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## Optimizing Strength and Versatility of Underwater Adhesion Polymer System

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**OPTIMIZING STRENGTH AND VERSATILITY OF UNDERWATER  
ADHESION POLYMER SYSTEM**

by

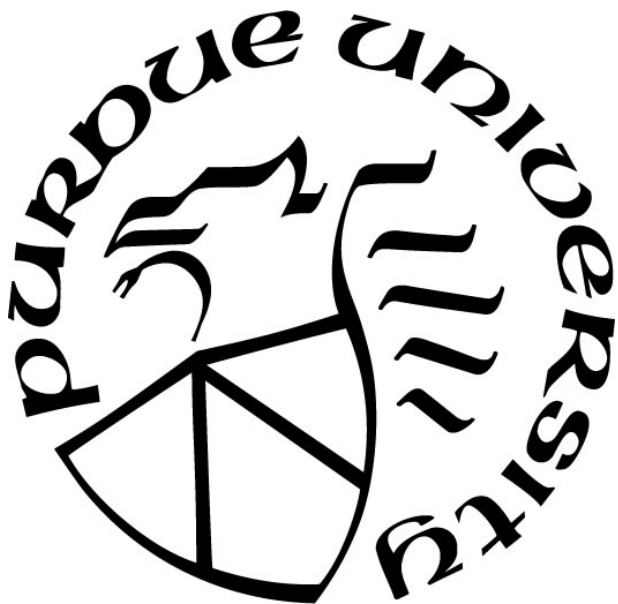
**Shannon R. Daily**

**A Thesis**

*Submitted to the Faculty of Purdue University*

*In Partial Fulfillment of the Requirements for the degree of*

**Master of Science**



Department of Chemistry

West Lafayette, Indiana

December 2017

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## LIST OF ABBREVIATIONS

DOPA	3,4-dihydroxyphenylalanine
Mfp	Mytilus edulis foot protein
g	gram
cm	centimeter
mL	milliliter
nm	nanometer
lb	pound
N	newtons
MPa	megapascal
<i>n</i> -BuLi	<i>n</i> -Butyllithium
BBr <sub>3</sub>	boron tribromide
DCM	dichloromethane
EDC	1,2-dichloroethane or ethylene dichloride
DMF	dimethylformamide
THF	tetrahydrofuran
DMSO	dimethyl sulfoxide
CCl <sub>4</sub>	carbon tetrachloride
PEG	polyethylene glycol
MeOH	methanol
CaCO <sub>3</sub>	calcium carbonate
NMR	nuclear magnetic resonance
GPC	gel permeation chromatography
3,4-DMS	3,4-dimethoxystyrene
M <sub>n</sub>	number average molecular weight
M <sub>w</sub>	weight average molecular weight
Đ	polydispersity index



## ABSTRACT

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Title: Optimizing Strength and Versatility of Underwater Adhesion Polymer System

Committee Chair: Jonathan Wilker

Among the hundreds of commercial adhesives available today, few, if any, have the ability to form strong adhesive bonds in an underwater system. Man-made adhesives have been shown to not work underwater because the glues interact with water rather than forming adhesive and cohesive bonds with the surface of the substrate. However, mussels and oysters naturally produce an adhesive that is unaffected by their watery environment. Examination of this adhesive has shown that shellfish use a mixture of proteins containing 3,4-dihydroxyphenylalanine (DOPA) that crosslink to form glue. Thus, an effort has been made to create a biomimetic polymer that includes DOPA with its unique adhesion mechanisms to increase adhesion underwater.

In the creation of a biomimetic adhesive, it is important to make it practical for use outside a controlled experiment. For an underwater polymer, this means the adhesive must be applied and cured completely underwater while demonstrating high adhesive strength. More specifically, during the application process, the polymer must stay in solution and display slightly negative buoyancy attributes as to not sink or float. To study these characteristics, solvent, crosslinker, and filler studies were completed to determine the best combination of additives for optimum adhesion. Experimentally, this was determined to be a solvent combination of 3:1 DMF:EDC with a filler additive of 5 wt.% 70 nm CaCO<sub>3</sub>. Testing involved changing the angle of the adherents to demonstrate the usefulness of the adhesive within a range of positions. Finally, a comparison study was completed to illustrate the superior adhesion strength of the created system.

## **CHAPTER 1. AN INTRODUCTION TO MUSSEL BIOMIMETIC ADHESIVES AND UNDERWATER ADHESION**

### 1.1 Nature's Prototype

Adhesives are a ubiquitous material in the modern world. From construction to the auto industry, technology to wood products, the world cannot stick together without glue.<sup>1</sup> While the first adhesives were produced from natural materials, the current market involves the mass production of synthetic glues. Many of these glues are incredibly commonplace (e.g. Super Glue, Gorilla Glue, Elmer's Glue).<sup>2</sup> With synthetically produced glues, the standard for adhesion has massively increased compared to natural products, and the amount of product available is enormous. However, for as commonplace and useful that adhesives are, there is a noticeable lack in innovation in the realm of underwater adhesion. In fact, it can be argued that there is not a reliable adhesive in existence that provides strong adhesive forces when applied and cured in a watery environment.<sup>3</sup> The difficulty surrounding this problem is that most synthetic adhesives lack the capability to displace water from the surface. In order for a strong bond to occur between two substrates, the adhesive must first create a strong bond between itself and the surface. With the introduction of water, this step often proves impossible. Instead, adhesives often end up interacting with the water rather than the surface they are meant to adhere to.<sup>4</sup>

When faced with an obstacle dealing with the world around us, it is useful to turn to nature to see how the problem is solved without technology. In this case, the ocean provides numerous of examples of marine animals that create their own adhesive to interact with the world around them. These animals include, but are not limited to, mussels, oysters, clams, and barnacles.<sup>5,6</sup> All of these sea creatures are able to adhere to rocks and even ships despite their wet conditions and the violent nature of the waves. This indicates a strong type of adhesive is at work that is

specifically designed to function underwater.<sup>6</sup> Among the adhesive-producing marine creatures, the most well-studied animal is the mussel.<sup>2,6</sup> In examining the adhesive that mussels naturally produce, a theory has been advanced to explain the chemistry behind their adhesion, and the creation of a synthetic alternative that mimics this chemistry has been developed.

### 1.1.1 The Muscle of Mussels

The glue a mussel creates is composed of a specific mixture of proteins that are secreted onto the surface the mussel is adhering to.<sup>7</sup> These proteins solidify into structures known as threads and plaques. The plaque is the primary adhesive layer while the threads connect the mussel to its adhesive plaque. The byssal threads incorporate a level of flexibility for the mussel and enable the animal to withstand the turbulence of the ocean environment while still remaining attached to the primary surface.<sup>7,8</sup> Meanwhile, the main adhesive properties lay within the plaque which can affix to a variety of surfaces.<sup>5,9</sup> Figure 1.1 shows a mussel adhering to a surface with its plaque and threads.

