Single-Molecule Studies

Probing the Influence of O_2 on Photoinduced Reversible Electron Transfer in Perylenediimide–Triphenylamine-Based Dendrimers by Single-Molecule Spectroscopy**

Mircea Cotlet, Sadahiro Masuo, Marc Lor, Eduard Fron, Mark Van der Auweraer, Klaus Müllen, Johan Hofkens, and Frans De Schryver**

Room temperature single-molecule fluorescence detection has evolved as a powerful tool to monitor dynamic processes in individual molecules.[1] Probing single molecules through their spectroscopic properties removes the inherent averaging factor that is present in ensemble experiments to render single-molecule detection as the method of choice to yield information at the most detailed level. Phenomena which are not necessarily predictable by ensemble experiments, such as fluctuations in the fluorescence intensity, lifetimes, or emission maxima, can be obtained by single-molecule detection. Here we use single-molecule detection to probe the influence of molecular oxygen (O_2) on the dynamics of photoinduced electron transfer (PET) in a dendrimer. PET is usually an efficient mechanism for fluorescence quenching, which renders single-molecule detection difficult, if not impossible. Therefore reports on PET in single molecules are rather limited.[2,3a,b]

A particular aspect of the electron-transfer (ET) process occurs in the case where the locally excited singlet (LES) state and the charge-separated state (CSS) lie close in energetic terms to each other (Figure 1a, with the CSS at level 2). Normally, upon excitation, the LES is deactivated mainly through forward electron transfer (FET) to the CSS. If the radiationless deactivation of the CSS to the ground state (GS) is slow and inefficient, the CSS decays mainly through the LES by a reverse electron-transfer (RET) process. If so, fluorescence is delayed but it is still characterized by a high quantum yield. This allows the electron-transfer process to be

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[*] M. Cotlet, S. Masuo, M. Lor, E. Fron, M. Van der Auweraer, J. Hofkens, F. De Schryver
Laboratory of Photochemistry and Spectroscopy
Katholieke Universiteit Leuven
3001 Heverlee (Belgium)
Fax: (+32) 16-327-990
E-mail: johan.hofkens@chem.kuleuven.ac.be
frans.deschryver@chem.kuleuven.ac.be

K. Müllen
Max Planck Institut für Polymerforschung
Ackermannweg 10
55128 Mainz (Germany)

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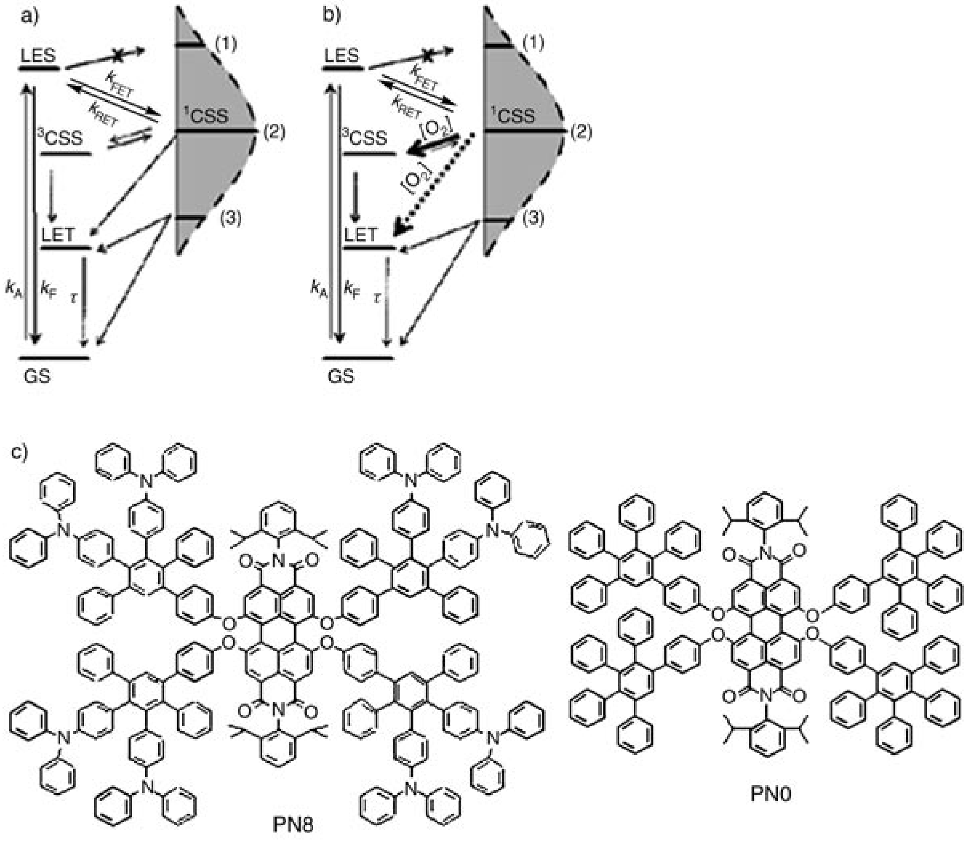

