Photochemical & Photobiological Sciences (2023) 22:2599–2606
https://doi.org/10.1007/s43630-023-00475-5

ORIGINAL PAPERS

Extraction of pure component spectra from ex situ illumination UV/Vis and NMR spectroscopy

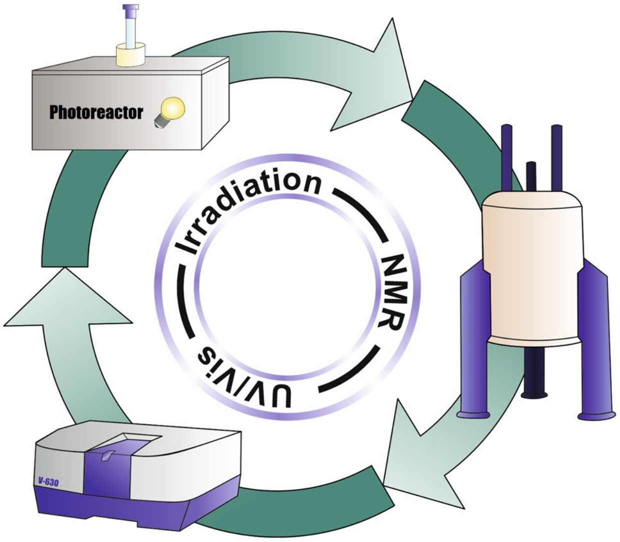
Dominik Herold[1] · **Jonas Kind**[1] · **Florian Frieß**[1] · **Christina M. Thiele**[1]

Received: 16 June 2023 / Accepted: 28 August 2023 / Published online: 26 September 2023

Abstract

Obtaining understanding of a photochemical reaction relies on the observation, identification and quantification of the compounds involved. The photochemical properties of the individual components are of particular importance, and their determination, however, is not always trivial. This is also true for the quantitative measure on the ability to absorb light, the extinction coefficient ε_i if more than one species i is present and two or more species absorb light of the same wavelength. In this work, it is demonstrated how pure component spectra can be obtained with a simple combination of successive and repeated *ex situ* illumination, UV/Vis and NMR spectroscopy. From the complementary information accessible, the wavelength-dependent extinction coefficients of all species can be calculated yielding the pure component spectra. A comparison with published data shows excellent agreement and thus proves that this approach is highly reliable.

Graphical abstract



Keywords Dithienylethene · NMR spectroscopy · Photochromism · Pure component · UV/Vis spectroscopy

1 Introduction

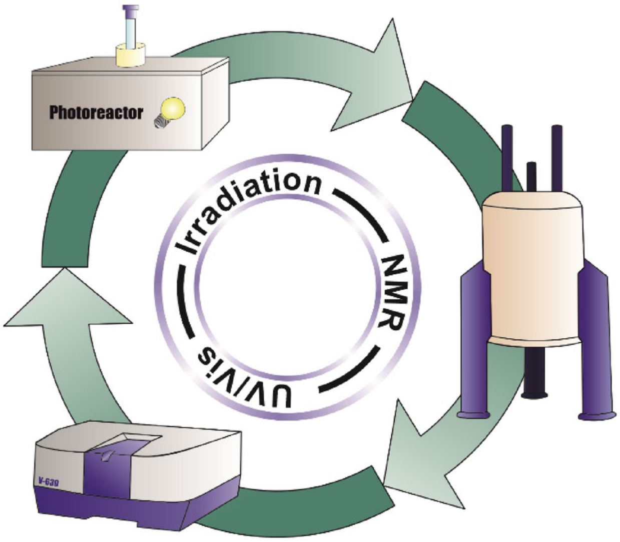
Photochemical reactions are of particular interest in many different disciplines of current research. This applies to catalyst-based synthesis and mechanistic studies [1–6], medical application [7–10], development of photo-reactors [11], new

---

✉ Christina M. Thiele
cthiele@thielelab.de

[1] Clemens-Schöpf-Institut für Organische Chemie und Biochemie, Technische Universität Darmstadt, 64287 Darmstadt, Germany