Within the plaque, there are six different mussel foot proteins (Mfp's) that are quite varied in terms of molecular weight and amino acid content. However, all of the Mfp's contain some amount of the amino acid 3,4-dihydroxyphenylalanine (DOPA).<sup>7,9</sup> Research has suggested that it is the presence of DOPA which provides one of the key adhering forces in the plaque.<sup>5,7,8</sup> The configuration of DOPA is shown to the side of Figure 1.1. This rare amino acid has an important catechol side chain that appears to be essential for mussel adhesion.<sup>9</sup>

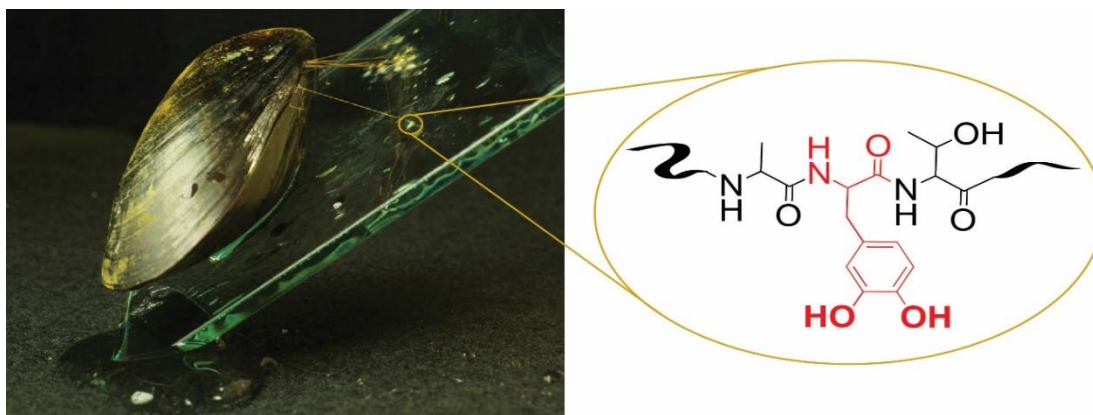


Figure 1.1 Mussel proteins<sup>9</sup>

Analysis of the Mfp's has shown a correlation between the molecular weights, DOPA percentages, and adhering properties. Those with higher DOPA percentages seem to be the main proteins associated with adhesion while those with higher molecular weights seem to be the proteins most involved in structural integrity of the plaques and threads.<sup>7,8</sup> Mfp's 3, 5, and 6 have the lowest molecular weights and highest DOPA contents and are the ones primarily responsible for the adhesive properties of the plaques. Conversely, Mfp's 2 and 4 have the highest molecular weights with lower DOPA contents, and they provide the structure for the plaque, including introducing the proper cohesive forces to keep the plaques and threads together. Mfp-1 also falls into this category of having high molecular weight but low DOPA content; however, it is responsible for providing a protective layer over all the other Mfp's to facilitate better adhesion.<sup>7,8</sup> For a more complete listing of the properties of the different Mfp's, refer to Table 1.1.

Table 1.1 Mussel foot protein properties<sup>7</sup>

Mfp #	Molecular Weight	DOPA content	% in plaque	Function
1	108 kDa	1%	5 wt%	protective layer
2	42-47 kDa	2-5%	25 wt%	inner cohesion
3	5-7 kDa	26%	15 wt%	adhesion
4	40 kDa	2-5%	18 wt%	inner cohesion
5	8.9 kDa	28%	5 wt%	adhesion
6	3 kDa	8%	10 wt%	adhesion

As discussed earlier, it is the DOPA content that is the main adhesive forces within the mussel plaque. To explain the chemistry as to why this occurs, it is important to look at the interactions DOPA forms (see Figure 1.2).<sup>10</sup> The hydroxyl backbones of the DOPA compound are particularly vital to the cross-linking interactions that happen within the molecule. There are primarily three reactions that occur to form the adhesive bond. To start, DOPA is oxidized to form quinone and semi-quinone groups. These groups can chelate to metal ions (primarily iron) found both in the surrounding water and within the mussel plaques themselves.<sup>10,11</sup> Additionally, radicals can form on these hydroxyl groups and bind to radicals on other DOPA molecules to result in covalent bonds between the DOPA groups. Between the iron-DOPA complexes and protein-protein coupling that occurs, these bonds provide a backbone for the cohesive nature of plaque.<sup>10,11</sup>

However, these types of bonds do not explain the adhesive properties of the plaques. In fact, it has been found that high levels of oxidation are actually detrimental to surface adhesion.<sup>10</sup> The way mussels compensate for this is the introduction of thiols and amines. Looking back at Mfp-6 which is vital to plaque adhesion but lower in DOPA content than the other similar Mfp's,

this protein is rich with thiols, which can reduce the oxidized DOPA molecules to promote binding to the surface.<sup>7,10</sup> This enables hydrogen bonds and covalent bonding between the DOPA molecule and any metals that are present on the surface of the adherent. Between these three interactions, an adhesive is formed that keeps the mussel glued together and stuck to almost any underwater surface.<sup>2,12</sup>

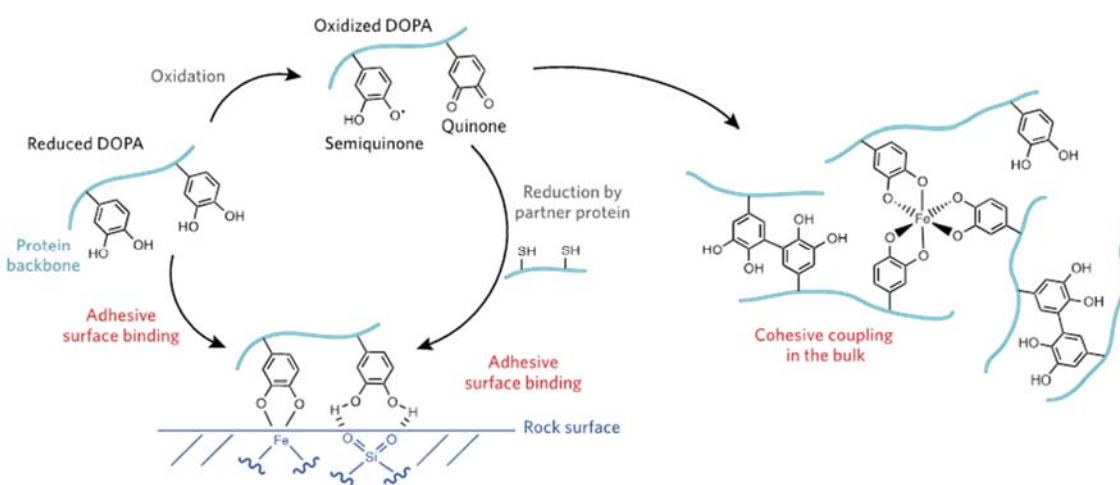


Figure 1.2 Surface binding of DOPA<sup>10,11</sup>

While mussels can utilize all these crosslinking mechanisms and adhesive bonds to adhere to a wide variety of surfaces in the ocean, commercial polymers are unable to do so. In particular, many cannot form the adhesive bonds to the surface. Instead of covalent bonds being formed between the adherent and the adhesive, the glue does not interact at all with surface particles due to the water present on the surface of the adherent.<sup>8,13</sup> Additionally, the internal chemistry of synthetic glues do not utilize redox chemistry with metals to maintain their structure.<sup>12</sup> This makes mussel biomimetic adhesives particularly unique in their adhesion forces.

### 1.1.2 Biomimetic Materials

With a natural solution present, the value of developing synthetic mimics could be questioned. However, it takes thousands of mussels producing reasonable quantities of adhesive to produce even 1 gram of adhesive proteins necessary for a glue.<sup>3,7</sup> Thus, it is not feasible to use mussels as a source for adhesives. By producing the glue synthetically, large quantities can be manufactured quickly, enabling the use of the glue across multiple markets.

