
AC 2012-3299: AN EXPERIMENT TO INTRODUCE PH-RESPONSIVE HYDROGELS FOR CONTROLLED DRUG DELIVERY

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**An Experiment to Introduce pH-responsive Hydrogels
for Controlled Drug Delivery**

ABSTRACT

Stimuli-responsive polymers are used in a variety of biomedical applications. For example, pH responsive hydrogels have been extensively investigated for controlled drug delivery. By responding to the pH environment in the body, which changes depending on location and metabolic state, a pH-sensitive drug dosage form is able to modulate drug delivery patterns to meet physiologic requirements and minimize side effects. This paper describes an experiment used to introduce freshmen engineering students to stimuli-responsive polymers for controlled release applications. Students produce a pH responsive hydrogel, made from polyethylene glycol grafted onto a polymethacrylic acid backbone p(MMA-EG) using free radical polymerization. These hydrogels were previously examined for oral delivery of insulin for diabetics by Nakamura et al¹. In our experiment, the swelling capabilities of the hydrogels in different pH environments are examined as a function of crosslink density. In future experiments, hydrogel mechanical properties and release properties, as a function of these variables, will be examined. In addition to learning about pH-responsive drug delivery, students will learn concepts of polymer chemistry, materials science, design of experiments, data analysis, and engineering design. An assessment plan will measure student mastery of learning outcomes specific to the field of biomaterials science and those set forth by ABET for undergraduate chemical engineering programs.

Keywords: pH Responsive Hydrogels, Oral Insulin Delivery, Diabetes, Controlled Drug Delivery

INTRODUCTION

Diabetes is a disease which affects millions of people around the world. It is classified into two major types. Type 1 Diabetes is an auto-immune disease in which, insulin-producing beta-cells within the pancreas are destroyed, resulting in insufficient insulin production by this organ. With type 2 diabetes, the body has developed a rejection to insulin and that glucose uptake cannot be regulated within a patient's muscle and fat cells. When glucose uptake cannot be regulated naturally, blood sugar levels can rise or fall to unacceptable levels and can lead to a life-threatening diabetic coma if not carefully monitored and treated with regular insulin doses.

Diabetes is a self-managed disease, and patients must administer insulin periodically by injection to maintain blood glucose levels. Injections are painful and inconvenient, and patient non-compliance is a serious concern. The mean rate of insulin usage has been reported to be 77.44% of the prescribed dose in patients with Type I diabetes who use periodic bolus injections². A convenient and painless oral dosage form could help increase patient compliance, however, insulin will be degraded by the acidic pH of the stomach before absorption into the blood stream. For this reason, researchers have focused on the development of pH responsive hydrogel drug delivery systems for oral delivery of insulin and other drugs^{3, 4, 5}.

Hydrogels are extremely hydrophilic crosslinked polymer networks that can absorb large amounts of water. They are largely used in many biomedical applications such as contact lenses, and the physical properties of hydrogels are very similar to living tissues in comparison to other synthetic biomaterials due to their high water content and rubbery properties. Hydrogels have been widely used for drug delivery systems since they allow molecules of

different sizes to diffuse into or out of the network for drug loading and release, respectively. Since the polymer chains of different hydrogels contain specific functional groups, hydrogels can be sensitive to changes in the surrounding environment, such as the changes in pH, temperature, and pressures. A successful pH-responsive hydrogel for insulin delivery would exhibit very little swelling in the low pH of the stomach, thereby restricting the release of the drug. In the higher-pH environment of the stomach, the hydrogel would swell, and the relaxation of the gel network structure would allow release of the drug by diffusion. Furthermore, mechanical properties of hydrogels can be modified by making relatively simple changes to the polymer structure, such as the crosslinked density.

