5-7-2015

TEMPO-Containing ROMP Polymers as a Component for Treatment of Traumatic Brain Injuries

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TEMPO-Containing ROMP Polymers as a Component for Treatment of Traumatic Brain Injuries

An Honors Thesis submitted in partial fulfillment of the requirements for Honors in Chemistry

By Cameron Feriante

Under the Mentorship of Dr. Hans-Joerg Schanz

ABSTRACT

During a traumatic brain injury (TBI) sustained in combination with hemorrhagic shock the brain undergoes anoxic conditions resulting from swelling and blood loss. Traditional intravenous solutions are used to replace blood volume, but are unable to replace the blood's oxygen carrying capacity. A proposed treatment based on isotonic solutions containing hemoglobin-based oxygen carriers (HBOC's) which were intended to provide readily available oxygen was developed. However, cell-free hemoglobin was shown to have excessive oxidative potential. Polynitroxylated Pegylated Hemoglobin (PNPH) was developed to address this shortcoming. During PNPH synthesis a combination of polyethylene glycol (PEG) and (2,2,6,6-tetramethylpiperidin-1-yl)oxidanyl (TEMPO), is bound to the cysteine and lysine residues on the protein. The PEG serves as a physical buffer, slowing oxygen release, while the TEMPO converts released superoxide radicals and counteracts their adverse effects. This antioxidant capability allows PNPH's to successfully serve in their role, however, the synthesis of PNPH's remains labor and cost intensive. This research focuses on the development of a TEMPO and PEG containing copolymer which can be bound to cell-free hemoglobin to serve in the same antioxidant capacity with additional benefits. These benefits include: increased PEG and TEMPO loading capacity, optimizability in the ratio between PEG and TEMPO, and a potentially easier synthesis pathway. This document focuses on the development of TEMPO containing ring-opening metastasis polymerization compatible monomers as a component of the target polymer and TBI treatment.

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Dr. Steven Engel

April 2015

Chemistry

University Honors Program

Georgia Southern University

Acknowledgements

First and foremost on this project, I want to thank Dr. Hans Schanz, my research mentor. The project was his idea, and the patient help and guidance he has given has been instrumental in my own research success. He has helped me with applications, presentations and posters, and has helped me to succeed in every way possible.

Thank you to the Georgia Southern Chemistry Department, the College Office of Undergraduate Research, and the Georgia Southern University College of Science and Mathematics for the funding that made this all possible.

I have to thank my fellow research students who worked and struggled and succeeded in the lab right beside me. The interesting conversations and helpful atmosphere you all fostered helped to make the lab the perfect place for learning and exploration. Thank you to Morgan, Andrea, Jessica, Sarah, Caleb, Koomi and all the others. And thank you to Dr. Li Ma, and the faculty at Armstrong University for their help in analysis.

I must express my gratitude to the many professors at Georgia Southern University that I have had the opportunity to learn from and interact with, inside the Chemistry Department and everywhere else in the school.

The Honors Program at Georgia Southern University deserves recognition, since without their programs and scholarships I would not have had the opportunity to experience all the opportunities available at Georgia Southern University. Their warm welcome helped me find my way here to GSU.

I now extend my gratitude to all the friends who have supported me, the teachers who have helped push me this far, and to the research group that worked beside me. And thank you Ashley Hayden for being the constant person beside me through all of my endeavors.

Introduction

Hypotension and the resulting hypoxia resulting from hemorrhagic stress have the potential to threaten internal organs through oxygen deprivation, resulting in cellular death. Particularly at risk to damage via this oxygen starvation is the brain. Traumatic brain injuries (TBI) occurring in conjunction with severe hemorrhagic stress have the potential to cause irreversible damage to neurons due to this oxygen starvation, resulting in irreversible damage ranging from mild to significant reduction of cognitive function, to death.[1] In order to mitigate the loss of fluids, intravenous (IV) isotonic solutions are administered by first responders; however, while this IV is intended to maintain blood pressure and thus oxygen supply during a serious injury, it does not replace the oxygen transport capacity of the lost fluids and leaves cells at the injury site vulnerable.[1-3] A proposed treatment based on isotonic solutions containing hemoglobin-based oxygen carriers (HBOC's) which were intended to provide readily available oxygen, was developed.

However, HBOC's were found to have excessive oxidative potential, resulting in oxidative damage to the injured cells from the highly reactive oxygen carried by the cell-free hemoglobin.[4-7] Hemoglobin demonstrated dose dependent cytotoxicity in rats, which was mediated through the addition of an antioxidant, radical scavenging compound.[8] This result indicates that antioxidant capability is critical to the effective use of HBOC's in real-world treatments. Additionally, cell-free hemoglobin demonstrates potent nitric oxide scavenging capabilities, causing a cascading series of physiological responses including systemic vasoconstriction, inflammatory response among other effects. These responses, combined with the decreased blood volume and swelling already present during a TBI could prove detrimental. Many developed HBOC's chemically modify the protein to reduce this effect, one common method being pegylation.[9] Pegylation is the chemical addition of polyethylene glycol chains to the protein, increasing its hydrodynamic radius and reducing the protein complex's activity within the bloodstream through physical interference.

Polynitroxylated pegylated hemoglobin compounds (PNPH's) represent the combination of both solutions to HBOC's cytotoxicity and neurotoxicity. PNPH's were developed by binding a combination of polyethylene glycol (PEG) and (2,2,6,6-

tetramethylpiperidin-1-yl)oxidanyl (TEMPO), a stable nitroxide radical, to the protein. PEG functions as a physical barrier, enveloping the protein, while TEMPO acts as a radical trap, reducing the formation of reactive oxygen species. This antioxidant activity was intended to prevent oxidative damage at the injury site while allowing the protein complex to deliver useful oxygen to the target, ideally reducing the likelihood of neuron death in the event of a TBI. While effective, PNPH's have drawbacks, including limited concentrations of TEMPO and PEG, strictly governed ratios between attached functional groups, as well as time and cost intensive synthesis procedures.[1]

