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Covalently Crosslinked Diels–Alder Polymer Networks

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Abstract

This project examines the utility of cycloaddition reactions for the synthesis of polymer networks. Cycloaddition reactions are desirable because they produce no unwanted side reactions or small molecules, allowing for the formation of high molecular weight species and glassy crosslinked networks. Both the Diels–Alder reaction and the copper-catalyzed azide–alkyne cycloaddition (CuAAC) were studied. Accomplishments include externally triggered healing of a thermoreversible covalent network via self-limited hysteresis heating, the creation of Diels–Alder based photoresists, and the successful photochemical catalysis of CuAAC as an alternative to the use of ascorbic acid for the generation of Cu(I) in click reactions. An analysis of the results reveals that these new methods offer the promise of efficiently creating robust, high molecular weight species and delicate three dimensional structures that incorporate chemical functionality in the patterned material. This work was performed under a Strategic Partnerships LDRD during FY10 and FY11 as part of a Sandia National Laboratories/University of Colorado–Boulder Excellence in Science and Engineering Fellowship awarded to Brian J. Adzima, a graduate student at UC-Boulder. Benjamin J. Anderson (Org. 1833) was the Sandia National Laboratories point-of-contact for this fellowship.

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1. Motivation

Cycloadditions are a broad class of reactions where unsaturated species combine to form a cyclic adduct.¹ Cycloadditions can be *click reactions* that produce high yields, tolerate other functional groups, and utilize simple reaction conditions. Unlike addition reactions ambient water and oxygen do not cause unwanted side reactions. Furthermore, unlike condensation reactions, small molecules are not produced, allowing for the formation of high molecular weight species and glassy crosslinked networks. Despite such inherent advantages and widespread use in small molecule synthesis,² cycloadditions have been seldom utilized by the polymer community as evidenced by their near or complete omission in the tomes of the field,^{3,4} and their rare use in commercial products.⁵ The unique reversibility of the Diels–Alder reaction received little mention until 2002,⁶ and the Huisgen cycloaddition was not widely utilized until the discovery of its copper catalysis in the same year.^{7,8}

In this project the utility of cycloaddition reactions for the synthesis of polymer networks was examined. Both the Diels–Alder reaction and the copper-catalyzed azide–alkyne cycloaddition (CuAAC) were studied. Spectroscopic experiments were conducted to demonstrate these reactions, and explore their mechanisms, while mechanical measurements examined the relationship between the underlying chemical reactions and the material properties. Both reactions were utilized in the fabrication of functional materials owing to efficient bond formation and tolerance of other functional groups, which allowed their modular use in a number of potential applications.

2. Accomplishments

2.1 Externally Triggered Healing of a Thermoreversible Covalent Network via Self-limited Hysteresis Heating

Previously, we had synthesized a reversibly crosslinked polymer network using the Diels–Alder reaction and demonstrated that upon heating the material readily reverted to a liquid composed of the initial monomers.⁹ While this material showed promise as a thermo-reversible adhesive and a self-healing material, remotely and rapidly heating optically thick and thermally sensitive materials is a non-trivial problem. Hysteresis heating,¹⁰ a process where an alternating magnetic field is used to heat magnetically susceptible materials, was evaluated as a method for heating, depolymerizing, and healing the material. Unlike other approaches such as photothermal particle heating¹¹ hysteresis heating allows for heating in optically thick samples such as composites and adhesives between opaque substrates. Chromium (IV) oxide particles were used as magnetic susceptor and were easily incorporated into the monomer mixture before polymerization. Upon activation of the radio frequency field generator heating occurred rapidly. Importantly, this process is inherently self-limiting, and ceases once the particles reach the Curie temperature of chromium (IV) oxide, approximately, 113 °C. This behavior prevented the material from heating to a temperature where destructive decomposition could occur via oxidative reactions. It was demonstrated that the material could be fractured and healed at least ten times with no change in either the modulus or ultimate strength. Healing was rapid, requiring only 100 seconds exposure to the field. Furthermore, while the full value of the strength took 12 hours to recover, a significant portion was recovered within 10 minutes. More details are included in [12].

2.2 Diels–Alder Based Photoresists

Typical photoresists undergo a change in solubility upon irradiation. This occurs either by polymerization in the case of negative photoresists, or by scission of polymer bonds in the case of some positive photoresists, such as polymethacrylate. Using the previously described Diels–Alder based polymer networks, we developed a photoresist based on the selective elimination of bond reversibility, and demonstrated its utility in 3D microstereolithography. In this approach selective irradiation is used to render the network structure permanent, or *fixed*, like any other thermoset. The unexposed regions retain their reversible behavior, and are later removed upon heating to produce an image. After pattern transfer the non-irradiated material is easily removed by the reversible nature of the chemical crosslinking. A chief advantage of this approach is that the reversible polymer matrix supports the fabrication of delicate three dimensional structures, such as logpile-type photonic crystal templates, during fabrication. This allows for the fabrication of structures not easily attainable by conventional methods. Photofixation also provides a method to eliminate thermoreversible behavior within reversible adhesives, self-healing materials,⁶ non-linear optics,¹³ and polymer encapsulates.¹⁴ Furthermore, this approach also allows for the incorporation of chemical functionality within the patterned material, and may eventually eliminate the need for a developing solvent.

Specifically, a thermoreversible crosslinked polymer network is formed by a Diels–Alder reaction between furan and maleimide-based monomers. Thiol and photoinitiating species are readily incorporated into the initial mixture, and upon irradiation the Diels–Alder adduct undergoes a thiol-ene reaction, rendering the bond irreversible. Release of the patterned structure in the exposed region is achieved by reversion of the remaining reversible crosslinks to monomer via the retro- Diels–Alder reaction. A manuscript is currently in preparation describing this process [15].