Nakamura et al. reported a hydrogel made with polyethylene glycol chains grafted onto a polymethacrylic acid backbone p(MMA-EG) that can be used to transport insulin through the stomach and into the near neutral pH environment of the intestines, where it can be effectively absorbed into the bloodstream¹. While this hydrogel has pH-responsive properties, there are other properties that need to be considered as well. The hydrogels must have the mechanical integrity necessary to make it through the stomach and reach the intestines intact. If the integrity of the gel structure is not maintained during transport through the GI tract, then some insulin could be released in the stomach. In the intestine, the hydrogel must release the drug gradually to avoid a burst of drug in the upper small intestine, which could result in decreased bioavailability by degradation.

Chemical engineers contribute to the design of controlled drug delivery systems which deliver a drug at a desired rate to a desired location in the body. Here, we describe an experiment in which students prepare pH-responsive hydrogels based on p(MMA-EG) based on the experiments outlined by Nakamura et al.¹ The properties of the hydrogels, such as swelling ratio, mechanical properties, and release characteristics are characterized as a function of pH and crosslink density. Students performing this experiment have the opportunity to vary the hydrogel structure in attempt to optimize the hydrogel for oral insulin delivery. Through this experiment, students will gain hands-on experience in an environment that mimics an undergraduate research experience. They will practice identifying important design variables, in this case, for drug delivery. The students will also practice translating quantitative laboratory measurements into data that can be used to evaluate a design. Lastly, they will learn aspects of polymer design and characterization, which is translatable to other areas of material science and engineering.

EXPERIMENT

In this experiment, students produce pH sensitive hydrogels by photo-polymerization of a monomer solution containing the photo-initiator dimethoxy propyl acetophenone, the cross-linker tetraethylene glycol dimethacrylate, and monomers methacrylic acid, and poly(ethylene glycol) (n) monomethyl ether monomethacrylate, as described by Nakamura et al¹. **Error! Bookmark not defined..**

Hydrogel Synthesis

Materials

- Dimethoxy propyl acetophenone (DMPA)
- Methacrylic Acid (MAA)
- Tetraethylene glycol dimethacrylate (TEGDMA)

- Poly(ethylene glycol) (n) monomethyl ether monomethacrylate (PEGMA)
- Ethanol (50wt%)
- Plain Glass Microslides (75x50 mm)
- Pasteur Pipette
- Micropipettes
- 50 mL Erlenmeyer flask
- Teflon sheet (0.8 mm thick)
- Small Binder Clips
- 365-nm ultraviolet light (Blak Ray, Upland, CA)

Procedure

1. In an Erlenmeyer flask, MAA, TEGMA, DMPA, PEGMA, ethanol were added in varying amounts described in Table 1. Compositions were chosen based on those proposed by Nakamura et al¹, but were modified to produce the hydrogels with the highest degree of mechanical integrity. The crosslink density of the hydrogels was modulated by varying the TEG crosslinker amount in the reaction mixture from low to high (1.04mL to 1.56mL to 2.08mL) in order to investigate the effects of hydrogel properties.

Table 1. Compositions of hydrogels made and their durability in pH 7.4

Mass of PEGMA(g)	Mass of MAA(mL)	Mass of TEGDMA(mL)	Mass of DMPA(g)	Mass of EtOH(g)
1	1.9233	1.04	0.0204	4.0881
1	1.9233	1.56	0.0233	5.0950
1	1.9233	2.08	0.0261	5.7163

2. The solution was mixed well, ensuring that all of the solid particles dissolved.
3. Strips of the Teflon sheet were cut and used to keep the two microscope slides spaced 0.8mm apart.
4. Binder clips were used to hold the microscope slides together.
5. Using the Pasteur pipette, the monomer solution was transferred between the microscope slides to completely fill the space between slides.
6. The microscope slides with solution were then placed underneath the UV lamp and the lamp was turned on.
7. The solution was left underneath the UV lamp for 30 minutes.
8. The UV lamp was turned off, and the hydrogel was removed from the microscope slides.
9. The hydrogel was placed in DI water for 24 hours to rinse off any possible residual monomer solution that did not polymerize.

