Photochemistry

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Spatial Structuring of a Supramolecular Hydrogel by using a Visible-Light Triggered Catalyst**

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Abstract: *Spatial control over the self-assembly of synthetic molecular fibers through the use of light-switchable catalysts can lead to the controlled formation of micropatterns made up of hydrogel structures. A photochromic switch, capable of reversibly releasing a proton upon irradiation, can act as a catalyst for in situ chemical bond formation between otherwise soluble building blocks, thereby leading to fiber formation and gelation in water. The use of a photoswitchable catalyst allows control over the distribution as well as the mechanical properties of the hydrogel material. By using homemade photomasks, spatially structured hydrogels were formed starting from bulk solutions of small molecule gelator precursors through light-triggered local catalyst activation.*

Low-molecular-weight gelators (LMWG) can trap the surrounding solvent by self-assembling into a three-dimensional network of fibers, thereby forming a gel.[1] Hydrogels may find application in cell culture,[2] as materials for controlled release,[3] and as actuators.[4] Hydrogels are commonly made as bulk, homogeneously distributed materials. Control over their distribution in space can be difficult to achieve but would be desirable for many applications.[5] We have recently described a system in which low-molecular-weight hydrogels are made in situ, under ambient conditions, through the reaction of two non-assembling building blocks to form the gelator.[6] The rate of gelator formation can be enhanced by using a catalyst, which has a pronounced effect on the rate of gelation, fiber morphology, and macroscopic gel properties. Spatial control over the rate of gelator formation should allow control over the spatial distribution of the gelator material and its properties. We reasoned that spatial control over this reaction rate may be achieved by controlling catalytic activity through an external stimulus. Recently, by employing an immobilized catalyst on a surface, we[7] and others[8] achieved spatial control over the formation of supramolecular gel fibers at the solid–liquid interface. Herein, we describe the use of a visible-light-triggered catalyst to control the spatial distribution of supramolecular hydrogel materials and their properties.

We recently described a low-molecular-weight hydrogelator that assembles upon formation from non-assembling aldehyde and hydrazide building blocks through the formation of a hydrazone bond. In aqueous environments, the rate of hydrazone formation can be enhanced by using either acid or aniline catalysis.[6,9] The use of a catalyst for which the activity can be changed by using light as an external trigger[10] would enable control over the rate of material formation in time and space. As an external stimulus, light has clear advantages over other stimuli such as pH or temperature since spatial variations in intensity can be applied easily, for instance, by using masks or focused beams. As such, this approach has been applied to photochromic gelators.[11] Light-triggered activation of a gelator precursor through photochemical deprotection has also been described,[12] as has photolysis of a photoacid generator (PAG) to induce protonation-triggered gelation.[13] The latter system has to be kept permanently at an acidic pH to maintain the gel state, which can be disadvantageous for certain applications. Likewise, the use of visible instead of ultraviolet light as a trigger can be desirable, especially when medium transparency plays a role. In our system, gelation is controlled by a visible-light-activated photoswitchable acid catalyst,[14] which allows a return to neutral pH upon deactivation of the catalyst.

We used LMWG **3** (Figure 1), which can be formed in situ through the acid catalyzed reaction of cyclohexane trishydrazide **1** and three molecules of aldehyde **2**.[6,8,15] Instead of the previously employed permanent mineral acid catalysts, we chose to use a photoswitchable organic acid to catalyze hydrazone bond formation upon irradiation. As a photoswitchable acid, we used the water soluble photochromic acid switch PAH (Figure 1).[14,16] In aqueous solution, stabilization of the polar, zwitterionic merocyanine form of this switch leads to predominant formation of the ring-opened structure. An aqueous solution of PAH ([PAH] $= 1.0$ mM) has a pH value of 6.6 owing to the slightly acidic phenol group. Upon irradiation with visible light ($400 < \lambda < 800$ nm; see the Supporting Information), ring closure to the spiropyran form of the switch occurs, thereby leading to the release of a proton into the medium. Thus, the pH of the solution drops to 5.4 upon irradiation with visible light. When the light is turned off, the pH of the solution rapidly returns to its original