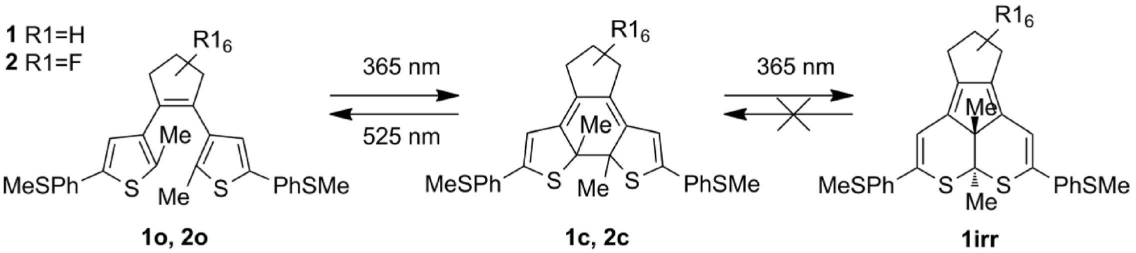
production processes of polymers and gels [12, 13], photoresponsive materials [14–18], optical memories [19–21] and even chemical rotors [22–24]. For this purpose, it is necessary to identify all components involved and to be able to track these components during the course of the reaction. For simple systems, like A → B reactions, where the absorption bands of the two components are well separated and full conversion can be achieved, it is an easy task to obtain the UV/Vis spectra of both pure components. For more complex systems, however, for example with an unknown number of reaction components, and/or if the species have overlapping absorption bands or for more complex reaction schemes, this task becomes more difficult. This is especially true when a large number of components are involved and it is therefore particularly challenging to distinguish them. However, in order to fully understand the photochemical reaction behaviour, it is necessary to obtain pure component spectra for each individual component separately. With respect to photochemical reactions, absorption spectra and the resulting photochemical properties like absorption coefficients ε are of particular interest. With the help of these, the absorption characteristics and the mechanisms can be described in detail and further photochemical values like quantum yields Φ become accessible [25]. Today, there still seems to be a problem in obtaining the absorption spectra of pure components for complex photochemical systems with negative consequences for further applications [26, 27]. For systems with a small number of stable components, separation of the involved species for example via HPLC can help obtain spectra of the pure components. However, this is often time-consuming or impeded by limited lifetimes. Thus, if there are more than two or an unknown number of components with overlapping absorption bands involved, other methods have to be used [28, 29]. If the number of components can be estimated mathematically [30], indirect hard modelling (IHM) can be used [31, 32]. If the number of components is known, a system of equations can be set up from rate equations of each component with the aid of a reaction model [33–35]. Multivariate curve resolution (MRC) and optimised variants have proven to be methods that correctly represent the reaction process for each individual component and the corresponding spectra of all pure components, independent of the selected measurement technique. These methods have been shown to lead to even superior results if complementary measurement techniques are used [36–39]. However, it is not always necessary to use such methods to obtain pure spectra of components of a photochemical reaction. As we will demonstrate herein (Scheme 1), even very simple approaches based on the complementary use of spectroscopic techniques (NMR and UV/Vis) lead to correct results with little mathematical effort.



Scheme 1 Schematic depiction of the iterative procedure to determine photochemical properties using consecutive illumination, UV/Vis and NMR spectroscopic measurements

Using a combination of UV/Vis and NMR spectroscopy for this purpose was described for azobenzene as an example, however, other approaches are also feasible [27, 40, 41]. The approach relies on the quantitative nature of NMR together with its structural information for obtaining the identity and concentrations of individual species, thus providing the complementary information missing in the UV–Vis spectra of complex systems. We propose here, that it is sufficient for many simple but also more complex photochemical systems, for example for photochromic substances or chemical actinometers, and that it provides reliable values for photochemical properties.

This will be demonstrated on two compounds belonging to the class of dithienylethenes (**DTEs**, Scheme 2). Previous investigations on both **DTE** systems describe their high potential as electrically and photochemically switchable systems [42]. In particular, **DTE1** (Scheme 2) shows an exceptional behaviour: its response to electrical stimuli is complete reversible without any signs of fatigue. From photochemical point of view, however, the cyclization reaction is not satisfactorily reversible and comes with irreversible formation of by-products [43]. In comparison, **DTE2** shows high reversibility with little or no photobleaching, decomposition or any by-products. Although both systems were previously investigated photochemically, no pure component spectra or characteristic photochemical properties were determined. Hence, to obtain pure component UV/Vis spectra of all species involved in the two photoreactions shown, the combination of illumination, UV/Vis and NMR spectroscopy was executed in this work. With this powerful combination, the photo-cyclization can be monitored, the species identified and quantified. Thus, pure component spectra as well as photochemical properties for each individual species



Scheme 2 Reaction scheme of **1** and **2** upon illumination with UV (365 nm) for photo-cyclization (reversible) and green light (525 nm) for photo-cyclo-reversion. The last reaction step only occurs for **1** upon continuing UV illumination of **1c** and yields an irreversible species **1irr**. This is not observed for **2** (no **2irr** is observed) and a PSS can be reached

become accessible. It is to note, however, that for systems with limited life-time, this stepwise approach will naturally lead to erroneous results and thus a simultaneous and quantitative measurement of UV–Vis and NMR spectra would be very beneficial. Alternatively, deconvolution of the UV-spectra can be employed assuming the absorption bands to be of Gaussian shape, e.g. [44].

