Antiwashout behavior of calcium phosphate cement incorporated with Poly(ethylene glycol)

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Abstract. The effect of powder-to-liquid ratio and addition of poly(ethylene glycol) on the antiwashout behavior of calcium phosphate cement has been investigated. Calcium hydroxide, Ca(OH)₂, and diammonium hydrogen phosphate, (NH₄)₂HPO₄, were used as precursors with distilled water as the solvent in the wet chemical precipitation synthesis of hydroxyapatite powder. Cement paste was prepared by mixing the as-synthesized powder with distilled water at certain ratios, varied at 1.0, 1.3, 1.5 and 1.6. Poly(ethylene glycol) was added into distilled water, varied at 1, 2, 3, 4 and 5 wt% using the powder-to-liquid ratio of 1.3. The antiwashout properties of the cement has been investigated by soaking in Ringer’s solution for 3 and 7 days. The evolution of compressive strength of calcium phosphate cement before and after soaking have been determined. After 7 days soaking, the strength of the cement increased by 94.4%, 2.98%, 11.39% and 111.29% for powder-to-liquid ratios 1.0, 1.3, 1.5 and 1.6 respectively. The addition of poly(ethylene glycol) up to 3% shows an increase in strength after 7 days soaking, with 57.75%, 16.4% and 19.97% increase for 1, 2 and 3% poly(ethylene glycol) contents respectively. The calcium phosphate cement produced in this current study shows excellent antiwashout behavior since no cement dissolution happened and the compressive strength of the cement increased with soaking time throughout 7 days soaking in Ringer’s solution.

1. Introduction

Calcium phosphate cement (CPC) is clinically accepted ceramic and has been widely used in biomedical application, predominantly in bone cement application. This is because CPC shows remarkable biological properties, injectability and potential to set in vivo [1]. CPC can be injected and molded to fill and take the shape of defect sites. Injectable CPC consisted of calcium phosphate powder of various phases and water mixed at certain ratios to set in vivo via dissolution-precipitation mechanism [2]. The mixing of phases such as tetracalcium phosphate (TTCP) and α–tricalcium phosphate (α-TCP) with water induced the formation of apatite cements of hydroxyapatite (HA) [3].

Biodegradability of CPC is significant to help the healing process through substitution of the hardened CPC by the newly forming bone [2]. Generally, the rate of cement resorption should be the same with the rate of new bone formation. Too fast resorption rate will cause collapse at the defect site [2]. However, disintegration of CPC tend to occur upon early contact with body fluids because of its
weak cohesion [1]. The solubility of CPC is affected by the surrounding pH value, and HA has the lowest solubility compared to that of dicalcium phosphate dihydrate (DCPD), octacalcium phosphate (OC) and β-tricalcium phosphate (β-TCP) [4].

The incorporation poly(ethylene glycol) (PEG) into CPC is one of the approaches to adjust the dissolution of CPC, so that the dissolution rate is similar to the rate of new bone formation. PEG is remarkably accepted in biomedical fields due to its non-toxicity, water solubility, flexibility and coagulant activity [5]. PEG also has been used as a thickening agent in premixed CPC to keep the cement paste stable [5,6]. Addition of PEG into CPC is expected to improve setting time and antiwashout performance, but lower injectability [6].

The evaluation of dissolution behavior of CPC has been done in vitro using Ringer’s solution. The composition of Ringer’s solution is similar to the inorganic components of extracellular body fluid to mimic the biological environment for bone, which aid the formation of new bone tissues. In this study, HA powder was synthesized via wet chemical precipitation method. The purpose of this study is to determine the effect of powder-to-liquid (P/L) ratio and addition of PEG on the antiwashout behavior of CPC.