Additionally, while mussels have one of the best underwater adhesive systems in existence, the strength of the system is directly tailored to the animal's properties.<sup>7,8</sup> In fact, since the adhesive is ultimately connected to the mussel's internal organs through the threads, this restrains the highest adhesive strength that can be provided from the system. If the adhesive was too strong, the mussels could potentially damage their internal organs.<sup>7,13</sup> Thus, by creating synthetic alternatives, it is possible to increase the adhesion strength even further to make the adhesive viable for industrial use. Previous studies have already demonstrated that synthetic alternatives can actually outperform the adhesive strength of mussel proteins.<sup>14-19</sup> However, in creating these synthetic alternatives, many more variables are introduced than if an exact copy of the mussel adhesive was created.

## 1.2 Polymer Synthesis

The main reaction for this project involves the polymerization of styrene into poly[(3,4-dihydroxystyrene)-*co*-styrene]. This reaction has been well-established and has functioned as the primary polymer used in multiple adhesion experiments within the Wilker lab.<sup>2,13</sup> The synthesis uses only flame-dried glassware and is performed utilizing Schlenk line techniques under argon. Characterization of the final product included <sup>1</sup>H-NMR (see Figure 1.3) which was performed

both before and after the deprotection step. The catechol percentage was determined using the ratio between the methoxy peaks at 3.0-4.0 ppm while deprotection was confirmed when these methoxy peaks disappeared. Deprotection of the hydroxyl groups was required for the polymer to have adhesive properties. Gel permeation chromatography (GPC) was used to determine the molecular weight of the polymer.

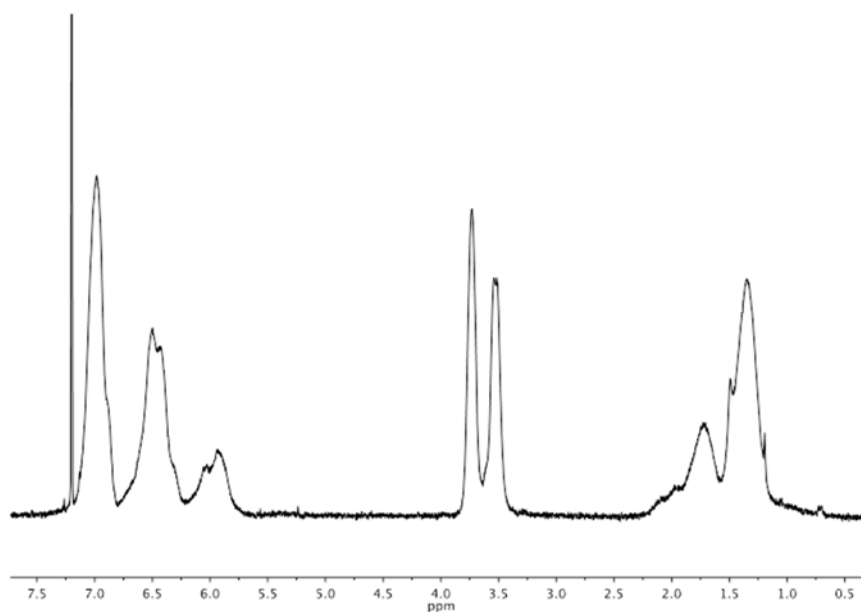


Figure 1.3 Proton NMR of protected polymer - poly[(3,4-dihydroxystyrene)-*co*-styrene]<sup>13</sup>

### 1.2.1 General Procedures

The synthesis of the polymers begins with purification of the starting materials – styrene and 3,4-dimethoxystyrene (3,4-DMS). Styrene is purified using a silica column while 3,4-DMS undergoes base filtration with DCM and a 1% NaOH solution. The two purified monomers are combined, and an anionic polymerization occurs with the introduction of the *n*-BuLi catalyst. After 24 hours, the reaction is quenched using methanol, and the polymer is precipitated out with cold methanol. This wash is repeated three times with chloroform used to re-dissolve the



polymer following each wash. Once dried, the resulting powder is the protected form of poly[(3,4-dihydroxystyrene)-*co*-styrene]. The polymer must be deprotected for it to function as an adhesive. This deprotection is accomplished using BBr<sub>3</sub> and a wash cycle with 1% HCl. This wash is also repeated three times using DCM to re-dissolve the polymer following each wash. The final product is dried using a rotary evaporator and high vacuum. Figure 1.4 illustrates the basic reaction scheme of the synthesis with 3,4-DMS/3,4-dihydroxystyrene shown in red and styrene shown in black. The first step is the synthesis and the second step is the deprotection.

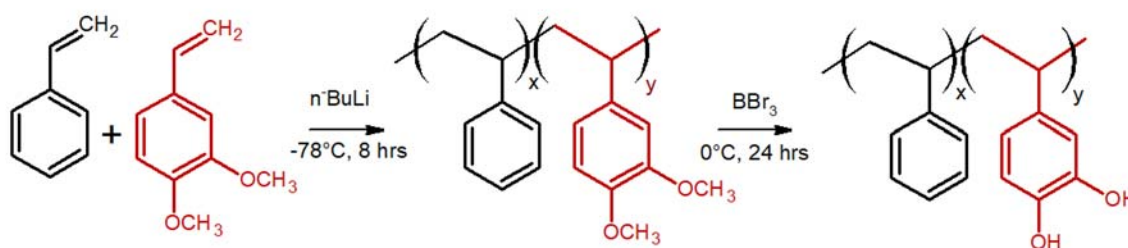


Figure 1.4 Reaction scheme of poly[(3,4-dihydroxystyrene)-*co*-styrene]

Once the polymer is deprotected (see chemical structure in Figure 1.4), it is ready for use in adhesion studies. However, the polymer will degrade in its deprotected form. Thus, to store the polymer for long periods of time, it should remain in its protected form. In both the protected and deprotected form, the polymer is light sensitive and should be covered to mitigate the possibility of oxidation. If oxidized, a color change will occur from white to brown/black.

### 1.3 Adhesion Studies

In order to evaluate the strength of each adhesive, adhesion testing was conducted using an Instron 5544 Materials Testing System at 2 mm/min with a 2 kN load cell. This instrument tests lap shear strength with the single lap-joint configuration. Figure 1.5 illustrates the set-up of the Instron instrument and the forces involved in lap shear testing. The adherents used for all adhesion tests were polished aluminum cut from type 6061 stock purchased from Farmer's Copper. These substrates were polished using a polishing machine and Mibro no. 3 and Mibro no. 5 polish.<sup>21</sup> After polishing, the adherents were subjected to a wash cycle of hexanes, methanol, acetone, and DI water.

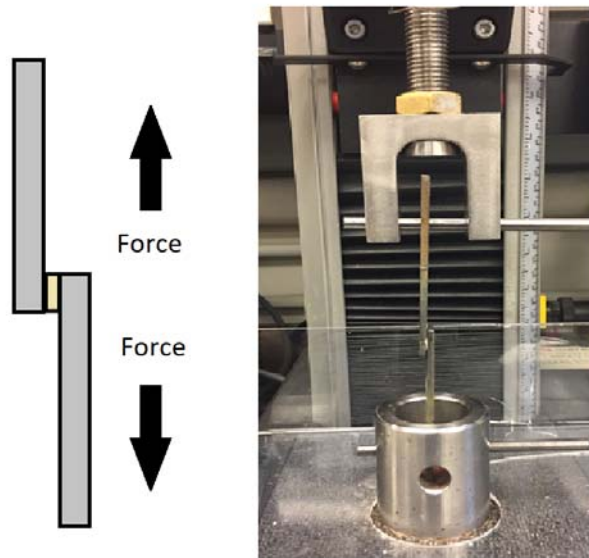


Figure 1.5 Instron machine loaded with aluminum adherents in single lap-joint configuration

Testing was conducted in plastic bins filled with artificial sea water. This water was prepared using Marine Environment dual phase formula and reverse osmosis water. Each test

began with the water at approximately 4°C and a salinity between 30-35 g/L. The bins were filled with enough water to completely cover the substrates during all aspects of set-up and cure time.

