Photochemistry of the amine-sensor dye 4-*N*,*N*-dioctylamino-4′-trifluoroacetylazobenzene

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Abstract

The chromoreactand 4-*N*,*N*-dioctylamino-4′-trifluoroacetylazobenzene (ETH^T 4001) has been developed for use in optical sensors to detect aliphatic and aromatic amines. In the present work, the photochemical behaviour of ETH^T 4001 and its effects on the sensor characteristics have been investigated. The azo dye exhibits a photochemical reactivity typically observed for donor–acceptor azo dyes with fully reversible photoisomerism. The *Z*-form exhibits a decrease in extinction and a shift to shorter wavelengths which is caused by the reduced planarity of the chromophore. The quantum yield of the *E*–*Z* isomerization was determined to be 0.16 in toluene at $-60\,^{\circ}$C. At room temperature, however, and if exposed to an irradiance as encountered in a spectrophotometer, the stationary amount of the *Z*-form is negligibly small. Flash photolysis experiments of ETH^T 4001 in toluene result in photochemically induced *E*–*Z* isomerization and show that the thermal isomerization is faster for ETH^T 4001 (lifetime 5.3 s at room temperature) than for unsubstituted azobenzene. While the kinetics of ETH^T 4001 dissolved in plasticised PVC were no longer single exponential, the lifetime of the *Z*-form was not enhanced in comparison to the toluene solution. The kinetics of the *Z*–*E* isomerization of ETH^T 4001 in toluene is significantly dependent on the presence of organic acids which is attributed to acid-assisted tautomerization of the *Z*-ammonium ion into a *Z*-azonium ion.
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1. Introduction

We have recently introduced a novel class of indicator dyes (chromogenic and fluorogenic reactands) that give colour changes based on their reversible chemical reaction with the analyte rather than by complexation or protonation/deprotonation. Chromo- and fluororeactands have been presented that allow the detection of alcohols [1], amines [2,3], aldehydes [4] and humidity [5]. The azo dye presented here, namely 4-*N*,*N*-dioctylamino-4′-trifluoroacetylazobenzene, is an amine sensitive dye because its trifluoroacetyl group reversibly interacts with amines to form a hemiaminal. Due to the weaker acceptor capacity of this hemiaminal group, a colour change from red to yellow is observed. The dye has previously been embedded in various polymer materials and the resulting optical sensors have been used to detect amines in aqueous solution [2,3]. The present work investigates the photochemistry of the chromoreactand in solution and discusses possible effects of photochemical

and thermal isomerism on the sensor performance. While the photochemistry is of general interest to discuss the characteristics of a dye, it is also an important parameter for practical applications, e.g. in optical sensing.

2. Experimental

2.1. Chemicals

The amine-sensitive chromoreactand 4-*N*,*N*-dioctylamino-4′-trifluoroacetyl-azobenzene was obtained from Dyomics GmbH, Jena, Germany (http://www.dyomics.com). All solvents were of analytical reagent grade.

Plasticised PVC membranes were prepared by dissolving 1.0 mg of ETH^T 4001 together with 40 mg of bis(2-ethylhexyl)sebacate and 80 mg of poly(vinyl chloride) in 0.8 ml of tetrahydrofuran and spin-coating 0.3 ml of the solution on rotating polyester foils with a thickness of 100 μm (serving as a mechanical support) [2]. The resulting sensor layers with a thickness of 3–5 μm were allowed to dry for 24 h at room temperature.

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2.2. Instruments and computation

All stationary photokinetic measurements were performed with a M400 spectrophotometer, a photokinetic accessory (Carl Zeiss Jena, Germany) and a thermostat. Fischer's method [6] was applied using a modified approach to calculate the spectrum of the Z-form of the azobenzene dye in that an average conversion ratio was calculated as a weighted mean from all spectral data points recorded every 2 nm from 300 to 600 nm. The square of the conversion ratio itself obtained at the corresponding wavelength was used as the statistical weight. Photostationary states obtained upon irradiation at 546, 436 and 313 nm were used to calculate the conversion ratios. The Z-spectrum (mean value of three evaluations using three combinations of photostationary states) is less accurate in the region near the maximum of the E-absorbance (error <50%); however, in the spectral region where Z-absorbance > E-absorbance, the error is less than 8%.