Figure 1. Scheme accounting for photoinduced reversible electron transfer in PN8 under a) deaerated (N_2; $\tau \approx 40$ μs) and b) aerated (O_2; $\tau < 1$ μs) atmospheric conditions; thick arrows indicate the pathways enhanced by the presence of O_2; τ represents the decay time; LES = locally excited singlet state; LET = locally excited triplet state; GS = ground state; ^{1}CSS = singlet charge-separation states; ^{3}CSS = triplet charge-separation states; k_A, k_F, k_{FET}, k_{RET} are the rate constants for absorption, fluorescence, forward-, and reverse-ET, respectively. ^{1}CSS is drawn at different levels within a Gauss-type density of states induced by polymer chain motion (see text); for simplicity, the ^{3}CSS in both schemes is shown together with ^{1}CSS at level 2. c) Structures of PN8 and PN0.

probed at the single-molecule level if the excited chromophore shows both a high quantum yield of fluorescence and long-term photostability. We have shown previously that single molecules of a perylenediimide–triphenylamine (PDI–TPA)-based dendrimer, which are embedded in polystyrene, undergo photoinduced reversible electron transfer upon excitation at $\lambda = 543$ nm.[3a,b] Herein we focus on a first generation rigid polyphenylene dendrimer, PN8, which contains a PDI core as an electron acceptor and eight TPA electron donor moities at the rim (Figure 1 c)—a system we have shown previously to be capable of undergoing photoinduced reversible electron transfer.[3b]

In an apolar solvent (methylcyclohexane), PN8 shows a quantum yield of fluorescence that approaches unity, and the fluorescence ($\lambda_{exc} = 543$ nm, $\lambda_{em} = 600$ nm) decays monoexponentially with a lifetime of 6.5 ns (see Supporting Information): a value that is characteristic for unquenched PDI. In this case, the CSS lies at a higher energy than the LES and no ET occurs (Figure 1 a, with the CSS at level 1). In aerated toluene, the fluorescence quantum yield of PN8 drops to 0.14 and the fluorescence decay becomes multiexponential with decay times of 0.07 (70 % amplitude), 1.2 (12 %), 6.5 (11 %), and 14 ns (7 %). Upon deaeration of the solution of PN8 in toluene, the fluorescence quantum yield increases to 0.38. The fluorescence decay of the deaerated solution of PN8 is also multiexponential, with decay times of 0.07 (62 % amplitude), 1.2 (10 %), 6.5 (9 %), and 22.2 ns (19 %) (see Supporting Information). For PN8 as well as for similar electron-donor–acceptor compounds, short decay times—that is, relative to the lifetime of unquenched PDI (6.5 ns)—reflect FET from the LES to the CSS, whereas longer decay times are associated with RET from the CSS to the LES.[3a–c] For such compounds, delayed fluorescence (long decay times) is a characteristic of RET (Figure 1 a, with the CSS at level 2). Upon deaeration of the solution of PN8 in toluene, the long decay time related to delayed fluorescence increases both in value and in contribution (see Supporting Information) as does the fluorescence quantum yield. This suggests that O_2 influences the deactivation of the CSS and therefore plays a role in the dynamics of RET in PN8. Nanosecond-transient absorption data of the deaerated and aerated solutions of PN8 in toluene strongly support the previous assumption (see Supporting Information).

