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Basic properties of calcium phosphate cement containing different concentrations of citric acid solution^①

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[Abstract] The properties of calcium phosphate cement consisting of α -tricalcium phosphate (α -TCP) and tetracalcium phosphate (TTCP) have been investigated by using a cement liquid that contained citric acid with concentration of 0.05 mol/L or higher. The relationship between the setting time of the system cement and the concentration of citric acid solution shows concave type curve. When solution concentration was 0.2 mol/L, the setting time was 8 min, which was the shortest. While the relationship between 24 h compressive strength of the cement and the citric acid concentration shows convex type curve. When solution concentration was 0.2 mol/L, the compressive strength was 39.0 MPa, which was the highest. Afterwards, the microstructure of the hardening product was observed by SEM, the effect of citric acid on the exothermic rate of hydrate reaction was studied by microcalorimeter, and the crushed specimens were subjected to X-ray diffraction. The results verified that the low citric acid concentration can accelerate the hydrate reaction rate of the α -TCP/TTCP system. However, the high citric acid concentration inhibited hydroxyapatite formation and retarded the rate of hydrate reaction of the α -TCP/TTCP cement.

[Key words] α -TCP/TTCP cement; citric acid; setting time; compressive strength; HAP formation

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1 INTRODUCTION

Calcium phosphate ceramics such as tricalcium phosphate (TCP) and hydroxyapatite (HAP) have been used for bone defect filling and oral surgery due to good biocompatibility and osteoconductivity^[1]. They are usually applied in the form of blocks or granules. There is difficulty in shaping block prior to surgery and in making them fit with the bone surface^[2]. These problems do not happen with granules, but secondary migration is often observed and a mechanical strength comparable to blocks cannot be expected^[3]. Recently, much attention has been paid to self-setting calcium phosphate cements, because they can be handled as a paste and set in situ, and they may overcome the practical disadvantages of blocks or granules^[4]. Their structure and composition, close to that of HAP, make them to be biocompatible materials^[5]. The conventional calcium phosphate cement (CPC) had some problems such as long setting time (30 ~ 60 min) and low compressive strength, etc^[6,7]. Fernandez et al^[8] reported that the addition of sulfate, citrate and pyrophosphate ions has proposed to increase setting time and tensile strength of CPC. Fresh wet bone contains citrate by about 1%^[9], which nearly corresponds to more than 70% of total citrate content in the body. Further, the concentration of citrate in bone and blood is increased

with the administration of vitamin D or parathyroid extract^[10]. It has been, therefore, conjectured that citrate ions play an important role in bone resorption/ossification through the formation of dissociated calcium citrate complexes in the surrounding body fluid^[11,12]. Thus, citric acid solution that had good biocompatibility was used as the mixing liquid. In our system, an α -TCP/TTCP powder mixture was mixed with water containing citric acid to control the setting time and compressive strength. The objectives of the present investigation are to study the effects of various citric acid solutions concentration on the properties of the cement.

2 EXPERIMENTAL

2.1 Materials

α -TCP was synthesized at 1250 °C from a 2:1 (molar ratio) mixture of analytical grade dicalcium phosphate dihydrate ($\text{CaHPO}_4 \cdot 2\text{H}_2\text{O}$) and calcium carbonate (CaCO_3). TTCP was prepared by heating an equimolar mixture of $\text{CaHPO}_4 \cdot 2\text{H}_2\text{O}$ and CaCO_3 at 1500 °C for 6 h and quenched at room temperature. α -TCP and TTCP were combined in an appropriate ratio and milled together down to the appropriate particle size ($d < 16 \mu\text{m}$) in an agate ball mill to prepare calcium phosphate cement (CPC, α -TCP/TTCP) powder. The citric acid solutions, with concentra-

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tions ranged from 0.05 to 0.35 mol/L, were used as the cement liquids. The solutions were prepared by dissolving analytical grade citric acid in CO₂-free double distilled water.