### 1.3.1 General Set-up

For each test, derivatives of poly[(3,4-dihydroxystyrene)-*co*-(styrene)] were dissolved in solvent to yield a 0.3 g/mL solution. Then, 45  $\mu$ L of the polymer solution was syringed onto an aluminum adherent already submerged in the artificial sea water. Once the polymer solution was dispensed, an additional 15  $\mu$ L of solvent was added before another adherent was placed on top of the first to form an overlapping joint of approximately 1.2 cm x 1.2 cm. A 0.4 lb weight was placed on top of the overlap joint of the adherents, and the adhesive was allowed to cure underwater for 24 hours. The adherents were then removed from the water and tested immediately on the Instron instrument. To determine the adhesion strength, the maximum force at the joint was divided by the area of overlap. Each adhesion test contained 5 samples, and these results were averaged together to determine the average adhesion for each condition. All error bars were created using 90% confidence intervals.

### 1.3.2 Previous Work and Results

All adhesion studies were conducted based on results and procedures from prior studies with this polymer system both dry and underwater. Additionally, the same polymer was used for each study to minimize adhesion changes due to variations in polymer composition. Finally, a majority of the studies utilized previously made protected polymers that only had to be deprotected before use. These polymers were recorded with their molecular weights and catechol

percentages. This enabled specific polymers to be chosen with a pre-determined molecular weight and catechol content. The catechol content and molecular weights were kept as consistent as possible and close to the recommended values for underwater adhesion. All of the polymers used are recorded in Table 1.2 and include what study each polymer was used in.

Table 1.2 Polymers used in studies

Catechol %	Styrene %	M <sub>N</sub>	M <sub>w</sub> (g/mol)	Đ	Study Used
24	76	20,000	42,000	2.2	solvent
25	75	31,000	41,000	1.4	solvent/covalent
27	73	37,000	54,000	1.5	filler

Both molecular weight and catechol percentage can dramatically alter the adhesive strength of a polymer. These characteristics have a direct effect on the cohesive and adhesive forces of the polymer.<sup>9</sup> When testing the strength of an adhesive, two types of bonds are important – cohesive bonds and adhesive bonds. Cohesive bonds refer to the internal bonding of the polymer to itself while adhesive bonds relate to how well the glue sticks to the surface of the adherent.<sup>22</sup> With higher molecular weights, cohesive bonding is strengthened because the increased bulk of the polymer creates more bonds and entanglements within itself. Thus, polymers with higher molecular weights typically result in higher adhesion.<sup>9,13</sup> Meanwhile, catechol content and its effect on adhesive force relates back to the original mussel system. The main Mfp's for adhesive properties are Mfp-3 and Mfp-5. These both have DOPA percentages of 20-30%, which are significantly higher than the other Mfp's. With this higher catechol content, the Mfp's are able to undergo more cross-linking and oxidation reactions to bind stronger to surfaces.<sup>7,10,15</sup>

However, as with any system, it is vital to determine exactly which molecular weight and catechol percentage is ideal. Previous underwater adhesion testing of this system determined the highest adhesion for both values.<sup>2,13</sup> The data from this study is presented in Figure 1.6. The best underwater adhesion values were with polymers that had a molecular weight of approximately 80,000 and a catechol percentage of about 22%.<sup>2,13</sup>

In comparison to this study, the polymers used were within the optimum catechol content range of 20%-30% but had lower molecular weights than typically ideal. This means that if these studies were conducted with polymers of higher molecular weights, it is likely the adhesion would increase.

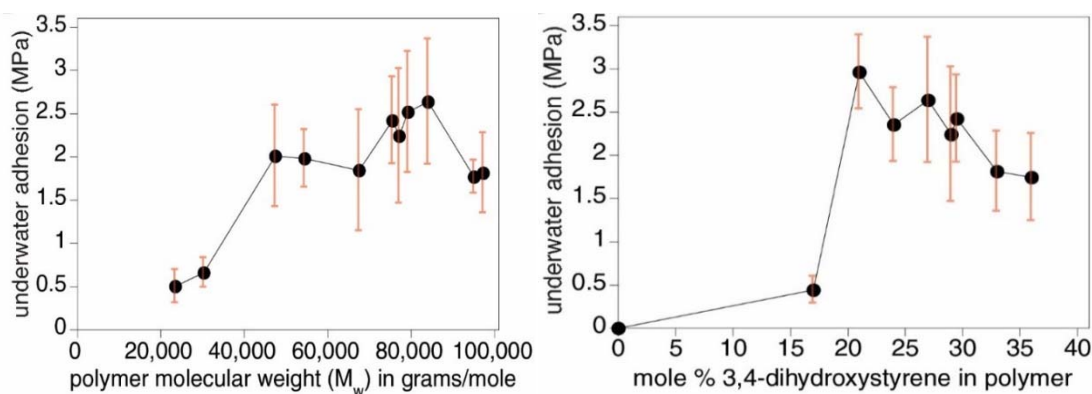


Figure 1.6 Molecular weight and catechol percentages<sup>2,13</sup>

The studies in this work are a continuation of previous work accomplished by Dr. North.<sup>2,13</sup> While this paper includes a solvent experiment, a crosslinker study, filler tests, slanted trials, and a comparison to commercial adhesives, his work included the previously mentioned molecular weight and catechol percent study, cross-linking experiments, filler tests, and comparison to both commercial adhesives and the natural mussel system. However, in those studies, the only solvent

used was chloroform. With the change in solvent, the cross-linker and filler tests needed to be repeated as the effectiveness of these additives varies with solvent. Thus, references will frequently be made to Dr. North's work as it is a solid comparison to the work accomplished here.

## CHAPTER 2. IMPROVING THE UNDERWATER ADHESION SYSTEM

### 2.1 Introduction

When testing a new adhesive, it is necessary to perform structure-function studies. While many structure-function studies have been done previously with poly[(3,4-dihydroxystyrene)-*co*-styrene] in both dry and wet conditions, many of these studies need to be redone to modify the adhesive for optimal adhesion and practicality.<sup>1,13</sup> In comparing to dry adhesion, many of the optimized conditions are not the same, as wet adhesion presents a barrier that many adhesives cannot adjust to. Thus, glues often fail to bind to the surface (adhesive failure) rather than a failure to bind together (cohesive failure). Good adhesion demonstrates a mixture of adhesive and cohesive forces.<sup>22</sup> For poly[(3,4-dihydroxystyrene)-*co*-styrene], some of this adhesive bonding problem is already resolved since the polymer contains DOPA. Thus, it is able to use unique mechanisms to avoid being stymied by underwater conditions.<sup>2,13</sup> The following structure-function tests include a solvent study to adjust density and miscibility, a cross-linking study to examine additional bonding capabilities, and a filler study to modify viscosity and internal binding characteristics. Once these were accomplished and an optimized polymer system was chosen, a slanted adherent study was conducted to test practicality of the system and a comparison study was completed to compare to current commercial adhesive products.