For PN8, because of a rather large edge-to-edge donor–acceptor distance,[3b] the singlet (^{1}CSS) and the triplet (^{3}CSS) charge-separated states lie close in energy, which allows intersystem crossing (ISC) to occur through hyperfine coupling interactions.[4,5] Moving from the unsubstituted PDI to PN8, the quantum yield of fluorescence in toluene drops from 1 to 0.38, which suggests that charge recombination in PN8 does not take place only through the LES state. From ^{1}CSS, charge recombination can also occur through ISC to the locally excited triplet (LET) state, either directly or, more likely, via ^{3}CSS (Figure 1 a).[3c,4] Nanosecond-transient absorption data (see Supporting Information) strongly suggest that the triplet and the charge separation states are interconnected in PN8. O_2 enhances ISC from the ^{1}CSS to the ^{3}CSS and LET states (Figure 1 b, pathways indicated by thick arrows) which leads to enhanced charge recombination and a decrease in the fluorescence quantum yield of PN8.[4,5]

According to time-correlated single-photon counting (TCSPC) data from the entire ensemble of PN8 molecules in solution in deaerated toluene, 9% of the molecules do not undergo ET processes—they have a fluorescence lifetime that is characteristic of that of unquenched PDI. By single-molecule detection, we can selectively identify molecules that do undergo RET because this phenomenon relates to the observation of delayed fluorescence.

Observation of single molecules on a longer timescale requires their immobilization. Here we immobilized single molecules of PN8 in polystyrene (PS) as this polymer is comparable to toluene in terms of polarity and polarizability.[3a,b] Figure 2 shows a fluorescence lifetime image of single PN8 molecules in PS ($\lambda_{exc} = 543$ nm, 20×20 µm^2, see Supporting Information). In the image, single PN8 molecules that undergo RET can be easily identified by their decay times, which are longer than the lifetime of unquenched PDI ($\tau \approx$ 6 ns). This allows the dynamics of RET in PN8 as well as the role of O_2 on the radiationless deactivation of the CSS to be probed at the single-molecule level.

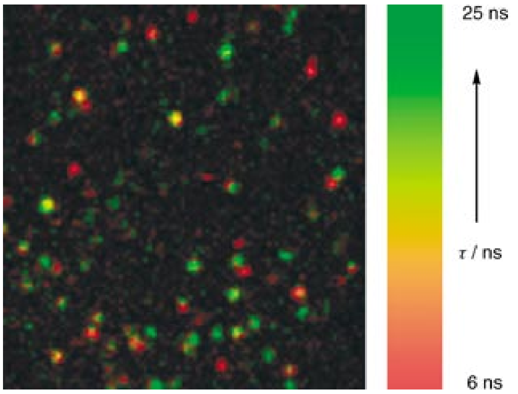

Figure 2. Fluorescence lifetime image ($\lambda_{exc} = 543$ nm; 20×20 µm^2, 100×100 pixels^2, 10 ms pixel^{-1}) of a polystyrene (PS) film that contains single molecules of PN8.