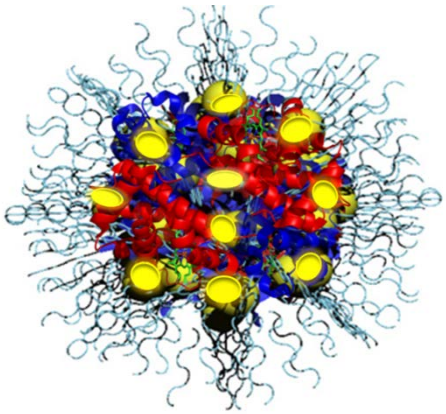


Figure 1: Conceptualization of PNPH, yellow groups represent TEMPO, green chains represent PEG

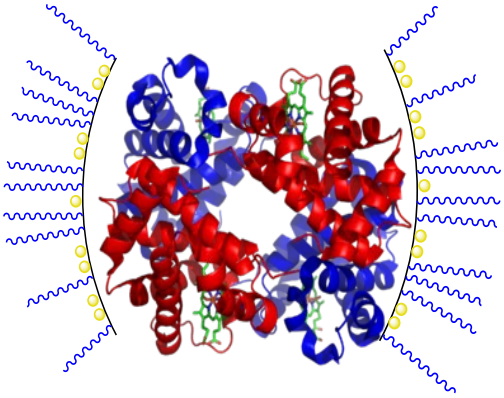


Figure 2: Conceptualization of a polymer modified hemoglobin complex. Yellow groups are TEMPO, blue is PEG, seen bound to a polymer backbone.

Our hypothesis is that these drawbacks could be addressed by combining TEMPO and PEG groups onto a polymer chain, which could then be bound to cell free hemoglobin to serve the same role as directly bonded TEMPO and PEG groups. This method should have the advantage of an easier synthesis route, more straightforward analysis of the resulting protein complex, and greater flexibility in the ratio between TEMPO and PEG groups bound to the protein.

Ring opening metastasis polymerization has proven compatible with TEMPO monomer derived from oxanorbornene and norbornene monomers. ROMP polymerization is a living polymerization using ring-strained monomers and a ruthenium alkylidene catalyst **figure 3**. As a living polymerization with a rapid initiation, the ROMP reaction will maintain molecular weight control and will continue effectively as long as unreacted monomer is present.[10] However, overly long reaction

times can lead to chain transfer reactions and loss of molecular weight control if the reaction is allowed to continue after monomer concentrations have depleted. To address this, the polymer reaction is capped with a terminating agent. An attractive feature of ROMP is the ability to add specific functionalities to the end of the polymer using a selected alkene as the terminating agent.

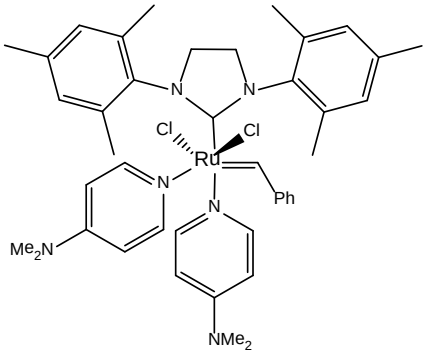
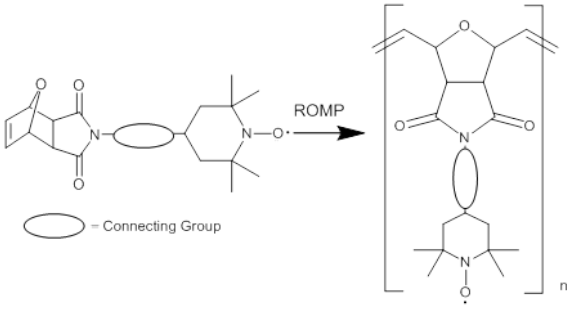
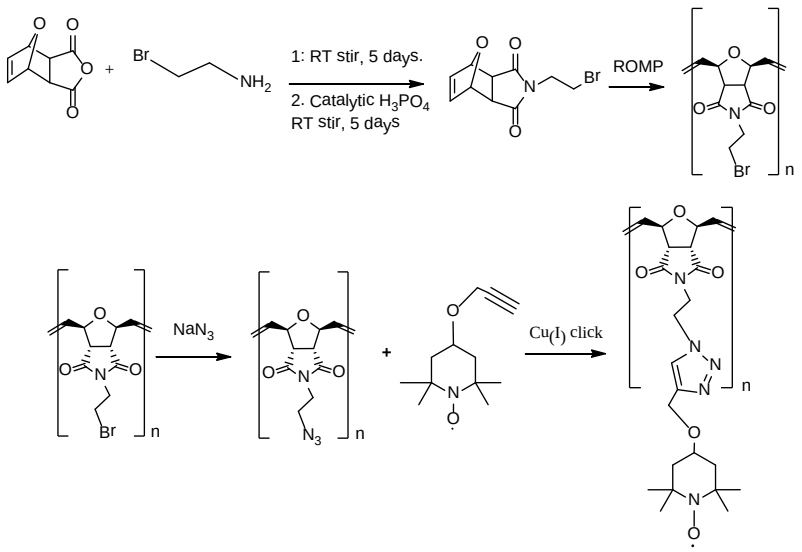


Figure 3: Structure of a Ruthenium catalyst used in ROMP polymerizations during this project.



Scheme 4: Generalized reaction of a TEMPO-functionalized oxanorbornene monomer yielding a TEMPO polymer.

The first step in synthesizing a TEMPO and PEG containing ROMP polymer, is monomer development. Several strategies are possible: A TEMPO containing monomer could be polymerized to create a TEMPO-functionalized homopolymer **scheme 4**, or co-polymerized with a PEG monomer to create the project's target polymer. Alternately, a functional polymer could be developed, then modified post-polymerization to attach the desired functional groups. The most likely reaction mechanism for this type of post-polymerization modification is the Huisgen [3+2] cycloaddition reaction, a cycloaddition reaction between an azide and an alkyne mediated by a copper (I) catalyst, otherwise known as the Click reaction. A potential synthesis procedure for this second strategy is presented in **scheme 5**.



Scheme 5: Potential synthesis strategy for TEMPO-functionalized ROMP polymers via an azide intermediate.