### 2.2 Solvent Study

While previous work with the underwater adhesion system utilized chloroform as the solvent, chloroform may not be the optimum solvent. It is possible by varying the solvents, the density and miscibility of the polymer solution can be adapted. There are key reasons for

adjusting both of these values. By adjusting density, the buoyancy of the solution can be altered. The ideal solution would be neutrally buoyant; however, the best alternative would be a slightly negative buoyancy if neutral buoyancy is not achievable. Both conditions would keep the solution at a consistent level for adhesion at all angles. This will increase the practicality of the system. With miscibility, there are two main considerations. With a slightly miscible solvent system, the solution could be more concentrated during the dispensing process without needing to increase the amount of polymer used. Additionally, it is possible that slight miscibility could help the polymer solution better interact with the surface of the adherent since the miscible solvent would be able to interact with the water on the surface of the substrate. Ideally, the solvent combination used should keep the polymer in solution while in water, have a buoyancy close to neutral (or slightly negative), and demonstrate good adhesion. Additional benefits would include high boiling point/low vapor pressure for long term storage and limited toxicity.

Previously chloroform was used in adhesion testing because it effectively dissolves the polymer and has a high density (1.49 g/mL) so it will sink in water. This makes it useful in laboratory testing of adhesion where substrates are laid at the bottom of a tub. However, in practical uses, chloroform based solutions will not be useful for slanted or upright surfaces. Additionally, chloroform has no miscibility with water. While this enables the polymer to stay completely in solution, it is possible for adhesion to increase with slight miscibility in water. Finally, chloroform is not ideal in terms of long term storage. It has a boiling point of 61.2 °C, which means it will easily evaporate if not properly sealed. Overall, while chloroform does work in an underwater system, there are several disadvantages. Thus, it is worth looking into a more suitable solvent or solvent combination.



To begin the solvent study, dry and wet adhesion tests were conducted with chloroform as the control. Initial testing of this polymer confirmed the previous studies results for dry and wet adhesion. The next step involved a normal solubility test to ensure the various solvents could dissolve the polymer. Afterwards, a simple water test was conducted to assess whether the polymer stayed in solution when injected into water. This process eliminated a majority of the chosen solvents, and those remaining were used to form solvent combinations. The same two steps were repeated with the solvent combinations. The most viable combinations were then used for the standard adhesion testing. All the results from this study are listed in Table 2.1.

The study began with fourteen solvents that were tested individually. These tests were carried out by dissolving the polymer in solution and then syringing the solution into 20 mL vials of salt water. Four solvents were singled out for combination using various ratios.

Table 2.1. Solvents tested for buoyancy

Solvent	Dissolved Polymer?	Color	Stayed in Solution?	Sink or Float?
Acetone*	yes	brown	no	float
Dioxane	yes	brown	no	sink
DMF*	yes	yellow	no	float
THF	yes	yellow	yes	float
Toluene	no	N/A	N/A	N/A
DMSO	yes	yellow	no	sink
Isopropanol*	yes	white	yes	float
Pyridine	yes	yellow	no	sink
Nitromethane	yes	yellow	yes	sink
Chloroform	yes	brown	yes	sink
Dichloroethane*	yes	brown	yes	sink
CCl <sub>4</sub>	no	clear		
Acetic Acid	yes	yellow	no	sink
PEG	yes	yellow	yes	sink

\* indicates future combination studies with varying ratios

Once the four best solvents were identified (acetone, DMF, isopropanol, EDC), varying ratios were tested. Since dichloroethane had the most similar response to chloroform with a slightly lower density (1.25 g/mL), it was chosen as the base for combining with the other three solvents. The three ratios initially tested for each solvent combination were 1:1, 3:1, and 1:3. This resulted in nine conditions that were tested with the same water test as listed above. Of the nine combinations, six showed potential as the solution both sunk and the polymer remained in

solution – the two main requirements for the polymer solution. These six ratios were then used for underwater adhesion. The results of these adhesions tests are shown in Figure 2.1.

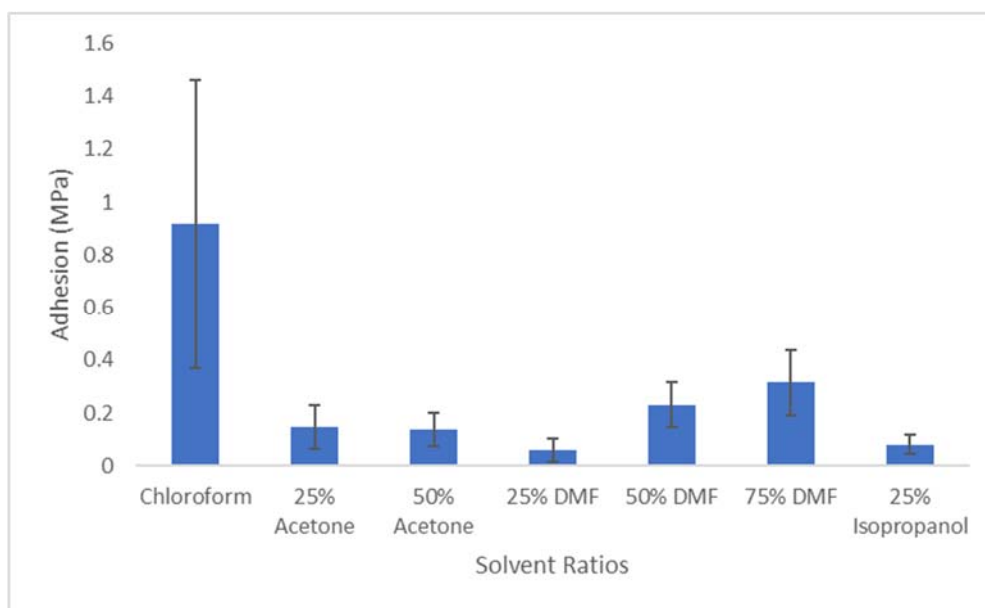


Figure 2.1 Results of solvent study

Both the 1:1 and 1:3 acetone:EDC test showed low adhesion values. However, the DMF:EDC combinations showed an increase in adhesion with an increase in DMF concentration. However, because the density of the solution must remain above 1 g/mL, even higher percentages of DMF were not tested as those would cause the solution to be less than 1 g/mL and thus would float when injected into salt water. While chloroform showed the highest adhesion, the 1:3 ratio of EDC:DMF was the next highest and thus was chosen as the solvent system to continue with for the remaining studies. It was anticipated that the adhesion might increase through the addition of crosslinkers and fillers. Additionally, even if the overall

adhesion of the solvent system was lower, the other properties could make it the better option for practical use.