2.2 Methods

Setting time of the CPC samples were measured according to the method of international standard ISO1566. The cement is considered to be set when a Gillmore needle with a tip diameter of 1mm and loaded with 3.9N fails to make a perceptible circular indentation on the surface of the cement at 37 °C and 100% relative humidity. CPC specimens for compressive strength measurements were mixed by the CPC powder and liquid at a powder-to-liquid (P/L) ratio of 1.50:1 (g/mL) on a glass slab for approximately 1min and the cement paste was packed into a split mould (10mm × 10mm × 10mm) and put in an incubator kept at 37 °C and 100% relative humidity. Specimens were removed from the moulds after sample prepared for 24h and the compressive strength was determined on Electro-hydraulic servocontrolled testing system (INSTRON 1341, Instron Ltd., Britain) with a crosshead speed of 0.5 mm/min. The compressive strength value and the setting time were the average values obtained from at least five specimens. Afterwards, the crushed specimens were quenched in liquid N₂, lyophilized and freeze-dried. The freeze-dried samples were ground into fine powders and identified by powder X-ray diffraction (XRD, D/MAX IIIA, Rigaku Ltd, Japan) in order to analyze the hydrated products and the progress of reaction. The samples were scanned from 3° to 60° in 2θ. The microstructure of the hardening product was observed by scanning electron microscopic (SEM, SX-40, Akashi Seisakusho Ltd., Japan). During the setting reaction differential-scanning calorimetry was carried out in micro calorimeter (C80, SETARAM, France) to estimate the rate of heat evolution at 37 °C. These studies were done in the following citric acid solutions: 0.1, 0.2 and 0.3 mol/L. Powder-to-liquid ratio of 1:1 (g/mL) was used. A datum point was collected every 3s and stored in a computer.

3 RESULTS

Fig. 1 shows that the relation between the setting time of the cement and the concentration of citric acid solution is in concave type curve, and the relation between the compressive strength of complexes and the concentration of citric acid shows in convex type curve after 24 h. With the addition of citric acid up to a concentration of 0.2 mol/L, the setting time shortens and the compressive strength values increase, at which the setting time shows minimal value (8 min), whereas the compressive strength reaches maximal value (39.0 MPa). Interestingly, the setting time

increases and the compressive strength value decreases as the citric acid concentration is increased further.

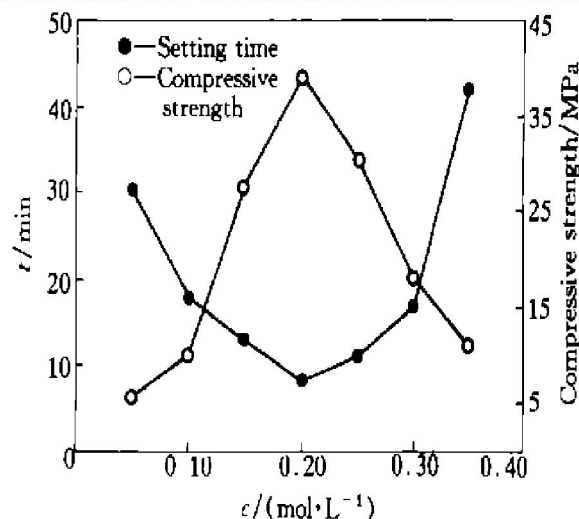


Fig. 1 Effect of various concentration citric acid solutions on setting time (final) and compressive strength (24 h)

X-ray diffraction patterns of α -TCP/TTCP containing various concentration of citric acid after 24 h are shown in Fig. 2. The patterns indicate that the main component is HAP after 24 h, and a small amount of untreated α -TCP and TTCP are observed. When the concentration of citric acid less than 0.2 mol/L, the diffraction peak of HAP heightens gradually, and the diffraction peak of α -TCP lowers by degrees with the citric acid concentration is increased. However, when the citric acid concentration is beyond 0.2 mol/L, the peak of HAP gently diminishes as the concentration is increased, and the base-lines of XRD patterns tend towards the diffraction peak of amorphous substance. The main elements constituting of α -TCP/TTCP (containing 0.35 mol/L citric acid) are still α -TCP and TTCP.

Fig. 3 shows typical SEM images of the α -TCP/TTCP with different concentrations of citric acid. Smaller crystals with larger pores are observed within the α -TCP/TTCP with 0.1 mol/L citric acid solution, the hydration product created a loose network structure (Fig. 3(a)). Larger crystals with smaller pores are observed within that with 0.2 mol/L citric acid solution, the hydration product formed compact construction (Fig. 3(b)). Larger amount of amorphous calcium citrate with pores and crystalline grain are observed within that containing 0.3 mol/L citric acid solution (Fig. 3(c)).