Figure 1. A logpile type structure formed of alternating layers of 10 μm beams. Fabrication of this complex structure was accomplished using two-photon direct laser writing in conjunction with a photofixation based photoresist.

2.3 Investigation of the Photochemical Catalysis of the Copper Catalyzed Azide–Alkyne Cycloaddition

The click reaction philosophy emphasizes reactions that are efficient and modular.¹⁶ In material science these attributes are desirable because they eliminate the difficult separation of macromolecular products, allow for the synthesis of highly functionalized materials, and permit the incorporation of reactions across different materials. While photochemistry has not been emphasized in the click chemistry literature, the development of photochemical click reactions offers the promise of efficient and modular reactions that can be spatially and temporally controlled, further improving their utility.

The thermal cycloaddition of azides and alkynes is typically sluggish; however, in 2002 it was discovered that the addition of Cu(I) accelerated the reaction rate by a factor 10^7 .^{7,8} Since then the copper catalyzed alkyne–azide cycloaddition (CuAAC) has become the preeminent click reaction, demonstrated by its use in a diverse assortment of materials.¹⁷ Most commonly, the Cu(I) catalyst is formed by the reduction of Cu(II) to Cu(I), using sodium ascorbate. As the generation of the catalyst is a bulk reaction, spatial control of the reaction is limited to techniques such as dip pen lithography and micro contact printing.^{18,19} These techniques are inherently limited to surfaces and lack the scope of more traditional photolithographic methods. Therefore a new approach for generating Cu(I) is required. We hypothesized that the Cu(I) generated by the radical mediated reduction of Cu(II) could be used to catalyze the CuAAC reaction and this process would enable spatial control of the CuAAC reaction.

The photo catalysis of the CuAAC reaction was first explored using with small molecules and Fourier transform infrared spectroscopy. Figure 2 shows the reaction of ethylazidoacetate and 1-hexyne in the presences of a copper (II) salt and a photoinitiator (bisphenyl phosphine oxide, Ciba). In the course of approximately 80 minutes all of the azide is consumed. ¹H-NMR and ESI+ mass spectroscopy both confirm the formation of the triazole produce and the absence of other coupling reactions (Eglinton coupling, etc.) Further verification of the mechanism is provided by control experiments where each component is eliminated. In the absence of any component or irradiation, no reaction occurs. The process is robust and does not require any purging of oxygen. For more details refer to [20].

The triazole linkages formed by the CuAAC reaction are both resistant to thermal and oxidative degradation making them useful polymer synthesis and modification.²¹ Spatial control of the reaction was then explored by the selective labeling of a fluorophore within a swollen hydrogel network. A hydrogel was first formed by the thiol-ene reaction between thiol and alkyne functionalized polyethylene glycol monomers (PEG) such that a small concentration of alkyne moieties remained (less than 40 mM). A mixture of copper sulfate, photo initiator (4-(2-hydroxyethoxy)phenyl-(2-hydroxy-2-propyl)ketone, Ciba) and azide functionalized fluorophore was then swollen into the gel. Selective irradiation using a standard chrome photomask produced the pattern shown in Figure 3.

Initial rate experiments were performed to gain an understanding of the reaction mechanism. These results suggest that the CuAAC reaction is likely to be the rate determining step under many experimental conditions, and undesirable side reactions of Cu(I) such as disproportionation, reduction of Cu(I) by radicals, and the reaction of Cu(I) with oxygen are

avoided, likely by ligand protection. Such interactions explain the high fidelity of patterning in systems where rapid diffusion of the photogenerated catalyst would otherwise be expected. These results demonstrate that this approach is an effective alternative to the use of ascorbic acid for the generation of Cu(I). These results are described in more detail in [22].

Figure 2. The CuAAC reaction of 1-hexyne (200 mM) and ethyazidoacetate (200 mM) occurs in the presence of 10 mM copper(II)sulfate and bisphenyl phosphine oxide (I819, 10mM) upon irradiation (10 mW/cm²). In the absence of any component, no reaction occurs.

Figure 3. The use of the photocatalyzed CuAAC reaction for the selective modification of a hydrogel is shown. A PEG hydrogel was first formed with an excess of alkyne functional groups using a thiol-ene reaction. A mixture of azide functionalized fluorophore, Cu(II), and photoinitiator was then swollen into the gel. Selective irradiation of the gel using a chrome photomask produced the image shown in the widefield fluorescent above (200 μ m scale bar). Features as small as 25 μ m were readily written into the swollen gels.

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Appendix A. List of Related Publications, Presentations, and Patent Applications

Publications

B.J. Adzima, C.J. Kloxin, C.A. DeForest, C.N. Bowman. “Photofixation: a new approach to photolithography,” (in preparation).

B.J. Adzima, C.J. Kloxin, C.N. Bowman. “Mechanism of the photo-mediated copper catalyzed azide—alkyne cycloaddition,” (submitted to *Chemistry of Materials*).

B.J. Adzima, Y. Tao, C.J. Kloxin, C.A. DeForest, K.S. Anseth, and C.N. Bowman. “Spatial and temporal control of the alkyne—azide cycloaddition by photoinitiated Cu(II) reduction,” *Nature Chemistry*, 2011, 3, 258-261.

B.J. Adzima, C.J. Kloxin, and C.N. Bowman. “Externally Triggered Healing of a Thermoreversible Covalent Network via Self-Limited Hysteresis Heating,” *Advanced Materials*, 2010, 22, 2784-2787.

Presentations

B.J. Adzima. “Click Polymerizations: Old and New Reactions for Material Synthesis.” University of Washington’s Distinguished Young Scientists Summer Seminar Series. July 2011

B.J. Adzima, Y. Tao, C.J. Kloxin, C.A. DeForest, K.S. Anseth, and C.N. Bowman. “Spatial and Temporal control of the azide-alkyne cycloaddition.” ACS Annual Spring Meeting, 2011

B.J. Adzima, C.J. Kloxin, C.N. Bowman. “Covalent Adaptable Networks.” The 32nd Australasian polymer symposium, 2011.