### 2.3 Crosslinker Study

Crosslinking involves the formation of bonds between polymer chains. Commercial adhesives use these types of bonds to strengthen the adhesive, as crosslinking generally makes polymers tougher.<sup>20</sup> As discussed previously, crosslinking is also present in mussel adhesives as one of the methods used to increase adhesion. There are multiple types of reaction mechanisms that cause crosslinking to occur, including redox chemistry, metal coordination, and hydrogen bonding.<sup>13,20</sup> Since the addition of crosslinkers has the potential to increase adhesion, it is useful to test a variety of crosslinkers to see if they have a positive effect. With dry adhesion of poly[(3,4-dihydroxystyrene)-*co*-(styrene)], tetrabutylammonium periodate was used as a crosslinker at a 3:1 catechol:periodate ratio, and it demonstrated a large increase in the strength of adhesion.<sup>9,12</sup> However, previous underwater adhesion studies with eight different crosslinking agents, including tetrabutylammonium periodate, showed no increase in adhesion.<sup>2,13</sup>

With a new solvent system, it is possible that crosslinking agents may have a different effect. To this regard, this study used the periodate crosslinker chosen based on the previous dry adhesion work. A 3:1 catechol:crosslinker ratio was tested first and had an average adhesion of 0.2 MPa (see Figure 2.2), which was much lower than the average adhesion of the control at 0.4 MPa. Additionally, the color of the adhesive was a brownish color compared to the normal white color present in previous adhesion tests. Based on this color and the work of adhesion graph, it was hypothesized that the 3:1 ratio might be over-crosslinked. This means there is more bonding occurring between the molecules of the polymer and less bonding between the adhesive and the

substrate. Therefore, a 6:1 ratio was attempted next to reduce the possibility of over-crosslinking. The 6:1 ratio demonstrated much higher adhesion with an average of 0.748 MPa. However, based on the high variability of this adhesion as seen with the higher standard deviation, it was determined that overall, the addition of periodate was not significant enough to carry over into future testing with fillers.

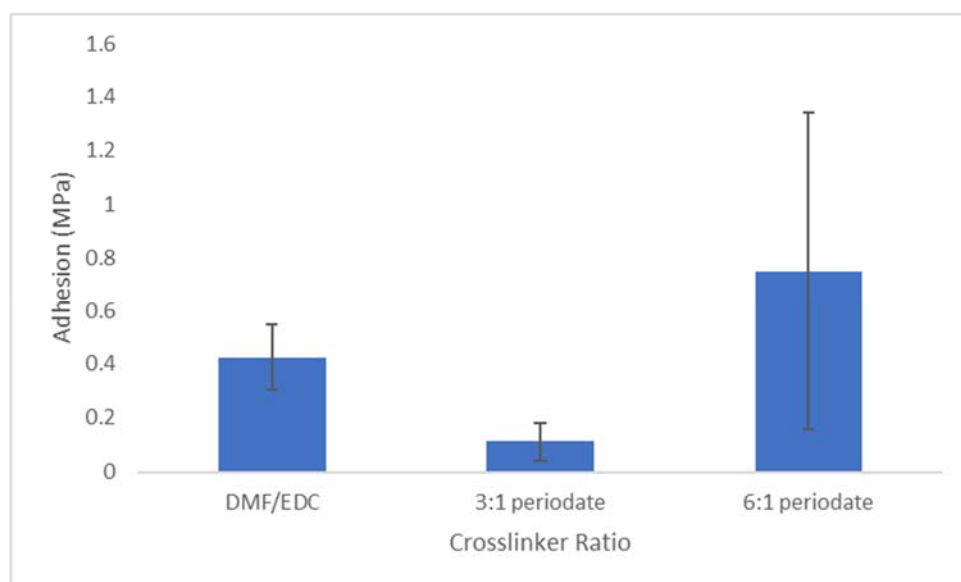


Figure 2.2 Crosslinker adhesion data

## 2.4 Filler Study

Another method for optimizing adhesives/adhesion involves the addition of fillers. Fillers are used in most commercial adhesives to reduce cost of the compound and increase the viscosity of the glue.<sup>12,23</sup> By increasing the viscosity, fillers can yield stronger and more rigid composites. However, there is a large degree of variability with fillers, like the type of material used, particle size, and the concentration in the compound.<sup>22,23</sup> All of these aspects can change the properties of the adhesive, which can impact the adhesion of the polymer either positively or negatively.<sup>12,13</sup>

Fillers were chosen based on previous studies with this polymer system that used limestone with two different particle sizes (70 nm and 30  $\mu\text{m}$ ), glass fibers, cellulose, and laponite. Since iron particles are known to help mussels with their adhesive properties, two different particle sizes of iron were tested as well (25 nm and 10  $\mu\text{m}$ ). Of the seven fillers tested,  $\text{CaCO}_3$ , glass fibers, and cellulose are the primary fillers used for commercial adhesives, typically in the range of 10-50% by weight. Within this study, the main percentages used were 5 wt.%, 10 wt.%, 20 wt.%, and 30 wt.% due to problems with solubility at the higher weight percentages. Additionally, based on poor adhesion during initial tests, some fillers were only done at 5-10 wt.%. All fillers were dissolved along with the polymer and vortexed to ensure even distribution of the filler throughout the polymer solution.

For the most part,  $\text{CaCO}_3$  fillers acted the same in solution with different particle sizes (see Figure 2.3). They visibly increased the viscosity prior to the use with the 20 wt.% turning into a gel-type solid mass. For this reason, 30 wt.% was not attempted. Additionally, with one notable exception, the filler ultimately lowered the adhesion of the polymer compared to the control. However, 5 wt.% of the 70 nm  $\text{CaCO}_3$  displayed vastly higher adhesion around 1.03 MPa. Based on this high adhesion, 1 wt.% was also attempted to determine if an even lower concentration would similarly increase adhesion, but there was no noticeable difference compared to the other concentrations with low adhesion.

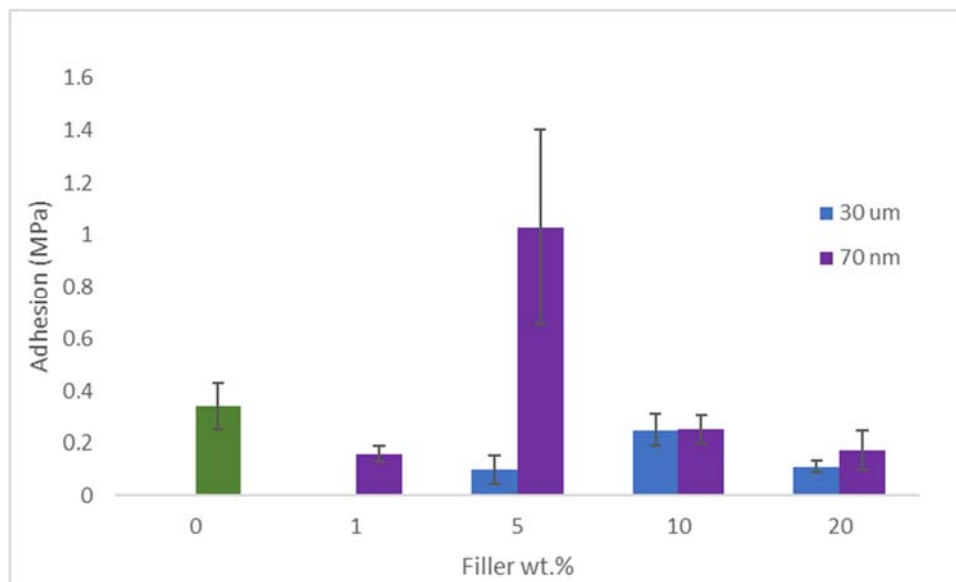


Figure 2.3 Calcium carbonate adhesion

Of the other fillers tested, almost none demonstrated an increase in adhesion (see Figure 2.4). In particular, cellulose had such poor adhesion at 10 wt.% that higher percentages were not attempted. Overall, laponite demonstrated the most uniform adhesion across all four concentrations, but most of the fillers peaked in adhesion around the 10 wt.% mark. The only filler that had any increase in adhesion strength was the iron nanopowder which achieved 0.49 MPa at 10 wt.%. However, when taking the 90% confidence intervals into account, there is no statistically significant difference compared to the control. Thus, it can be concluded that the addition of most fillers does not help with underwater adhesion in this polymer system.