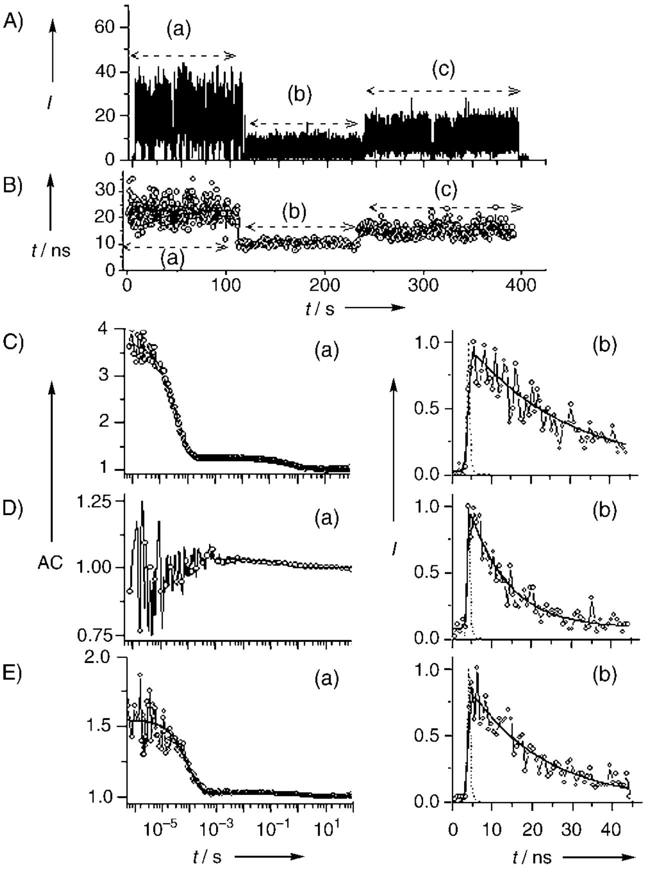

Figure 3. A) Fluorescence intensity (*I* represents the number of counts per 10 ms) and B) decay time (τ) trajectories recorded from a single molecule of PN8 in PS by alternating the atmosphere from (a) N_2 to (b) air and (c) back to N_2. C), D), and E) show the autocorrelations (AC) of the fluorescence intensity (parts (a), ○) and the fluorescence decays (I_F (a.u.), parts (b), ○, 1000 photons) that correspond to part A) (a), (b), and (c), respectively. Exponential fits (——) are also shown.

Figure 3, A and B, show the fluorescence intensity and decay time trajectories, respectively, of a single molecule of PN8 (immobilized in PS) undergoing reversible photoinduced electron transfer. The data were recorded by alternating a stream of either nitrogen (N_2) or air over the sample of the single molecule. Initially, under N_2 atmosphere (Figure 3 A and B, a), an average photon count rate of 30 counts per 10 ms and a decay time of 22 ns were detected. After switching to an air atmosphere (Figure 3 A and B, b), decreases in both the count rate, to 10 counts per 10 ms, and the decay time, to 10 ns, were observed Switching back to a N_2 atmosphere (Figure 3 A and B, c) resulted in a partial increase in both the count rate, to 20 counts per 10 ms, and the decay time, to 16 ns.[6] Typical single-molecule fluorescence decay profiles (1000 photons) that correspond to the regions a, b, and c are shown in Figure 3 C–E, parts b, respectively. Data for a single PN8 molecule that undergoes RET in an alternating atmosphere prove that O_2 influences the decay of the CSS by enhancing ISC, which leads to radiationless deactivation of the CSS to the ground state. Indeed, both the fluorescence intensity (Figure 3 A) and the decay times of RET decrease (or increase) when the concentration of O_2 is increased (or decreased).

For the single molecule of PN8 under an alternating atmosphere, on/off dynamics can be seen in the fluorescence intensity trajectory, especially under a N_2 atmosphere (Figure 4a). To capture the timescale of such dynamics, we computed the autocorrelation (AC) of the fluorescence intensity $AC = \langle I(t)I(t+\Delta t)\rangle / \langle I \rangle^2$ with the intensities $I(t)$ and $I(t+\Delta t)$ at times t and $t+\Delta t$, respectively (see Supporting Information). Figure 3 C–E, parts a, show the autocorrelation of the fluorescence intensity (AC) for the regions a, b, and c, respectively. For region a, the AC decays biexponentially with offtimes of 0.4 ms (90% weight) and 700 ms (10%). When O_2 is present (b), the AC shows only a slow and low contributing component of 900 ms. Restoring the N_2 atmosphere (c), we again detect fast on/off dynamics with an offtime of 0.35 ms (90% weight), together with slow on/off dynamics with an offtime of 1000 ms (10%).

