Research Article

Electrochemical Processes for the Formation of Hydroxyapatite Powders

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Abstract

Electrochemical synthesis of hydroxyapatite particles was performed from a homogeneous solution of $Na_2H_2EDTA.2H_2O$, KH_2PO_4 and $CaCl_2$ without stirring to investigate the mechanism of hydroxyapatite formation. We found that OH^- ions generated by water reduction at the cathode play an important role in the formation of hydroxyapatite particles. The OH^- ions induce the liberation of Ca^{2+} ions and the dissociation of phosphoric acid, which serve as the reactants for the formation of hydroxyapatite particles. Two layers with a clear boundary were formed during electrolysis. The upper layer comprises the produced particles and the lower layer is a homogeneous solution. The produced particles were held in the region between the electrodes mainly due to the electrostatic interactions of charged particles in an electric field. The hydroxyapatite particles are agglomerates consisting of spherical particles. Aging the suspension for 24 h after electrolysis leads to the transformation of hydroxyapatite to brushite. Thus, if producing hydroxyapatite is desired, the particles should be continuously removed from the system. This method appears to be promising as a continuous process to produce hydroxyapatite particles using an electrochemical method. © 2014 BCREC UNDIP. All rights reserved

1. Introduction

Hydroxyapatite (HA) is one of the calcium phosphate-based bioceramic materials that compose the majority of inorganic components in human bones and teeth. This compound is thermodynamically the most stable crystalline phase of calcium phosphate in body fluid. Because of the similarity between HA and the mineral component of human bones and teeth, HA has great potential for use as a bone substitute material.

Several methods have been developed to produce HA powders, including chemical precipitation [1, 2], hydrothermal [3, 4], emulsion [5], sol-gel [6] and mechano-chemical [7] methods. The electrochemical method is beginning to emerge as a valuable option for preparing HA powders [8, 9]. Using the electrochemical method, the particle size can be controlled easily by simply adjusting the imposed current or potential of the system [10, 11]. Montero *et al*.

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[8] and Djošić *et al.* [9] assumed that HA was formed by consuming OH⁻ ions produced by reduction of water at the cathode. However, because the initial pH of the solution was high, i.e., pH > 9, it is not clear whether the OH⁻ ions were consumed or produced during the precipitation of HA. Moreover, the change in pH during the electrolysis was not measured, which makes it difficult to understand the processes that occurred. Therefore, it is necessary to clarify the role of OH⁻ ions in the formation of HA particles during electro-synthesis.

In this paper, we report on the electrolysis of a solution containing Ca^{2+}, EDTA and PO_4^{3-} to prepare HA particles. The electrolysis was carried out without stirring to investigate the mechanism of particle formation. In the work of Montero *et al.* [8] and Djošić *et al.* [9], the electro-synthesis was carried out under stirring and at a high pH, making it difficult to determine the mechanism of particle formation. By conducting electrolysis without stirring, the progress of the reactions could be observed carefully to understand the parameters influencing the formation of HA particles.

2. Materials and Methods

The homogeneous solution for the electrosynthesis of HA powders was composed of $Na_2H_2EDTA \cdot 2H_2O$ (Dojindo Molecular Technologies, Inc.), KH_2PO_4 (Wako Pure Chemical Industries, Ltd.) and $CaCl_2$ (Kanto Chemical Co. Inc.) at a concentration relationship of Ca^{2+}/EDTA/PO_4^{3-} of 0.25/0.25/0.15 M, following the works of Montero *et al.* [8] and Djošić *et al.* [9]. The electrolysis was carried out in an electrochemical cell consisting of two platinum foil electrodes with a dimension of (20x10) mm. The electrodes were set parallel with 3 cm between the electrodes. The electrodes were connected to the
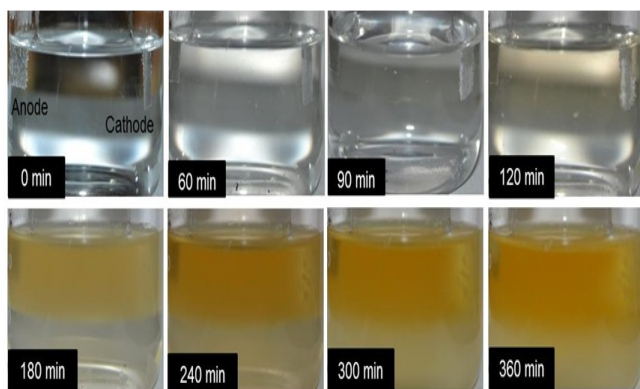
positive and negative terminals of a DC power supply with an imposed current density of 53 mA/cm², corresponding to the applied voltage of 4.3 V. The solution pH at three different positions (near the cathode, near the anode and in the bulk solution) was recorded by a pH meter (Orion 2-Star, Thermo Scientific).

The resulting suspension was either filtered, and dried directly in an oven at 110 °C for 24 h or aged at room temperature for 24 h, and then filtered and dried at 110 °C for 24 h. The progress of the reaction was observed by periodically photographing the electrochemical cell for later inspection.

A cyclic voltammetry measurement was performed for the reaction system using a potensiostat/galvanostat instrument (Autolab PGSTAT 302, Metrohm). The measurement was carried out at a voltage range from -1000 to 1,500 mV vs. Ag/AgCl with a scan rate of 0.5 mV/s. The morphologies of the particles were observed using scanning electron microscopy (JSM 6510 JEOL). The X-ray diffraction (XRD) pattern of the particles was detected using an X-ray diffractometer (RINT2100PC/N Rigaku).