The quantum yield of the E–Z isomerization was measured at $-60\,^{\circ}$C in toluene as a solvent. At this temperature no thermal reverse reaction (Z–E) is observed during the irradiation period. The irradiation intensity was determined with the help of the ferrioxalate actinometer applying the same irradiation geometry, cuvette and sample volume (V) as in reference [7]. The molar absorptivity was determined to be 39980 $M^{-1}\,cm^{-1}$ at $\lambda_{max} = 498$ nm. The concentration was corrected for volume contraction.

Conventional flash photolysis experiments of the trifluoroacetyl and the hemiaminal forms of ETH^T 4001 in toluene were carried out in a home-made cylindrical cell of 10 cm length and 10 mm diameter in a double elliptical reflector with two flash lamps [8]. When investigating the sensor layers, three layers were fixed in a hollow glass tube (of similar dimensions as the cylindrical cell) at an angle of about 45° relative to the axis of the cell. Cut-off filters were used to avoid UV excitation (filter for excitation: Schott BG7). The electrical energy of the flashes amounted to 900 J at a duration of 14 µs. Kinetic data analysis was performed by non-linear regression using the ALAU program [9] in a modified PASCAL version. The measurements were performed at 25 °C.

3. Results and discussion

3.1. Absorbance of the Z-form and efficiency of E–Z isomerization

The exocyclic double bond of azobenzene dyes can provide both photo-induced and thermal isomerization (Fig. 1). This is of importance for sensors that use such dyes as optical transducers of analyte recognition because it may compromise the signal stability of the sensor. Two different mechanisms of isomerization have been discussed for azobenzene dyes, namely E–Z isomerization via rotation
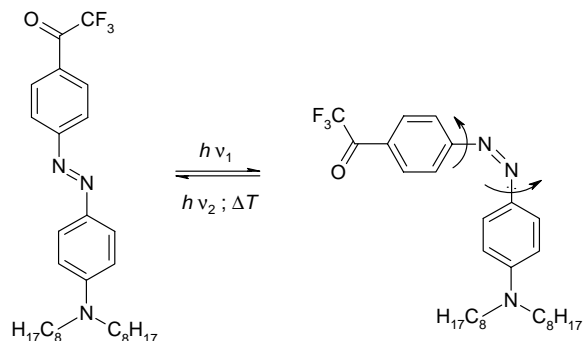

Fig. 1. E–Z isomerization of ETH^T 4001.

about the –N=N– bond or via inversion of one of the nitrogen atoms (by means of an sp-hybridized transition state) [10]. ETH^T 4001 exhibits reversible photoisomerization (Fig. 2). The Z-form exhibits a significant decrease in extinction and shift to shorter wavelengths which is caused by the reduced length and planarity of the chromophore.

The true differential photoisomerization quantum yield $\phi_{E \to Z}$ for E–Z isomerization of the azobenzene dye ETH^T 4001

$$\phi_{E \to Z} = \frac{d[Z]}{I_{abs}\,dt} \quad \text{with } I_{abs} = I_0(1 - 10^{-A}) \tag{1}$$

was obtained by extrapolating the integral apparent quantum yield

$$\varphi_{app}(t) = \frac{\Delta[Z](t)}{I_{abs}(t)\,\Delta t} \quad \text{with } \Delta[Z](t) = \frac{\Delta A_{498}}{\varepsilon_{498}} \tag{2}$$

to $t = 0$, where [Z] denotes the concentration of the Z-form, A the absorbance at a defined wavelength, and ε the extinction coefficient of the Z-form. The experimentally obtained integral apparent quantum yield $\phi_{app}(t)$ was found to decrease exponentially with time. The quantum yield ϕ_{E-Z} was calculated to be 0.16.