B.J. Adzima, C.J. Kloxin, C.N. Bowman. “Fracture healing of a polymer network via hysteresis heating.” Polymer networks group, 2010.

Patent applications

C.J. Kloxin, B.J. Adzima, and C.N. Bowman. “Photoinitiated Alkyne—Azide Click Reactions.” (CU2703B-61/417,785; 2010-2011).

C.J. Kloxin, B.J. Adzima, and C.N. Bowman. “Photolithography via photofixation of a Diels-Alder reversible network scaffold.” (CU2612B-61/417,594; 2010-2011).

B.J. Adzima, C.J. Kloxin, and C.N. Bowman. “Radio frequency magnetic field responsive polymer.” (CU2386B-61/266,767; 2009-2011).

Appendix B. Kinetics and Mechanism of the Photo-Catalysis of the Copper Catalyzed Azide–Alkyne Cycloaddition

Kinetics and mechanism of the photo-catalysis of the copper catalyzed azide–alkyne cycloaddition

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KEYWORDS *Click chemistry, CuAAC reaction, kinetics*

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ABSTRACT Photo-mediated catalysis of the copper catalyzed azide–alkyne cycloaddition (CuAAC) reaction using a photoinitiator was studied using infrared spectroscopy and initial rate experiments. It was found that the CuAAC reaction is the rate determining step, and the undesirable side reactions of Cu(I), such as disproportionation, further reduction by radicals, and reactions with oxygen are largely avoided, likely due to ligand interactions. The lack of these reactions, and the protection of Cu(I) by ligands also explains both the propensity for dark reaction and high fidelity spatial control of the reaction. These results demonstrate that the photo-mediated catalysis of the CuAAC reaction has all of the advantages of using sodium ascorbate to reduce Cu(II), while enabling spatiotemporal control of the reaction.

INTRODUCTION

The copper catalyzed azide–alkyne cycloaddition (CuAAC reaction) represents one of the most powerful and most implemented reactions in recent materials science research. The ability to photochemically control the reaction will prove useful across the range of developed substrate materials for the CuAAC reaction which include surfaces,¹⁻³ polymers,^{4,5} and particles.^{6,7} While the 1,3-dipolar cycloaddition is a straightforward “no mechanism” reaction,⁸ its popular variant, the copper catalyzed azide–alkyne cycloaddition (CuAAC), exhibits reaction kinetics that suggest a more complex mechanism.^{9,10} The Cu(I) catalyst is typically generated *in situ* by the reduction of Cu(II)¹¹ or by comproportionation of Cu(II)/Cu(0),¹² although the direct addition of Cu(I) salts is also employed.¹³ Sodium ascorbate is the most common reductant; however, the photochemical generation of reductants^{14,15} or use of light sensitive Cu(II) complexes is also possible.¹⁶ Our recent report demonstrating the spatial control of radical mediated reduction of Cu(II) produced two seemingly contradictory findings.¹⁵ While the reaction exhibited a significant dark reaction, that is the CuAAC reaction continued long after irradiation was ceased, masked images were reproduced with high fidelity despite the potential for Cu(I) to diffuse into

the masked area. Here, through a series of initial rate experiments a more thorough understanding of the kinetics and mechanisms involved in the photo-mediated catalysis of the CuAAC reaction is developed and discussed with its implications.

The CuAAC reaction mechanism is understood as involving three general steps: **(I)** the Cu(I)-acetylide formation, **(II)** the formal cycloaddition, and **(III)** catalyst regeneration (see **Figure 1**).^{10,12} Under saturated conditions, where catalyst turnover is not required, the CuAAC reaction shows first order dependence on both azide and alkyne concentrations, consistent with an elementary bimolecular reaction.⁹ In contrast, the kinetics are highly dependent on any ligands, buffer, salts, and substrates present when catalytic concentrations of Cu(I) are utilized.¹⁰ This behaviour has been interpreted as the consequence of a diverse family of copper coordination complexes that are formed *in situ*.¹⁰ In general, these copper species are highly reactive as evidenced by the consistent copper catalysis of the reaction for a large variety of terminal alkynes, but the reactivity of these species varies subtly to influence which step is rate-limiting.

Cu(I) is typically generated using sodium ascorbate as a reductant, which is used in large excess (10:1) to compensate for oxidation and disproportionation of Cu(I).^{12,17} As such, nearly quantitative reduction of Cu(II) is assumed to occur. It is perhaps surprising, given the CuAAC reaction's susceptibility to other species, that the ascorbate anion appears to have no effect on the reaction kinetics.¹⁰ While the radical mediated reduction of Cu(II) to copper metal has been observed for nearly 100 years,^{18,19} it has recently attracted renewed interest for both removal of hazardous wastes²⁰ and generation of copper nanoparticles.²¹ Due to Cu(II)/Cu(I)'s half reaction (reduction) potential of 0.16 V, a variety of organic radicals are capable of reducing it, e.g. ketyl, phosphinoyl, and semi-pinacol radicals generated by common photoinitiators such as bis(acyl)phosphines, α -hydroxyl ketones, and benzophenone. The reaction is typically described as occurring via the reduction of Cu(II) to Cu(I), followed by subsequent reduction to copper metal (reactions **R1** and **R2**, respectively in **Figure 1**).^{19,22} Besides further reduction to copper metal, Cu(I) is both air sensitive and prone to disproportionation (reaction **D** in **Figure 1**).²³ It would be expected that all three of these reactions could play a complicated role in the photo-mediated catalysis of the CuAAC reaction, as shown in **Figure 1**.

