**Rapid Visible Light-Mediated Controlled Aqueous Polymerization with In Situ Monitoring**

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**Supporting Information**

**ABSTRACT:** We report a simple procedure for rapid, visible light-mediated, controlled radical polymerization in aqueous solutions. Based on the photoelectron transfer reversible addition–fragmentation chain transfer (PET–RAFT) polymerization, fast chain propagation at room temperature in water was achieved in the presence of reductant and without prior deoxygenation. A systematic study correlating irradiation intensity and polymerization kinetics, enabled by in situ nuclear magnetic resonance spectroscopy, provided optimized reaction conditions. The versatility of this procedure was demonstrated through a rapid triblock copolymer synthesis, and incorporation of water-labile activated esters for direct conjugation of hydrophilic small molecules and proteins. In addition, this technique boasts excellent temporal control and provides a wide range of macromolecular materials with controlled molecular weights and narrow molecular weight distributions.

The development of controlled radical polymerization (CRP) techniques has enabled the synthesis of well-defined functional polymers with predefined molecular weight and low dispersity (D). In particular, CRP techniques that operate in water have attracted significant attention, since they grant access to hydrophilic monomers and serve as a more environmentally friendly and biocompatible alternative to organic solvents. Nevertheless, traditional aqueous CRP techniques often rely on either metastable Cu(I) catalysts that readily undergo disproportionation in water or operation at elevated temperatures, resulting in diminished control and a loss in chain end-fidelity during the polymerization.

Recently, there has been interest in the development of visible-light mediated CRP techniques that offer spatiotemporal control and room temperature operation. For example, Hawker, Miyake, Yagi, and others have developed visible light-mediated atom transfer radical polymerization (photoATRP) strategies, while Cai and Boyer expanded the platform to photoinitiated and photoelectron transfer reversible addition–fragmentation chain transfer (PET–RAFT). These advances provide polymers with low D and excellent chain-end fidelity. Concurrently with this work, PET–RAFT conditions were developed that allow for room temperature aqueous CRPs, though reaction rates are slow, resulting in significant irradiation times. Faster reaction rates are highly desired as they would reduce exposure time, making the methodology compatible with hydrolytically stable monomers and in situ bioconjugation. Alternatively, the photoinitiated RAFT system developed by Cai and co-workers showed impressive reaction rates, but required deoxygenation prior to polymerization. In this report, we address this challenge through the design of simple, room temperature-based aqueous CRP procedures that reach high conversion within minutes. Optimal reaction conditions that balance propagation rate and polymerization control are systematically identified using a recently developed in situ NMR monitoring technique. Given that high conversion and chain-end fidelity is achievable with this platform, the rapid fabrication of triblock copolymers, and incorporation of hydrolytically unstable activated ester building blocks for in situ bioconjugation of small molecules and proteins are explored.

Initial studies examined tris(2,2-bipyridyl)dichlororuthenium(II) (Ru(bpy)_3Cl_2) as a water-soluble photocatalyst for PET–RAFT, which, upon photoexcitation, undergoes electron/energy transfer to induce polymerization (Figure S3a). However, in this mechanism the presence of trace oxygen and the generation of singlet oxygen greatly retards chain propagation (Figure S3b). It was hypothesized that the addition of a reductant would mitigate this issue by preferential...
reaction with singlet oxygen, while simultaneously serving as an electron source to improve catalyst turnover (Figure S3c). To this end, the polymerization of N,N′-dimethylacrylamide (DMA) as the monomer, 2-(butylthiocarbonothioyl) propionic acid (BTPA) as the chain transfer agent (CTA), and sodium ascorbate (NaAsc) as the reductant was studied under ambient conditions in water (Figure 1a). The initial experiments were screened inside an NMR spectrometer using a fiber-coupled 470 nm light emitting diode (LED; Figure 1b), revealing fast and controlled polymerization kinetics. Specifically, targeting a degree of polymerization (DP) of 400 at a light intensity of 26 mW/cm² provided high monomer conversion (∼85%) in less than 40 min. We attributed the short induction period of ∼3 min to oxygen consumption; deoxygenation of the reaction mixture prior to polymerization eliminated this initial inhibition (Figure S4). The evolution of the experimental number-average molecular weight (Mn), as measured by size exclusion chromatography (SEC), with respect to monomer conversion is linear during the course of the reaction, indicating good control over polymerization (Figure S5). Additionally, replacing NaAsc with sodium acetate (NaOAc) led to no observable polymerization over a period of 4 h (Figure S6), which attests to the importance of a reducing agent to quench singlet oxygen. The role of oxygen in polymerization inhibition was further probed via a method established by Boyer and co-workers, where anthracene-9,10-dipropionic acid disodium salt (ADPA) is used as a singlet oxygen sensitizer. Using this technique, we observed a significant reduction of singlet oxygen evolution in the presence of NaAsc being observed (Figure S7). While the presence of NaAsc proved critical to achieve rapid and controlled aqueous PET-RAFT polymerizations without the need for deoxygenation, a deeper understanding of the effects of light intensity was particularly desirable for further development of this system.

