

ARTICLE

Photolabile ROMP Gels Using *ortho*-Nitrobenzyl Functionalized Crosslinkers

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This paper describes a series of four *ortho*-nitrobenzyl substituted bis(norbornene) crosslinkers that are suitable for the preparation of photoresponsive organogels and hydrogels through ring-opening metathesis polymerization. As measured by several techniques—visual inspection, rheology, and release of network-bound polymers—organogels prepared using these four crosslinkers varied in their sensitivity to UV irradiation by about two orders of magnitude. The reactivity of the gels show qualitative correlation with the stability of the intermediate benzylic radicals. Gels with larger crosslink densities required longer UV irradiation time to dissolve than gels with smaller crosslink densities. Together, these results demonstrate how rational changes to the structures of crosslinkers and gels can tune photosensitivity. Finally, we show a proof-of-concept photochemical release experiment of a physically entrapped fluorescent polymer from a photolabile ROMP hydrogel.

Introduction

Stimuli-degradable gels are a promising material platform for a number of applications.¹ A common strategy for achieving on-demand dissolution of gels is to design the physical or chemical crosslinks that hold the polymeric network together to break upon introduction of a specific stimulus. Examples of stimuli that other groups have reported to induce the degradation of crosslinks include enzymatic catalysis, temperature, pH, and light.^{2–9} The use of light to induce gel degradation has a number of advantages, such as the ability to pattern gels spatially,^{10–12} and using the different photosensitivities of different photoreactive groups to release multiple trapped payloads sequentially, or perform other functions.^{13–17} Drug delivery systems, which release payloads trapped within the gel upon application of a stimulus, are an example of an application of light-responsive gels finding significant interest.^{5,18} These materials also show promise as tunable and degradable scaffolds for tissue engineering and for culturing and releasing cells.^{15,19–21}

Although a number of photocleavable moieties, such as coumarins, have found application in the design of light-degradable crosslinkers,²² the *o*-nitrobenzyl (ONB) group remains a highly popular choice for integration into photocleavable linkers.^{21,23,24} The ONB group has the following advantages: 1) it is readily installed and functionalized, 2) the radical mechanism of photocleavage does not require the presence of a highly polar solvent to stabilize ionic intermediates, and 3) the variety of functional groups that ONB groups can reveal upon photolysis.²⁴ In an important step in understanding structure-property relationships of this

photocleavable moiety, a recent paper by Bally, Bochet, and coworkers showed that the quantum yields of photolyses of ONB groups correlated inversely with the strength of the benzylic C-H bond, with an ONB ether showing the largest quantum yield of all compounds tested.²⁵ We are interested in understanding how these types of changes in ONB structure, and therefore photolytic efficiency, translate into controlling new light-responsive materials. Kasko and coworkers used differences in reactivity of different ONB esters, including methylating the benzylic positions and varying the number and regiochemistry of alkoxy substituents on the nitroarene, to demonstrate staged release of two different populations of stem cells,¹⁵ and have also developed a library of ONB-macromers for incorporation into gels prepared by vinyl polymerization.²⁶ In addition, our group has demonstrated how the structures of ONB groups, including ONB ethers, dictate the photoinduced reactivity of conjugated oligomers and polymers with photolabile solubilizing chains.²⁷

Our group has also been interested in the use of ring-opening metathesis polymerization (ROMP) for the construction of functional, photoreactive materials.^{28–31} Commercially available ruthenium-based catalysts for ROMP have the advantages of wide functional group tolerance, polymerization at room temperature, the capacity for living and controlled polymerizations, and efficacy in the presence of air and water.³² Using ROMP, our group has demonstrated the preparation of ONB-containing block copolymers,³⁰ photodegradable layer-by-layer films,²⁸ and photochromic random copolymers that show reversible switching of the sign of static charging.²⁹ Following the logic of studies that have

demonstrated that molecules bearing more than one norbornene unit could yield insoluble polymer networks using ROMP reactions,³³⁻³⁶ we also published a preliminary study that reported crosslinkers comprising two norbornene units bound together through photolabile ONB esters: both organogels and hydrogels prepared using these crosslinkers dissolved upon irradiation with UV light, with those gels using secondary ONB esters with benzylic methyl groups yielding gels that required shorter irradiation times to dissolve than those with primary ONB esters.³¹ Herein we report a detailed study of this approach centered on the effect of ONB structure on the efficiency of photodisruption of gels prepared by ROMP as measured by the release of covalently bound fluorophores into solution, light-induced changes in viscoelastic properties, and the release of trapped fluorescent guests.