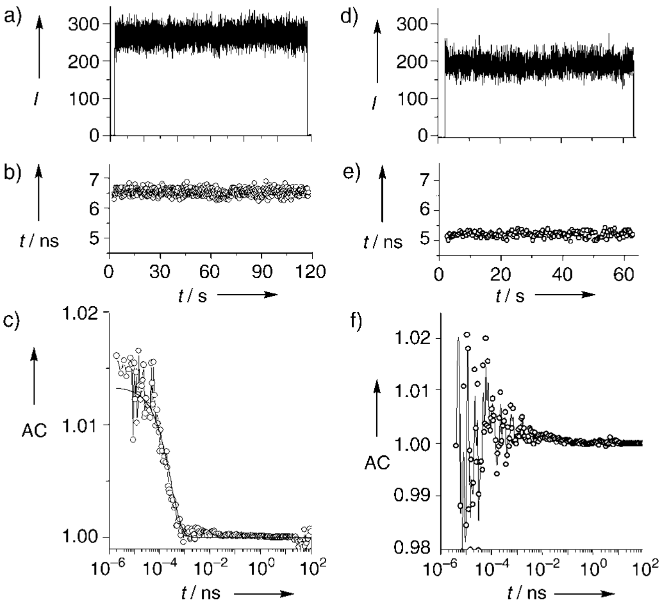

Figure 4. a) Fluorescence intensity (I = counts per 10 ms) and b) lifetime (1000 photons per decay) trajectories from a single molecule of PN0 in polystyrene under a N_2 atmosphere; c) the autocorrelation (AC) of the single molecule which corresponds to parts a–b); d) fluorescence intensity and e) lifetime (1000 photons per decay) trajectories from a single molecule of unsubstituted PDI in polystyrene under an O_2 atmosphere; f) the AC of the single molecule that corresponds to panels d–e.

The fast on/off dynamics that were detected in the fluorescence intensity of single PN8 molecules that undergo RET, which is only observed under a N_2 atmosphere, relates to the population of the LET state (Figure 1a). Similar fast on/off dynamics were detected in the fluorescence intensity of single molecules of a model compound, which did not have rim-substituted amine groups (PN0) and which was embedded in PS, again only under N_2 atmosphere (Figure 4a–c). Under N_2 atmosphere, a single molecule of PN0 emits fluorescence with one stable intensity level (Figure 4a, average count rate: 280 counts per 10 ms) and a lifetime of ≈6.5 ns (Figure 4b), and the AC from such a single molecule of PN0 decays monoexponentially with an offtime of 0.3 ms, which relates to the population of the triplet state (Figure 4c). In the presence of O_2, a small decrease in both the fluorescence intensity and lifetime of a single PN0 molecule was detected (Figure 4, d and e) as well as the disappearance of the fast on/off dynamics in the related AC (Figure 4f). The decay of the triplet was faster than the time resolution of the experiment (μs). However, the contribution of the off-events was significantly smaller in single molecules of PN0 (Figure 4c) relative to single molecules of PN8 (Figure 3C, a), which suggests that triplet formation in PN8 (in PS) is strongly enhanced through the formation of a radical-ion-pair that can recombine through ISC to the LET state (Figure 1a). In an aerated atmosphere, because O_2 is an efficient triplet quencher, the off-events become shorter than the experimental bin time (μs) and do not contribute to the decay of the AC (Figure 3D, a) for PN8; Figure 4f for PN0). On/off dynamics on the timescale of 0.1–1 ms with offtimes that are sensitive to the concentration of O_2 were reported previously for single dye molecules, which were embedded in polymers, and these were related to the population of the triplet.[7] The data reported here demonstrate, on a single-molecule level, that 1) O_2 influences the dichotomy between the return from the CSS to the LES or to the GS and 2) owing to the formation of a radical anion, the population of the triplet state and hence the quantum yield for ISC are strongly enhanced. From the data for single molecules under a N_2 atmosphere, we estimate quantum yields for the ISC process of 10^{-4} and 10^{-2} for PN0 and PN8, respectively.[8]