2 Experimental

2.1 Concept

As some knowledge of the reaction system is required to fully describe the total reactions process and absorption behaviour, we assume here that, the photoreactions of **DTEs** can be written as consecutive $A \rightleftharpoons B(\rightarrow C)$ reactions and that the species do not possess limited life-times. As a first step, the species occurring in the sample have to be identified and quantified (requirement 1). Second, the absorbance of the sample at each time/composition point needs to be known to obtain the individual absorbances of the species (requirement 2). Both spectroscopic techniques, UV/Vis and NMR, reveal important and complementary information for the photochemical reactions. Using NMR spectroscopy, all occurring species can be identified and with the knowledge of the total concentration in the sample, concentrations of each individual species can be calculated from the ratio of the corresponding NMR signals (fulfilling requirement 1). From UV/Vis series, time-dependent absorbances of the total sample are accessible. If all possible species i in the system are known, the total absorbance at a specific wavelength $A_{\lambda,\mathrm{tot}}$ can be written as the sum of the absorbances of each pure species $A_{\lambda,i}$. Further following Lambert–Beer's law, $A_{\lambda,\mathrm{tot}}$ can be calculated with Eq. 1 as linear combination of the pathlength d, the concentrations c_i and the wavelength-dependent extinction coefficients $\varepsilon_{\lambda,i}$ of each species.

$$A_{\lambda,\mathrm{tot}} = d \bullet \sum c_i \bullet \varepsilon_{\lambda,i} \qquad (1)$$

The total absorbance $A_{\lambda,tot}$ in Eq. 1 is dependent on the illumination time t giving a system of linear equations (in which the concentration from NMR is used). With the measurements of the absorbance at different time points $A_{\lambda,\mathrm{tot}}(t)$, the solution of the system of linear equations results in corresponding absorbance of each species (and thus fulfilling requirement 2). This allows the calculation of extinction coefficients at all wavelengths $\varepsilon_{\lambda,i}$ and thus represents pure component spectra for each species in the photo-cyclization of compounds **1** and **2**.

2.2 Characterisation and measurement methods

UV/Vis spectra were recorded on a V-630 spectrophotometer (JASCO) with 1 mm quartz cuvettes and standard grade solvents. UV/Vis data were processed with JASCO Spectra Manager and exported as ASCII files. All ^{1}H-NMR spectra were recorded on a 400 MHz proton resonance frequency (Bruker AVANCE III HD, equipped with a 5-mm Bruker PABBO broadband probe (BB/^{1}H-^{19}F/D) with z-gradient). 5 mm economy NMR tubes (Wilmad LabGlass, WG-5-E7) were used for the preparation of those samples. DMF-d_7 (99.5 atom % D) was purchased from Sigma-Aldrich in separate 0.75 mL ampoules and used as received. All measurements were performed at 300 K without spinning of the sample. Detailed acquisition parameters can be found in the SI. Bruker TopSpin (Version 3.5 pl 2) was used for processing NMR data. All spectra were referenced to the DMF signal at 8.03 ppm. After Fourier transformation, phase and baseline corrections were executed automatically using TopSpin. To obtain spectra with different conversion ratios, ex situ illuminations with LEDs of specific wavelengths were performed for successive time intervals. For this purpose, a custom-made photo-reactor with UV (365 nm) and green (525 nm) LEDs featuring a socket for a 5 mm economy NMR tube was used [45], ensuring constant and reproducible illumination. The approach described consists of sequentially recording NMR and UV/Vis spectra alternating with periods of illumination. Prior to the start of the measurement series, green light (525 nm) was used to ensure that the highest possible proportion of open species **o** is present in the respective sample. At first, both measurements (NMR and UV/Vis) were performed on the solution of either **2** or **1** in DMF-d_7 before illumination with UV light (365 nm) was conducted. The sample was prepared in an NMR tube and a ^{1}H NMR was recorded. For UV/Vis spectroscopy, the solution was transferred into a quartz cuvette. After retransferring the solution into the NMR tube, the ring-opening photoreaction was initiated upon illumination with UV light (365 nm) in a custom-made photo-reactor [45] and the measurement procedure was repeated, resulting in ^{1}H-NMR and UV/Vis spectra series.

3 Results and discussion

To demonstrate the beauty of this approach for the determination of spectra of pure components, two DTE systems are considered in which the number of components is known and follows a $A \rightleftharpoons B$ or $A \rightleftharpoons B \rightarrow C$ reaction scheme. These are the identical compounds we used in our previous study. They were synthesised by the Gallei group. Details on the synthetic procedure can be found in our previous publication [42]. The validity of our approach is first demonstrated with **2** due to its simple reaction system with separated absorption bands for the ring-opened and ring-closed form of **2** and is subsequently applied to the more complex system of **1**, with overlapping absorption spectra for all isomers of **1** (Scheme 2). It is to note, that we did not investigate the conformational preferences of the open compounds in detail as we assume the interconversion of the respective conformers to be fast (as was previously observed for similar compounds) [46] so that no influence of the interconversion of conformers on the photo-cyclization is expected. The measurements start at pure **2o/1o** (obtained via green light) and follow the iterative procedure NMR, UV/Vis and illumination (see Scheme 1) with UV light.