RESULTS AND DISCUSSION

Synthesis of Crosslinkers

Chart 1 shows the structures of the four ONB-containing bis(norbornene) crosslinkers we designed and prepared in this study. The key differences between these molecules are the structures of the ONB units, which are either: ONB esters (**CL-1**), ONB esters methylated in the benzylic positions (**CL-2**), ONB ethers (**CL-3**), or ONB ethers methylated in the benzylic positions (**CL-4**). Both methylation and substitution of electron donating ethers on the benzylic positions are designed to stabilize the benzylic radical intermediate of the first step of ONB photolysis. Our expectation in this study was that either of these two modifications would increase the photoinduced sensitivity of the resulting gels, and that combining these modifications (as in **CL-4**) would lead to gels with the fastest response to UV light.

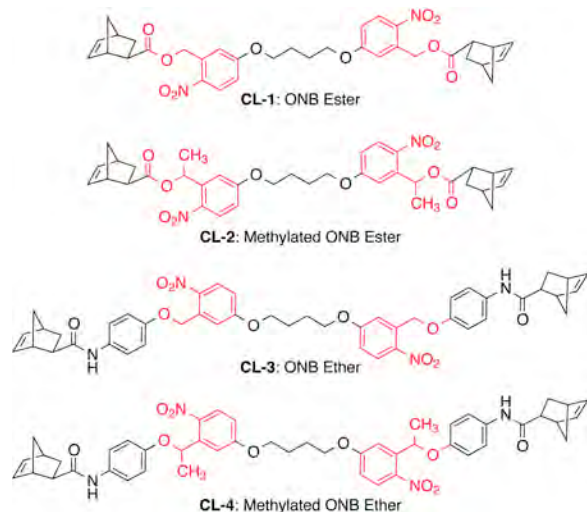
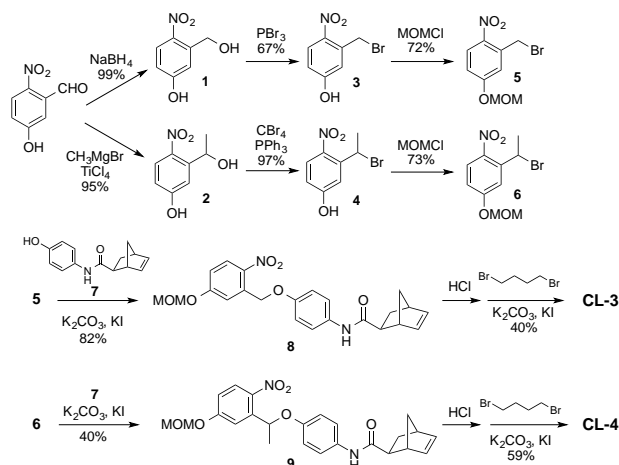


Chart 1: Structures of photolabile ROMP crosslinkers. The different photosensitive ONB moieties are highlighted in red.

Crosslinkers **CL-1** and **CL-2** were available from our previously published study on photoreactive ROMP gels.³¹ Preparations of ONB ether crosslinkers **CL-3** and **CL-4**

followed similar paths, with reduction of 5-hydroxy-2-nitrobenzaldehyde with either NaBH_4 or $\text{CH}_3\text{MgBr}/\text{TiCl}_4$ followed by bromination to yield the corresponding primary or secondary bromides **3** and **4**. Protection of the phenols with chloromethyl methyl ether (MOMCl) followed by substitution of the bromides with the phenol-functionalized norbornene **7** yielded compounds **8** and **9**. MOM deprotection followed by alkylation with 1,4-dibromobutane yielded the target ONB ether crosslinkers. The UV/vis absorbance spectra of all crosslinkers in tetrahydrofuran showed absorbance maxima at approximately 300 nm with molar extinction coefficients of $\sim 2 \times 10^4 \text{ M}^{-1}\text{cm}^{-1}$, with absorbance confined to the ultraviolet region of the spectrum.



Scheme 1: Synthesis of ONB ether-containing bis(norbornene) crosslinkers **CL-3** and **CL-4**.