The quantum yield of the reverse photoreaction can be obtained from the Zimmermann equation for the concentration ratio at a photostationary state with the conversion ratio α obtained from Fischer's evaluation procedure [6]

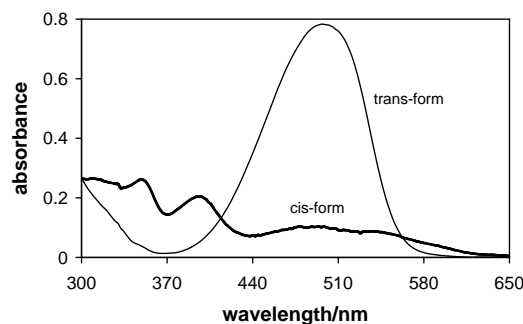$$\phi_{Z \to E} = \frac{\varepsilon_E \phi_{E \to Z}(1 - \alpha)}{\varepsilon_Z \alpha} \tag{3}$$

Fig. 2. Absorbance of the Z- and E-forms of ETH^T 4001 obtained via the Fischer evaluation [6].

Due to the uncertainty of the molar absorptivity ε_Z at 436 nm, $\phi_{Z \to E}$ is significantly less accurate and values in the range from 0.7 to 1.1 are obtained for the different photostationary states. Consequently, with respect to the quantum yield of the reversible photoisomerization, ETH^T 4001 is comparable to azobenzene ($\phi_{E \to Z, i\text{-octane}} = 0.24$ and $\phi_{Z \to E, i\text{-octane}} = 0.55$, respectively) [7].

The photostationary state of a photo- and thermally reversible system:

$$E \underset{h\nu_2; \Delta T}{\overset{h\nu_1}{\rightleftharpoons}} Z$$
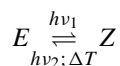
is described by Eq. (4)

$$\frac{I_0(1 - 10^{-A})}{A}(\varepsilon_E[E]\phi_{E \to Z} - \varepsilon_Z[Z]\phi_{Z \to E}) = k[Z],$$
$$\text{where } A = \varepsilon_E[E] + \varepsilon_Z[Z] \quad (4)$$

With the known extinction coefficients, ε_E, ε_Z, the quantum yields $\phi_{E \to Z}$, $\phi_{Z \to E}$, the rate constant for the thermal reverse reaction, k, and a given concentration ratio [Z]/[E] to be tolerated (set equal to 0.01) we can readily calculate an upper limit for the light intensity I_0 which does not disturb the sensing process. The numerical calculation results in a light intensity $I_0 = 3.2 \times 10^{-7} \, \text{M s}^{-1}$ which is orders of magnitude above the light intensity in the sample compartment of a spectrophotometer and even compared with the irradiance of normal ambient illumination. Hence, we can safely conclude that there is no significant disturbance of the sensing properties due to photoisomerization.

3.2. Kinetics of Z–E isomerization

Flash photolysis experiments of the trifluoroacetyl form of ETH^T 4001 in toluene result in photochemically induced E–Z isomerization. Due to the fact that extinction coefficients of Z-isomers are lower than the corresponding ones of E-isomers, instantaneous bleaching is observed. Complete recovery of the initial absorbance of the solution is observed during thermal Z–E isomerization, indicating that the entire process is fully reversible. Furthermore, the transient absorbance spectra are essentially mirror images of the corresponding ground-state absorbance spectra of the dye in solution. In detail, a reversible absorption depletion from 410 to 570 nm and a weak transient absorption above 570 nm and below 410 nm (Fig. 3) is found. The thermal reverse reaction follows strictly first order kinetics and the rate constants are independent of the recording wavelength. The lifetime of the Z-form is 5.8 s at 25 °C, which is shorter than for azobenzene (122 h) [11]. This does not come unexpected because the thermal isomerization of donor–acceptor azobenzenes (such as ETH^T 4001) involves both the inversion and the rotation processes, and proceeds more quickly than in the case of azobenzenes without strong donor or acceptor groups [12,13]. The kinetics were analysed in the temperature region from 15 to 70 °C in toluene. Fig. 4 shows an Arrhenius
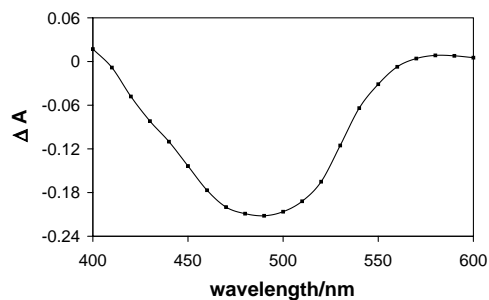