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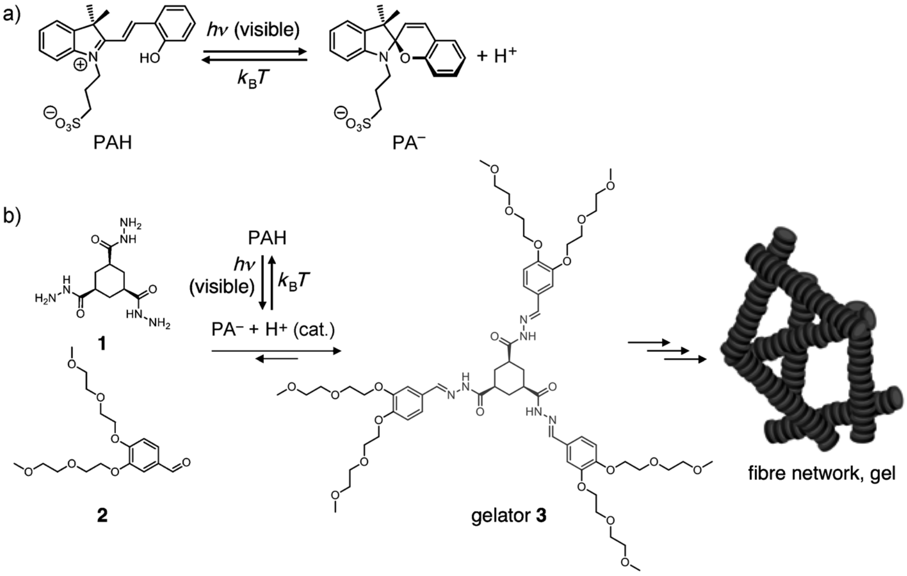

Figure 1. Supramolecular gelation using a photoswitchable catalyst. a) Light-driven conversion of the merocyanine (PAH) to the spiropyran (PA$^-$) form of the photoswitchable acid catalyst yields a proton (H$^+$). Thermally activated conversion back to PAH removes the proton from solution. b) Light-triggered acid-catalyzed formation of trishydrazone gelator **3** from trishydrazide **1** and benzaldehyde **2** leads to gelation through the formation of a crosslinked fiber network.

value since the spiropyran reverts back to the protonated merocyanine form at room temperature. Typical time scales for the photochemical forward and thermal back reactions are 2 min and 5 min, respectively. The process is reversible and thus the pH can be simply controlled within a certain range by switching a light on and off. Furthermore, because of the efficient thermal back reaction, diffusion of protons to non-irradiated zones in a sample is limited. It is important to note that the current LMWG system will remain in the gel state across the entire pH window of PAH and it thus differs from those gelator systems[13a,17] that dissolve with increasing pH values.

Previous studies have shown that the reaction between **1** and **2** yields mainly the trishydrazone gelator **3** and a minor amount of the bishydrazone ($<5\%$) derivative, irrespective of catalyst content. However, catalysts increase the rate of gelator formation, which results in faster gelation and the formation of stiffer gels.[6,15] In the current system, mixing clear aqueous solutions of **1** and **2** (pH 7.1) at room temperature without a catalyst resulted, after 6 h reaction time, in the formation of a colorless turbid suspension that was unable to support its own weight (Figure 2c). The failure of the uncatalyzed system to gel is in line with previous results and occurs because the slow reaction leads to severe bundling and a low degree of branching of the formed fibers, thereby leading to a poor network structure.[6] In the rheometer, this mixture did show the formation of a gel ($G' > G''$) with a storage modulus in the kPa range, an inconsistency that is often observed in gel systems and can be attributed to the different length scales of the experimental setups.[18] Next, the performance of PAH as a switchable acid catalyst was investigated. Irradiating aqueous solutions of **1**, **2**, and PAH (1 mM, pH^{irr} 5.8) afforded a pale yellow stable gel (Figure 2a). In the absence of irradiation (pH 6.7), the same mixture still resulted in a gel (Figure 2b), however, this gel appeared much weaker, as was confirmed by rheology (Figure 2d). Light irradiation also resulted in reduction of the gelation time, which changed from 200 min for the non-irradiated PAH sample to 100 min for the light-irradiated PAH sample, while the gelation time for the sample without PAH was 350 min. These results indicate that the rate-determining step in the gelation process is the formation of gelator molecule **3**. PAH-catalyzed gels quickly (5–10 min) regained their original pH value (pH 6.6) upon removing irradiation, as the spiropyran form of PAH reverted to the ring-opened merocyanine form through the thermally activated back reaction.