Figs. 4 and 5 show the effects of citric acid on the kinetics of α -TCP/TTCP hydrate reaction. The rates curves in Fig. 4 show the instantaneous rates of heat evolution and the rates of HAP formation in various citric acid solutions at a liquid-to-solid ratio of 1:1 (g/mL). The maximum of the first reaction peak increases as the citric acid concentration increases. Two reaction peaks are observed at

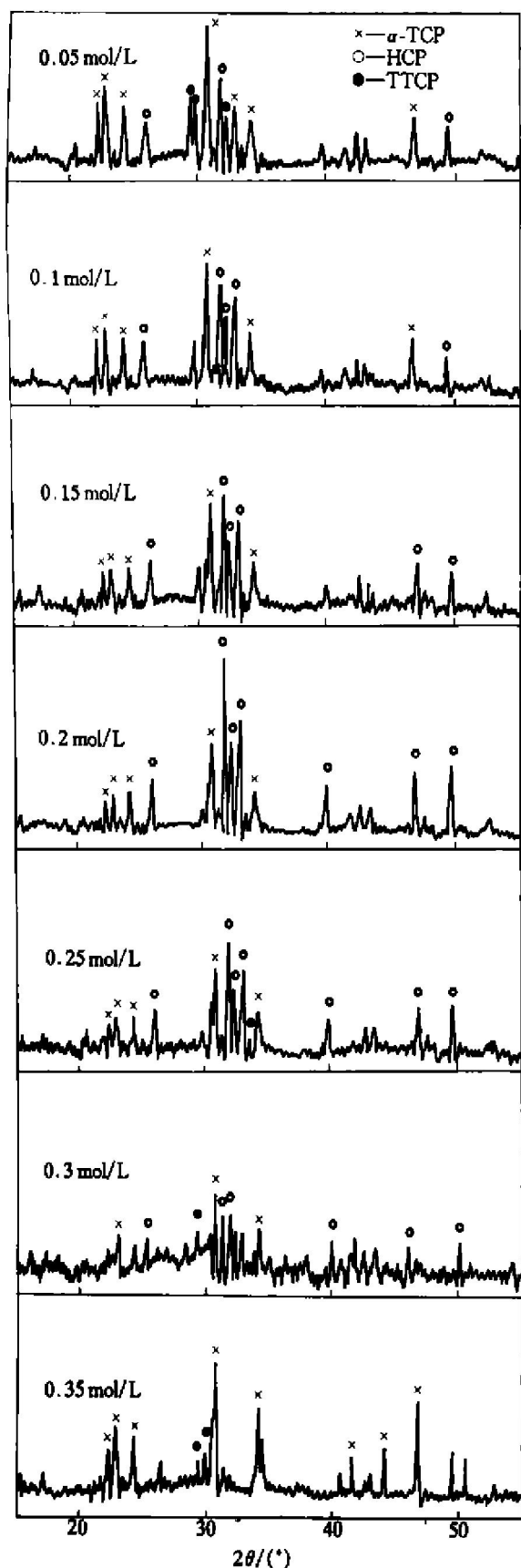


Fig. 2 X-ray diffraction patterns of α -TCP-TTCP- $\text{C}_6\text{H}_8\text{O}_7\text{-H}_2\text{O}$ complexes

citric acid concentration below 0.2 mol/L. The second reaction peak reaches a maximum at 250 min for concentration of 0.1 mol/L. When citric acid concentration is 0.2 mol/L, reaching the maximum of the second reaction peak needs 180 min, the