Fig. 3. Transient absorbance spectrum of ETH^T 4001 in toluene obtained via flash photolysis experiments. The lifetime of the Z-form is 5.8 s.
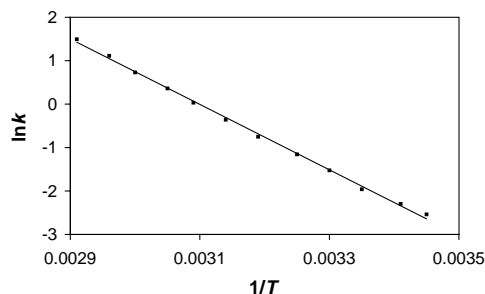

Fig. 4. Arrhenius plot for ETH^T 4001 in toluene.

plot which yields an activation energy of $61.2 \pm 0.7 \, \text{kJ mol}^{-1}$ and $\log A = 10.0 \pm 0.1$ (mean values from two independent measurement series). With the help of the Eyring relation an activation enthalpy of $58.7 \pm 0.7 \, \text{kJ mol}^{-1}$ and an activation entropy of $-62 \pm 2 \, \text{J mol}^{-1} \, \text{K}^{-1}$ is obtained. Removal of dissolved oxygen does not influence the kinetics. No photodegradation was observable after repeated flashing (20–25 flashes).

Similar results are obtained with sensor layers containing ETH^T 4001, bis(2-ethylhexyl)-sebacate and PVC. Whereas the spectral changes upon irradiation are very close to those of the toluene solution (Fig. 5, see also Fig. 3), the kinetics are more complicated as frequently observed in polymer solutions [14]. The absorption recovery may be fitted to a bi-exponential decay function with a main component of 100 ms lifetime and a minor one with 1 s. Since it was
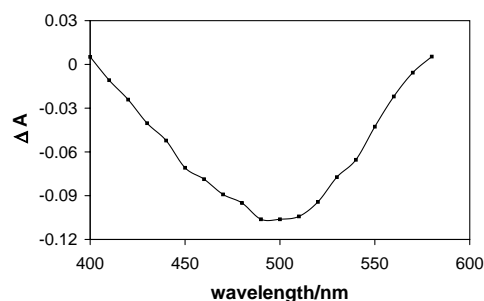

Fig. 5. Transient absorbance spectrum of ETH^T 4001 in plasticised PVC obtained via flash photolysis experiments. The spectrum is similar to the one in toluene (see Fig. 3).
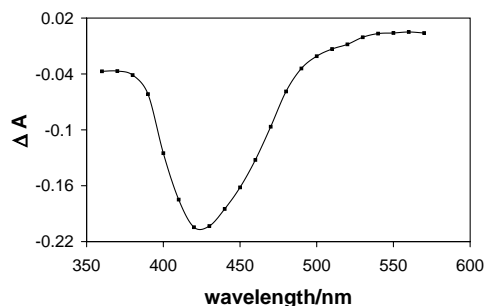
Fig. 6. Transient absorbance spectrum of the hemiaminal form of ETH^T 4001 (obtained by addition of 1-butylamine) in toluene.