Distinct rheological differences were observed between the irradiated and non-irradiated samples (Figure 2d). Irradiation-triggered gelation of the PAH samples ([**1**]/[**2**]/[PAH] = 16:96:1, at millimolar concentrations)

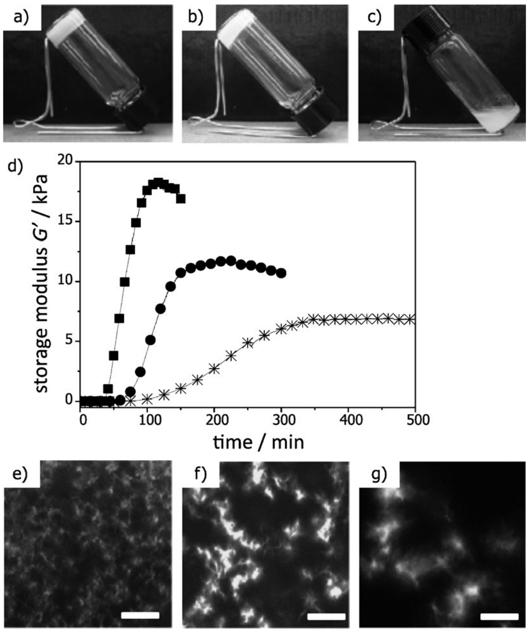

Figure 2. Light controlled gelation. Gelation a) with PAH (1 mM) after visible light irradiation, b) with PAH (1 mM) in the dark, and c) without PAH in the dark. d) Gel formation measured by rheology: the evolution of G′over time (■ PAH, light; ● PAH, dark; ⋆ no PAH, dark). CLSFM of network morphology (scale bar: 10 μm): e) with PAH after visible light irradiation, f) with PAH in the dark, and g) in the dark without PAH. General conditions: [**1**]/[**2**] = 16:96 at millimolar concentrations). An aldehyde-functionalized fluorescein fluorescent probe was used in the CLSFM experiments.[15]

resulted in an increase of the maximum obtained storage modulus (G') to 18.5 kPa, where without irradiation, the PAH-sample reached a 11.7 kPa G'. The sample without PAH only reached a 6.9 kPa G'. A control experiment in which a sample without PAH was irradiated did not show an increased G' compared to the non-irradiated sample, thus ruling out any other irradiation-induced effects, such as heating of the sample. Using confocal laser scanning fluorescence microscopy (CLSFM), we analyzed the resulting fiber morphology. The irradiated PAH sample showed the formation of dense networks made up of heavily branched interconnecting fibers (Figure 2 e). The non-irradiated PAH sample consisted of branched but poorly connected bundles of fibers (Figure 2 f), whereas the system without PAH was made up of isolated fiber structures (Figure 2 g). These results show that the rate of gelator formation, and with that the structure and mechanical properties of the assembled materials, can be controlled by using light-triggered catalysis.

Being able to control the rate of gel formation by using a light-triggered catalyst, we then focused on patterning gels by locally changing the reaction rates. Irradiation of a partially covered vial only gave gelation in the irradiated area. Smaller, repetitive gel patterns were obtained by irradiating a gelation solution covered by a mask fabricated by using conventional laser printing on a transparency (Figure 3 a–d). In an imaging chamber (0.6 mm thickness), a solution of **1**, **2**, and PAH was irradiated with visible light ($400 < \lambda < 800$ nm) through the mask for 100 min to yield a gel pattern following the dimensions and shape of the mask (Figure 3). Using this method, we were able to fabricate diverse shapes (squares, circles, hexagons, and letters) with variable spacing, as confirmed by CLSFM (Figure 3). For instance, when using a mask with 500 × 500 µm square holes, spaced 500 µm apart, we obtained similarly sized square gel structures (490 ± 16) µm (Figure 3 a,e,i and the Supporting Information) spanning the entire chamber as pillars. As in the bulk system (see above), a branched interconnected fibrous network was observed in the irradiated regions of the pattern, while there was a poorly connected network in the non-irradiated regions of the pattern (see the Supporting Information). We then used CLSFM to investigate possible mass transfer across the sample. After irradiation, samples were allowed to react further in the dark for an additional 12 h to ensure complete conversion.[6a] The fluorescence intensity of the bound fluorescent probe can then be taken as a measure of local fiber density. From measured intensity differences between irradiated and dark regions, we calculated a more than 50 % degree of mass transfer from the non-irradiated region to the light-exposed region during photopatterning (see the Supporting Information). This observation supports the concept of generating photo-induced concentration gradients between light and dark zones through locally changing reaction rates by using the photoswitchable PAH catalyst. The large difference in time scale between the switching process (forward and back) and the gelation process is instrumental in controllably achieving the observed spatial resolution, since the catalyst will quickly and spontaneously deactivate in the non-irradiated zones.