Figure 1. The copper catalyzed azide-alkyne reaction is shown to occur in three steps. The alkyne (**a**) reacts with Cu(I) to form the copper-acetylide (**b**). This species then reacts with the azide (**c**) to form the cycloaddition product (**d**). The catalytic cycle is completed when Cu(I) is regenerated and the triazole product (**e**) formed. In the radical mediated process Cu(I) is generated by reduction of Cu(II) (reaction **R1**). Once generated, Cu(I) can potentially disproportionate into Cu(II) and Cu(0) (reaction **D**), or it could be expected to be further reduced to copper metal by radical reaction (reaction **R2**). Any ligands present are omitted for clarity.

While a variety of photoinitiators are capable of reducing Cu(II), it is worth noting that organic azides can photochemically decompose to nitrene when irradiated with UV light below 400 nm.²⁴ We found decomposition to be slow relative to the CuAAC reaction when using a 365 nm light source; however, by utilizing a visible light photoinitiator decomposition is entirely avoided. As such, we utilized a visible light source (400-500 nm) and a visible light photoinitiator, phosphine oxide phenyl bis(2,4,6-trimethyl benzoyl) (Irgacure 819, Ciba), to generate copper-reducing radicals. The phosphinoyl radicals generated via photolysis are expected to participate in electron transfer reactions with Cu(II).²⁵

EXPERIMENTAL INFORMATION

All of the following compounds 1-dodecyne (Sigma Aldrich), dimethylformamide (ACS or Biological grade, Sigma Aldrich), copper sulfate pentahydrate (Sigma Aldrich), Irgacure 819 (Bis(2,4,6-trimethylbenzoyl)-phenylphosphineoxide, Ciba), were used without further purification.

A Nicolet Magna-IR 750 Series II FTIR spectrometer, outfitted with a horizontal transmission stage, was used in conjunction with a variable pathlength liquid cell with calcium fluoride windows (International Crystal Laboratories). The samples were irradiated by an EXFO Acticure high pressure mercury vapor short arc lamp equipped with a 400-500 nm bandgap filter. Light intensities were measured using an International Light IL1400A radiometer equipped with a GaAsP detector and a quartz diffuser. In a typical experiment, a solution of dimethylformamide (DMF) with 200 mM ethyl azidoacetate, 200 mM 1-hexyne, 10 mM copper sulfate pentahydrate, and 10 mM I819 was injected into the liquid cell. More details are provided in^[15]. Samples that were noted as being sparged had either ultra high purity argon or oxygen bubbled through them for 15 minutes.

RESULTS AND DISCUSSION

The CuAAC reaction was monitored using Fourier transform infrared (FTIR) spectroscopy, which allows for *in situ* measurement of azide and alkyne concentrations. Throughout these experiments model compounds were used in solution to simplify the analysis and avoid problems such as the onset of vitrification, which regularly occurs during the polymerization of materials with sub-ambient glass transition temperatures. The click nature of the CuAAC reaction suggests that the behaviour observed from this model pair of reactants may be extrapolated to a wide range of azides and terminal alkynes. However, it should be noted that the specific substitution of the triazole may play some role.¹⁰ Initial rate experiments performed with ethylazidoacetate and 1-dodecyne at catalytic concentrations of copper(II) and photoinitiator (ca. 20 to 1 molar ratios) verified the overall reaction rate to be nearly independent of both azide and

alkyne concentrations (**Figure 2**). Consequently, zero order kinetics are observed until approximately 40% to 60% of the limiting reagent is consumed (refer to supporting information). Beyond this regime non-linear behaviour is observed, consistent with the mechanism of the reaction shifting to a different rate law as the concentrations of the reactants and copper catalyst become similar. The zero order kinetics and the shift in the reaction mechanism is consistent with previous measurements of benzylazide and phenylacetylene.⁹ Such behaviour is believed to be the general case for the CuAAC reaction in the absence of added accelerating ligands.¹⁰ The independence of the reaction rate on alkyne and azide concentrations would tentatively imply that the regeneration of the Cu(I) is typically the rate-limiting step (reaction **III** in **Figure 1**). This behavior then suggests that if the generation of Cu(I) is rapid, as in the case of reduction by sodium ascorbate, that the CuAAC reaction mechanism will not be influenced by the method used to reduce Cu(II).

The reaction rate dependence on irradiation intensity is nearly zero-order (0.097 ± 0.02) at irradiation intensities greater than approximately 0.3 mW/cm^2 , as shown in **Figure 3**. The irradiation intensity is proportional to the photolysis rate, and therefore the rate at which radicals are generated. Consequently, the independence of the reaction rate on the light intensity implies that the generation of radicals is fast relative to the rate limiting step. As in the case of reduction by sodium ascorbate, the cycloaddition appears to be the rate-limiting step. This outcome is not surprising as the half-life of I819 at 20 mW/cm^2 is approximately 36 seconds. Comparatively, over the first 36 seconds less than two percent of the azide is consumed by the CuAAC reaction. When the timescales of photolysis and the CuAAC reaction become comparable, i.e., here at much lower light intensities, it is expected that the reaction rate would no longer be independent of the light intensity. In Figure 3 it is observed that the scaling value of the light intensity does indeed increase at irradiation intensities less than 0.3 mW/cm^2 , which corresponds to when the half-life of the photoinitiator is calculated to be of the same order as that of the CuAAC reaction itself.

Figure 2. The dependence of the reaction rate on the initial concentrations of 1-dodecyne , ethylazidoacetate, and triazole product reveals that the rate is approximately independent of all three (the slopes are 0.08 ± 0.07 , 0.16 ± 0.07 , and -0.03 ± 0.03 , respectively) The concentration of

the non-varying component is 200 mM for all experiments. The photoinitiator and copper sulphate concentrations were both 10 mM, and the irradiation intensity was 20 mW/cm² for all experiments.