This method would require the development of an azide-functionalized polymer and a TEMPO derivative containing an alkyl substituent. Both strategies were explored during the course of this research. This document details the synthesis, purification and characterization of several monomers variants and their resulting polymers.

Discussion and Results

TEMPO-Containing monomers: The initial strategy for synthesizing TEMPO-contianing ROMP polymers focused on the development of a ROMP monomer with a TEMPO group. Monomer **1** was synthesized according to a literature procedure.[11] This was then analyzed using a thermo spray mass spectrometer, yielding the spectra seen in **figure 6**. The mass spectrum shows a clear molecular ion peak at 448, exactly the expected value for monomer **3** without its bromine counter anion. The positive mass spectrum allows us to continue with polymerization of this TEMPO monomer.

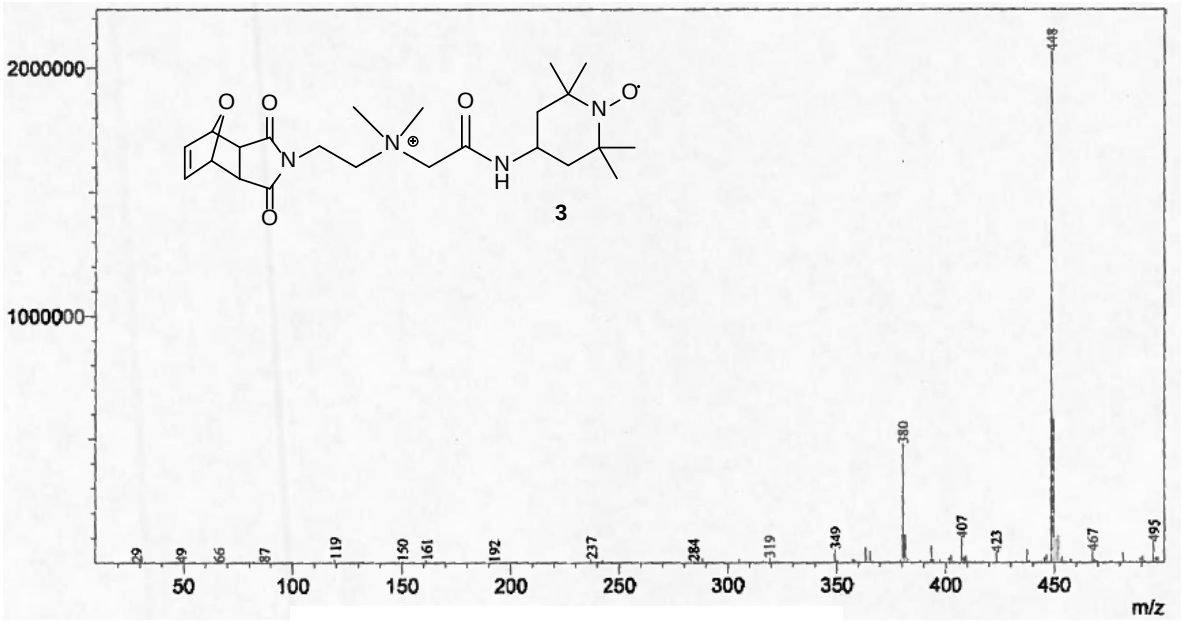
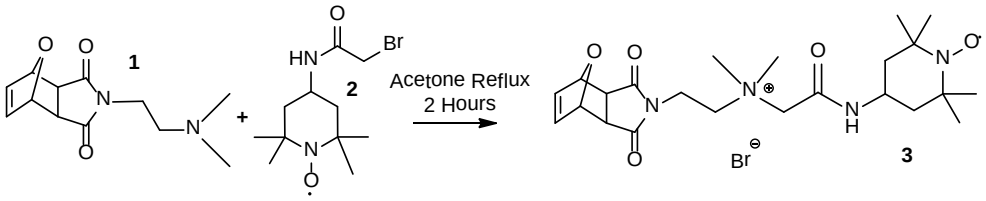


Figure 6: Structure and mass spectrum of monomer **3** showing the molecular ion at m/z=448

This monomer was then polymerized using Grubb's 1st generation catalyst, resulting in polymer **3**. Electron paramagnetic resonance spectroscopy (EPR) spectra were taken on compound **2**, monomer **3**, and polymer **4**, resulting in the spectra shown in **figure 8**. The spectrum demonstrates that the radical functionality of TEMPO survives the coupling reaction unchained. Additionally, the loss of fine structure in the polymer EPR spectra due to spatially close unpaired electrons demonstrates that polymerization has occurred, and more importantly, that the radical functionality is compatible with ROMP polymerization. This polymer was later analyzed using matrix assisted laser desorption ionization time-of-flight mass spectrometry (MALDI-ToF),

however, due to the cationic monomer unit, the mass spectrum was unable to yield useful data. The difficulty in further analyzing this monomer and resultant polymer prompted further monomer development.



Scheme 7: Synthesis of a cationic TEMPO monomer

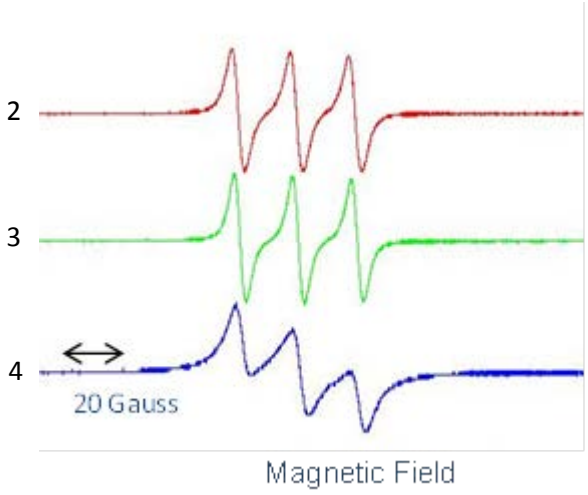


Figure 8: EPR results of compound 2, monomer 3, and polymer 4. The loss of fine structure in the polymer spectra indicates polymerization.