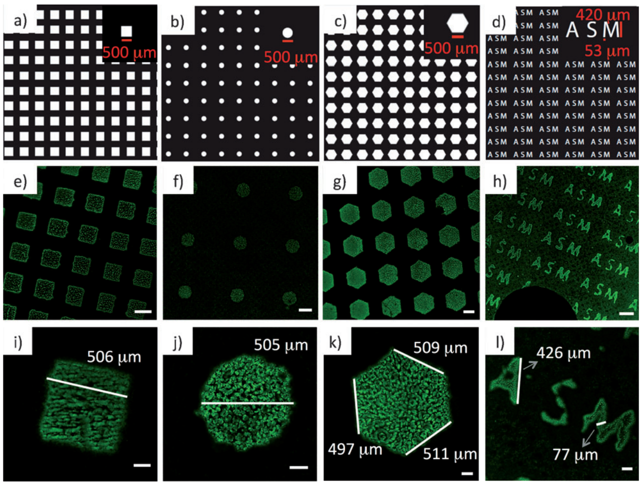

Figure 3. Light-triggered gel patterning: a–d) mask layouts. Inset: a single pore of each mask. e–h) CLSFM of gel patterns formed after irradiation through the mask. Scale bar: 500 µm. i–l) shape and size of the pattern features. Scale bar: 100 µm. General conditions: ([**1**]/[**2**]/[PAH] = 16:96:1 at millimolar concentrations).

In conclusion, we have shown how, by using a visible light triggered catalyst, it is possible to control the rate of formation and the mechanical properties of a low-molecular-weight hydrogel, as well as its distribution in space. A photochromic switch, capable of reversibly releasing a proton upon irradiation, can act as a catalyst for hydrazone formation between otherwise soluble building blocks, thereby leading to fiber formation and gelation in water. By using a photomask, spatially structured hydrogels were formed starting from isotropic bulk solutions of small molecule gelator precursors. The use of a reversible photochromic switch allows a return to ambient pH value after irradiation. The concept of light-controlled catalytic pattern formation opens up the possibility of using spatially controlled self-assembly in the future to make patterned channels of gels for use in microfluidics,[19] biosensors[20] and synthetic biomaterials;[21] as structured microgels[22] for use in tissue engineering;[23] or as stimulus-responsive materials.[24]

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[1] a) M. de Loos, B. L. Feringa, J. H. van Esch, *Eur. J. Org. Chem.* **2005**, 3615–3631; b) N. M. Sangeetha, U. Maitra, *Chem. Soc. Rev.* **2005**, *34*, 821–836.
[2] M. He, J. Li, S. Tan, R. Wang, Y. Zhang, *J. Am. Chem. Soc.* **2013**, *135*, 18718–18721.
[3] J. Boekhoven, M. Koot, T. A. Wezendonk, R. Eelkema, J. H. van Esch, *J. Am. Chem. Soc.* **2012**, *134*, 12908–12911.
[4] L. Ionov, *Adv. Funct. Mater.* **2013**, *23*, 4555–4570.
[5] a) I. A. Eydelnant, B. B. Li, A. R. Wheeler, *Nat. Commun.* *5*, 3355; b) C. A. DeForest, K. S. Anseth, *Nat. Chem.* **2011**, *3*, 925–931; c) D. J. Cornwell, B. O. Okesola, D. K. Smith, *Angew. Chem.*

Int. Ed. **2014**, *53*, 12461—12465; *Angew. Chem.* **2014**, *126*, 12669—12673; .

[6] a) J. Boekhoven, J. M. Poolman, C. Maity, F. Li, L. van der Mee, C. B. Minkenberg, E. Mendes, J. H. van Esch, R. Eelkema, *Nat. Chem.* **2013**, *5*, 433 – 437; b) R. Eelkema, J. H. van Esch, *Org. Biomol. Chem.* **2014**, *12*, 6292 – 6296.

[7] R. J. Williams, A. M. Smith, R. Collins, N. Hodson, A. K. Das, R. V. Ulijn, *Nat. Nanotechnol.* **2009**, *4*, 19 – 24.

[8] A. G. L. Olive, N. H. Abdullah, I. Ziemecka, E. Mendes, R. Eelkema, J. H. van Esch, *Angew. Chem. Int. Ed.* **2014**, *53*, 4132 – 4136; *Angew. Chem.* **2014**, *126*, 4216 – 4220.