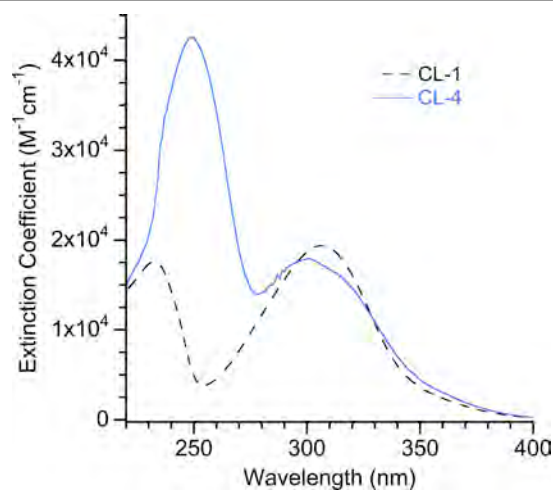
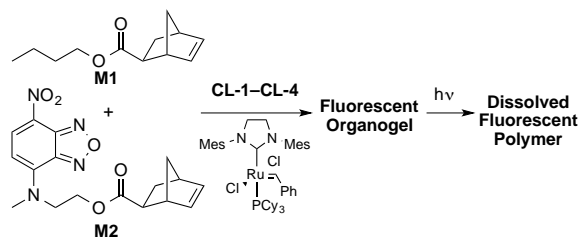


Figure 1: UV/vis spectrum of ONB ester crosslinker **CL-1** and methylated ONB ether crosslinker **CL-4** in tetrahydrofuran.

Preparation of Organogels:

All the organogels reported herein, regardless of crosslinker structure, were prepared using the same procedure (Scheme 2): (1) dissolution of 40 mg of ROMP monomer **M1** and 0.5 mg fluorescent ROMP monomer **M2** containing the nitrobenzofurazan (NBD) dye, together with the crosslinker of

interest (**CL1-CL4**), into 0.2 mL DMF, (2) dissolution of the Grubbs 2nd generation ruthenium carbene catalyst into 0.2 mL DCM, and (3) mixing the two solutions together. The final concentrations of monomer (0.51 M) and catalyst (0.007 M) in the mixed solutions were the same in all polymerizations. Under these conditions, all four of the crosslinkers yielded gels within 30s. To study the effect of the gel composition on the properties of the photoresponsive gels (*vide infra*), we prepared gels with molar ratios of crosslinker:catalyst of 1.3 and 1.5. We purified all gels using Soxhlet extraction with dichloromethane for two days to remove soluble polymer chains and monomers. As expected, gels prepared from crosslinker **CL-4** had a higher storage modulus (45 Pa) when the ratio of crosslinker:catalyst molar ratio was 1.5 than gels prepared with a ratio of 1.3 (27 Pa). We determined that the somewhat different structures of the ether and ester-based crosslinkers did not affect the ratio of crosslinker to monomer incorporated into the gel: the yield of dry xerogel for gels prepared with **CL-2** and **CL-4** were the same, within experimental error (89-91%), and ¹H NMR analysis of the soluble fractions isolated from Soxhlet extraction showed no evidence of either of the crosslinkers. Further evidence for similar crosslink densities for gels prepared with either ONB-ether and ONB-ester crosslinkers is that their gravimetric swelling ratios— 22 ± 1 for gel prepared with **CL-2**, 20 ± 2 for gel prepared with **CL-4** at crosslinker:catalyst ratio of 1.3—are not distinguishable within experimental error.



Scheme 2: Overall design of photoresponsive gels prepared using crosslinkers **CL-1** – **CL-4**. Fluorescent monomer **M2** was used in gels at a loading of 1.4 mole percent relative to **M1**.

Photodegradation of Gels

We used several approaches to characterize how the structure of the crosslinker and the crosslinker:catalyst ratio affects the sensitivities of gels to UV light; as a function of irradiation time, we visually monitored the presence of the gel in solution and tracked the release of a fluorescent dye covalently attached to a norbornene monomer (**M2**) that was incorporated into the gels. In these experiments, we irradiated gels with a 200 W Hg/Xe lamp equipped with a BG40 bandpass filter to induce the degradation of gels. The power density incident on samples in these experiments was 25 mW/cm². We also measured the viscoelastic properties of several gels as a function of irradiation time with a portable UV lamp emitting 365 nm with power density incident on samples of 9 mW/cm². In all examples, leaving gels in the dark in parallel with our

irradiation experiments did not result in any significant degradation as determined by any of our three approaches.