Table 1 Calculated extinction coefficients at the maxima and excitation wavelengths of all species in both dithienylethenes in DMF

		ε (λ [nm]) [10^3 M^{-1} cm^{-1}][a]		
Comp	Signal	Open form	Closed form	Irreversible. form
1	S	~34 (295)	~17 (286)	
			~17 (369)	
	M	41.14 (325)	33.94 (325)	35.60 (320)
			21.58 (537)	11.03 (525)
	E	0.65 (365)	18.00 (365)	2.70 (365)
		0.28 (525)	20.46 (525)	11.03 (525)
2	S		~13 (280)	
	M	56.65 (320)	28.79 (384)	
			22.00 (610)	
	E	2.16 (365)	25.57 (365)	
		0.15 (525)	8.45 (525)	

[a] *S* = Shoulder, *M* = Maximum, *E* = Excitation wavelengths

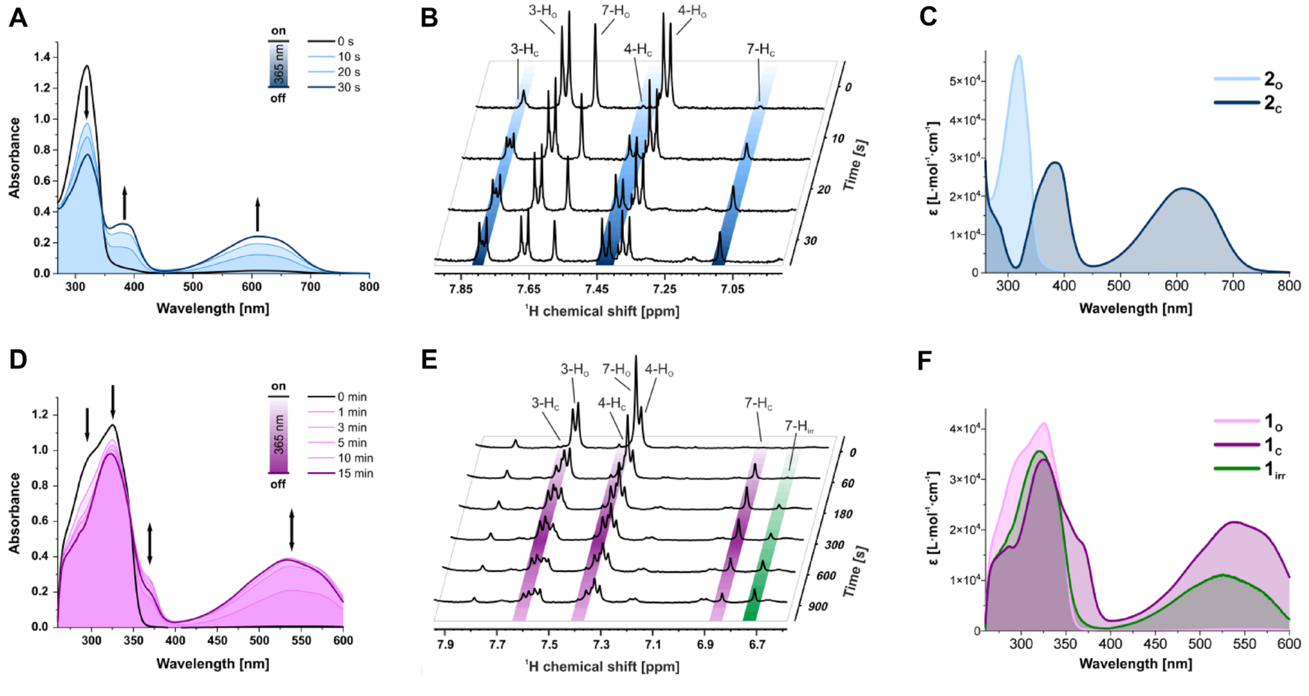


Fig. 1 UV/Vis spectra series (left, **A** and **D**) and ^{1}H NMR (400 MHz) spectra series (middle, **B** and **E**) of **2** and **1** in DMF-d_7 at 300 K upon UV illumination (365 nm) in a custom-made photo-reactor equipped with a NMR tube socket. Resulting pure component spectra of **2o** and **2c** (right, **C**) as well as **1o**, **1c** and **1irr** (right, **F**)

The photo-induced cyclization of both DTEs with UV light can be monitored in the respective spectra series either by NMR signals (**B**, **E**) in the aliphatic and aromatic region or by the absorbance maxima in UV/Vis spectra (**A**, **D**). The absorbance maxima for **2o** are at 320 nm, for **2c** at 610 nm, for **1o** at 325 nm and for **1c** at 537 nm. For both DTEs, a decrease of the open form and a simultaneous increase of the closed form are observed during illumination with UV light. Additionally, during illumination of **1** with UV light, a third species appears within the first minutes, which is structurally characterised and confirmed to be **1irr** [42] (absorbance and concentration profiles are shown in the SI Figure S5). Due to this irreversible rearrangement reaction, no photo-stationary state can be reached and the illumination was stopped after 900 s (a continued illumination followed by UV/Vis spectroscopy is given in the SI Figure S6). In comparison, applying the same procedure to a sample of **2**, the illumination with UV light leads to a photo-stationary state of 70% **2c** within a few minutes of illumination confirming the results of our previous investigation [42]. Reaching a photo-stationary state indicates an equilibrium state without consecutive reactions, decomposition or photobleaching. This can be verified by NMR and UV/Vis spectroscopy in which no change of signal intensity in ^{1}H or absorption spectra is observed. By integration of baseline separated ^{1}H NMR signals (**2**: 7-H$_o$, thiophene proton for **2o** and 7-H$_c$, thiophene proton for **2c** as well as 13-H$_o$, methyl group at thiophene for **1o**, 7-H$_c$, thiophene proton for **1c** and 7-H$_{irr}$, thiophene proton for **1irr**), concentrations of each species can be calculated. Following EQ1 using time-dependent absorbances and these concentrations, the system of linear equations can be solved with the method of least squares. This yields extinction coefficients for both DTEs and each respective species for the measured wavelength range (Fig. 1, Table 1).