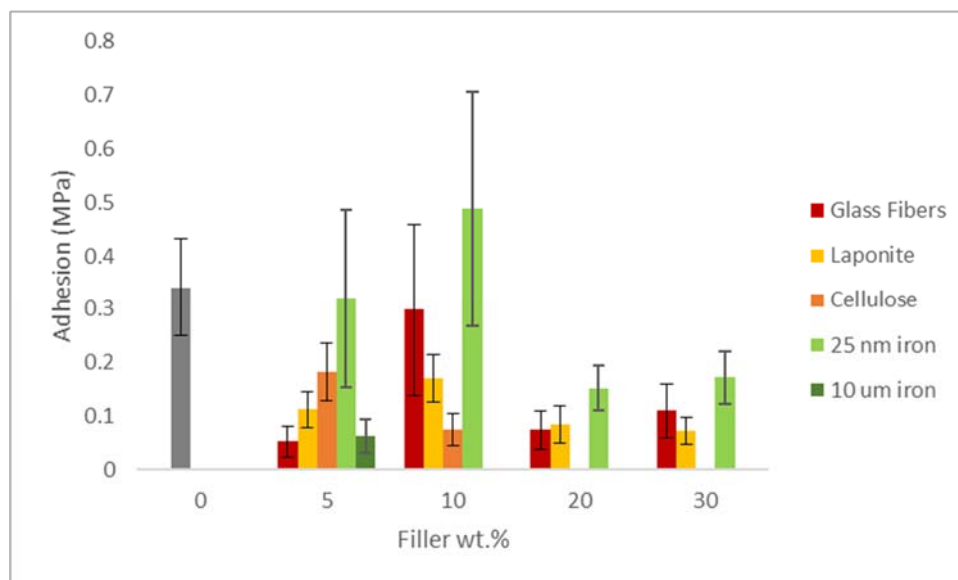


Figure 2.4 Industrial filler adhesion

Since 5 wt.% 70 nm  $\text{CaCO}_3$  was the only filler to demonstrate a significant adhesion increase, it was chosen to use with the remainder of the studies. However, it was first combined with the crosslinker to see if the two worked in sync to increase adhesion or if they resulted in lower adhesion due to competing forces. In the end, the two did not work cooperatively, having an average adhesion of only 0.28 MPa and showing evidence of over-crosslinking. Thus, only the  $\text{CaCO}_3$  was added to the DMF:EDC solvent system. The final study of this paper, slanted adhesion, was done using this system.

## 2.5 Slanted Adhesion

One of the key advantages to using DMF:EDC rather than the previously used chloroform is related to the density of the solution in salt water. Chloroform has a higher density of 1.4 g/mL, making it difficult to use on slanted or upright surfaces due to its tendency to slide off the material before curing can occur. However, the combination of DMF:EDC has a density very



close to salt water at 1.1 g/mL. With only a slightly negative buoyancy, it increases the possibility of using the adhesive on varying surface angles. The initial tests to confirm this ability was done in 20 mL glass vials of salt water. Pieces of aluminum were cut to 2.5 mm and 3.7 mm to create angled surfaces of 35° and 65° respectively. Polymer was then dissolved in solution and applied to these surfaces. Observations were made initially and at 24 hours to determine how much of the polymer remained on the slanted surfaces. The ratios tested included the six ratios tested in the previous solvent study (sec. 2.2). The DMF:EDC samples were the only ones that showed polymer still on the surfaces both initially and after 24 hours. Comparatively, the chloroform control sample rolled off both angles as soon as the solution was applied.

Next, a new bin was set up with a top support, mid support, and base support for proper adhesion testing. Clamps were used to keep the adherents together for the 24-hour cure time. The initial set up tested a 35° angle for slanted adhesion, but a setup was also included utilizing rubber bands to test a completely upright 90° condition. Because the clamps put increased pressure on the system, the 0° slant conditions were redone with clamps rather than the weighted tubes to properly compare to the flat trials. Finally, a slanted study with the system from the flat trials that yielded the highest adhesion (addition of 5 wt.% 70 nm CaCO<sub>3</sub> filler) was also completed.

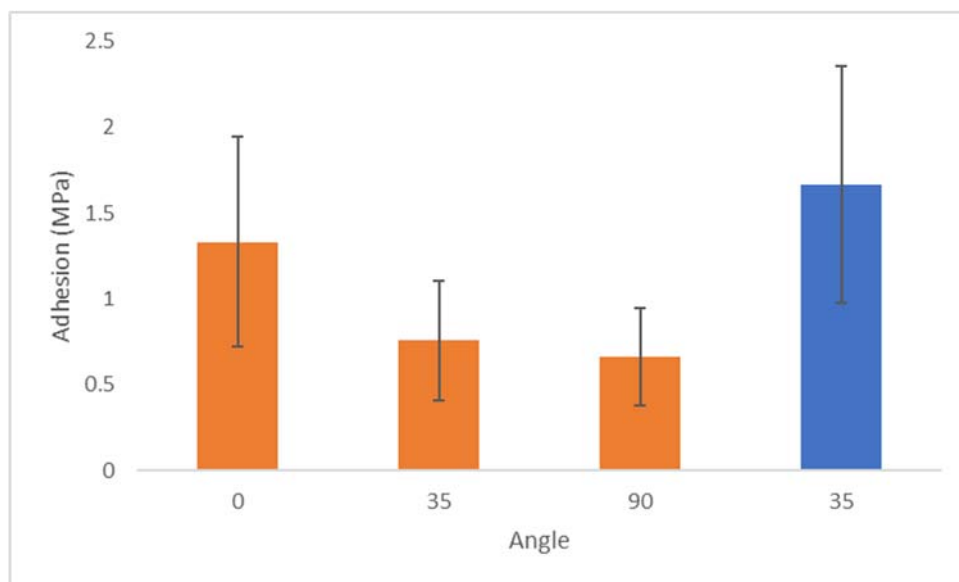


Figure 2.5 Effect of angle on adhesion

As seen in Figure 2.5 above, the adhesion decreased with greater angles. However, this decrease was not significant between the 35° slant and the 90° slant. These had an average adhesion of 0.76 MPa and 0.66 MPa, respectively. Due to these close values, the remainder of the slanted tests were carried out at the 35° angle only. Meanwhile, the addition of the CaCO<sub>3</sub> yielded a comparable increase in adhesion even when slanted, resulting in an average adhesion of 1.7 MPa, which was greater than even the flat adhesion without the filler. Based on the evidence from this test, it can be concluded that this polymer system with the DMF:EDC solvent system can be used on a wide range of slanted surfaces, thereby increasing the practical use of the adhesive.

## 2.6 Commercial Comparisons

As with any new material, it is important to compare to the current products that are available. In this case, there are multiple adhesives on the market that are designed specifically to

work in underwater or wet systems. For this polymer adhesive to be considered useful, it should outperform the current commercial adhesives, or at least demonstrate useful properties that the commercial adhesives do not. However, there tends to be difficulties in making such comparisons due to the varied nature of the commercial products available.

