Bioactivity of Amorphous Bioactive Glass and Glass-Ceramic in Simulated Body Fluid

Casey Lewis, Janet Krevolin.
Bio2 Technologies Inc.

Statement of Purpose: *In vitro* testing of biomedical devices is very desirable for companies and researchers in order to gain valuable knowledge in a short period of time and for minimal capital. It has been shown that various materials bind to living bone through a layer of hydroxyapatite (HAp), and this layer can be produced *in vitro* in an acellular and protein-free environment by means of simulated body fluid (SBF) which has ion concentrations nearly equal to human blood plasma[1]. Therefore, hydroxyapatite formation on the surface of implant material after having been soaked in SBF is a good indicator of bioactivity of the implant and how the implant will react *in vivo*. This experiment was designed to evaluate differences in bioactivity due to sample crystallinity and bulk porosity.

Methods: The samples were produced by sintering bioactive glass similar to 13-93 in a fiber and powder form, using organic poreformers as sacrificial porosity enhancements and as a binder during formation. Various relative quantities of the ingredients and sintering temperatures were used to produce five different test batches: 700 Low, 700 High, 800 Low, 800 High, and 900 High, where the number is the sintering temperature (˚C) and the Low/High is the relative porosity at that temperature. Samples were machined under non-aqueous conditions on a lathe to 4mm diameter cylinders; the ends were cut using a diamond blade circular saw making the length 10mm. Samples were submerged in 50mL of Simulated Body Fluid (SBF) at 36.5˚C and placed on a shake plate at 90 rpm. The SBF had an initial pH of 7.40. Five samples were soaked for each time period of 0, 1, 7, 14, and 24 days, resulting in a total of 125 samples. Samples were compression tested in house using a Satac (Grove City, PA) Ultimate Testing Machine (UTM). Scanning Electron Microscopy (SEM), Energy-Dispersive X-Ray (EDX) Spectroscopy, and X-Ray Diffraction (XRD) analysis done at Alfred University, Alfred, NY.

Results: XRD analysis of each batch showed the 700˚C batches were completely amorphous while the 800˚C and 900˚C batches had crystal phases. The major phases in the 800˚C batches were β-Wollastonite ($CaSiO_3$) and Combeite ($Na_4Ca_3Si_6O_{16}(OH)_2$) while the 900˚C batch had only β-Wollastonite. The crystallinity of the 800 Low is: 20% Wollastonite, 11% Combeite, 10% minor phases, total 41%; 800 High is: 13% Wollastonite, 12% Combeite, 9% minor phases, total 34%; 900 High is: 24% wollastonite. SEM/EDX was used to visually and chemically observe hydroxyapatite formation. Figure 1 is an example of chemical changes with the increases of CaO and P_2O_5 showing HAp formation and visibly in Figure 2. The other batches had similar SEM images showing HAp formation. The EDX data for the other batches was less correlative to the SEM images suggesting formation of HAp was less than the 800 High batch. The compressive strength of the samples ranged from 102 MPa for 700 Low at 16% porous to 39 MPa for 900 High at 59% porous both at time 0. The pH of the SBF increased from 7.40 to about 7.85 after 4 weeks.

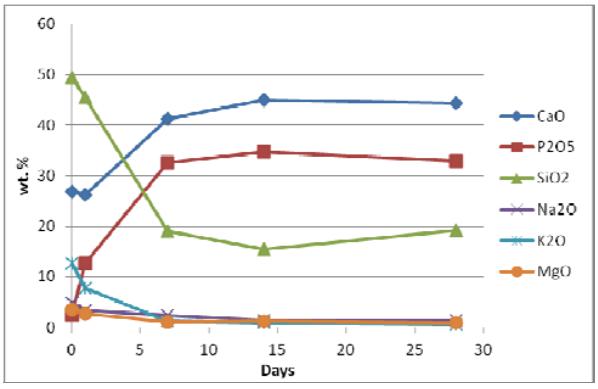

Figure 1. EDX results of 800 High batch showing changes in sufrace oxide weight percentages over the duration of the study due to leaching and HAp formation.

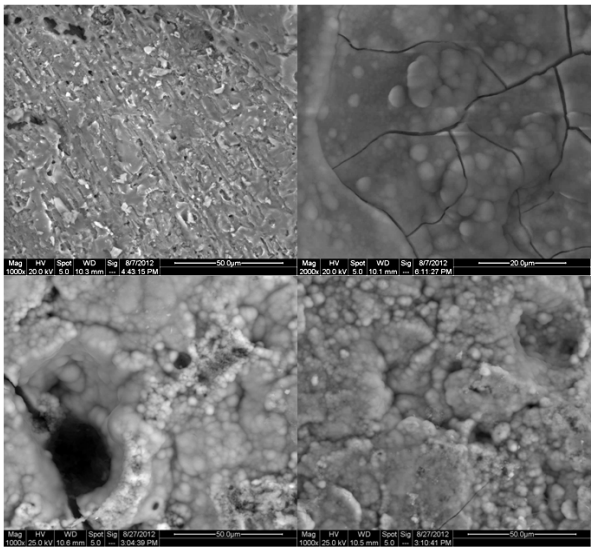

Figure 2. SEM images at a) 0 day, b) 1 day, c) 2 weeks, and d) 4 weeks showing surface HAp formation.

Conclusions: Each of the five batches tested exhibited bioactivity through hydroxyapatite formation on the surface of the samples. Porosity of the samples did not affect surface HAp formation but did have an effect on sample crystallinity. The 800˚C High porosity batch had very good hydroxyapatite formation while still having nearly a 40 MPa compressive strength after 4 weeks. This shows that the overall increased crystallinity does not have adverse effects on hydroxyapatite formation in SBF, but actually appears to increase the hydroxyapatite formation due to more bioactive crystal phases and a less weight percentage of wollastonite compared to the 900˚C High porosity batch. However, this does not suggest how the material will perform *in vivo* nor does it suggest how quickly the material will resorb.

References: [1] Kokubo T. Biomaterials 2006; 27:2907–15.