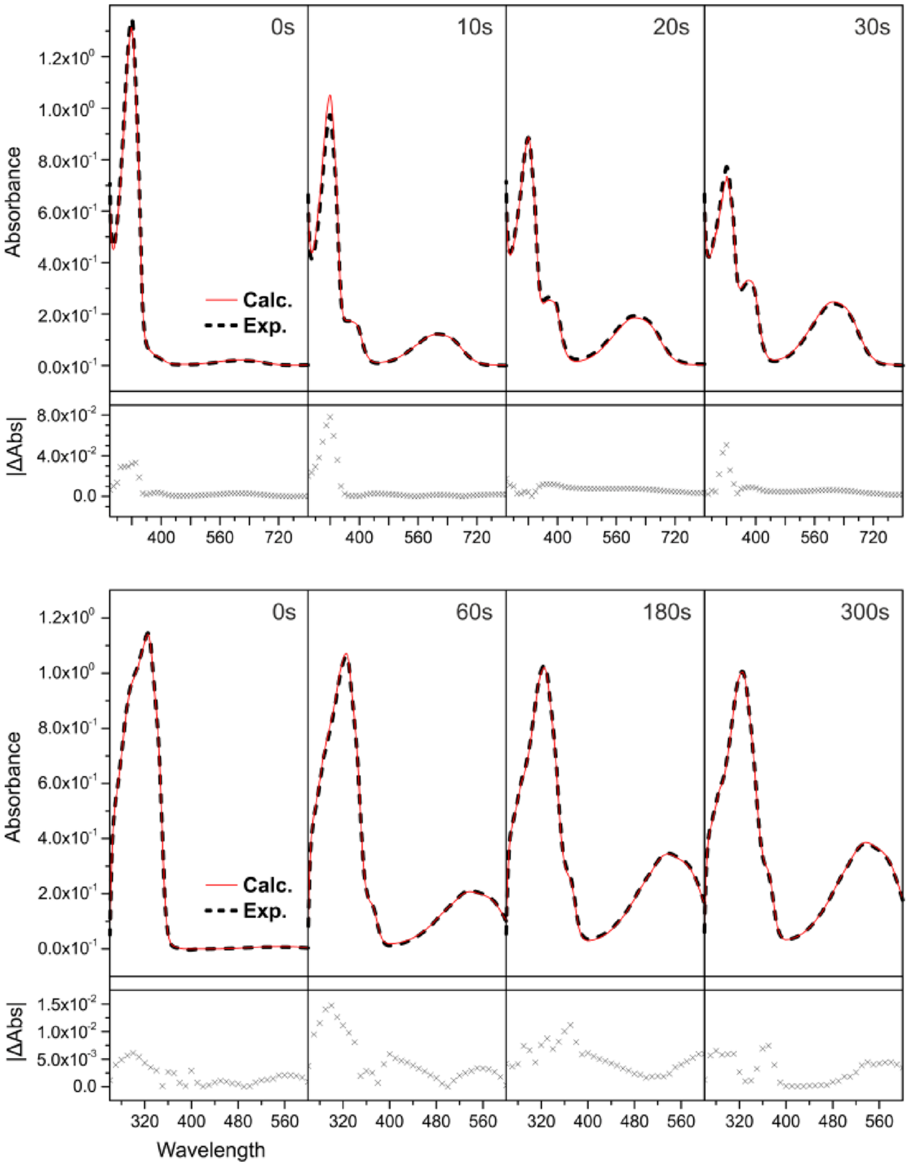


Fig. 2 Calculated and experimental UV/Vis spectra series of the photo-cyclization reaction of **2** (top) and **1** (bottom) upon illumination with UV light (365 nm) in DMF-d_7 at room temperature. The absolute difference $|\Delta Abs|$ is given at the bottom part of each spectrum separately for every wavelength. Illumination times are set as specified in the legends

These represent the pure component spectra, which provide information about the compound's photochemical ability to absorb light at specific wavelengths and are essential for understanding and applying these systems. As last step, in order to be able to assess whether the results obtained are of a correct order of magnitude, we compared our results with results from previous studies and used (back-)calculated absorption spectra to quantify the error. Since **2** was only investigated in our previous study [42], comparability to literature is limited and thus we can only compare our values to related compounds (for further information see SI Figure S3 and Table S4). **1** has already been synthesised, investigated and extinction coefficients were determined by Browne et al. [47] However, a different solvent (CH_3CN (ACN)) was used resulting in slightly different and shifted values at the maxima (M), isosbestic points (I) and shoulders (S). In both cases, extinction coefficients agree very well and also wavelengths are reproduced reliably. The same is demonstrated by calculating the absorption spectra with the extinction coefficients obtained (Fig. 2, for whole UV/Vis series, see SI, Figure S4).