VISUAL OBSERVATION OF GEL DISSOLUTION. This approach, in which we note the time of irradiation required for the gel to no longer be visible to the naked eye in benzene, although imprecise, is a quick, low-tech, and direct method for identifying large differences in photosensitivity between gels of different composition. We used this simple method in our initial communication on this subject to establish that gels prepared with **CL-2** are more reactive than gels prepared with **CL-1**. Figure 2 summarizes the irradiation time required for dissolution of ~ 2 mg samples of ROMP organogels prepared with a 1.3 crosslinker:catalyst molar ratio with each of the four different ONB crosslinkers. The most reactive gels are those prepared with the methylated ether ONB crosslinker, **CL-4**, which combines the radical stabilizing features of a tertiary benzylic radical with an electron donating ether functional group, and dissolve in about 10 minutes.

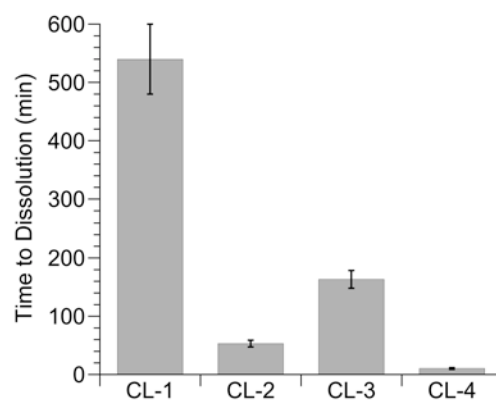


Figure 2. UV irradiation time required to dissolve 2 mg of organogel (prepared using a 1.3 molar ratio of crosslinker to catalyst as shown in Scheme 2) in benzene. Error bars are the standard deviations of three measurements for each crosslinker.

MONITORING DISSOLUTION OF FLUORESCENT POLYMER CHAINS.

As a number of potential applications of photolabile gels focus on light causing the release of guests, we developed a simple method to follow the release of solubilized polymer chains released from the insoluble polymer network upon UV irradiation. This approach uses a small percentage of norbornene monomer (**M2**) labeled with a fluorophore—nitrobenzoxadiazole (NBD) in this case, see Scheme 2—in the synthesis of gels; only upon breaking the ONB crosslinks with UV light can these fluorophores, as part of soluble polymer chain photoproducts, be released into solution. The fluorescence intensity of the NBD dye in the fluid solution as a function of irradiation time reflects the quantity of soluble polymer chains released by photolysis. In these experiments, we used approximately the same mass (2 mg) of dry xerogel swollen in 3 mL of benzene, and irradiated all gels under the conditions described above. The fluorescent side-chains, although they compete for absorbance of UV photons with the photocleavable ONB groups (See ESI Figure S4 for UV/vis

absorbance spectrum), the molar ratio of monomer **M2** in all polymerizations was the same, such that any differences in competitive absorbance between gels prepared with different crosslinkers are small.

Figure 3 summarizes the results of these experiments for gels prepared using a 1.3 crosslinker:catalyst ratio and the four different ONB-containing crosslinkers. In all experiments, the quantity of NBD present in solution before UV irradiation of the gels, as detected by fluorescence, was not distinguishable from background. Fluorescence from the NBD, however, increases with irradiation of the gel suspended in benzene at $\lambda > 320$ nm, consistent with UV-induced dissolution of the gel. In all cases, the intensity of fluorescence from the probe reaches a plateau at durations of irradiation generally consistent with the time required for dissolution of the gel as determined visually; to compare the reactivity of different gels, we set the average integrated fluorescence intensity of the last three data points, corresponding to this plateau in each experiment, to 1.0.

