Development of Biomimetic Alendronate Sodium loaded CDHA coating

Huan Zhou[1], Ahmed Touny[2], Joseph Lawrence[1], Darcy Wagner[1], Sarit Bhaduri[2]
[1]Department of Bioengineering, [2]Department of Mechanical, Industrial & Manufacturing Engineering, University of Toledo, Toledo, OH, 43606

Statement of Purpose: The main objective of this paper is to develop a dual purpose biomimetic coating of Carbonated Calcium Deficient Hydroxyapatite (CDHA) loaded with Bisphosphonates (BPs). There are two interesting aspects of this project. First, since CDHA resembles the bone mineral as opposed to Hydroxyapatite (HA), CDHA is more bioactive and enhances osteoblast proliferation [1]. Second, Bisphosphonates (BPs) such as Alendronate Sodium (AS) have already been used in treatment of Osteoporosis [2]. There are drawbacks related to conventional oral administrations of BPs described elsewhere [3]. Hence, controlled local delivery of BPs eluting from implants such as CDHA is an alterative way of administering the drug. In this paper, AS loaded CDHA coatings were produced by a co-precipitation technique in Simulated Body Fluid (SBF). The release kinetics of AS and proliferation of osteoblasts on coatings with different AS loading were studied.

Methods: Polished titanium squares were pretreated by 5M NaOH and distilled water [4], dried in air and then finally soaked in 1.5× SBF solution herein referred to as t-SBF [5], which was replenished every day. After 15 days, samples were soaked in 1.5× t-SBF containing different concentrations of AS (0, 10^{-6}, 10^{-5} and 10^{-4} M) and the solution was replenished every other day. Then the coated samples were dried in air after 6 days. In addition, some of the substrates loaded without AS were soaked in 3×10^{-4} M AS solution for 24 hours, then they were dried in air. Osteoblast proliferation studies were evaluated for all coated samples formed from t-SBF. AS amount released from CDHA coating in m-PBS solution was measured by using HPLC technique [6].

Results: i) Figure 1 shows X-Ray diffraction patterns from samples prepared with different concentrations of AS. There is no significant effect for AS on the crystallization of CDHA.

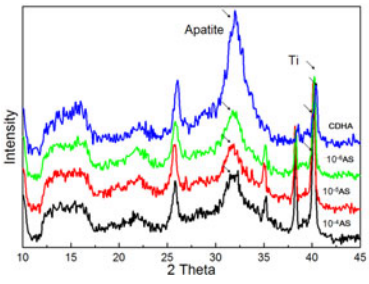

Figure 1. XRD patterns of samples soaked in 1.5× t-SBF solution with different concentrations of AS (0 M, 10^{-6}M, 10^{-5}M and 10^{-4}M)

ii) Figure 2 and Figure 3 show as the amount of AS in the substrates increases, the growth rate of osteoblast cells decreases.

iii) Figure 4 shows AS loading through water produces a fast and unstable release rate in m-PBS solution compared to that of 1.5× t-SBF solution.

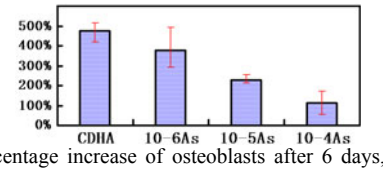

Figure 2. Percentage increase of osteoblasts after 6 days, Percentage= (Number of osteoblasts after 6 days - Number of osteoblasts after 24 hours)/Number of osteoblasts after 24 hours)

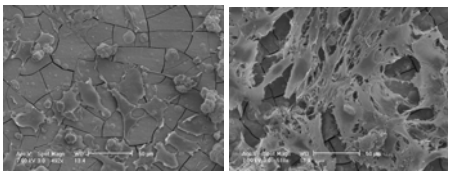

Figure 3.SEM micrographs of proliferation of osteoblasts after 6 days on the surface of samples soaked in 1.5× t-SBF with10^{-4} M AS (left) and without AS (right)

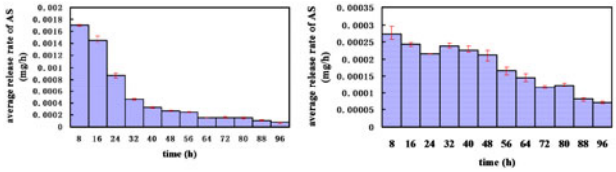

Figure 4.Average release rate of AS from the sample soaked in water with 3×10^{-4} M AS (left) and soaked in 1.5× t-SBF with 10^{-4} M AS (right)

Conclusions: AS did not produce any significant effects on the formation of CDHA coatings. However, the CDHA peak becomes broader in the presence of higher amount of AS in 1.5× t-SBF. This may be related to the high affinity of BPs for calcium ions of CDHA [7]. In previous studies, it has been shown that CDHA formed from t-SBF can promote the growth of osteoblasts [8], but AS produces adverse effect on the proliferation of osteoblasts if its concentration is too high [9]. Our result is close to these reports. The amount of loaded AS will be optimized based on the culture of osteoclasts on these substrates. Substrates loaded AS through 1.5× t-SBF solution can provide a stable release rate of AS *in vitro*, although AS loading efficiency for this approach needs to be improved in future.

References:

1. Sergey V.D. et al. Angew. Chem. Int. Ed. 2002: 41: 3130-3146.
2. H. Fleisch. Bisphosphonates in Bone Disease: From the Laboratory to the Patient, 3rd edition. New York: The Parthenon Publishing Group, 1997.
3. Migliorati C. et al. J. AM. Dent. Assoc. 2005: 136: 1658-68
4. Jalota S et al. J. Mater. Res. 2007: 22: 1593-1600
5. A.C. Tas. Biomaterials. 2000: 21: 1429-1438.
6. Şamdancıoğlu, S et al. FABAD J. Pharm. Sci. 2003: 28: 183-192.
7. Nancollas GH et al. Bone. 2006: 38: 617-627.
8. Jalota S et al. Mater. Sci. and Eng. C: Biomimetic and Supramolecular Systems. 2007: 27: 432-440.
9. Gun-Il Im et al. Biomaterials. 2004: 25: 4105-4115.

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