Fig. 7. Boenstedt plot of the logarithm of the isomerization rate constant ($\log k$) versus pK_a of organic acids (see text).

not possible to control the temperature of the sensor layer (which changed from experiment to experiment) no further experiments were performed. However, it is important to notice that the lifetime is not enhanced in the polymer compared to the toluene solution. It should, furthermore, be noted that the lifetimes of Z-isomers of donor–acceptor azobenzenes are strongly dependent on the polarity of the solvent or the polymer matrix and are much shorter in polar matrices [12,13]. The hemiaminal form of ETH^T 4001 also exhibits a reversible bleaching in the region of the longest wavelength absorption band and a very weak transient absorbance peak at 560 nm (Fig. 6). The reaction is again fully photoreversible. Due to the comparatively long lifetime of the instable isomer (the hemiaminal is a less strong acceptor than the trifluoroacetyl group) it was not possible to follow the kinetics without significantly enhancing the reaction rate by absorption of measuring light.

3.3. Effects of acids on the rate of Z–E isomerization

The rate constant k of the Z–E isomerization of ETH^T 4001 in the presence of varying amounts of several acids of various pK_a (chloroacetic acid, salicylic acid, formic acid, 3-chlorobenzoic acid, acetic acid, pivalic acid) using toluene as the solvent was investigated. A quadratic dependence of the rate constant from the acid concentration was found which indicates an intermediate double protonation. Fitting of the rate constants to a polynomial

$$k = k_0 + a[\text{acid}] + b[\text{acid}]^2 \tag{5}$$

gave (mostly negative) values for the linear coefficients a with an error always larger than 100%. Hence, a was set to zero. Whether k_0 was fitted as a parameter or kept fixed to the experimental value did not significantly change the results for the coefficients b.

In Fig. 7, $\log k$ is plotted against the pK_a of the acids (Brønsted-plot). Even if interpreted as the sum of two Brønsted-coefficients of two consecutive reactions, the slope is remarkably large [10]. The catalysis is attributed to acid-assisted tautomerization of the Z-ammonium ion (formed by rapid protonation of Z-ETH^T 4001 generated upon photochemically induced E–Z isomerization) into a
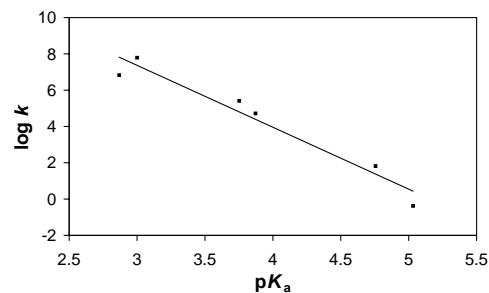
Z-azonium ion. The latter can easily isomerize via rotation around the –N=N– bond as a result of the concomitant decrease in the double bond character. However, the performance of sensor layers is not compromised by the effect of acids on the rate constant of Z–E isomerization. When the dye is dissolved in polymer matrices, a significant dielectric constraint against protonation of the dye (in absence of lipophilic anionic additives) is observed [2,3]. Consequently, protonation can only take place in the presence of, e.g. concentrated hydrochloric acid. Furthermore, measurements of aliphatic amines are performed at a pH around 13.0 in order to avoid the conversion of amines into non-reactive ammonium ions. Even in the case of aromatic amines, where a near-neutral pH is sufficient to provide amino groups, no protonation of the azobenzene dye is possible.

4. Conclusion

A wide range of publications on the application of dyes in DNA-sequencing, labelling of biomolecules, the investigation of membrane and cell characteristics, or in optical sensing can be found that describe the absorbance characteristics, changes in luminescence (lifetime), or sensing performance. In most cases, however, no details are given on the photochemistry of the dyes and how it may affect their performance. In the present investigation, we have shown that the amine-sensitive indicator dye 4-N,N-dioctylamino-4'-trifluoroacetylazobenzene is virtually not affected by photochemical and thermal isomerism. Thus, it can be used for sensing applications without the risk of false positive or negative results due to photochemistry.

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