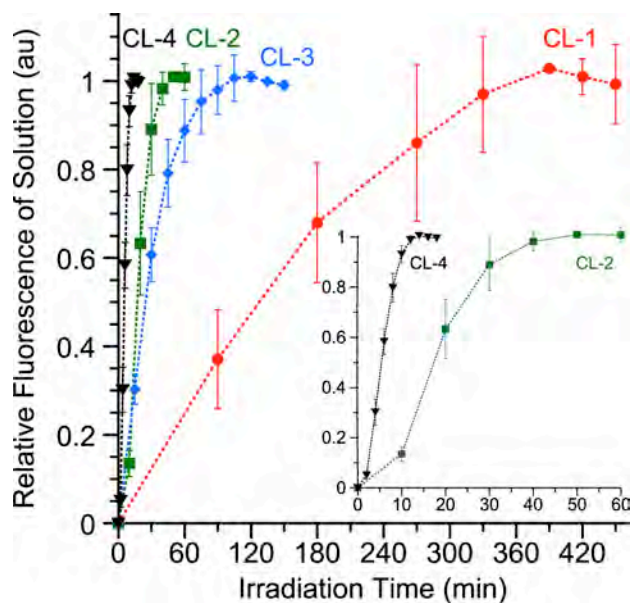


Figure 3: The dependence of fluorescence intensity of benzene-soluble portion of NBD-containing ROMP gels crosslinked with each of the ONB crosslinkers on duration of UV irradiation using a BG40 bandpass filter. In each experiment, we used 2 mg of dry xerogel. The wavelength of excitation for fluorescence spectroscopy was 465 nm, while the fluorescence intensities were determined by integrating between $\lambda_{em} = 500$ nm and $\lambda_{em} = 550$ nm. The data points are the average of three measurements of relative fluorescence intensity in separate irradiation experiments; the error bars represent standard deviations of these measurements.

The order of reactivity of the gels prepared with different crosslinkers, determined using fluorescence spectroscopy, was the same as determined by simple visual inspection: gels prepared with **CL-4**, the methylated ONB ether crosslinker, were most reactive, releasing all fluorophores into solution within 10 minutes, while gels with **CL-1**, the unmethylated ONB ester crosslinker, were the least reactive, taking up to 6 hours to reach a plateau of fluorescence intensity of soluble fractions; even after this protracted time of irradiation, some quantity of **CL-1** gel still remained insoluble. As negative

control experiments, identical samples of gels that were not exposed to UV light showed no fluorescent signal discernible from background. This ordering of reactivity is consistent with our initial hypothesis, as combining both i) ether-for-ester substitution and ii) increasing the number of alkyl groups on the benzylic position stabilize the benzylic radical intermediate, increasing the efficiency of photolysis more than either of these two substitutions in isolation.

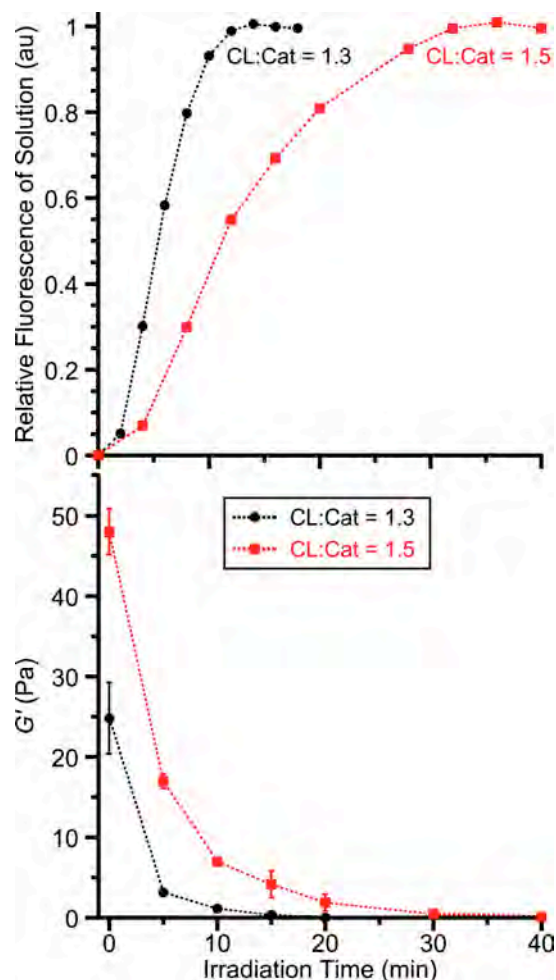


Figure 4. Dependence of dissolution of bound fluorescent polymers (a) and storage modulus (b) of gels on ratio of crosslinker (**CL-4**) to catalyst.

