement with those theoretical results. The Stark measurements of Krawczyk et al.¹⁴ indicated that the Chl *a* absorption bands behave like those of monomeric Chl *a*, which is more in line with the current results. Pieper et al.¹⁵ concluded from the hole-burning measurements at 4.2 K that the three lowest-energy states in LHCII all have a dipole strength close to 1 and are only weakly coupled to other Chl molecules.¹⁵ Therefore, this work and previous reported experimental studies show that the transition to the lowest state has a dipole strength close to that of monomeric Chl *a* and that there are no indications for strong delocalization; exciton interactions are present, but they are relatively small compared to the homogeneous and inhomogeneous broadening. Moreover, we have found very little temperature dependence for the dipole strength of the states that contribute to the spontaneous emission. Therefore, it seems justified to view the energy transfer in LHCII as a process where the excitation hops from one pigment to another, which can be described with the localized Förster mechanism, as was done by Visser et al., Gradinaru et al., and Trinkunas et al.,^{44,45,55,56} and not by a coherent mechanism.⁵⁷ This contrasts with the energy transfer in the LH2 and LH1 antenna complexes of purple bacteria, where the determined radiative decay rates indicate significantly delocalized excitations.²⁵

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References and Notes

- (1) van Grondelle, R.; Dekker, J. P.; Gillbro, T.; Sundström, V. *Biochim. Biophys. Acta* **1994**, *1187*, 1.
- (2) Kühlbrandt, W.; Wang, D. N. *Nature* **1991**, *350*, 130.
- (3) Kühlbrandt, W.; Wang, D. N.; Fujiyoshi, Y. *Nature* **1994**, *367*, 614.
- (4) van Amerongen, H.; van Grondelle, R. *J. Phys. Chem. B* **2001**, *105*, 599.
- (5) Jansson, S. *Biochim. Biophys. Acta* **1994**, *1184*, 1.
- (6) Yang, C.; Kosemund, K.; Cornet, C.; Paulsen, H. *Biochemistry* **1999**, *38*, 16205.
- (7) Rogl, H.; Kühlbrandt, W. *Biochemistry* **1999**, *38*, 16214.
- (8) Remelli, R.; Varotto, C.; Sandonà, D.; Croce, R.; Bassi, R. *J. Biol. Chem.* **1999**, *274*, 33510.
- (9) Ide, J. P.; Klug, D. R.; Kühlbrandt, W.; Giorgi, L. B.; Porter, G. *Biochim. Biophys. Acta* **1987**, *893*, 349.
- (10) Hemelrijk, P. W.; Kwa, S. L. S.; van Grondelle, R.; Dekker, J. P. *Biochim. Biophys. Acta* **1992**, *1098*, 159.
- (11) Kwa, S. L. S.; Groeneveld, F. G.; Dekker, J. P.; van Grondelle, R.; van Amerongen, H.; Lin, S.; Struve, W. S. *Biochim. Biophys. Acta* **1992**, *1101*, 143.
- (12) Nussberger, S.; Dekker, J. P.; Kühlbrandt, W.; van Bolhuis, B. M.; van Grondelle, R.; van Amerongen, H. *Biochemistry* **1994**, *33*, 14775.
- (13) Reddy, N. R. S.; van Amerongen, H.; Kwa, S. L. S.; van Grondelle, R.; Small, G. J. *J. Phys. Chem.* **1994**, *98*, 4729.
- (14) Krawczyk, S.; Krupa, Z.; Maksymiec, W. *Biochim. Biophys. Acta* **1993**, *1143*, 273.
- (15) Pieper, J.; Rätsep, M.; Jankowiak, R.; Irrgang, K.-D.; Voigt, J.; Renger, G.; Small, G. J. *J. Phys. Chem. A* **1999**, *103*, 2412.
- (16) Voigt, J.; Renger, Th.; Schödel, R.; Schrötter, Th.; Pieper, J.; Redlin, H. *Phys. Status Solidi B* **1996**, *194*, 333.
- (17) Renger, Th.; May, V. *Phys. Rev. Lett.* **2000**, *84*, 5228.
- (18) Peterman, E. J. G.; Pullerits, T.; van Grondelle, R.; van Amerongen, H. *J. Phys. Chem. B* **1997**, *101*, 4448.
- (19) Spano, F. C.; Mukamel, S. *J. Chem. Phys.* **1989**, *91*, 683.
- (20) Meier, T.; Zhao, Y.; Chernyak, V.; Mukamel, S. *J. Chem. Phys.* **1997**, *107*, 3876.
- (21) Zhao, Y.; Meier, T.; Zhang, W. M.; Chernyak, V.; Mukamel, S. *J. Phys. Chem. B* **1999**, *103*, 3954.
- (22) Spano, F. C.; Kuklinski, J. R.; Mukamel, S. *Phys. Rev. Lett.* **1990**, *65*, 211.
- (23) Spano, F. C.; Kuklinski, J. R.; Mukamel, S. *J. Chem. Phys.* **1991**, *94*, 7534.
- (24) Potma, E. O.; Wiersma, D. A. *J. Chem. Phys.* **1998**, *108*, 4894.
- (25) Monshouwer, R.; Abrahamsen, M.; van Mourik, F.; van Grondelle, R. *J. Phys. Chem.* **1997**, *101*, 7241.
- (26) Fidler, H.; Knoester, J.; Wiersma, D. A. *J. Chem. Phys.* **1991**, *95*, 7880.
- (27) Leegwater, J. A. *J. Phys. Chem.* **1996**, *100*, 14403.
- (28) Jimenez, R.; Dikshit, S. N.; Bradforth, S. E.; Fleming, G. R. *J. Phys. Chem.* **1996**, *100*, 6825.
- (29) Zucchelli, G.; Garlaschi, F. M.; Jennings, R. C. *Biochemistry* **1996**, *35*, 16247.
- (30) Hübner, M.; Kuhl, J.; Stroucken, T.; Knorr, A.; Koch, S. W.; Hey, R.; Ploog, K. *Phys. Rev. Lett.* **1996**, *76*, 4199.
- (31) Oberli, D. Y.; Vouilloz, F.; Kapon, E. *Phys. Status Solidi A* **1997**, *164*, 353.
- (32) Fidler, H.; Knoester, J.; Wiersma, D. A. *Chem. Phys. Lett.* **1990**, *171*, 529.
- (33) De Boer, S.; Wiersma, D. A. *Chem. Phys. Lett.* **1990**, *165*, 45.
- (34) Fidler, H.; Wiersma, D. A. *Phys. Status Solidi B* **1995**, *188*, 285.
- (35) Monshouwer, R.; van Grondelle, R. *Biochim. Biophys. Acta* **1996**, *1275*, 70.
- (36) Peterman, E. J. G.; Dukker, F. M.; van Grondelle, R.; van Amerongen, H. *Biophys. J.* **1995**, *69*, 2670.
- (37) Weber, G.; Teale, F. W. J. *Trans. Faraday Soc.* **1957**, *53*, 646.
- (38) Vladkova, R. *Photochem. Photobiol.* **2000**, *71*, 71.
- (39) Barzda, V.; Gulbinas, V.; Kananavicius, R.; Cervinskis, V.; van Amerongen, H.; van Grondelle, R.; Valkunas, L. *Biophys. J.* **2001**, *80*, 2409.
- (40) Vasil'ev, S.; Irrgang, K.-D.; Schrötter, T.; Bergmann, A.; Eichler, H.-J.; Renger, G. *Biochemistry* **1997**, *36*, 7503.
- (41) Seydack, M.; Redlin, H.; Voigt, J. In *Photosynthesis: From light to Biosphere*; Mathis, P., Ed.; Kluwer Academic Publishers: Dordrecht, The Netherlands, 1995; Vol. 1, p 283.
- (42) van Amerongen, H.; Valkunas, L.; van Grondelle, R. *Photosynthetic Excitons*, 1st ed.; World Scientific: Singapore, 2000; Chapter 2.
- (43) Knox, R. S. PS2001 Proceedings. 12th International Congress on Photosynthesis; CSIRO Publishing: Melbourne, Australia, 2001.
- (44) Gradinaru, C. C.; Özdemir, S.; Gülen, D.; van Stokkum, I. H. M.; van Grondelle, R.; van Amerongen, H. *Biophys. J.* **1998**, *75*, 3064.

