The Problem: Pathologic demineralization destroys bones and teeth, and it affects humans of all ages. Biomineralization of these tissues is the ultimate solution, but the behavior of precursors that can induce mineralization is not completely understood. Therefore, the purpose of this project is to test the biomineralization ability of Hydroxyapatite (HA) and two bioglasses (40S5 and 45S5) at different concentrations for different periods of time. The extent of mineralization based on the size of the particles and crystal structure of the precursors is also examined. Finally, the calcium release after mineralization is assessed.

Introduction:

- Biomineralization is the process of minerals deposition in a biological matrix. Here, collagen is used as a matrix to be mineralized by precursors such as synthetic hydroxyapatite (HA) and bioglasses (40S5 and 45S5).
- HA has nanoparticles (less than 100 nm) that are highly crystallized. 40S5 is also nanoparticles (120 nm), although not completely amorphous, it is not as crystallized as HA. 45S5 share similar crystallinity as 40S5, but its particles are microparticles (10 µm).
- HA and 45S5 precursors have been tested in vivo and in clinical studies, but their particle size and crystallinity limit their mineralization ability. Thus, 40S5, which shares similar chemical composition as 45S5, but differs in size and in the method of synthesis, is being tested against HA and 45S5 to determine if better collagen mineralization can be obtained.

Methods:

- Two different TRIS buffer solutions were prepared (Calcium TRIS buffer and Phosphate TRIS buffer) to simulate bodily fluid with a pH of 7.4
- Collagen tape was used in the mineralization process by placing a strip (10 mm x 3 mm) of collagen in a 50:50% vol Ca buffer and PO4 buffer.
- The control did not contain any of the precursors.
- The samples contained a precursor with the following concentrations: 0.5 mg/mL, 1 mg/mL, and 2 mg/mL.
- Each concentration had 4 replicas.
- Samples were sonicated to dissolve particles. Then, placed on the shaker for 1 hour.
- 1/3 of the collaged tape strip was cut and placed in ethanol, then dried in the oven.
- The process was repeated at the 2nd and 3rd hour.
- Collagen mineralization was assessed by means of FT-IR.
- The same process was repeated to assess calcium release, but after 1 hour on the shaker, collagen was degraded with 0.25N HCl, then buffered with 0.5N NaOH.
- Calcium released was analyzed spectrophotometrically.

Results:

- Figure 3. For all concentrations and for the 3 hours, 40S5 showed The most mineralization followed by HA followed by 45S5. Mineralization is assessed based on the intensity of PO4²⁻ present at 1045 cm⁻¹.
- Figure 4. 40S5 and 45S5 did not show improvement of mineralization as time progresses. However, HA increased mineralization with time.

Discussion:

This study lays down the foundation of in vitro mineralization of collagen. Next step will be applying the knowledge about speed of mineralization and the minimum concentration of precursor needed to cause mineralization to demineralized teeth.