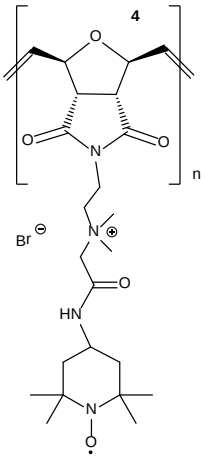
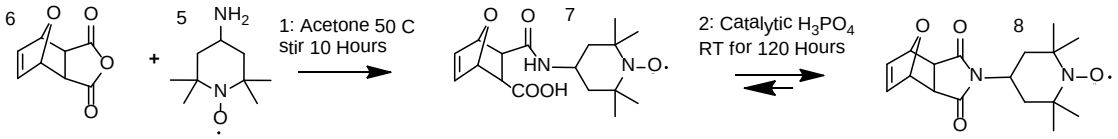


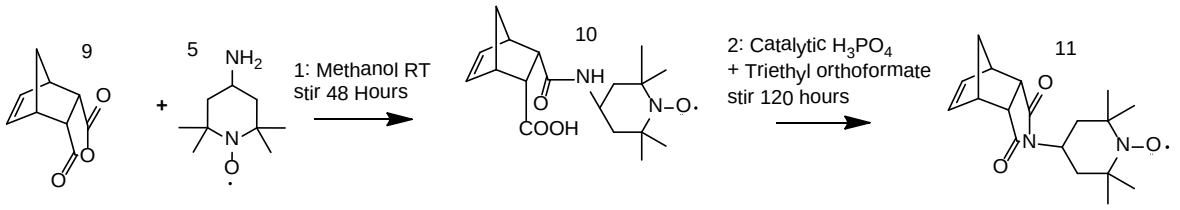
Figure 9: The poly-cationic TEMPO polymer from monomer 3.

The difficulties in analyzing the finished polymer and the poor solubility of the polymer prompted further research into TEMPO-containing monomers in order to develop a new monomer that is neutral, and displays better solubility. To address these goals a second monomer synthesis method shown in **Scheme 10** was performed. Two commercially available reactants were reacted to produce a TEMPO-containing monomer.



Scheme 10: Two step synthesis of an TEMPO monomer from 4-amino TEMPO and 3,6-epoxy-1,2,3,6-tetra-hydrophthalic anhydride.

Scheme 10 shows the synthesis of a TEMPO containing monomer from two commercially available components. The reaction occurs in two steps, the second of which is an equilibrium reaction where water is produced in a ring-closing reaction. The reaction yield was low at 51.8%, likely due to the equilibrium mentioned above. Improvement was attempted by increasing the reaction time, to little effect. Monomer **8** is promising because of its appreciable solubility when compared to monomer **3**, as well as its commercially available precursors, but at this point is difficult to isolate and purify while maintaining acceptable reaction yields. This prompted exploration into a new monomer, again using two commercial reagents, as well as exploration of methods to remove water from the reaction mixture, forcing the equilibrium to the right and ideally increasing product yield. The new experimental synthesis, shown in **scheme 11** utilizes a different commercially available norbornene, and incorporates triethyl orthoformate as a water scavenger in the second step of the reaction.



Scheme 11: synthesis of TEMPO monomer (11) from carbic anhydride and 4-amino TEMPO

In this new monomer synthesis, carbic anhydride **9**, an endonorbornene was reacted with 4-amino TEMPO **5** in methanol. The reactants were stirred for four days at room temperature prior to the addition of catalytic phosphoric acid and excess triethyl orthoformate. The triethyl orthoformate was added to remove water from the methanol solvent through hydrolysis, yielding formic acid and ethanol in the process. This addition helps to force the equilibrium between the intermediate monomer **10** and the final monomer **11**, to the right by removing water as the reaction progresses. These byproducts were removed along with the methanol after eight days. After purification the final yield of this reaction was improved from the earlier monomer **8** synthesis. The synthesis method yielding monomer **11** will be applied to monomer **8**

as well, since both effectively follow identical reaction pathways and the addition of triethyl orthoformate should positively affect the product yield for monomer **8** as well. A sample from the solid was reduced for NMR using ascorbic acid, the resultant spectra is shown in **Figure 12**.

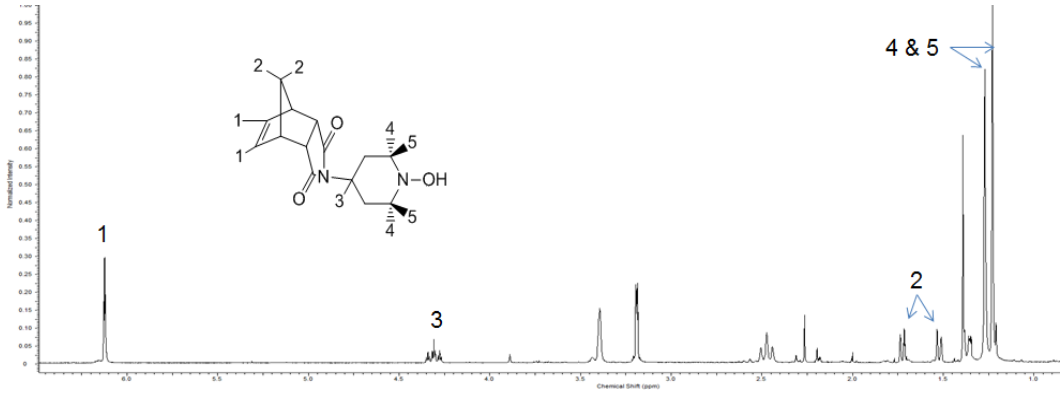


Figure 12: Structure and H^1 spectra of reduced TEMPO monomer **11'** in $CDCl_3$.

Monomer **11** was collected in sufficient purity for polymerization and was polymerized using Grubb's 3rd generation catalyst in toluene. **Figure 13** shown below displays an NMR spectrum of this polymer after reaction with ascorbic acid to reduce the pendant TEMPO groups. Monomer signals are still visible, but new, broader signals signifying polymerization have also appeared. Integration of the H^1 signals corresponding to hydrogen on the double bonds of the polymer backbone are shown integrated in detail in **figure 14**. The ratio between these polymer signals and the remaining monomer signal show a conversion of 80.9% based on NMR integration. The incomplete reaction indicates that a longer polymerization time will be needed in future polymerizations with this monomer.