Figure 4 shows that the reaction kinetics for samples containing equimolar concentrations of photoinitiator and Cu(II) prepared under ambient conditions possesses a similar reaction rate to those sparged with argon or oxygen. When the ratio of photoinitiator to Cu(II) is reduced, oxygen is then seen to significantly retard the CuAAC reaction. Oxygen is highly reactive towards phosphinoyl and benzoyl radicals,²⁵ and the resulting peroxy radicals are incapable of reducing Cu(II).²⁶ In contrast, equimolar mixtures of Cu(II) and photoinitiator do not exhibit an induction period. The lack of inhibition is explained by the excess of radicals from the photoinitiator which ensures complete consumption of Cu(II), even though a fraction of the radicals are scavenged by oxygen. Unlike some systems utilizing Noorish type II photoinitiators, the excess radicals eliminate the need for rigorous purging of oxygen.¹⁴ Importantly, the data also indicates that oxygen does not irreversibly consume Cu(I). If oxygen did irreversibly consume Cu(I) then the reaction rates of all the equimolar cases would not be similar. Whether Cu(I) reactions with oxygen are prevented by ligand coordination or effectively reversed by the excess of radicals is unclear from this data alone.

Figure 3. The effect of irradiation on the CuAAC reaction rate is shown. For irradiation intensities greater than 0.3 mW/cm² the effect of light intensity on the reaction rate is negligible with a scaling constant equal to 0.097±0.02 while for lower light intensities, the scaling exponent was found to be approximately 1.5. The azide and alkyne concentrations were 200 mM each, and the photoinitiator and copper sulphate concentrations were both 10 mM for all experiments.

Figure 4. When the concentration of photoinitiator and Cu(II) are both 10 mM, sparging with oxygen is seen to have no effect on the reaction kinetics. When the concentration of photoinitiator is reduced to 1 mM, radical scavenging by oxygen significantly retards the CuAAC reaction. The azide and alkyne concentration were each 200 mM, and the irradiation intensity was 20 mW/cm² for all experiments.

The dependence of the reaction rate on copper and photoinitiator is significantly more complicated. When the photoinitiator concentration is held constant at 10 mM the reaction rate shows an abrupt change in scaling at an initial Cu(II) concentration of approximately 15 mM (**Figure 5**). Above the threshold value of 15 mM of Cu(II), the reaction rate becomes largely independent of the initial Cu(II) concentration. The concentration of Cu(I) resulting from the reduction of Cu(II) could either be limited by the amount of photoinitiator available to reduce Cu(II), or from the resulting disproportionation/comproportionation equilibrium. The independence of the reaction rate from the initial Cu(II) concentration implies that while disproportionation of Cu(I) is highly thermodynamically favorable,²⁷ and typically rapid,²⁸ it is not occurring in this system. If disproportionation were occurring, the initial addition of excess Cu(II) would shift the equilibrium to produce more Cu(I) and the reaction rate would increase. Ligands that bind Cu(I) can protect it from disproportionation by shifting the equilibrium from favoring disproportionation ($\log K_{\text{Disp}} = 4.26$)²⁷ to comproportionation ($\log K_{\text{Disp}} = -2.20$).²⁹ In this system whether the ligands responsible for this behaviour are the triazole products, which are known to protect Cu(I),¹⁷ or the reactants themselves is unclear. The lack of an inhibition time and the absence of autocatalysis, could suggest that the protecting ligand is present at the beginning of the reaction, implying the alkyne reactant is the protecting agent. However, as the triazole concentration is greater than that of Cu(I) after 5% conversion, the resolution of these experiments may not be sufficient to detect a slight inhibition during the timeframe that the protecting ligand is formed.

Likewise, when the initial Cu(II) concentration is held constant at 10 mM, the reaction rate shows an abrupt change in scaling at a photoinitiator concentration of 5mM. Above this

threshold concentration of photoinitiator, the reaction rate is independent of photoinitiator concentration. If the reduction of Cu(I) to copper metal were occurring, it would be expected that adding more photoinitiator would result in more radicals and less Cu(I). As the CuAAC reaction rate typically shows a second order dependence on Cu(I) concentration,^{9,10} it would be expected that increasing the initial photoinitiator concentration would reduce the CuAAC reaction rate. The independence of the CuAAC reaction rate at higher photoinitiator concentrations implies that the reduction of Cu(I) to copper metal is not occurring to a significant extent under these conditions in this system. This behavior is somewhat surprising as radical reduction has previously been used to synthesize copper nanoparticles.²¹ However, the alkyne or triazole ligands appear to protect Cu(I) from each of these three side reactions: oxidation by dissolved oxygen, disproportionation, and further reduction by radicals. Indeed the only time we have noted the formation of copper metal is when working with very dilute hydrogels where the concentration of Cu(II) was nearly equimolar with the reactants.

In both the case of Cu(II) and the photoinitiator the threshold behavior appears to result solely from a change in the limiting reagent. The threshold values suggest that each molecule of I819 results in the reduction of 1.5 to 2 molecules of Cu(I). As I819 can produce two benzoyl and two phosphinoyl radicals this would suggest that either one of the radicals is ineffective in reducing Cu(II), or that the initiator fragments are no longer photoactive to visible light and cannot generate a second pair of radicals.

Below the threshold initial Cu(II) concentration of 15 mM, the reaction rate displays a second order dependence on the initial concentration of Cu(II). In the absence of disproportionation and reduction of Cu(I) to copper metal, it would be expected that all of the Cu(II) would be reduced to Cu(I) by the excess radicals that are generated. Indeed, the scaling constant is measured to be 2.0 ± 0.3 , consistent with previous measurements.^{9,10,16} Below the threshold photoinitiator concentration of 5 mM, the reaction rate shows a reaction order of 1.58 ± 0.01 . When Cu(II) is in excess, it would be expected that the amount of Cu(II) reduced to Cu(I) would be proportional to the initial photoinitiator concentration, which would result in a second order dependence of the CuAAC reaction rate on the photoinitiator concentration. The scaling exponent of 1.58 instead of 2.0 implies that as the photoinitiator concentration is increased, unproductive radical reactions such as primary radical recombination, i.e., radical-radical termination, become more prevalent. This value is similar to the scaling exponent of 1.5 that the light intensity appears to approach at its lower extreme (Figure 3). It is expected that both the light intensity and initial radical concentration should have similar scaling constants under these conditions.

