Reactions and Surface Transformations of a Bone-Bioactive Material in a Simulated Microgravity Environment

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A comprehensive program to investigate the expeditious *in vitro* formation of three-dimensional bone-like tissue is currently underway at the University of Pennsylvania. The study reported here forms a part of that program. Three-dimensional bone-like tissue structures may be grown under the simulated microgravity conditions of NASA designed Rotating Wall Bioreactor Vessels (RWV's). Such tissue growth will have wide clinical applications. In addition, an understanding of the fundamental changes that occur to bone cells under simulated microgravity would yield important information that will help in preventing or minimizing astronaut bone loss, a major health issue with travel or stay in space over long periods of time.[1] The growth of three-dimensional bone-like tissue structures in RWV's is facilitated by the use of microcarriers which provide structural support. If the microcarrier material additionally promotes bone cell growth, then it is particularly advantageous to employ such microcarriers.

We have found that reactive, bone-bioactive glass (BBG) is an attractive candidate for use as microcarrier material.[2] Specifically, it has been found that BBG containing Ca- and P- oxides upregulates osteoprogenitor cells to osteoblasts.[2] This effect on cells is preceded by BBG reactions in solution which result in the formation of a Ca-P surface layer. This surface further transforms to a bone-like mineral (i.e., carbonated crystalline hydroxyapatite (c-HA)). At normal gravity, time-dependent, immersion-induced BBG reactions and transformations are greatly affected both by variations in the composition of the milieu in which the glass is immersed and on the immersion conditions.[3] However, the nature of BBG reactions and phase transformations under the simulated microgravity conditions of RWV's are unknown, and have to be understood in order to successfully use BBG as microcarrier material in RWV's. In this paper, we report some of our recent findings in this regard. We have employed experimental and numerical methods.

BBG behavior in physiological solutions was tested in simulated microgravity and compared with that at normal gravity. On the basis of our numerical study,[4] we have chosen the BBG granule size to be in the range 40-70 µm, and a RWV rotational speed of 10 rpm. Changes in the solution composition and the material surface were analyzed after immersion.

Materials and Methods

Immersion experiments were conducted in buffered solution (pH 7.4 at 37 °C) both in the simulated microgravity environment of a High Aspect ration Vessel (HARV-50 ml, Synthecon, Houston, Texas) and in unit gravity. BBG granules (40-70 µm) were immersed at 1 mg/ml weight-to-solution volume ratio for 1, 3, 6, 10 and 24 hours. After immersion, the solutions were analyzed for changes in the Si, Ca and P-concentrations using atomic absorption spectroscopy and colorimetry. BBG surface was analyzed prior to and after immersion using Fourier transform infrared technique (FTIR).

Results and Discussion

Temporal changes in the Ca, P and Si-concentrations were observed upon the immersion of BBG granules into the buffered solution, both in the simulated microgravity environment of the HARV and at unit gravity. The data suggests that the following events have occurred in both environments: an initial increase in the concentrations of all ions, indicative of dissolution reactions; subsequent uptake of P-ions suggesting precipitation of Ca-P phases, and leveling of Si-concentration, indicative of the solution saturation with Si. The data also reveals that although the sequence of events was similar under both conditions, the kinetics of these reactions were significantly affected by simulated microgravity as indicated by a remarkable increase in the rate and the amount of ion release. The precipitation of CaP phases was also enhanced. FTIR analysis of immersed BBG granules detected the formation of bone-like crystalline apatite (c-HA) after 6 hours of immersion in both environments; however, the apatite layer was significantly thicker on granules which reacted under simulated microgravity in comparison to those in unit gravity.

The study has demonstrated that the simulated microgravity environment remarkably enhances the kinetics of BBG reactions that are associated with bone-bioactive behavior.

References

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