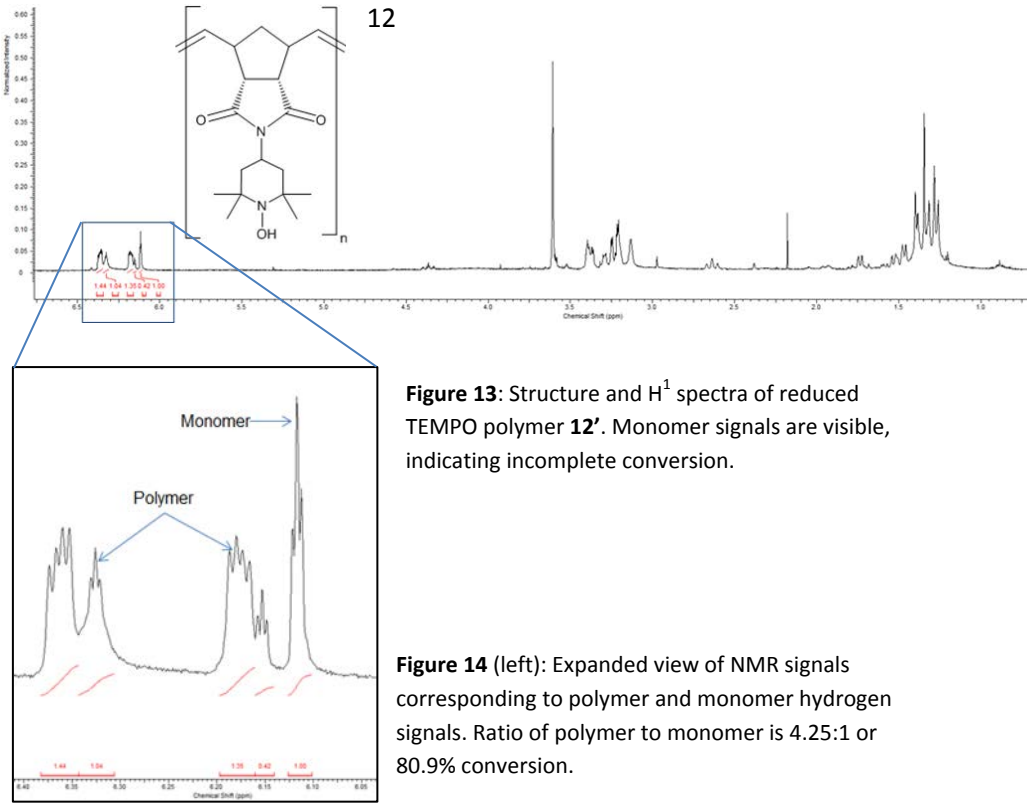
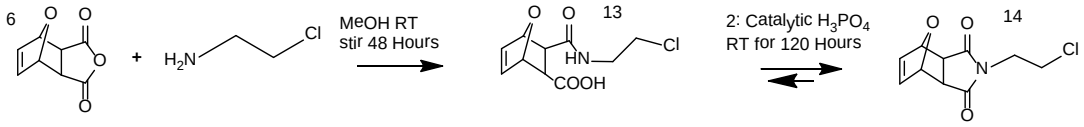


Figure 13: Structure and H^1 spectra of reduced TEMPO polymer **12'**. Monomer signals are visible, indicating incomplete conversion.

Figure 14 (left): Expanded view of NMR signals corresponding to polymer and monomer hydrogen signals. Ratio of polymer to monomer is 4.25:1 or 80.9% conversion.

Monomers and Polymers for Post-Polymerization Modification: The alternate strategy for developing TEMPO-containing polymers focuses on developing monomers with active functional groups. The first monomer developed towards this strategy was synthesized by reacting 3,6-epoxy-1,2,3,6-tetra-hydrophthalic anhydride with 2-chloroethylamine hydrochloride, both of which are commercially available reagents. The 2-chloroethylamine was deprotonated with potassium hydroxide. This synthesis, shown in **scheme 15** resulted in monomer **14**, an ROMP-compatible monomer with a chloro group available for subsequent coupling reactions.



Scheme 15: Synthesis producing a chloro group containing ROMP monomer.

Monomer **14** was analyzed via NMR in $CDCl_3$, yielding the spectra shown in **figure 16.** The monomer was then polymerized using Grubb's 1st generation catalyst, using ethyl vinyl ether as a terminating agent. This polymer was then analyzed via NMR in $CDCl_3$, yielding the spectra shown in **figure 17**. The broadening of signals associated with polymerization is clearly visible in the polymer spectra, providing supporting evidence of successful polymerization.

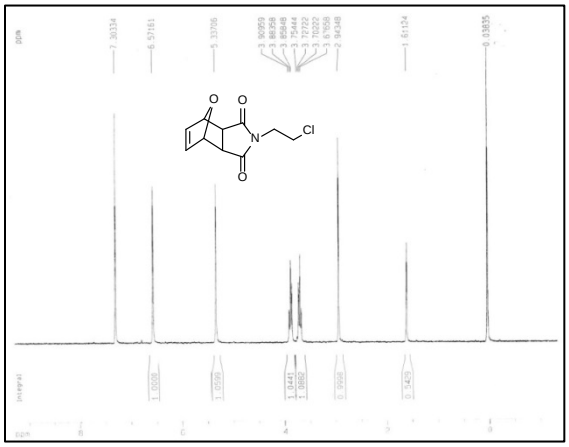


Figure 16: H^1 spectra of chloro monomer **14** in $CDCl_3$.

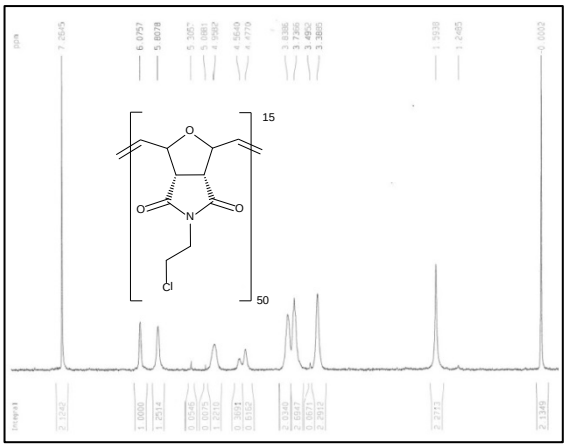


Figure 17: H^1 spectra of chloro polymer **15** in $CDCl_3$.