Figure 5 The average reaction rate versus the initial concentrations of Cu(II) and photoinitiator (red circles and black squares, respectively), revealing two distinct regimes. When Cu(II) is in excess, Cu(II) shows a scaling constant of 0.01 ± 0.03 and the photoinitiator scales with an exponent of 1.58 ± 0.004 . When the photoinitiator is in excess, the Cu(II) shows a scaling constant of 2.0 ± 0.3 , and the photoinitiator scales with an exponent of -0.05 ± 0.04

CONCLUSIONS

These results suggest that the photo-catalyzed CuAAC reaction is controlled not by the rate of generation of Cu(I), but rather the amount of Cu(I) generated. Like sodium ascorbate, conventional photoinitiators rapidly reduce Cu(II) under typical conditions. When used in a polymerization reaction, these attributes would make the photo-mediated CuAAC reaction quite different from the typical radical photopolymerization where the rate of polymerization depends on the rate of active species generation. Further, we have shown results that suggest oxidation by dissolved oxygen, disproportionation, and further reduction of Cu(I) by radicals are all suppressed in these systems. This likely occurs because of Cu(I) coordination with either the alkyne or triazole ligands that are present. This behaviour explains both the dark cure reaction previously noted, and the high fidelity of the patterning previously observed, as Cu(I) spends much time bound to less diffusive species.¹⁵ As these side reactions are suppressed, this demonstrates that large excesses of conventional photoinitiators are not required to continually regenerate Cu(I), as is the case when sodium ascorbate is used as the reductant. Thus, when designing a reaction, workers need only to consider the tendency of the azides to decompose when exposed to short wavelength UV radiation and choose a photoinitiator appropriately. Finally, these experiments indicate that understanding catalyst turnover could be key to reducing catalyst concentration in real systems such as bulk polymerizations, surface modifications, and particle functionalizations.

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Supporting Information Available: The data from which the initial rates were obtained in Figures 2 and 5 is available in the supporting information. This material is available free of charge via the Internet at <http://pubs.acs.org>.

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Appendix C. Photofixation Lithography in Diels–Alder Networks

Photofixation lithography in Diels–Alder Networks

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ABSTRACT

While nano imprint lithography and a variety of other techniques have emerged for the fabrication of two dimensional (2D) structures, the top-down construction of three dimensional (3D) structures still presents significant challenges. Arbitrary 3D shapes incorporating overhanging and free standing features are difficult, or impossible, to fabricate using current methodologies. Importantly, such structures are key to developing photonic crystals,¹ elastic metamaterials,² and other critical micro devices. To overcome this difficulty, we developed a light sensitive, reversibly crosslinked polymer network scaffold, which allows subsequent 2D and 3D patterning of complex structures. Specifically, a thermoreversible crosslinked polymer network is formed by a Diels–Alder reaction between multifunctional furan and maleimide-based monomers, and a subsequent spatio-selective *photofixation* thiol-ene reaction creates well-defined irreversible regions within the material. Release of the patterned structure in the exposed region is achieved by reversion of the remaining reversible crosslinks to monomer. This novel materials approach employing a reversibly crosslinked scaffold facilitates the fabrication of complex three dimensional shapes, which are otherwise intractable by current patterning methodologies.

MAIN TEXT

The ability to carry out photochemical reactions selectively and in a spatiotemporally controlled manner plays a key role in many modern technologies, including microfluidic devices,³ metamaterials,⁴ artificial tissues,⁵ parallel protein synthesis techniques,⁶ three dimensional prototyping,⁷ optical device fabrication, and microchip fabrication.⁸ Conventional photolithography begins with the preparation of a photoactive thin film cast onto a substrate, typically using spin coating techniques, followed by the application of heat to evaporate the solvent. A pattern is then transferred to the photoresist using actinic masked or focused laser light. The resulting image is developed by immersion in a solvent that selectively removes the undesired material. Photoresists are generally classified as being negative or positive tone

resists.⁸ Positive tone resists are rendered soluble by irradiation, typically due to degradation or modification of the polymer polarity, while negative tone resists utilize polymerization or crosslinking reactions to render them insoluble after irradiation.⁸

3D structures are fabricated by tracing out a 3D pattern using modern optical direct write lithographic techniques,⁹ or through a tedious cycle of patterning, development, planarization, and alignment of multiple, sequential layers each of which possesses its own 2D pattern.^{10,11} While direct writing reduces the total number of steps, it still presents significant challenges. First, 3D construction of non-contiguous objects in a liquid negative tone photoresist is often problematic. Unlike 2D lithography, 3D features must be built up layer by layer in a bath of liquid monomer when using a negative tone resist.^{12,13} Accordingly, each underlying layer must support the next layer, or overhanging features will sediment from the densification associated with the polymerization. Sedimentation is slowed by utilizing higher viscosity photoresists; however, this approach complicates the later removal of the unreacted resist while aiming to preserve fragile features. Alternative ablative techniques such as focused ion beam lithography share similar structural limitations, and freeform fabrication techniques, such as laser sintering, are limited in resolution and restricted to rapid prototyping of macroscopic objects such as machine tooling and artificial bone constructs.¹⁴ Notably, positive tone photoresists in conjugation with 2-photon processes overcome this challenge to write channels and other hollow interconnected structures where minimal material needs to be removed.¹⁵

We overcome the aforementioned limitations to conventional 3D photolithography by developing a novel materials approach in which it is possible to photolithography create arbitrary 3D structures within a thermoreversible polymer scaffold. While conventional chemically-crosslinked networks (*i.e.*, thermosets) are insoluble, infusible, and cannot be readily manipulated, networks formed using reversible reactions can be triggered to be readily able to depolymerize. Utilizing a *photofixation* reaction (*vide infra*), these reversible crosslinks are transformed into irreversible junctions. Thus, pattern transfer is performed within a reversible scaffold that supports arbitrarily complex 3D structures, and the desired 3D image is developed simply by triggering the depolymerization reaction in the unexposed regions.