The comparison between the calculated and the experimental absorption spectra results in very small errors, which further supports the accuracy of the procedure used.

4 Conclusion

In this work, we present an approach to extract the pure component spectra of photochemical systems with more than one absorbing species and overlapping absorption bands using a combination of consecutive illumination, UV/Vis and NMR spectroscopy and simple mathematical tools. NMR spectroscopy allows the identification of all species together with their quantification. Thus, by obtaining the concentration from NMR and absorbance time profiles from UV/Vis spectroscopy, the wavelength-dependent extinction coefficients of all individual species can be determined. This iterative procedure is applicable not only to simple but also to more complex photochemical/chromic reactions. This allows obtaining pure component spectra. We further used (back-)calculated absorption spectra to confirm the accuracy of the extracted data and to quantify errors. The approach described here thus provides a method to reliably determine photochemical properties of pure components in complex mixtures.

Supplementary Information The online version contains supplementary material available at https://doi.org/10.1007/s43630-023-00475-5.

Acknowledgements We want to thank Dr. Jonas von Irmer from the group of Prof. Dr.-Ing. Markus Gallei for synthesizing and providing the DTE compounds, Dr. Henning Schröder from the group of Prof. Dr. Klaus Neymeyr for discussions and initial interpretation of the measurement data.

Funding Open Access funding enabled and organized by Projekt DEAL. This work was financially supported by the German Research Foundation DFG under grant TH 1115/9–1.

Declarations

Conflict of interest The authors declare no conflict of interest.

References

1. Buzzetti, L., Crisenza, G. E. M., & Melchiorre, P. (2019). Mechanistic studies in photocatalysis. *Angewandte Chemie (International ed. in English), 58*, 3730–3747. https://doi.org/10.1002/anie.201809984
2. Howe, R. F. (2008). Recent developments in photocatalysis. *Developments in Chemical Engineering and Mineral Processing, 6*, 55–84. https://doi.org/10.1002/apj.5500060105
3. Göstl, R., Senf, A., & Hecht, S. (2014). Remote-controlling chemical reactions by light: Towards chemistry with high spatio-temporal resolution. *Chemical Society Reviews, 43*, 1982–1996. https://doi.org/10.1039/c3cs60383k
4. Tay, N. E. S., Lehnherr, D., & Rovis, T. (2022). Photons or electrons? A critical comparison of electrochemistry and photoredox catalysis for organic synthesis. *Chemical Reviews, 122*, 2487–2649. https://doi.org/10.1021/acs.chemrev.1c00384
5. Marzo, L., Pagire, S. K., Reiser, O., & König, B. (2018). Photokatalyse mit sichtbarem Licht: Welche Bedeutung hat sie für die organische synthese? *Angewandte Chemie, 130*, 10188–10228. https://doi.org/10.1002/ange.201709766
6. Marzo, L., Pagire, S. K., Reiser, O., & Konig, B. (2018). Visible-light photocatalysis: does it make a difference in organic synthesis? *Angewandte Chemie (International ed. in English), 57*, 10034–10072. https://doi.org/10.1002/anie.201709766
7. Gao, S., Tang, G., Hua, D., Xiong, R., Han, J., Jiang, S., Zhang, Q., & Huang, C. (2019). Stimuli-responsive bio-based polymeric systems and their applications. *J Mater Chem B, 7*, 709–729. https://doi.org/10.1039/c8tb02491j
8. Olejniczak, J., Carling, C. J., & Almutairi, A. (2015). Photocontrolled release using one-photon absorption of visible or NIR light. *Journal of Controlled Release, 219*, 18–30. https://doi.org/10.1016/j.jconrel.2015.09.030
9. Wang, D., & Wang, X. (2013). Amphiphilic azo polymers: Molecular engineering, self-assembly and photoresponsive properties. *Progress in Polymer Science, 38*, 271–301. https://doi.org/10.1016/j.progpolymsci.2012.07.003
10. Heinze, K. (2017). The quest for mononuclear gold(II) and its potential role in photocatalysis and drug action. *Angewandte*

Chemie (International ed. in English), 56, 16126–16134. https://doi.org/10.1002/anie.201708349