Additionally, the polymer was analyzed via MALDI-ToF, using 5-chloro-2-mercaptobenzothiazole as a matrix, the polymer was dissolved in 1:9 methanol to dichloromethane at 1 mg/ml, while the matrix was 1:1 MeOH:DCM. The resulting MALDI-ToF spectrum is shown in **figure 18**. In the MALDI-ToF spectrum, peaks separated by the monomer mass are clearly visible, overall the MALDI-ToF analysis procedure could benefit from additional optimization. The MALDI-ToF data taken on this polymer shows that there is good control of the polymer molecular weight, as the bulk of the polymer detected are within a few repeating units of each other. However, the spectra is not of sufficient quality for precise calculations.

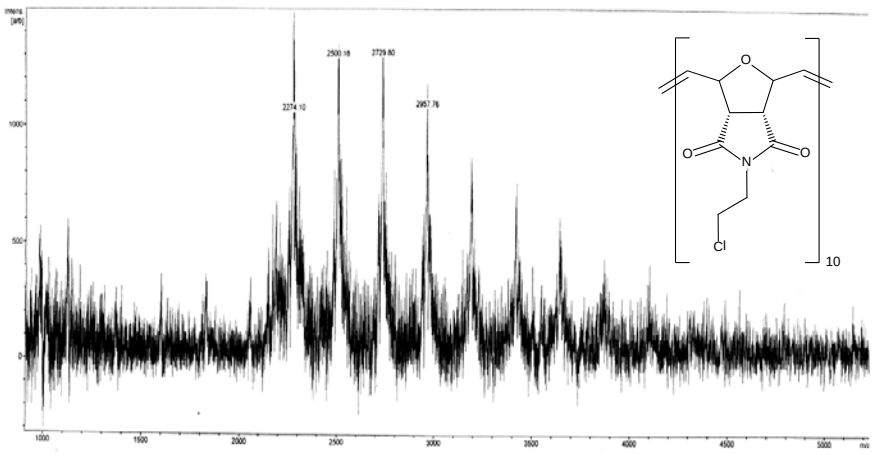


Figure 18: MALDI-ToF spectrum of a ROMP polymer with pendant chloro groups.

The purpose of developing a functional polymer was for post-polymerization modification, in this case, modification with an azide for later use in the the Huisgen [3+2] cycloaddition reaction. The chloro polymer, and later the monomer were both reacted with sodium azide in an attempt to substitute the azide for the chloride through a simple nucleophilic substitution reaction. However, even under a variety of conditions these chloro groups remained unreacted with sodium azide. This prompted the development of a new bromo group containing monomer based off monomer **14**, which was prepared through a synthesis similar to **scheme 15**, the only difference being the use of 2-bromoethyl amine hydrobromide rather than 2-chloroethylamine hydrochloride. This monomer was analyzed via H^1 NMR, resulting in the spectra shown in **figure 19**.

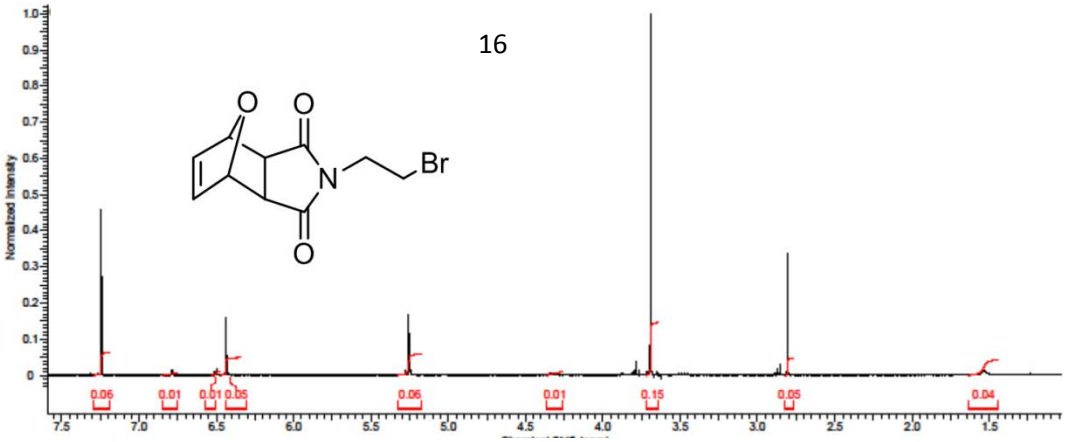


Figure 19: Structure and H^1 spectra of monomer **16**
a bromo containing ROMP monomer in $CDCl_3$.

This new bromo group containing monomer was polymerized using Grubb's 1st generation catalyst in dichloromethane. The monomer polymerized successfully, and was analyzed via NMR in $CDCl_3$, the resulting spectra is shown in **figure 20**.

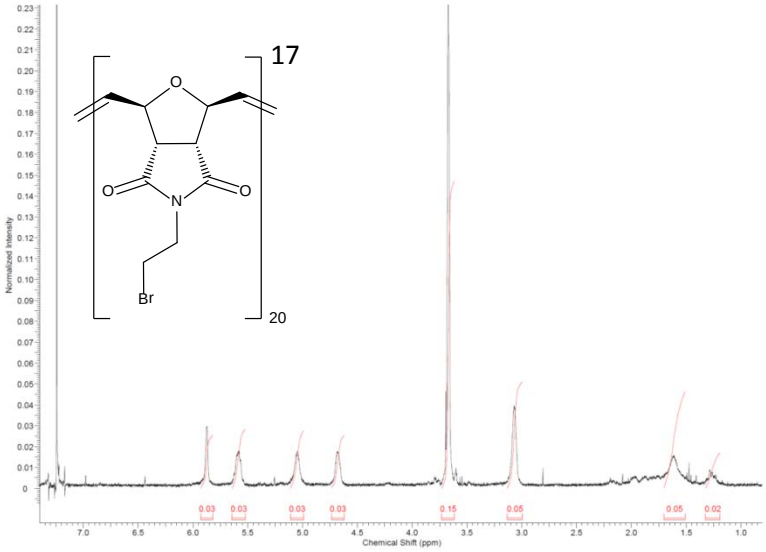


Figure 20: Structure and H^1 spectra of polymer **17** bromo containing ROMP polymer in $CDCl_3$.