A digitally controlled LED coupled with in situ NMR monitoring offers the capability to simply and systematically study the effect of light intensity on these PET-RAFT systems. Tuning the light intensity (470 nm) from 8 to 139 mW/cm² revealed a direct linear relationship with k_p, while maintaining log−linear kinetics for all intensities measured as well as narrow molecular weight distributions (≤1.4) at high monomer conversion (≥85%), consistent with controlled polymerization processes (Figure 1c,d). In addition, for faster polymerization speeds at higher light intensities, induction times are shortened (Figure S8). However, it is noteworthy that a slight compromise between light intensity and D exists (i.e., higher intensity leads to larger D), as seen in Figure 1d and in the SEC traces of the resulting polymers (Figure S9), which may be attributed to an increase in side reactions at high k_p, such as photolysis of the trithiocarbonate chain-end (Figure S10 and S11) and radical termination by chain−chain coupling. This systematic study revealed a desirable balance between k_p and D at a light intensity of 26 mW/cm², and as such, this intensity was used in all subsequent studies, unless otherwise indicated.

Temporal control was then investigated using in situ NMR monitoring with automated “on”/“off” cycling of the LED (Figure 1d). Rapid chain propagation during illumination is significantly decreased when the light is turned “off” and increases quickly with the next “on” cycle with minimal inhibition and similar rates of polymerization. This “on”/“off”
cycling was repeated four times, exemplifying the excellent temporal control that is achievable under these conditions.

To examine the versatility of this technique, the targeted molecular weights for DMA polymerizations were varied and the monomer scope was expanded. Reactions with DP ranging from 25 to 400 were conducted at a constant monomer concentration and CTA/catalyst/reductant ratio, along with light intensity (26 mW/cm², Table 1, entries 1–5). Additionally, at low DPs (i.e., high CTA concentrations) 20 vol % DMSO was added as cosolvent to solubilize BTPA. Significantly, for a wide range of targeted molecular weights, monomer conversions of greater than 80% could be achieved within ~15–30 min. This reaction rate is markedly higher than previously reported PET-RAFT systems, which take 5–10× longer to reach the same monomer conversion.26,37 Additionally, polymerizations of N,N'-diethyacrylamide (DEA) and N-acryloylimorpholine (NAM) with a targeted DP = 40 were successfully conducted without deoxygenation, achieving >95% monomer conversion within 15 min. Both polymers exhibited low D and molecular weights that were consistent with theoretical values (Table 1, entries 6 and 7).

One of the hallmarks of a CRP process is the ability to generate well-defined architectures, such as multiblock copolymers. To achieve such structures, high polymer chain-end fidelity is essential, which was confirmed with electrospray ionization mass spectrometry (ESI-MS) for a poly(NAM) sample prepared using the presented approach (Figure S12). To take advantage of the rapid PET-RAFT system and the resulting excellent chain-end fidelity, one-pot block copolymerizations were conducted, which mitigates the need for intermediate purification as it relies on quantitative monomer conversion for each consecutive block. First, NAM was polymerized to nearly quantitative conversion (97%) after 15 min of irradiation (26 mW/cm²), followed by DEA (97%, 15 min) and DMA (94%, 20 min). The shift to higher molecular weight after each block addition was tracked using SEC, revealing a low D (<1.3) that was maintained throughout (Figure 2). The small high molecular weight peak observed for poly(NAM) in Figure 2 is attributed to chain–chain coupling at high conversion, indicated by its continued presence in poly(NAM-b-DEA) and poly(NAM-b-DEA-b-DMA). Notably, the triblock copolymer was synthesized in less than 1 h, without the need for deoxygenation. These results clearly demonstrate the efficiency of the fast PET-RAFT process, and highlight its utility to prepare well-defined water-soluble triblock copolymers.