11. Boyjoo, Y., Sun, H., Liu, J., Pareek, V. K., & Wang, S. (2017). A review on photocatalysis for air treatment: From catalyst development to reactor design. *Chemical Engineering Journal, 310*, 537–559. https://doi.org/10.1016/j.cej.2016.06.090
12. Ravetz, B. D., Pun, A. B., Churchill, E. M., Congreve, D. N., Rovis, T., & Campos, L. M. (2019). Photoredox catalysis using infrared light via triplet fusion upconversion. *Nature, 565*, 343–346. https://doi.org/10.1038/s41586-018-0835-2
13. Yeo, H., & Khan, A. (2020). Photoinduced proton-transfer polymerization: a practical synthetic tool for soft lithography applications. *Journal of the American Chemical Society, 142*, 3479–3488. https://doi.org/10.1021/jacs.9b11958
14. Emoto, A., Uchida, E., & Fukuda, T. (2012). Optical and physical applications of photocontrollable materials: azobenzene-containing and liquid crystalline polymers. *Polymers, 4*, 150–186. https://doi.org/10.3390/polym4010150
15. Wei, Y. B., Tang, Q., Gong, C. B., & Lam, M. H. (2015). Review of the recent progress in photoresponsive molecularly imprinted polymers containing azobenzene chromophores. *Analytica Chimica Acta, 900*, 10–20. https://doi.org/10.1016/j.aca.2015.10.022
16. Khoo, Z. X., Teoh, J. E. M., Liu, Y., Chua, C. K., Yang, S., An, J., Leong, K. F., & Yeong, W. Y. (2015). 3D printing of smart materials: A review on recent progresses in 4D printing. *Virtual and Physical Prototyping, 10*, 103–122. https://doi.org/10.1080/17452759.2015.1097054
17. Sun, S., Liang, S., Xu, W.-C., Xu, G., & Wu, S. (2019). Photoresponsive polymers with multi-azobenzene groups. *Polymer Chemistry, 10*, 4389–4401. https://doi.org/10.1039/c9py00793h
18. Hirschmann, M., Soltwedel, O., Ritzert, P., von Klitzing, R., & Thiele, C. M. (2023). Light-controlled lyotropic liquid crystallinity of polyaspartates exploited as photo-switchable alignment medium. *Journal of the American Chemical Society, 145*, 3615–3623. https://doi.org/10.1021/jacs.2c12760
19. Berkovic, G., Krongauz, V., & Weiss, V. (2000). Spiropyrans and spirooxazines for memories and switches. *Chemical Reviews, 100*, 1741–1754. https://doi.org/10.1021/cr9800715
20. Irie, M., Fukaminato, T., Matsuda, K., & Kobatake, S. (2014). Photochromism of diarylethene molecules and crystals: Memories, switches, and actuators. *Chemical Reviews, 114*, 12174–12277. https://doi.org/10.1021/cr500249p
21. Yokoyama, Y. (2000). Fulgides for memories and switches. *Chemical Reviews, 100*, 1717–1740. https://doi.org/10.1021/cr980070c
22. Feringa, B. L. (2017). The art of building small: from molecular switches to motors (nobel lecture). *Angewandte Chemie (International ed. in English), 56*, 11060–11078. https://doi.org/10.1002/anie.201702979
23. Kottas, G. S., Clarke, L. I., Horinek, D., & Michl, J. (2005). Artificial molecular rotors. *Chemical Reviews, 105*, 1281–1376. https://doi.org/10.1021/cr0300993
24. Filatov, M., Paolino, M., Min, S. K., & Kim, K. S. (2018). Fulgides as light-driven molecular rotary motors: computational design of a prototype compound. *Journal of Physical Chemistry Letters, 9*, 4995–5001. https://doi.org/10.1021/acs.jpclett.8b02268
25. Gauglitz, G., Goes, R., Stooß, W., & Raue, R. (1985). Determination of partial photochemical quantum yields of reversible photoisomerizations of stilbene-1 derivatives. *Zeitschrift für Naturforschung A, 40*, 317–323. https://doi.org/10.1515/zna-1985-0403
26. Wriedt, B., & Ziegenbalg, D. (2020). Common pitfalls in chemical actinometry. *Journal of Flow Chemistry, 10*, 295–306. https://doi.org/10.1007/s41981-019-00072-7
27. Vetrakova, L., Ladanyi, V., Al Anshori, J., Dvorak, P., Wirz, J., & Heger, D. (2017). The absorption spectrum of cis-azobenzene. *Photochemical & Photobiological Sciences, 16*, 1749–1756. https://doi.org/10.1039/c7pp00314e
28. Antonov, L., & Petrov, V. (2002). Quantitative analysis of undefined mixtures - "fishing net" algorithm. *Analytical and Bioanalytical Chemistry, 374*, 1312–1317. https://doi.org/10.1007/s00216-002-1600-8
29. Antonov, L. (1997). Drawbacks of the present standards for processing absorption spectra recorded linearly as a function of wavelength, TrAC. *Trends in Analytical Chemistry, 16*, 536–543. https://doi.org/10.1016/s0165-9936(97)00064-2
30. Elbergali, A., Nygren, J., & Kubista, M. (1999). An automated procedure to predict the number of components in spectroscopic data. *Analytica Chimica Acta, 379*, 143–158. https://doi.org/10.1016/s0003-2670(98)00640-0
31. Kriesten, E., Mayer, D., Alsmeyer, F., Minnich, C. B., Greiner, L., & Marquardt, W. (2008). Identification of unknown pure component spectra by indirect hard modeling. *Chemometrics and Intelligent Laboratory Systems, 93*, 108–119. https://doi.org/10.1016/j.chemolab.2008.05.002
32. Kriesten, E., Alsmeyer, F., Bardow, A., & Marquardt, W. (2008). Fully automated indirect hard modeling of mixture spectra. *Chemometrics and Intelligent Laboratory Systems, 91*, 181–193. https://doi.org/10.1016/j.chemolab.2007.11.004
33. Antonov, L., & Stoyanov, S. (1995). Resolution of overlapping UV-visible absorption bands: Quantitative analysis of tautomeric equilibria. *Analytica Chimica Acta, 314*, 225–232. https://doi.org/10.1016/0003-2670(95)00281-4
34. Antonov, L., & Nedeltcheva, D. (2000). Resolution of overlapping UV–Vis absorption bands and quantitative analysis. *Chemical Society Reviews, 29*, 217–227. https://doi.org/10.1039/a900007k
35. Antonov, L., & Stoyanov, S. (1993). Analysis of the overlapping bands in UV-Vis absorption spectroscopy. *Applied Spectroscopy, 47*, 1030–1035.
36. de Juan, A., & Tauler, R. (2021). Multivariate Curve resolution: 50 years addressing the mixture analysis problem - a review. *Analytica Chimica Acta, 1145*, 59–78. https://doi.org/10.1016/j.aca.2020.10.051
37. Ruckebusch, C., & Blanchet, L. (2013). Multivariate curve resolution: A review of advanced and tailored applications and challenges. *Analytica Chimica Acta, 765*, 28–36. https://doi.org/10.1016/j.aca.2012.12.028
38. Garrido, M., Rius, F. X., & Larrechi, M. S. (2008). Multivariate curve resolution-alternating least squares (MCR-ALS) applied to spectroscopic data from monitoring chemical reactions processes. *Analytical and Bioanalytical Chemistry, 390*, 2059–2066. https://doi.org/10.1007/s00216-008-1955-6
39. Ruckebusch, C., Vitale, R., Ghaffari, M., Hugelier, S., & Omidikia, N. (2020). Perspective on essential information in multivariate curve resolution, TrAC. *Trends in Analytical Chemistry., 132*, 1160. https://doi.org/10.1016/j.trac.2020.116044
40. Roseau, M., De Waele, V., Trivelli, X., Cantrelle, F. X., Penhoat, M., & Chausset-Boissarie, L. (2021). Azobenzene: A visible-light chemical actinometer for the characterization of fluidic photosystems. *Helvetica Chimica Acta*. https://doi.org/10.1002/hlca.202100071
41. Roseau, M., Chausset-Boissarie, L., Gremetz, S., Roth, P. M. C., & Penhoat, M. (2022). Multiple wavelength (365–475 nm) complete actinometric characterization of corning((R)) lab photo reactor using azobenzene as a highly soluble, cheap and robust chemical actinometer. *Photochemical & Photobiological Sciences, 21*, 421–432. https://doi.org/10.1007/s43630-022-00171-w
42. von Irmer, J., Frieß, F., Herold, D., Kind, J., Thiele, C. M., & Gallei, M. (2019). Photochromic dithienylethenes characterized by in situ irradiation NMR-spectroscopy and electrochemically induced responsiveness on gold substrates. *Journal of Materials*