[9] V. T. Bhat, A. M. Caniard, T. Luksch, R. Brenk, D. J. Campopiano, M. F. Greaney, *Nat. Chem.* **2010**, *2*, 490 – 497.

[10] R. S. Stoll, S. Hecht, *Angew. Chem. Int. Ed.* **2010**, *49*, 5054 – 5075; *Angew. Chem.* **2010**, *122*, 5176 – 5200.

[11] a) J. J. D. de Jong, L. N. Lucas, R. M. Kellogg, J. H. van Esch, B. L. Feringa, *Science* **2004**, *304*, 278 – 281; b) J. J. D. de Jong, P. R. Hania, A. Pugzlys, L. N. Lucas, M. de Loos, R. M. Kellogg, B. L. Feringa, K. Duppen, J. H. van Esch, *Angew. Chem. Int. Ed.* **2005**, *44*, 2373 – 2376; *Angew. Chem.* **2005**, *117*, 2425 – 2428; c) S.-C. Wei, M. Pan, K. Li, S. Wang, J. Zhang, C.-Y. Su, *Adv. Mater.* **2014**, *26*, 2072 – 2077; d) J. K. Sahoo, S. K. M. Nalluri, N. Javid, H. Webb, R. V. Ulijn, *Chem. Commun.* **2014**, *50*, 5462 – 5464.

[12] T. Muraoka, C. Y. Koh, H. Cui, S. I. Stupp, *Angew. Chem. Int. Ed.* **2009**, *48*, 5946 – 5949; *Angew. Chem.* **2009**, *121*, 6060 – 6063.

[13] a) J. Raeburn, T. O. McDonald, D. J. Adams, *Chem. Commun.* **2012**, *48*, 9355 – 9357; b) V. Javvaji, A. G. Baradwaj, G. F. Payne, S. R. Raghavan, *Langmuir* **2011**, *27*, 12591 – 12596.

[14] Z. Shi, P. Peng, D. Strohecker, Y. Liao, *J. Am. Chem. Soc.* **2011**, *133*, 14699 – 14703.

[15] J. M. Poolman, J. Boekhoven, A. Besselink, A. G. L. Olive, J. H. van Esch, R. Eelkema, *Nat. Protoc.* **2014**, *9*, 977 – 988.

[16] R. Klajn, *Chem. Soc. Rev.* **2014**, *43*, 148 – 184.

[17] a) C. Tang, A. M. Smith, R. F. Collins, R. V. Ulijn, A. Saiani, *Langmuir* **2009**, *25*, 9447 – 9453; b) R. C. T. Howe, A. P. Smalley, A. P. M. Guttenplan, M. W. R. Doggett, M. D. Eddleston, J. C. Tan, G. O. Lloyd, *Chem. Commun.* **2013**, *49*, 4268 – 4270; c) D. J. Cornwell, B. O. Okesola, D. K. Smith, *Soft Matter* **2013**, *9*, 8730 – 8736.

[18] P. Terech, C. Rossat, F. Volino, *J. Colloid Interface Sci.* **2000**, *227*, 363 – 370.

[19] D. Steinhilber, T. Rossow, S. Wedepohl, F. Paulus, S. Seiffert, R. Haag, *Angew. Chem. Int. Ed.* **2013**, *52*, 13538 – 13543; *Angew. Chem.* **2013**, *125*, 13780 – 13785.

[20] O. Andersson, A. Larsson, T. Ekblad, B. Liedberg, *Biomacromolecules* **2009**, *10*, 142 – 148.

[21] a) I.-C. Liao, F. T. Moutos, B. T. Estes, X. Zhao, F. Guilak, *Adv. Funct. Mater.* **2013**, *23*, 5833 – 5839; b) A. Mata, L. Hsu, R. Capito, C. Aparicio, K. Henrikson, S. I. Stupp, *Soft Matter* **2009**, *5*, 1228 – 1236.

[22] M. Zourob, J. E. Gough, R. V. Ulijn, *Adv. Mater.* **2006**, *18*, 655 – 659.

[23] A. Khademhosseini, R. Langer, J. Borenstein, J. P. Vacanti, *Proc. Natl. Acad. Sci. USA* **2006**, *103*, 2480 – 2487.

[24] X. He, M. Aizenberg, O. Kuksenok, L. D. Zarzar, A. Shastri, A. C. Balazs, J. Aizenberg, *Nature* **2012**, *487*, 214 – 218.