Given the significant implications of polymer bioconjugates in biotechnology and medicine,43,42 it was desirable to investigate whether common bioconjugation approaches were compatible with the presented technique. For example, activated esters, such as N-hydroxysuccinimidyl (NHS) and pentfluorophenyl (PFP) esters, have been extensively used for bioconjugation of amine-containing compounds. Although these activated ester monomers have been incorporated into copolymers,43–47 their poor water solubility, and propensity to hydrolyze over time limits their utility in aqueous systems. To overcome these deficiencies, a water-soluble monomer, N-acryloxysulfosuccimide (NASS), was synthesized and its incorporation in hydrophilic copolymers was examined using the present aqueous PET-RAFT procedure. It is noteworthy that the rapid nature of the polymerization enabled the incorporation of NASS with minimal degradation, thereby allowing for subsequent in situ conjugation (Figure 3a). To further quantify the rate of hydrolysis, NASS was placed in an analogous polymerization environment (100 mM NaAsc(aq), pH = 8) and monitored with 1H NMR spectroscopy, revealing a half-life of ~60 min (Figure S13). Therefore, a 15 min copolymerization should afford ≥85% of the activated ester groups intact. To demonstrate this ability, the polymerization

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**Table 1. Summary of Polymerization Conditions and Results**

<table>
<thead>
<tr>
<th>entry</th>
<th>[M]/[BTPA]/[catalyst]/[red]</th>
<th>monomer</th>
<th>solvent</th>
<th>catalyst concn (mM)</th>
<th>time (min)</th>
<th>α (%)</th>
<th>M&lt;sub&gt;chain&lt;/sub&gt;&lt;sup&gt;b&lt;/sup&gt;</th>
<th>M&lt;sub&gt;end&lt;/sub&gt;&lt;sup&gt;c&lt;/sup&gt; (SEC)</th>
<th>D</th>
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<td>400:1:1 × 10⁻¹:2</td>
<td>DMA</td>
<td>water</td>
<td>6.25 × 10⁻³</td>
<td>45</td>
<td>90</td>
<td>39900</td>
<td>43200</td>
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<tr>
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<td>DMA</td>
<td>water</td>
<td>1.25 × 10⁻²</td>
<td>30</td>
<td>90</td>
<td>18100</td>
<td>22100</td>
<td>1.24</td>
</tr>
<tr>
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<td>DMA</td>
<td>water</td>
<td>2.5 × 10⁻³</td>
<td>30</td>
<td>80</td>
<td>8150</td>
<td>7010</td>
<td>1.28</td>
</tr>
<tr>
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<td>DMA</td>
<td>water + 20 v% DMSO</td>
<td>6.25 × 10⁻²</td>
<td>20</td>
<td>95</td>
<td>4000</td>
<td>6000</td>
<td>1.21</td>
</tr>
<tr>
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<td>25:1:1 × 10⁻¹:2</td>
<td>DMA</td>
<td>water + 20 v% DMSO</td>
<td>1.0 × 10⁻¹</td>
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<td>90</td>
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<td>1500</td>
<td>1.30</td>
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<tr>
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<td>40:1:1 × 10⁻¹:2</td>
<td>DEA</td>
<td>water + 20 v% DMSO</td>
<td>6.25 × 10⁻²</td>
<td>15</td>
<td>96</td>
<td>5100</td>
<td>7020</td>
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<tr>
<td>7</td>
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<td>NAM</td>
<td>water + 20 v% DMSO</td>
<td>6.25 × 10⁻²</td>
<td>15</td>
<td>95</td>
<td>5600</td>
<td>6100</td>
<td>1.22</td>
</tr>
</tbody>
</table>

"Monomer conversion (α) determined by 1H NMR spectroscopy. "Theoretical molecular weight calculated based on monomer conversion. "Molecular weight and D (M<sub>ω</sub>/M<sub>n</sub>) determined using SEC (DMF with 0.1% LiBr as eluent, relative to PS standards). Light intensity: 26 mW/cm².