The spectrum in **figure 20** shows the same broadening as seen in **figure 17**, again indicating successful polymerization. However, when this polymer was reacted with sodium azide, it similarly remained unreactive, and substitution was not observed in subsequent NMR spectra. Additional work may allow substitution by finding favorable reaction conditions for this substitution reaction, however, the low heat tolerance of the ROMP polymer backbone exacerbates the difficulty of this to some extent. The method of coupling an amine into a norbornene or oxanorbornene allows for further examination of this synthesis strategy, and despite the challenges, this method still holds substantial promise if modified to utilize a more active leaving group, or a larger spacing structure between the monomer and the leaving group. More research is needed to successfully develop the azide polymer desired for this synthesis strategy.

Conclusions

During the course of this project, three distinct TEMPO containing monomers were developed and synthesized. Monomer **3** was simple to synthesize, however the monomer displayed poor solubility, and produced a poly-cationic polymer which was not conducive to mass spectrometry. This prompted the development of monomer **8**, which demonstrated improved solubility in organic media, but was difficult to isolate due to the equilibrium in the second step of the reaction. To address this, monomer **11** was developed utilizing triethyl orthoformate as a water scavenger to drive the equilibrium in the second reaction step closer to completion. This was effective, raising the yield by 13.23%, however more work will be done to improve the yield.

These three TEMPO monomers were polymerized via ROMP, resulting in TEMPO-containing ROMP polymers, a crucial step on the way to the projects target TEMPO and PEG containing co-polymer. However, before the project can move forward a more effective and efficient monomer synthesis is desired.

Two new monomers were developed in pursuit of an azide functionalized polymer for post-polymerization coupling of TEMPO functionality onto a ROMP polymer backbone. These monomers, **14** and **16** were polymerized successfully into polymers **15** and **17,** however, as of yet a reaction procedure able to substitute an azide onto the chloro or bromo groups of these monomers and polymers at the temperatures they are able to withstand has been elusive. The potential for a readily customizable polymer backbone is enticing both for this project, as well as other potential applications, and work will continue in pursuit of an azide functionalized polymer.

Overall the project was able to successfully synthesize three TEMPO monomers, two of which show potential to move forward in the overall project. Additionally, two monomers and polymers were synthesized as a step towards a post-polymerization capable polymer for use in coupling either TEMPO or PEG functional groups onto a ROMP backbone. As the project moves forward, these developments are expected to be useful in the overall development of a PEG and TEMPO containing polymer for use as an antioxidant modification for cell-free

hemoglobin, which would then find similar applications to existing PNPH's while offering a more practical synthesis route.

Experimental Data

Synthesis of Cationic TEMPO-Oxanorbornene monomer 3: Initially, 0.3094 mmol of compound **2** was weighed and added to 0.3728 mmol of monomer **1**. These were dissolved in 25 ml of acetone and refluxed for 2 hours. A precipitate formed after 1 hour, and was filtered and dried. The filter was dried under reduced pressure for three days. The solid was scraped from the filter and weighed. Dry yield was 0.1650 mmol, 53.33% yield.

Synthesis of Cationic TEMPO-Oxanorbornene polymer 4: To start, 0.1913 mmol of monomer **3** was dissolved in 20 ml of 1:1 isopropyl alcohol to dichloromethane under and inert nitrogen atmosphere. 0.0251 mmol of Grubbs 1st generation catalyst was added. The reaction was stirred at room temperature for 62 minutes. At 62 minutes, 0.06944 mmol of ethyl vinyl ether was added to terminate the reaction. This stirred for a further 30 minutes before the reaction was dried. Final polymer mass was 0.0872 grams, composed of 0.1651 mmol of monomer, reaction yield is therefore 86.33%.

Synthesis of TEMPO-Oxanorbornene monomer 8: Initially, 0.9173 mmol of 4-Amino-TEMPO was weighed and added to a clean flask with 0.9631 mmol of 3,6-epoxy-1,2,3,6-tetra-hydrophthalic anhydride then dissolved in 6.0052 grams of methanol. This reaction was stirred at room temperature for 9.5 hours prior to the addition of one drop of concentrated ortho-phosphoric acid (H_3PO_4). The reaction was then stirred for 5 days at room temperature. The reaction solvent was removed using a rotavapor, then the resultant residue was separated in dichloromethane and deionized water, the organic phase was collected. Additional DCM was added, and the biphasic mixture was made basic with sodium bicarbonate ($NaHCO_3$). The organic phase from this was collected and added to the previous organic phase. The DCM solution was dried using solid anhydrous sodium sulfate (Na_2SO_4) and decanted. The DCM was then removed using a rotovapor, followed by several hours in a room temperature vacuum oven. The dry yield was 0.475 mmol or 51.8% yield.

Synthesis of TEMPO-endo-norbornene monomer 11: Initially, 2.599 mmols of carbic anhydride (endo-cis-5-Norbornene-2,3-dicarboxylic anhydride) **9** was reacted

with 2.570 mmol of 4-amino TEMPO **5** in 10 ml of methanol. The reactants were stirred for 96 hours at room temperature prior to the addition of three drops of ortho-phosphoric acid (H_3PO_4) and 11.191 mmols of triethyl orthoformate. The reaction was stirred for eight days at room temperature. All solvent was removed from the reaction after eight days. The dried reaction was sonicated in methyl *tert*-butyl ether for 1.5 hours, and the solid was filtered and collected. Ligroin was added to the orange filtrate solution, prompting additional precipitation. This was filtered, and the solution was dried on the rotavapor to remove the methyl *terti*-butyl ether, prompting further precipitation. This was filtered, and the pale orange solid was dried in a vacuum oven under very low heat. Final solid yield was 1.69 mmol (65.03% yield).

