Developing Hands-On Biomaterials Education Modules Using Alginate Hydrogels

Victoria G. Muir, Kayla Brown, Julio A. Rivera De Jesus, Dr. Jason A. Burdick
Department of Bioengineering, University of Pennsylvania, Philadelphia, PA

Statement of Purpose: Hands-on learning experiences are known to greatly enhance students' understanding of STEM concepts at every age, from K12 to university education. Here, we present four integrated hands-on education modules for teaching hydrogel biomaterials concepts using safe, cost-efficient resources (Fig. 1). The activities are designed for a high school audience, drawing on concepts from high school STEM education and introducing the concepts of biomaterials, tissue engineering, and mechanical testing in engineering.

Module 1: *Alginate hydrogel formation.* In this activity, students use alginate to form a hydrogel, through crosslinking in the presence of divalent cations (i.e., Ca^{2+}). This straightforward ionic crosslinking mechanism has led to the widespread use of alginate hydrogels in the biomaterials field[1], including for education tools to teach concepts in polymer chemistry and fluid dynamics.[2,3] The learning objectives for this module are 1) define "hydrogel" and "crosslinking", and 2) explain how an alginate hydrogel forms with ionic crosslinking. For setup, mix food-grade alginate (Modernist Pantry) in water (~2g/100mL) with a hand mixer, and allow it to dissolve overnight. Food dye can be added to the alginate solution to aide in visualization. Just before the activity, dissolve food-grade calcium chloride ($CaCl_2$, Modernist Pantry) in water (~5g/100mL). In a separate container, dissolve food-grade sodium chloride (NaCl) in water (~5g/100mL). In the activity, students will use a squeeze bottle to add alginate solution to either 1) water, 2) NaCl solution, or 3) $CaCl_2$ solution. The students can record their observations, noting that alginate hydrogels only form when added to $CaCl_2$ solution due to the divalent cation crosslinking the alginate polymers. To demonstrate the concepts of diffusion, students can compare alginate hydrogels that have been immersed in $CaCl_2$ solution for either 10 s or 2 min, noting how longer immersion in $CaCl_2$ leads to hydrogels with increased stiffness due to diffusion of Ca^{2+} further into the hydrogel (observed qualitatively by squishing hydrogel). This module can be accompanied by video demonstrations of other crosslinking methods (i.e., photocrosslinking).

Module 2: *Compression testing of hydrogels*. The behavior of hydrogels and tissue under compressive and tensile loading is important for many biomaterials applications.[4] In this module, students use a simple Arduino-based compression testing device to quantitatively characterize stiffness of hydrogels.[5] The learning objectives are to define and explain compression testing, stiffness, and force-displacement relationships, as well as explaining how varied crosslinking density affects hydrogel stiffness. A simple compression testing device can be assembled using an Arduino microcontroller, force sensitive resistor, and a few electronic parts. For setup, prepare discs (~1 cm diameter, ~1 cm tall) of alginate hydrogels by immersing a cylindrical mold full of alginate solution into a bath of $CaCl_2$ for 1 hour. To vary the stiffness of alginate hydrogel samples, prepare $CaCl_2$ baths of various concentrations (0.1-10 wt.%). Using the force sensor, students can compress the alginate hydrogel discs and record the maximum force sustained before the hydrogel ruptures. Comparing the force across testing groups allows for discussion around varying the stiffness of hydrogels.

Module 3: *Tensile testing hydrogels.* In this activity, students create an alginate hydrogel cylinder (or, "worm") by extruding alginate solution into a bath of $CaCl_2$. The students can then take the hydrogel cylinder and mark two dots about 1 cm apart using permanent marker onto the hydrogel. Using a slow-motion phone camera, students can capture stretching the alginate hydrogel with the dots and a ruler in view until the hydrogel breaks in the center. Using the videos and images, students can determine the percent stretching of the hydrogel before breaking by measuring the initial and final distance between dots on the hydrogel cylinder. The learning objectives of this module are to define and explain tensile testing and material failure. Both Modules 2 and 3 can be accompanied by videos of mechanically testing hydrogels and tissue in lab.

Module 4: *Applying hydrogels for biomaterials applications.* This module introduces the applications of bioprinting and drug delivery using hydrogels. For setup, mix a dark food dye color (blue, purple) in excess in the alginate solution. First, to introduce the concept of 3D printing, students will use a squeeze bottle to extrude an alginate design onto a Styrofoam plate ("hand" 3D printing), which is subsequently submerged in a bath of $CaCl_2$ to stabilize the structure. Then, using a phone camera, students will capture the food dye diffusing into the surround $CaCl_2$ bath over a period of 10-20 minutes. Using a free online color picker, students can quantify the intensity of food dye in the bath over time, creating a release curve. This module can be accompanied by videos of 3D printing and data on drug release from hydrogels.

Conclusions: Here we present four integrated hands-on education modules for teaching hydrogel biomaterials concepts designed for a high school audience. These activities can also be adapted for broader use in K12 education and university coursework.

References: 1) Lee+, *Progress in Polymer Science*. (2012). 2) Erdal+, *J. Chem. Educ.* (2019). 3) Boyd, *ACS Polym. Au.* (2021). 4) Guimarães+, et al. *Nat. Rev. Mat.* (2020). 5) Adafruit, "Using an FSR." (2021).

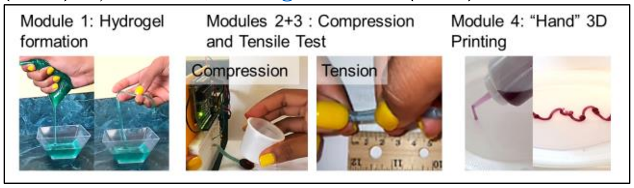

Figure 1. *Hands-on education modules for hydrogel biomaterials concepts, including 1) crosslinking an alginate hydrogel with $CaCl_2$, 2) compressing a hydrogel with an Arduino-based force sensor apparatus, 3) tensile testing a hydrogel using markers on the gel, and 4) "hand" 3D printing with hydrogels.*