**Figure 2.** SEC analysis of the poly(NAM-b-DEA-b-DMA) one pot triblock copolymer synthesis. Samples were withdrawn from the reaction after each block and SEC was performed with DMF as the eluent and molecular weights are relative to polystyrene standards.

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was performed with a 9:1 monomer mixture of NAM/NASS and an irradiation intensity of 80 mW/cm², affording monomer conversions of 95% and 87% for target DPs of 40 and 400, respectively. NAM was chosen as the comonomer due to its similar reactivity ratio with NHS-containing acrylate monomers. Both polymers demonstrated low $D$ (≤1.3; Figure S14), suggesting good control. Monitoring the reaction kinetics with in situ NMR spectroscopy indicates that NASS incorporates slightly faster than NAM (Figure 3b), where $^1$H NMR resonances at 2.58, 2.80, and 3.92 ppm correspond to NASS, indicating successful incorporation (∼12 mol %; Figure 3c, top spectrum). To further confirm the existence of intact active esters and demonstrate the feasibility of in situ conjugation, amine-containing small molecules and proteins were added to the crude postpolymerization mixture. Both L-phenylalanine (Phe) and 5-[(2-aminoethyl)amino]-naphthalene-1-sulfonic acid sodium salt (EDANS) were reacted in molar excess with poly(NAM-co-NASS) over the course of 4 h at room temperature. $^1$H NMR spectroscopy revealed 3% and 8% incorporation of Phe and EDANS, respectively (Figure 3c, middle and bottom spectra, and Figure S15). The grafting density for the EDANS–polymer conjugate was also quantified using fluorescence spectroscopy, finding 11.7 mol %, which was in reasonable agreement with NMR and with the original feed ratio (Figure S16). The difference in grafting efficiency is attributed to the reduced steric hindrance and higher nucleophilicity of the amino group of EDANS relative to Phe. Additionally, the in situ grafting-to conjugation of proteins was tested using bovine serum albumin (BSA). Significantly, this was shown to proceed with high efficiency using sodium dodecyl sulfate-polyacrylamide gel electrophoresis (SDS-PAGE), affording 88% yield of conjugation (Figure S17). Overall, the in situ postpolymerization conjugation of small molecules and proteins highlights the utility of rapid PET-RAFT polymerizations for the preparation of functional materials and bioconjugates.

In summary, a visible light-mediated, rapid, and controlled radical polymerization strategy in aqueous solution at room temperature is reported. In situ NMR spectroscopy was used to systematically correlate light intensity to propagation rate, dispersity, and inhibition time, while simultaneously demonstrating temporal control during "on"/"off" cycling. The utility of this rapid polymerization technique is highlighted by the facile one-pot fabrication of triblock copolymers in less than 1 h. Furthermore, the synthesis of a water-soluble activated ester monomer and subsequent in situ incorporation into copolymers and conjugation with amine-containing compounds in water was achieved with high levels of functionalization and only minimal hydrolysis. Given the versatility and fast kinetics of the presented approach, a wide range of hydrophilic and functional polymers are now available to nonexperts with exciting potential in biorelated applications.

**ASSOCIATED CONTENT**

**Supporting Information**

The Supporting Information is available free of charge on the ACS Publications website at DOI: 10.1021/acsmacrolett.7b00587.

Additional experimental details (PDF).

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**Figure 3.** In situ postpolymerization conjugation. (a) Reaction scheme. The light intensity was increased to 80 mW/cm² to increase the reaction rate. (b) Reaction kinetics of the copolymerization of NAM/NASS = 9:1, target DP = 400. (c) $^1$H NMR characterization of the small molecule conjugation. From top to bottom: the purified polymer with NASS incorporated (top); the polymer conjugated with Phe (middle); and the polymer conjugated with EDANS (bottom). The insets show the magnified characteristic peaks of the conjugates.
The authors declare no competing financial interest.

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