Synthesis of TEMPO-endo-norbornene polymer 12: To start, 0.628 mmols of monomer **11** was weighed under and inert nitrogen atmosphere along with 0.0103 mmol of Grubb's 3rd Generation catalyst. To this 10 mL of toluene was added. The solution was stirred at room temperature for 95 minutes prior to the addition of two drops of ethyl-vinyl ether. The toluene volume was reduced on the rotavapor. 30 mL of a 1:6 toluene to ligroin mixture was added and the polymer was sonicated for 1 hour. The resulting solid was filtered. A 1:1:8 mixture of methanol, dichloromethane and ligroin was added and sonicated, forming a colloidal solution. This was filtered through the same filter. 80 ml of DCM was washed through the filter, then the filter was dried under reduced pressure for 2 days.

Reduction of TEMPO monomers and polymers with *l*-ascorbic acid: Prior to NMR analysis all TEMPO monomers and polymers must be reduced with *l*-ascorbic acid. This is accomplished in a biphasic mixture of 2-5 ml each of DCM and deionized water to which several mg of sample, and excess *l*-ascorbic acid is added. The reaction mixture is stirred for at least an hour, or until the characteristic orange color of the TEMPO is no longer apparent. The reaction mixture is made basic with sodium bicarbonate, the amount necessary being dependent on the amount of acid present. The two phases are then separated. The organic DCM phase is washed two additional times with sodium bicarbonate and deionized water, before being dried with anhydrous sodium sulfate. This is decanted, then dried on a rotavapor. The yield is generally small, since the N-OH helps to solubilize the TEMPO

derivatives in water. The residue left on the drying vessel is dissolved in $CDCl_3$ for NMR analysis. Precise weight control of *l*-ascorbic acid is not necessary so long as a twofold excess or more is used.

Synthesis of chloro-endo-norbornene monomer 14: Initially, 34.88 mmol of potassium hydroxide was dissolved in 60 ml of methanol. This solution was chilled to 0 degrees Celsius in an ice bath prior to the addition of 34.94 mmol of 2-chloroethylamine hydrochloride. This mixture was stirred until the amine had fully dissolved, then stirred for an additional ten minutes before being removed from the ice bath. 14.11 mmol of 3,6-epoxy-1,2,3,6-tetra-hydrophthalic anhydride was then added and allowed to react at room temperature for 48 hours. After 48 hours, 51.02 mmol of ortho-phosphoric acid was added to the reaction and the reaction was stirred for an additional five days. After 5 days, the methanol was removed using a rotavapor, and the leftover solid dissolved in 30 ml deionized water and 30 ml dichloromethane. This was then separated, and the organic DCM phase was washed an additional 2 times with deionized water. The organic phase was dried fully with anhydrous sodium sulfate, decanted, and dried on a rotavapor followed by several hours in a vacuum oven at room temperature. The dry yield was 4.38 mmol (31.1%).

Synthesis of chloro-endo-norbornene polymer 15: To start, 1.317 mmol of monomer **14** was dissolved in 30 ml of DCM under an inert nitrogen atmosphere. 0.0263 mmol of Grubb's 3rd generation catalyst was added, and the reaction was stirred at room temperature for 40 minutes prior to the addition of 0.06944 mmol of ethyl vinyl ether. This was then stirred for five minutes before the reaction was dried on a rotavapor. The reaction was dissolved in 3 ml of dichloromethane, and loaded onto a flash column. 500 ml of ethyl acetate was passed through the flash column, however, when dried this yielded only 0.0522 grams of polymer. The flash column was rewashed with 100 ml of DCM, then with 100 ml of 2.5 % methanol in dichloromethane. These washes were combined and dried yielding 0.1195 grams of polymer, corresponding to a 40.14% yield.

Synthesis of bromo-endo-norbornene monomer 16: To synthesize monomer 16, 24.08 mmol of potassium hydroxide was dissolved in 30 ml of methanol. This solution was chilled to 0 degrees Celsius in an ice bath prior to the addition of 12.07 mmol of 2-bromoethylamine hydrobromide. This mixture was stirred until the amine had fully dissolved, then stirred for an additional 15 minutes before being removed from the ice bath. 12.05 mmol of 3,6-epoxy-1,2,3,6-tetra-hydrophthalic anhydride was then added and allowed to react at room temperature for 48 hours. After 48 hours, 30.30 mmol of ortho-phosphoric acid was added to the reaction and the reaction was stirred for an additional nine days. After 9 days, the methanol was removed using a rotavapor, and the leftover solid dissolved in 30 ml deionized water and 30 ml dichloromethane with 9.434 mmol of sodium carbonate. This was then separated, and the organic DCM phase was washed an additional 2 times with deionized water with additional sodium chloride to promote cleaner phase separation (approximately 1 gram per wash). The organic phase was dried fully with anhydrous sodium sulfate, decanted, and dried on a rotavapor followed by several hours in a vacuum oven at room temperature. The dry yield was 9.596 mmol (79.6% yield).

Synthesis of bromo-endo-norbornene polymer 17: To polymerize monomer 17, 0.972 mmol of monomer 16 was dissolved in 20 ml of dichloromethane under an inert nitrogen atmosphere. 0.048584 mmol of Grubb's 1st generation catalyst was added and the reaction was stirred at room temperature for one hour prior to the addition of 1.5 ml of ethyl-vinyl ether. The reaction was stirred for five minutes, then removed from under nitrogen and dried on a rotavapor. The reaction was dissolved in 20 ml each of dichloromethane and deionized water, this was separated, and the organic phase washed an additional two times with deionized water. The organic phase was dried with anhydrous sodium sulfate, decanted, and dried on a rotavapor. A flash column was run on the polymer. The polymer was dissolved in 3 ml of DCM, and loaded onto a flash column. The polymer was washed through with 100 ml of DCM, then dried on a rotavapor. Final polymer yield is 0.0582 grams, corresponding to a yield of 22.01%.

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