Swelling Study

Materials

- Synthesized hydrogels
- Phosphate buffered saline (PBS) solution, pH 2.2, 5.0 or 7.4.
- 3-50mL containers

- Stopwatch

Procedure

1. Hydrogels were broken into approximately 1x1” pieces and dried under vacuum.
2. The dry mass was recorded.
3. Each piece was placed into a separate 50mL container containing PBS solution.
4. The wet mass was recorded every 10 minutes for the first hour, then at 2, 4 and 24 hours.

Experimental Calculations

The mass swelling ratio (MSR) of the hydrogels at any time point can be calculated by using Equation 1.

$$\text{MSR} = \frac{M_g}{M_d} \quad [1]$$

where MSR is mass swelling ratio, M_g is mass of gel in air, and M_d = initial mass of the dried gel in air. Values for MSR as a function of time, TEGMA concentration, and pH were compared statistically with a student’s t-test and a 95% confidence interval to determine significance.

RESULTS AND DISCUSSION

Figure 1 shows the mass swelling ration (MSR) as a function of pH for the hydrogels prepared with 2.08 mL of TEGDMA in each pH environment. After 24 hours, the hydrogels in the pH of 7.4 had higher MSR than those in pH 2.2 and 5.0 ($p < 0.05$). Similar results were seen for the TEGMA concentrations.

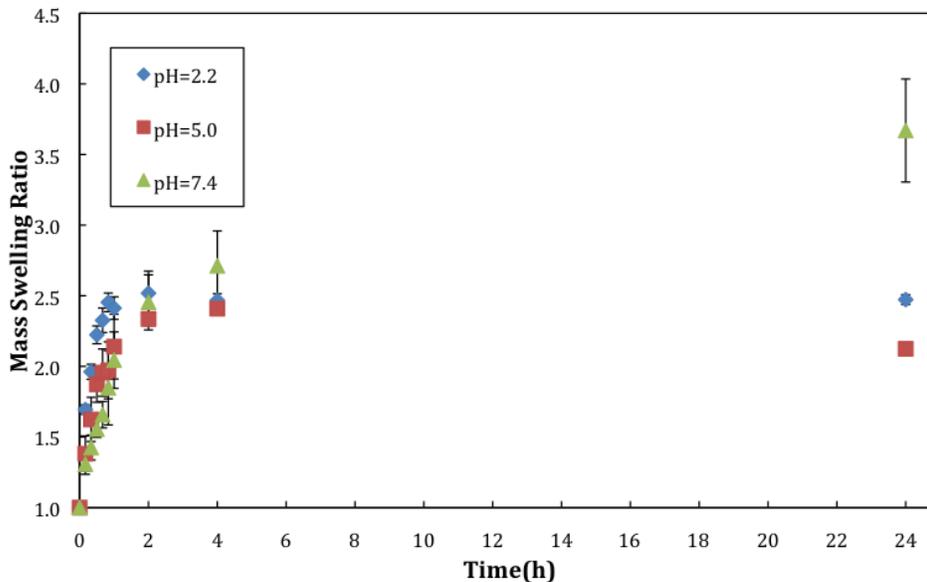


Figure 1. MSR vs. Time for 2.08 mL TEGDMA formulation in various pH environments. The error bars are plus or minus one standard deviation for each set.

This result is consistent with what was observed previously by researchers who characterized this system. At low pH values, interpolymer complexes form between the carboxylic acid protons on the PMMA and oxygen on the PEG chains. At higher pH values, the acid groups on the PMMA ionize, causing the polymer complexes to dissociate due to ionic

repulsions, allowing the network to accommodate more water⁶. At 24 hours, hydrogels in a pH of 2.2 had an average MSR of 2.27, hydrogels in a pH of 5.0 had an average MSR of 2.32, and hydrogels placed into a pH of 7.4 had an average MSR of 3.95.