2. Experimental procedures

2.1. Synthesis of HA powder
HA powder has been synthesized via wet chemical precipitation method following the procedure reported elsewhere [7]. This route employed calcium hydroxide, Ca(OH)$_2$, and diammonium hydrogen phosphate, (NH$_4$)$_2$HPO$_4$, as the precursors and the solvent used was distilled water. The calcium-to-phosphorus (Ca/P) ratio was fixed at 1.67. Each precursor was dissolved in distilled water to prepare calcium and phosphorus solution. The two solutions were mixed together through drop wise addition of calcium solution into the phosphorus solution. 25% ammonia solution was added to adjust the pH of the mixture at pH 11. Then, the mixture was refluxed at 90°C. The precipitate formed underwent overnight (18 hours) aging at room temperature, washing with distilled water and filtering. The filtered precipitate was then dried overnight at 85°C, and finally crushed.

2.2. Preparation of CPC
Cement paste was prepared by mixing the synthesized HA powder with distilled water. Two types of cement were prepared, without and with PEG addition. Cement paste without PEG was prepared at different P/L ratios, varied at 1.0, 1.3, 1.5 and 1.6. Cement paste incorporated with PEG (MW300, Sigma) was prepared with the P/L ratio of 1.3. PEG was added into the liquid phase, varied at 1, 2, 3, 4 and 5 wt%. The cement paste was put into a Teflon mold and left for 48 hours to dry. The sample dimension used is 10 mm diameter x 15 mm length.

2.3. Antiwashout test
The antiwashout behavior of CPC was investigated by soaking the cement in a Ringer’s solution for up to 7 days. The mechanical strength of CPC after the immersion was measured at 3 and 7 days. At each interval, samples were washed with distilled water and dried before compression test. The compression test of CPC was done by using Lloyd LR 10 K+ Universal Testing Machine on the 10 mm diameter x 15 mm length specimen at 1 mm/s crosshead rate.

2.4. Porosity measurement
The porosity of CPC was calculated using the following equation:

\[
\% \text{Porosity} = 100\% \times \frac{\rho_{\text{app}}}{\rho_{\text{th}}} \times 100\%
\]

Relative density $= \frac{\rho_{\text{app}}}{\rho_{\text{th}}}$
Where, $\rho_{\text{app}}$ is the apparent density measured by densitometer and $\rho_{\text{th}}$ is the theoretical density of HA taken as 3.156 g/cm$^3$ [8],[9].

3. Results and Discussion

The dissolution behavior of CPC was investigated by soaking in Ringer’s solution for 7 days. The effect of P/L ratio and PEG content on the dissolution properties of CPC was investigated. CPC specimens were prepared using the P/L ratio of 1.0, 1.3, 1.5 and 1.6. The addition of PEG into the liquid phase of CPC was varied at 0, 1, 2, 3, 4, and 5%. From the observation, all CPCs show excellent integration properties since no cement dissolution shown throughout 7 days soaking in Ringer’s solution.

The evolution of compressive strength of CPC before and after soaking have been evaluated in this present work. Figure 1 shows the result of compressive strength of CPC with different P/L ratios up to 7 days soaking in Ringer’s solution. All cements show increasing compressive strength with the increase in soaking period. The strength increased by 94.4%, 2.98%, 11.39% and 111.29% for CPC with the P/L ratios 1.0, 1.3, 1.5 and 1.6 respectively, with the range of 0.416 MPa to 1.344 MPa before soaking and increased to the range of 0.879 MPa to 1.384 MPa after 7 days soaking. This might be attributed to the formation of apatite via biomineralisation mechanism. As reported by Mohammadi et al. [10], CPC was immersed in human blood plasma and Ringer’s solution revealed that the compressive strength of CPC increased with increasing soaking time. The formation of apatite also enhanced throughout the soaking period. Study done by Verma and Sinha [11] reported the cohesiveness of CPC in distilled water for different P/L ratios, (1:1, 1:1.1 and 1:1.2 g/ml). Their study revealed that no structural degradation of CPC after 7 days immersion for CPC after 20 min incubation. However, structural disintegration was observed when the paste was directly injected into the distilled water and the decrease in P/L ratio increased the rupture time.