- (45) Trinkunas, G.; Connelly, J. P.; Müller, M. G.; Valkunas, L.; Holzwarth, A. R. *J. Phys. Chem. B* **1997**, *101*, 7313.
- (46) Gruszecki, W.; Grudzinski, W.; Banaszek-Glos, A.; Matula, M.; Kernen, P.; Krupa, Z.; Sielewiesiuk, J. *Biochim. Biophys. Acta* **1999**, *1412*, 173.
- (47) Groot, M. L.; Peterman, E. J. G.; van Stokkum, I. H. M.; Dekker, J. P.; van Grondelle, R. *Biophys. J.* **1995**, *68*, 281.
- (48) Ruban, A. V.; Dekker, J. P.; Horton, P.; van Grondelle, R. *Photochem. Photobiol.* **1995**, *61*, 216.
- (49) Barzda, V.; Vengris, M.; Valkunas, L.; van Grondelle, R.; van Amerongen, H. *Biochemistry* **2000**, *39*, 10468.
- (50) Barzda, V.; Jennings, R. C.; Zucchelli, G.; Garab, G. *Photochem. Photobiol.* **1999**, *70*, 751.
- (51) Razi Naqvi, K.; Melø, T. B.; Raju, B. B.; Javorfi, T.; Simidjiev, I.; Garab, G. *Spectrochim. Acta, Part A* **1997**, *53*, 2659.
- (52) *Handbook of Chemistry and Physics*, 77th ed.; Lide, D. R., Ed.; CRC Press: 1997; Chapter 8.
- (53) Jackson, J. D. *Classical Electrodynamics*, 2nd ed.; John Wiley & Sons: 1975; Chapter 7.
- (54) Valkunas, L.; Cervinskas, V.; Trinkunas, G.; Muller, M. G.; Holzwarth, A. R. *J. Chem. Phys.* **1999**, *111*, 3121.
- (55) Visser, H. M.; Kleima, F. J.; van Stokkum, I. H. M.; van Grondelle, R.; van Amerongen, H. *Chem. Phys.* **1996**, *210*, 297.
- (56) Visser, H. M.; Kleima, F. J.; van Stokkum, I. H. M.; van Grondelle, R.; van Amerongen, H. *Chem. Phys.* **1997**, *215*, 299.
- (57) Renger, Th.; May, V. *J. Phys. Chem. B* **1997**, *101*, 7232.