While there are many reversible covalent chemical reactions,^{16,17} few are as robust or possess the unique behavior of the Diels–Alder reaction between furan (1) and maleimide (2) (**Figure 1**). Upon heating, the Diels–Alder adduct (3) readily undergoes a retro-Diels–Alder reaction to recover (1) and (2). This reversible behavior has been utilized in self-healing materials¹⁸ and cyclic polymer synthesis.¹⁹ Polymer networks formed by the Diels–Alder reaction are reversibly depolymerized by increasing the temperature;²⁰ however, side-reactions can render the Diels–Alder adduct irreversible. By intentionally and spatio-selectively triggering a side-reaction, the location of this adduct within the scaffold is permanently *fixed*. The oxy-norbornene adduct (3) exhibits excellent reactivity within the thiol–ene and other radical-mediated reactions.²¹ By introducing a photoinitiator and a thiol-functionalized species into the formulation, the Diels–Alder scaffold is readily photofixed upon irradiation *via* the thiol–ene reaction. The material in the unexposed region remains unreacted and can easily be removed with sample heating, leaving the irreversibly-crosslinked user-defined structure. **Figure 1** illustrates this negative tone photofixation process and the accompanying chemical reactions, while **Figure 2** shows the specific species examined in this manuscript.

While the addition of the thiol-functionalized species enables photofixation, it is worth noting that the thiol can potentially undergo the base-catalyzed Michael addition reaction with the maleimide. This reaction is often employed in protein functionalization owing to its ease and

high yield.²² However, in the absence of a nucleophile or base, the thiolate anion is never formed, which effectively prevents the Michael addition from occurring. The addition of a small amount of acid is sufficient to prevent the unwanted thiol–maleimide reaction. In the present case, the thiol utilized in these studies (**6**) contains sufficient trace levels of acid impurities, to retard the thiol–maleimide reaction. Infrared spectroscopy reveals that a stoichiometric mixture of (**5**), (**6**), (**7**), and (**8**) equilibrated at 80 °C yields a consumption of 86% furan and 88% maleimide *via* the Diels–Alder reaction, while less than 1% of the thiol was reacted (**Figure 3a**). Consequently, bulk samples and films as thin as 10 µm were readily prepared by mixing the components, spin coating a substrate, and then baking the sample. Thinner films require the use of a solvent to reduce the viscosity. As the thiol–ene reaction is often considered part of the ‘click’ chemistry family of reactions, the chemical details of the thiol-bearing species are of minor importance,²³ and this approach could be also be used to incorporate additional chemical functionality within the polymer network with spatial control of its addition.

Upon actinic irradiation, photolysis of the photoinitiator produces radicals that are capable of initiating a variety of reactions. The strained nature of the olefinic bond in the oxy-norbornene makes it a preferable *ene* for the radical-mediated thiol–ene addition, while the electrically activated maleimide is less reactive towards the thiol–ene reaction and has a greater tendency to homopolymerize.²¹ Infrared spectroscopy reveals the complete consumption of the free maleimide and oxy-norbornene groups, accompanied by 70% conversion of the thiol (**Figure 3a**). This result indicates that the thiol–ene reaction dominates over homo- and co-polymerization of the oxy-norbornene and maleimide. Further, the complete reaction of the maleimide and oxy-norbornene suggests that no reversible bonds remain after irradiation. The network is permanently crosslinked and incapable of reverting to a monomeric liquid state (**Figure 3c**).

The multifunctional nature of the thiol (**7**) results in a large increase in the crosslinking density (**Figure 3 b**). Applying rubber elasticity theory,

$$G \propto \nu T, \quad (1)$$

the elastic modulus (G) is proportional to the temperature (T) and crosslinking density (ν). The ratio of the storage modulus before and after irradiation shows that the crosslinking density increases nearly fivefold during photofixation. In doing so the material obtains an ambient modulus of nearly 6 GPa, which lies near the upper limit of polymers lacking higher modulus filler materials [ref]. The photofixation reaction is also accompanied by a modest increase in the glass transition temperature, which rises from 31 to 42 °C. Before photofixation, the glass transition temperature lies approximately 50°C below the curing temperature, indicating that thermodynamic equilibrium rather than kinetic frustration *via* vitrification limits the Diels–Alder reaction from obtaining near-quantitative conversion. After photofixation, the breadth of the glass transition temperature does not increase, supporting the hypothesis that the primary mode of polymerization is *via* the step-growth thiol–ene reaction, whereas chain growth mechanisms tend to produce a more heterogeneous network structure and therefore would be expected to broaden the glass transition.²⁴

Selective irradiation of the material using masked light and subsequent heating results in the development of an image in only the illuminated regions. Image development is considerably aided by heating the material for 25 minutes at 95°C in the presence of furfuryl alcohol. Furfuryl alcohol is a naturally derived,²⁵ high boiling point solvent that additionally traps free maleimide and prevents crosslink reformation. Using a simple photolithographic set up (*i.e.*, collimated

light from a medium pressure mercury lamp through a chrome mask), features as small as 2 μm were readily fabricated on thiol functionalized glass surfaces (**Figure 4**).

Direct writing of 3D patterns into the bulk material presents two additional challenges. First, light must be transmitted deep into the material to initiate radical generation. Second, out-of-focus light must be minimized to prevent unconfined reactions away from the focal plane.

