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### UV-crosslinking of PhotoHA hydrogel solubilized using solvents of varying pH

Solvents Used: 1x PBS (typical), 0.08 NaOH, 20 mM Acetic Acid

#### Procedure

1. 5 mL solvent added to 50 mg methacrylated HA powder
2. Mixed on shaker table in the fridge (between 2-10C) for 60 min
3. Photo initiator solubilized by adding 1 mL methanol to 100 mg C12H16O4 (Irgacure photoinitiator)
4. 50 microliters added to each HA solution
5. 4 mL of each solution pipetted into a foil cup for crosslinking
6. Crosslinked under 365 nm UV light for 5 min

Observations (as compared to the typical solvent - 1xPBS)

#### 0.08 NaOH

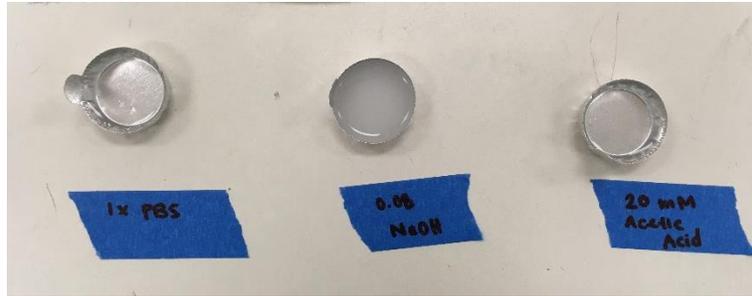
- Solubilization: NaOH solution seemed to solubilize the HA powder well and final solution was clear and comparable to 1xPBS. However, the solution was significantly less viscous than 1xPBS solution and once the solution was pipetted into the foil cup the solution began to cloud and produced a small ring of bubbles around the edge.
- Crosslinking: NaOH solution did not crosslink. However, approximately 10-20 min following exposure to UV light the solution went clear again.

#### 20 mM Acetic Acid

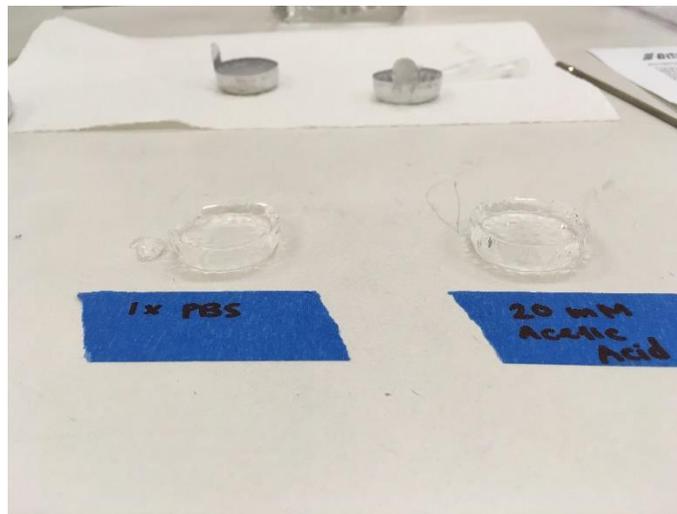
- Solubilization: Acetic Acid solution solubilized comparably to 1xPBS. Both solutions were clear although the Acetic Acid solution was slightly more viscous.
- Crosslinking: Acetic Acid solution likely crosslinked more than 1xPBS. While both looked visually identical, the formed photogel from Acetic Acid was more firm and retained its shape better under pressure. Acetic Acid gel also retained its liquid better. On touch, the acetic acid gel left little to no liquid residue on the glove while 1xPBS left a clear coating of liquid. This was also seen when the gels were removed from the foil cups: the 1xPBS gel was slippery and left a liquid film underneath while the acetic acid gel did not.



*Figure 1. Before UV exposure, left to right, 1xPBS, 0.08 NaOH, 20 mM Acetic Acid.*



*Figure 2. After UV exposure, left to right, 1xPBS, 0.08 NaOH, 20 mM Acetic Acid.*



*Figure 3. Formed photogels of 1xPBS and 20 mM Acetic Acid solvents*

Future tests need to be performed to determine if the pH affected the ability of the HA to crosslink, or if the pH affected the functionality of the photoinitiator (irgacure 2959).

It is recommended to repeat the tests using LAP (water soluble photoinitiator) at 405nm light.