The first key issue involves the form of the product. Poly[(3,4-dihydroxystyrene)-*co*-(styrene)] adhesives are a solvent based system whereas all the commercial adhesives tested are not. Therefore, in order to conduct an accurate comparison study between the two, a change was made in the application of the adhesive while keeping as many parameters as possible constant. Based on calculations, the amount of polymer in 45  $\mu\text{L}$  of the 0.3 g/mL polymer solution is 13.5 mg. Thus, the amount of adhesive applied for all the commercial samples was approximately 13.5 mg. The cure time (24 hours), cure temperature, substrate material, and overall underwater conditions were kept constant. The only change was the method of application. With the high viscosity and tacky nature of the commercial samples, it was impossible to apply via a syringe. Therefore, all the samples were applied using the edge of a needle with weight measurements conducted before and after to confirm the amount of adhesive applied to each sample.

The previous study with this particular polymer system identified 11 commercial adhesives that were tested underwater. Of these 11, only four showed any significant underwater adhesion, as some were completely unable to cure underwater. These four were retested with the slanted system described above. The results of this adhesion study can be seen in Figure 2.6. All tests had 5 samples per test, were done at a 35° slant and were clamped to keep the adherents in place.

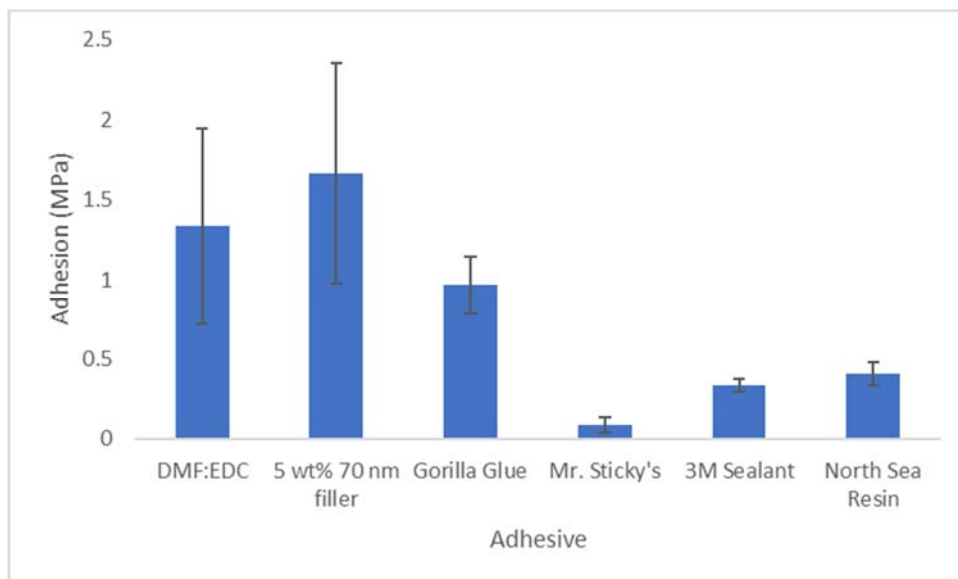


Figure 2.6 Commercial adhesion

Overall, the biomimetic polymer with filler yielded the highest adhesion as compared to all the other commercial adhesives. However, none of the commercial adhesives were able to match even the control polymer system without the filler added. Of the commercial samples, Gorilla Glue displayed the highest adhesion with the other four showing adhesion values of all below 0.5 MPa. Thus, based on this study and the one done previously, it appears the poly[(3,4-dihydroxystyrene)-*co*-(styrene)] adhesives outperform a large number of commercial adhesives available on the market.

## 2.7 Conclusions and Future Work

Based on the work done for this thesis, the previously created poly[(3,4-dihydroxystyrene) *co*-(styrene)] adhesive has been improved by changing the solvent from chloroform to 3:1 DMF:EDC and adding a CaCO<sub>3</sub> filler with a particle size of 70 nm at 5 wt.%. The first change

enables the adhesive to be used more effectively on slanted surfaces and allows the possibility of longer-term storage without evaporating. The addition of the filler merely increased the adhesion for this solvent combination. While the final adhesion of the system was not as high as the previous study, it more than surpassed commercial alternatives and offers better parameters for practical use. Additionally, since lower molecular weight polymers were used in these studies, it is likely that adhesion would increase even more with polymers of a higher molecular weight, thus bringing the adhesion values even closer to the high values obtained with chloroform as the solvent.

While progress was made toward the eventual use of this system in the field, there is still work to be done. Currently, this biomimetic polymer shows high levels of adhesion on one specific substrate – polished aluminum. However, in the field, steel is most often used rather than aluminum, and it may not always be possible to polish the metals to the quality used in the lab. Therefore, it would be useful to test and possibly adapt the polymer system to be able to function on sanded aluminum and either sanded or un-sanded steel. By increasing the number of conditions the adhesive can be used, the more effective it will be solving wide-spread issues.

The next potential issue is the application method. Currently, the polymer solutions are dispensed using glass syringes. This method is not feasible for almost any underwater adhesive application. Therefore, the next step in increasing the practicality of the system would be changing to a plastic syringe application method. Plastic syringes can withstand much more damage without breaking, which is necessary if the polymer is being applied underwater by divers. Additionally, plastic syringes can be used to store adhesives as well. Thus, the solution can be premade (or partially premade) and placed directly into the application device for later use. With this change, it would be important for a specific amount of polymer to be dispensed

accurately and for there to be no contamination or leeching between the polymer solution and the plastic syringe.

Finally, storage considerations need to be taken into account. Most locations would be unable to create the polymer system on site. Thus, the solution needs to be able to be created and stored long term. In particular, this means the solvent cannot evaporate at room temperature. With the change in solvent from chloroform to DMF/EDC, this problem is partially solved as both of these solvents have higher boiling temperatures than chloroform. However, it remains to be determined the shelf life of a solution created with these solvents. More experimentation is needed to see how quickly the polymer degrades or dries out in a variety of conditions.

These are just a few examples of problems that still need to be explored and potential solutions. Without knowing the exact conditions this adhesive will be used for, it is hard to optimize to cover all potential issues with the system. However, any future work will undoubtedly improve the poly[(3,4-dihydroxystyrene) *co*-(styrene)] adhesive even further and continue to make it the strongest underwater adhesive in existence.

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## VITA

Shannon Rebecca Daily came into this world at a ridiculously early hour on the 28<sup>th</sup> of January to parents Sherrill and Allyson Daily. From an early age, she discovered a love of reading and of learning and quickly found multiple avenues to challenge herself. After being accepted into a magnet school for math, science, and research at the start of high school, Shannon attended the Academy of Science in Northern Virginia for the next four years and found her passion in biochemistry with her first major research project on synthetic skin. Graduating from high school in 2012, Shannon made the seemingly crazy decision to pursue a bachelor's degree in biochemistry at the United States Air Force Academy. During her grueling 4 years there, Shannon massively added to her practical skills both in the lab with a wide variety of experiments, and outside of the lab with land and water survival training, sky-diving lessons, and a slew of standard military procedures. Graduating in 2016, Shannon also simultaneously commissioned into the US Air Force as a 2<sup>nd</sup> Lieutenant. With her job as a chemist for the Air Force, Shannon was able to finagle her way to graduate school at Purdue University to complete a master's degree in Chemistry. Having left the majestic Rocky Mountains for the cornfields of Indiana, Shannon became attached to the lab of Dr. Jonathan J. Wilker and worked as free labor on an underwater adhesive study improving a biomimetic glue. Over the course of 18 months, Shannon learned graduate school is not for weak-willed, and it is with this knowledge and the support from her labmates and peers that Shannon graduated from Purdue University with her graduate degree. Her next assignment will be working a joint mission for the Defense Intelligence Agency doing research and development. It remains to be seen if she will continue on in the future for a Ph.D.