While researchers have demonstrated a number of techniques to reduce out of focus reactions,²⁶⁻²⁹ both problems are diminished by utilizing two-photon optical direct write lithography. Two-photon absorptions typically occur at longer wavelengths, where single-photon absorption, with its associated photochemical reactions, is reduced. This enables deeper penetration of light into the sample and minimizes out-of-focus reactions from single-photon processes. Additionally, as two-photon absorption is proportional to the square of the light intensity, rather than directly proportional, two-photon initiation is more readily confined to the focal plane.

The material formulation is the same for both 2D and 3D lithography, as the UV photoinitiator (**8**) possesses an adequate 2-photon cross-section. Irradiation with a 740 nm femtosecond pulsed laser produced a refractive index change within the sample, allowing immediate visualization of written shapes (Figure 5a), prior to any heating and image development. After heating the sample in furfuryl alcohol, the unexposed material depolymerized and the written 3D structures were carefully recovered from the developing liquid. Complex structures that are difficult, at best, to produce by other techniques, such as freely rotating interlocked rings or log pile-type structures, are easily fabricated using this 3D patterning approach (**Figure 5**).

Photofixation lithography in a reversible polymer network scaffold represents a powerful new approach to fabricate arbitrarily complex 3D structures. The use of a reversibly crosslinked polymer network as a photoresist has a number of potential advantages. It is expected that diffusion of the active species in such systems will be significantly retarded by the polymer network, enhancing the feature resolution. Furthermore, thin films can be prepared without the use of volatile organic solvents, because the starting materials are low viscosity monomers, rather than linear polymers that are employed in conventional photoresists (*i.e.*, SU-8 popularized by soft-lithography³). Moreover, it is also possible that the use of solvents could be eliminated in the image development step as well. Photofixation provides a method to eliminate thermoreversible behavior within reversible adhesives, self-healing materials,¹⁸ non-linear optics,³⁰ and polymer encapsulants.¹ Finally, the utilization of photofixation lithography in conjunction with thiol-functionalized materials, such as cysteine-terminated peptides, thiol functionalized nanoparticles, and thiol-functionalized sensing moieties, would enable the simultaneous fabrication of complex 3D structures with uniquely defined chemical functionalization for an array of applications.

Figure 1| Schematic of the photofixation process and associated chemical reactions. First, multi-functional furan (**1**) and maleimide (**2**) monomers (bubble A) form a crosslinked polymer (bubble B) by the Diels–Alder reaction (reaction I). In the irradiated areas (bubble C) the resulting oxy-norbornene groups (**3**) are then selectively converted to irreversible crosslinks (**4**) by radical reactions with thiol molecules freely suspended in the polymer network (reaction II). The pattern is then developed by selectively removing the remaining material in the non-irradiated areas by shifting chemical equilibrium such that the retro-Diels–Alder reaction (reaction III) occurs and the material depolymerizes (bubble D).

Figure 2| Specific chemical structures. The chemical structures of the species used throughout this manuscript: tetrakis furfurylthiopropionate (**5**); 1,13-bismaleimido 4,7,10-trioxatridecane (**6**); pentaerythritol tetrakis 3-mercaptopropionate (**7**); and 2,2-dimethoxy-1,2-diphenylethanone (**8**).

Figure 3| Spectroscopic and mechanical characterization. a) The initial FTIR spectrum of a stoichiometric mixture of (5), (6), (7), and (8) shows infrared absorption peaks characteristic of maleimide, furan, and thiol (696 , 1012 , and 2571 cm^{-1}). After curing for 12 hours at $80\text{ }^{\circ}\text{C}$ a peak indicative of the oxynorbornene appears at 911 cm^{-1} , while the maleimide and furan peaks have decreased in intensity. Comparison to the initial spectrum suggests 86% of the furan and maleimide have reacted, while less than 1% of the thiol remains is consumed (inset). After irradiation with 365 nm light at an intensity of 10 mW/cm^2 for 10 minutes the oxy-norbornene and maleimide peaks vanish, and 70% of the thiol is consumed. b) Dynamic mechanical analysis shows that when heated to temperatures greater than $100\text{ }^{\circ}\text{C}$, the unreacted material can relax and flow via breakage of the Diels–Alder adducts. Once the thiol–ene reaction occurs, this behavior largely ceases, although a slight decrease in the modulus is noted at approximately $150\text{ }^{\circ}\text{C}$. The reaction of the thiol also results in an increase in crosslinking density and an elevation of the glass transition temperature(inset).

Figure 4| Two dimensional Patterning. The increased crosslinking density in the irradiated areas results in the development of an image after the unreacted material is removed by the retro-Diels–Alder reaction. Features as small as 2 μm were readily produced in both thin films ($< 1 \mu\text{m}$) that were stripped completely to the glass, and in thicker films (approximately 10 μm) where the etching proceeds 1-2 μm into the film (scale bars 200 μm main image, 20 μm inset).

Figure 5| Patterning of interlocked rings and a logpile type structure. (a) An SEM micrograph shows a set of 8 freely rotating rings written using two-photon techniques and developed by heating in furfuryl alcohol (200 μm scale bar). An inset optical micrograph demonstrates that the index change can be immediately visualized as seen by two sets of

interlocking rings still contained in the surrounding polymer scaffold (200 μm scale bar). (b) An SEM micrograph of a five layer log pile type structure (40 μm scale bar).

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AUTHOR CONTRIBUTIONS

CNB, CJK, KSA, and BJA conceived the concept and experiments. BJA performed the patterning experiments and the characterization of samples and materials, with the exception of the two photon lithography which was performed by CAD. CNB and BJA wrote the manuscript with input from all authors.

ADDITIONAL INFORMATION

The authors declare no competing financial interests. Supplementary information accompanies this paper on www.nature.com/naturematerials. Reprints and permissions information is available online at <http://www.nature.com/reprints>. Correspondence and requests for materials should be addressed to CNB.

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