THE EFFECT OF CROSSLINK DENSITY ON GEL PHOTOREACTIVITY:

To characterize the effect of crosslink density on the sensitivity of these gels to UV light, we compared the photoinduced dissolution of gels prepared with the most reactive crosslinker, **CL-4**, with crosslinker:catalyst molar ratios of 1.3 and 1.5. Figure 4 shows the results of two experiments designed for this purpose, which in both cases conformed to our expectations that the gels with larger concentrations of crosslinkers take longer to dissolve than those with smaller concentrations. When we monitored the release of soluble NBD-labelled soluble polymer chains upon ONB photolysis using fluorescence spectroscopy, fluorescence of the solutions containing more highly crosslinked gels took 2-3

times longer to reach a plateau than the solutions containing less highly crosslinked gels (Figure 4a). As a complement to this approach, we also determined the storage moduli of gels with different concentrations of crosslinkers as a function of UV irradiation (Figure 4b). As expected, the gels with larger crosslink densities showed larger storage moduli and required more time of UV irradiation to approach storage moduli of zero than gels with smaller crosslink densities.

Photoactivated Release of Fluorescent Guest From Hydrogel

To demonstrate the potential utility of these materials in aqueous environments, we also prepared photoresponsive ROMP hydrogels using a monomer previously reported by Tew and coworkers (**M3**),³³ and shown to be effective in our preliminary report on this class of photoresponsive gels.³¹ To prepare these gels, we used a procedure similar to the procedure for organogel preparation (*vide supra*). We first combined i) hydrophilic monomer **M3**, ii) crosslinker **CL-4**, and iii) a fluorescently-labeled random copolymer of poly(ethylene glycol)methyl ether methacrylate and 7-nitro-2,1,3-benzoxadiazole methacrylate (NBDMA) in dichloromethane, then to this solution added Grubbs second generation ruthenium carbene catalyst to yield a crosslinker:catalyst molar ratio of 1.5. The gel that resulted after approximately 30 minutes was purified by Soxhlet extraction, resulting in a fluorescent hydrogel due to physically trapped polymer chains bearing NBD fluorophores as pendants. This hydrogel was photolabile, as photolysis with UV light using irradiation conditions identical to those used with the organogels (*vide supra*) in water resulted in the dissolution of the gel and release of the fluorescent polymeric guest (Figure 5).

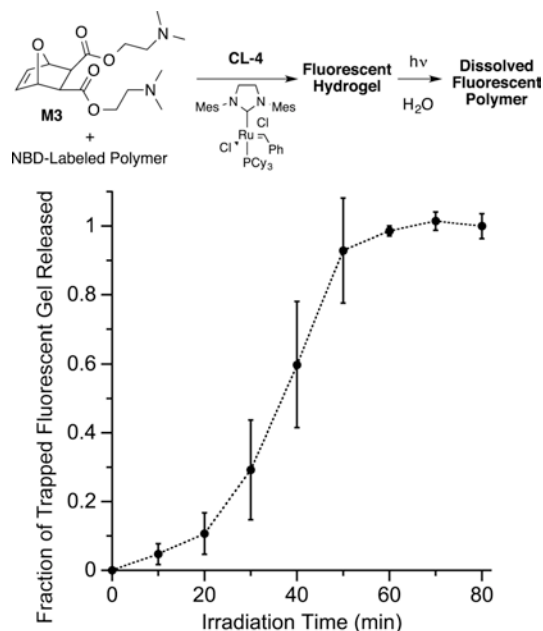


Figure 5. UV-induced release of fluorescent NBD-labeled methacrylic polymer from hydrogel prepared with hydrophilic monomer **M3** and crosslinker **CL-4**.

CONCLUSION

This work combines the structurally tunable nature of the ONB photocleavable groups, the photolysis of which that can occur in many environments, with advantages of ROMP—simple protocols, fast polymerization rates, and high functional group tolerance—for the preparation of photosensitive organogels and hydrogels. Work described in this paper also expands the repertoire of photolabile crosslinkers available for photoresponsive gels. The dependence of gel photosensitivity on crosslinker structure is consistent with the previously determined correlation of stability of the intermediate benzylic radical of the mechanism of nitrobenzyl group cleavage on the quantum yield of photolysis. Gels prepared with ONB methyl ether crosslinkers showed the fastest response of all samples investigated, highlighting the potential of this substitution pattern on photocleavable groups in this and other platforms. Photolabile ROMP hydrogels also allow for light-induced release of physically trapped guests. Finally, our approach opens up the possibility of combining different photoreactive moieties in ROMP gels to achieve wavelength-selective photoresponsive properties.³⁷

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Notes and References

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