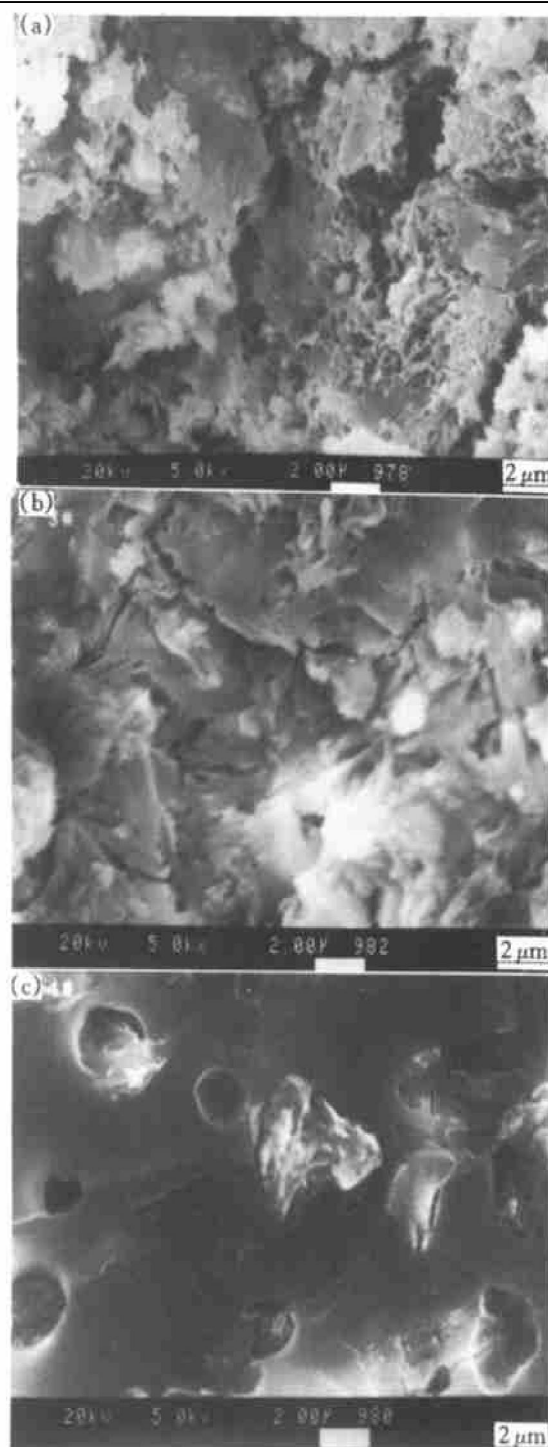


Fig. 3 SEM images of α -TCP-TTCP- $\text{C}_6\text{H}_8\text{O}_7\text{-H}_2\text{O}$ complexes
(a) α -TCP/TTCP with 0.1 mol/L citric acid solution;
(b) α -TCP/TTCP with 0.2 mol/L citric acid solution;
(c) α -TCP/TTCP with 0.3 mol/L citric acid solution

induction period shortens, and the reaction peak increases evidently. Three heat evolution peaks are observed for concentration of 0.3 mol/L. The second and the third reaction peaks significantly overlap the first peak and appear as a broad shoulder. Fig. 5 gives the normalized integral heat evolution as a function of the reaction time. Complete hydration needs 36, 24, and 30 h for concentration of 0.1, 0.2, and 0.3 mol/L. This indicates that the rate of hydrate

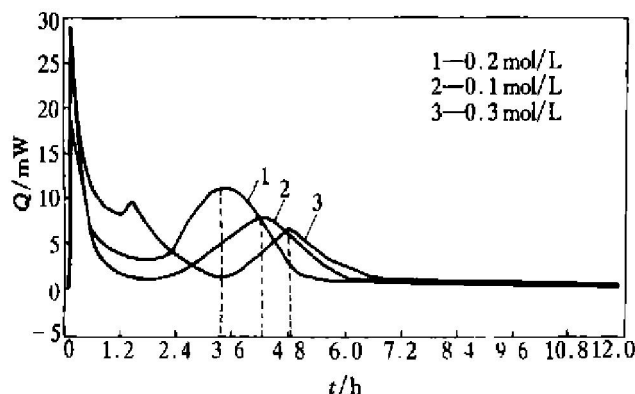


Fig. 4 Hydrate heat evolution curves of α -TCP/TTCP with various citric acid concentration

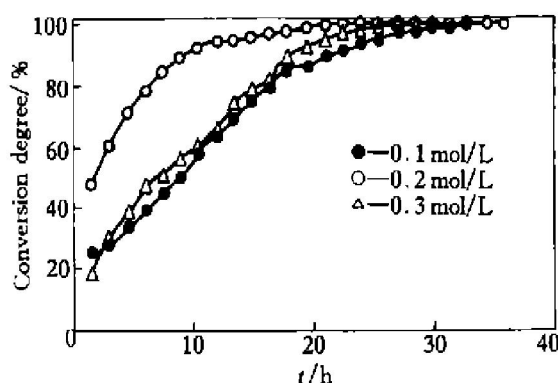


Fig. 5 Normalized heat evolution of α -TCP/TTCP cement at 37 °C

reaction is accelerated as the citric acid concentration up to 0.2 mol/L, but higher concentration retard the hydrate reaction.