Chemistry C, 7, 14088–14097. https://doi.org/10.1039/C9TC04495G

43. Irie, M., Lifka, T., Uchida, K., Kobatake, S., & Shindo, Y. (1999). Fatigue resistant properties of photochromic dithienylethenes: by-product formation. *Chemical Communications.* https://doi.org/10.1039/A809410A
44. Rustler, K., Nitschke, P., Zahnbrecher, S., Zach, J., Crespi, S., & Konig, B. (2020). Photochromic evaluation of 3(5)-Arylazo-1H-pyrazoles. *Journal of Organic Chemistry, 85*, 4079–4088. https://doi.org/10.1021/acs.joc.9b03097
45. Niedek, D., Erb, F. R., Topp, C., Seitz, A., Wende, R. C., Eckhardt, A. K., Kind, J., Herold, D., Thiele, C. M., & Schreiner, P. R. (2020). In situ switching of site-selectivity with light in the acetylation of sugars with azopeptide catalysts. *The Journal of Organic Chemistry, 85*, 1835–1846. https://doi.org/10.1021/acs.joc.9b01913
46. Fredersdorf, M., Göstl, R., Kolmer, A., Schmidts, V., Monecke, P., Hecht, S., & Thiele, C. M. (2015). Exploring the conformational space of bridge-substituted dithienylcyclopentenes. *Chemistry - A European Journal, 21*, 14545–14554. https://doi.org/10.1002/chem.201501842
47. Browne, W. R., de Jong, J. J. D., Kudernac, T., Walko, M., Lucas, L. N., Uchida, K., van Esch, J. H., & Feringa, B. L. (2005). Oxidative electrochemical switching in dithienylcyclopentenes, part 2: Effect of substitution and asymmetry on the efficiency and direction of molecular switching and redox stability. *Chemistry – A European Journal, 11*, 6430–6441. https://doi.org/10.1002/chem.200500163