Single molecules of PN0 in PS do not show slow on/off dynamics (Figure 4, c and f). Therefore, the slow on/off process present in the fluorescence emission of single molecules of PN8 that undergo reversible photoinduced electron transfer must be related to the interaction between the PDI acceptor and the rim-substituted TPA donors—that is, to the presence of the radical anion. RET occurs in a single molecule of PN8 only if the local environment—that is, the polymer—has a polarity such that the CSS and the LES lie close in energy (Figure 1a, with the CSS at level 2). We have shown previously in a single-molecule study on RET that motion of the polystyrene chain can induce fluctuations on the second timescale in the RET-related decay times of single molecules of PN8.[3b] The polymer can therefore modulate the local polarity of the rim-substituted donors through its chain motion to influence the feasibility of charge separation. This can be regarded as a density of states in which the energy of the CSS of the single molecule fluctuates with the motion of the polymer (Figure 1a, with the density of states for the CSS drawn as a Gauss envelope). If the CSS becomes more stabilized than the LES owing to polymer chain motion (Figure 1a, with the CSS at level 3), charge recombination from the CSS will take place only to the ground state (GS) either directly or via the LET state; that is, only through a nonradiative pathway. If so, the fluorescence of a single molecule of PN8 will be interrupted as long as the CSS lies at lower energy than the LES, with a time interval in which the molecule can undergo many optical excitation cycles. If the polymer chain reorients such that the CSS and the LES state become close again in energy, the pathway for charge recombination through the LES state becomes available again and fluorescence is restored. Polymer chain motion will impose, with its own timescale,[9] the length of the dark periods in the fluorescence emission of a single molecule of PN8 that undergoes reversible photoinduced electron transfer. Indeed, the timescale of the slow dynamics is independent of the presence/absence of O_2, within experimental error (Figure 3C–E, a), as it is dictated by the polymer motion. Only the frequency of such off-events will be influenced by O_2 whose presence enhances ISC from the ^{1}CSS to the ^{3}CSS and LET states. Slow on/off dynamics were observed in the fluorescence intensity of single perylenediimide dimers, and this observation was suggested to be connected to the population of a nonfluorescent ion-pair state.[2g]

In summary, we have shown that the deactivation of the CSS either to the LES state or to the GS for a single molecule can be influenced in a reversible fashion[6] by the modification of the local O_2 concentration. Furthermore, we have shown that polymer chain motion can induce "off" periods in the emission of a single molecule that undergoes ET; the length of such periods reflects the timescale of the polymer motion. This suggests that electron donor–acceptor systems such as PN8 can be used as single-molecule probes for polymer dynamics. To our knowledge, this is the first report, which involves single molecules, that demonstrates the influence of molecular oxygen on the radiationless deactivation of the CSS to the GS for an electron donor–acceptor system as well as of the role of polymer motion on the dynamics of fluorescence of a single molecule that undergoes electron transfer.

Experimental Section

PN8 and PN0 were prepared according to reported methods.[10] Ensemble experiments were performed on aerated and degassed (freeze–pump–thaw cycles) of solutions (10^{-5} M) in toluene. Details on bulk-phase stationary and time-resolved experiments are provided in the Supporting Information. For single-molecule detection, a solution of either PN8 or PN0 (10^{-10} M) in chloroform was mixed with the polymer (PS), and the mixture was spin-coated onto a cover glass. Single-molecule experiments were performed upon 543-nm pulsed (laser) excitation on a scanning stage confocal microscope.[11] Experimental details and data analysis are provided in the Supporting Information.

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