The effect of PEG addition on the compressive strength of CPC after soaking in Ringer’s solution is shown in figure 2. After 7 days immersion, the compressive strength of CPC increased with the increase in immersion time for CPC with 0, 1, 2 and 3% PEG contents. The strength increased by 2.98%, 57.75%, 16.4% and 19.97% for the CPC with 0, 1, 2 and 3% PEG content, respectively, with the range of 1.167 MPa to 1.786 MPa before soaking and increased to the range of 1.384 MPa to 2.079 MPa after 7 days soaking period. However, CPC with 4 and 5% PEG contents show an increase in compressive strength up to 3 days soaking with 7.18% and 3.89% increase respectively, and then the
compressive strength decreased by 25.5% and 40% respectively after 7 days soaking. The strength decreased from 1.371 MPa to 1.1 MPa and from 1.285 MPa to 0.801 MPa for 4 and 5% PEG contents respectively. This might be because of hydrophilicity of PEG, which causes PEG to dissolve in the liquid medium leading to the increase in porosity and hence reducing the compressive strength of CPC. Roy et al. [12] has reported that addition of PEG slightly delayed the nucleation and formation of DCPD crystals since its compressive strength was high initially but dropped significantly after 3 days soaking in PBS and increased again after 7 days soaking.

![Figure 2](image-url)  

**Figure 2.** Effect of PEG addition on the dissolution behavior of CPC after soaking in Ringer’s solution.

Porosity of CPC before soaking have been determined. The average porosity of CPC ranged from 39.2% to 47.1%. Their apparent density ranged between 1.67 to 1.92 g/cm³. From table 1, the porosity of CPC decreases with the increase in P/L ratio. This is attributed to the increase in powder content in the cement paste. High porosity of CPC is presumably due to water entrapped between the crystals and porosity introduced during cement preparation [13]. When PEG is added into CPC, the porosity of is in the range of 39.6% to 51.4% and the apparent density is between 1.53 to 1.90 g/cm³. The result shows that incorporation of PEG into CPC increases porosity of CPC as tabulated in table 2. The higher the PEG content, the lower its porosity because PEG particles filled up the pores in CPC. This is true only for 1% to 4% PEG content. The porosity started to increase again when 5% PEG was added. This indicates that an appropriate amount of PEG is needed to reduce porosity of CPC.
Table 1. Porosity of CPC with different P/L ratios.

| P/L ratio | Porosity (%) |
|-----------|--------------|
| 1.0       | 47.1         |
| 1.3       | 45.2         |
| 1.5       | 40.8         |
| 2.0       | 39.2         |

Table 2. Porosity of CPC with different PEG contents for the P/L ratio of 1.3.

| PEG content (wt%) | Porosity (%) |
|-------------------|--------------|
| 0                 | 45.2         |
| 1                 | 51.4         |
| 2                 | 48.5         |
| 3                 | 43.8         |
| 4                 | 39.6         |
| 5                 | 45.7         |

4. Conclusion

This present work successfully investigated the effect of P/L ratio and addition of PEG on the antiwashout behavior of CPC. The prepared CPC shows remarkable antiwashout properties because no cement dissolution was observed throughout the 7 days soaking in Ringer’s solution. In addition, the evolution of compressive strength of CPC after soaking shows strength improvement with the increase in soaking period. The strength of CPC before soaking ranged from 0.416 MPa to 1.344 MPa and increased to the range of 0.879 MPa to 1.384 MPa after soaked for 7 days. PEG addition up to 3% showed an increase in strength by 2.98%, 57.75%, 16.4% and 19.97% for CPC with 0, 1, 2 and 3% PEG contents, respectively, which ranged from 1.167 MPa to 1.786 MPa before soaking and increased to the range of 1.384 MPa to 2.079 MPa after 7 days soaking.

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