MSR vs. time as a function of TEGMA concentration is shown in Figure 2 at the neutral pH of 7.4. It was expected that increasing TEGMA concentration, or increasing crosslink density, would decrease the amount of water that can be accommodated in polymer network at equilibrium.⁷ However, for each pH, no statistical differences ($p < 0.05$) were seen in MSR for the hydrogels at any time point. In future work, a broader range of crosslinker concentrations will be studied.

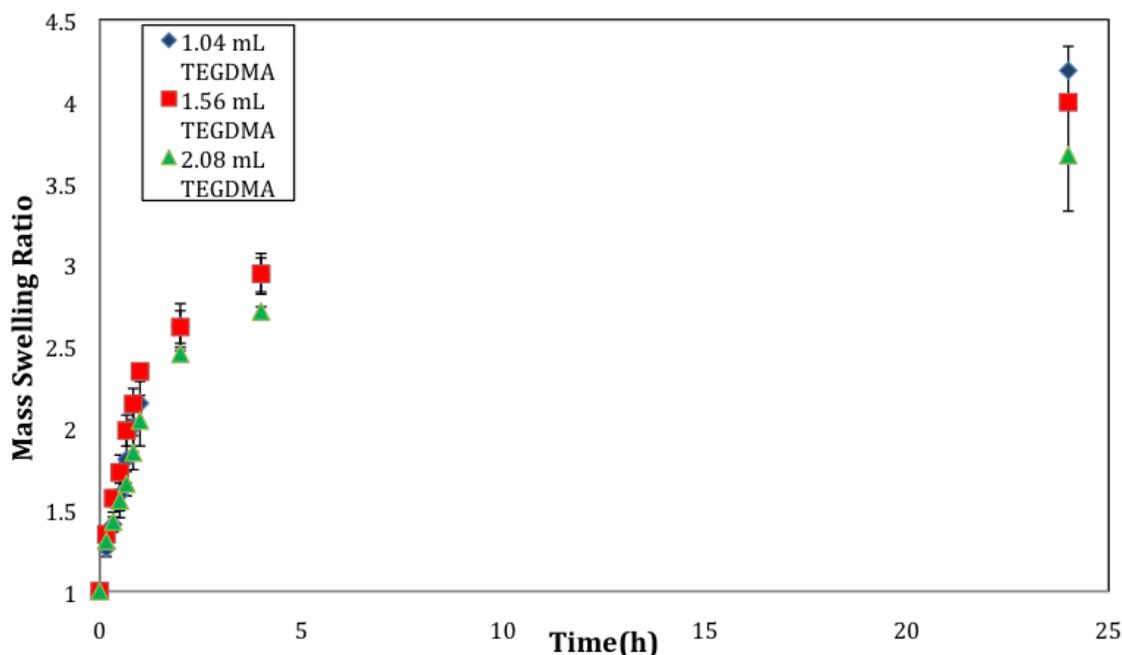


Figure 2. MSR vs. time for each hydrogel composition at pH=7.4

SUMMARY AND RECOMMENDATIONS

In this paper, we present a laboratory activity on biomaterials and drug delivery for undergraduate engineering students. This aspect of our project focused on structure-property relationships in pH sensitive hydrogels for oral insulin delivery. In upcoming work, the activity will be expanded to include rubber elasticity experiments for the calculation of network mesh size as a function of pH and TEGMA concentration. In addition, drug release and mechanical properties will be evaluated as a function of these variables. Through this hands-on activity, students will not only develop skills specific to drug delivery and biomaterials, but in data acquisition and analysis and engineering design. The laboratory will be implemented into a freshman-level laboratory during Spring 2012. At this time, pre and post-tests will be used to gauge student mastery of learning outcomes specific to the field of biomaterials science and those set forth by ABET for undergraduate chemical engineering programs.

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