4 DISCUSSION

The hydrate process of α -TCP/TTCP includes dissolution and precipitation at the same time, when α -TCP and TTCP are mixed with solution, Ca^{2+} and PO_4^{3-} dissolve firstly. The concentration of Ca^{2+} and PO_4^{3-} ions increase with α -TCP/TTCP grains dissolving continually. HAP crystallizes and precipitates when the ions reach supersaturation in the solution. In this way, the reactants dissolve and the resultants deposit continually until hydrate reaction finishes. Because complexes that have little solubility can be formed from citrate and dissociative Ca^{2+} ions in the solutions, the dissolution rates of α -TCP and TTCP are accelerated, the concentration of Ca^{2+} and PO_4^{3-} ions are increased. When $Q_c = [\text{Ca}^{2+}]^5 [\text{OH}][\text{PO}_4^{3-}]^3$ is beyond the K_{sp} of HAP in the solution, it contributes to HAP precipitation that the balance tend to form HAP. H ion concentration is increased. The dissolution rate of α -TCP/TTCP is accelerated and the concentration of Ca and P ions are increased with increase of citric acid concentration up to 0.2 mol/L.

As far as HAP is concerned, the time that reaches $Q_c > K_{sp}$ is shortened further, the crystallizing rate of HAP and the rate of hydrate are accelerated, thus the setting time decreases. On the other hand, because three carboxyl groups of citric acid and the calcium ions in calcium phosphate salt can form chelating-ligand, which constitute $[\text{COO}]-\text{Ca}^{2+}-[\text{OPO}_3]$ type space network structure. They form amorphous setting substance because of disordered structure^[13]. As the distance between grains is short enough to form crystal combine, the space between calcium phosphate salt grains is increased from zero to certain distance by this structure^[14], which reduces the total amount of hydrate product when they bring chemical bond connection, accordingly, the setting time is decreased. With the citric acid concentration is increased, the rate of hydrate reaction is quickened, and the dissolution of α -TCP/TTCP is accelerated, that benefit HAP precipitate and reticulation structural formation. The compressive strength trend to increase gradually as the concentration is greater than before.

However when the concentration is beyond 0.2 mol/L, great deals of calcium citrate complex that have little solubility form because of higher concentration of citric acid. Since the unreacted α -TCP/TTCP grains are wrapt by abundant complexes, the diffusion of ions and water are hindered, and the hydrate exothermic rate of α -TCP/TTCP is reduced. Thereafter, those unstable products self-decompose, and the unreacted α -TCP/TTCP grains are exposed again. The reaction comes into speed hydration phase, the second reaction peak results from the remaining α -TCP/TTCP reacting with citric acid in solution. The unreacted grains of α -TCP/TTCP are wrapped by a mass of new-formed complex over again, that make the wrapt layers of complex thicken, the hydrate exothermic reaction turns into an all-time low. Thenceforward, the rate of reaction is accelerated gradually with the unstable complex decomposes by degrees, and the third reaction peak appears. Finally, the hydrate reaction enters into a basal stable phase, whose heat evolution rate is quite low. It can be seen that the hardening acceleration effect of citric acid is reduced obviously owing to great deals of complex formation as the concentration is beyond 0.2 mol/L. The unreacted α -TCP/TTCP grains are wrapt badly by great deals of complex occurred, in which water is wrapt and form network by hydrogen bond association. The infiltration of water and the diffusion of ions are hindered, and the retarded action of HAP formation is enhanced. That brings about certain inhibitory action and retards the hardening of α -TCP/TTCP. Thus, the rate of hydrate reaction becomes slow, and the setting time increases, and the HAP formation decrease accordingly. That results in the compressive strength appearing downtrend.

5 CONCLUSION

The results indicate that the α -TCP-TTCP- $\text{C}_6\text{H}_8\text{O}_7\cdot\text{H}_2\text{O}$ complexes exhibit good cement-like properties. Citric acid accelerates the rate of hydrate reaction and HAP formation when the concentration is less than 0.2 mol/L. The setting time reduces and the compressive strength increases with the addition of citric acid up to a concentration of 0.2 mol/L. But hydrate reaction and HAP formation are retarded by citric acid concentrations above 0.2 mol/L owing to forming great deals of calcium citric complex which wraps α -TCP/TTCP grains and forms network with water and then hinders water infiltration and ion diffusion. Thus, the setting time is 8 min, which is the shortest; the compressive strength is 39.0 MPa, which is the highest when the concentration is 0.2 mol/L. These results indicate that citric acid can be used to control the setting time and the compressive strength if clinic desire. The results also show that this cement paste with a good consistency can be placed easily on any variously shaped defects. It is useful as a bone graft material as well as HAP. In a word, a calcium phosphate cement made from α -TCP, TTCP and citric is promising.

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