3. Results and Discussion

Figure 1 shows the photographs taken during the electrolysis of a solution containing Ca^{2+}, EDTA and PO_4^{3-} at various elapsed times. The solution was clear at the beginning of the experiment when neither current nor potential had yet been applied. Gas bubbles appeared immediately at the surface of both electrodes (anode and cathode) after the current or potential was applied to the system. The most likely redox reactions occurring at the anode and cathode were, respectively, the oxidation and reduction of water. The reactions may be
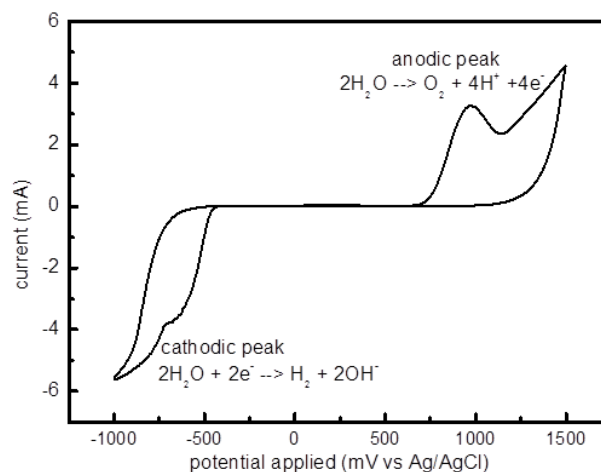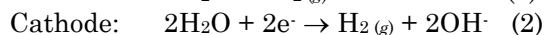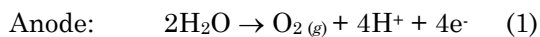

Figure 1. Time-resolved pictures of particle growth and behavior during electrolysis.

Figure 2. Cyclic voltammetry of the solution to synthesize HA

written as:

Anode: $2H_2O \rightarrow O_{2\,(g)} + 4H^+ + 4e^-$ (1)
Cathode: $2H_2O + 2e^- \rightarrow H_{2\,(g)} + 2OH^-$ (2)

The cyclic voltammogram shown in Figure 2 corroborates these reactions. An anodic peak at approximately 972.6 mV vs. Ag/AgCl indicates water oxidation (Equation 1) and a cathodic peak at approximately -682 mV vs. Ag/AgCl indicates water reduction (Equation 2). Thus, the gases observed at the electrodes were oxygen at the anode and hydrogen at the cathode. The generation of OH⁻ ions at the cathode was confirmed by the increase in pH at the region near cathode (Figure 3).

Particles started to appear at the cathode after approximately 90 min. The amount of particles generated at the cathode increased with the elapsed time and they moved away from the cathode region because of the concentration gradient. At an elapsed time of approximately 180 min, a suspension of particles appeared in the bulk solution. Surprisingly, the particles only moved in the region between electrodes, and there almost no particles were observed in the region below the bottom edge of the electrodes. From the photographs, it can clearly be observed that there are two layers with a clear boundary. The upper layer is a suspension containing particles produced at the cathode, and the lower layer is a homogeneous solution.

The formation of the two layers during electrolysis may be explained by considering the phenomena of particles suspended in an electrolyte solution in the presence o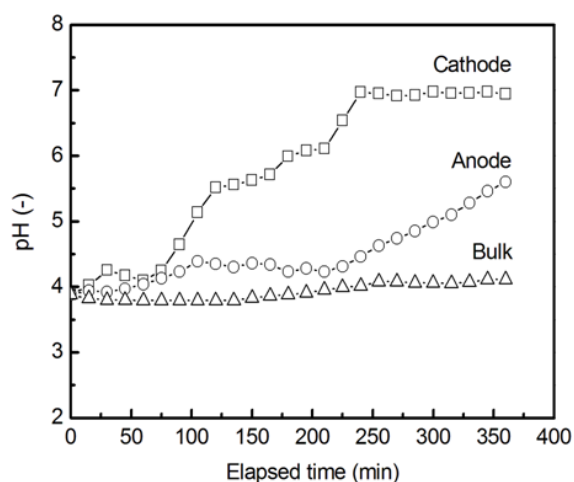f an electric field: When particles were first formed at the cathode, they tended to move away from the cathode region to the bulk solution due to the concentration gradient. The particle concentration around the cathode was higher than that of the bulk solution; therefore, the particles diffused away from the cathode. The movement of particles to the region of lower particle concentration due to the concentration gradient can be in any direction. However, when the particles were suspended in an electrolyte solution, an electrical double layer was formed on the surface of particles that make the particles carry electrical charge. The charged particles were then influenced by the electric field that existed in the region between the electrodes. In addition, the charged particles also influenced each other due to the electrostatic repulsion that tends to keep the particles in their positions. It seems that the electrostatic interaction among the charged particles dominates the particle movement rather than neutral diffusion and gravity. Hence, the particles were held in the region between the electrodes, and there were no particles below the bottom edge of the electrodes. The above phenomena may be illustrated schematically in Figure 4.

The amount of particles continued to increase as time elapsed. It can be observed that the boundary of the two layers gradually became lower due to the increase in the amount and size of the particles. When the amount of particles increases, the total mass in the upper layer also increases and the effect of gravity on the bulk of the particles becomes significant. Some particles also started to diffuse from the high-concentration upper layer to the low-concentration lower layer, especially from the
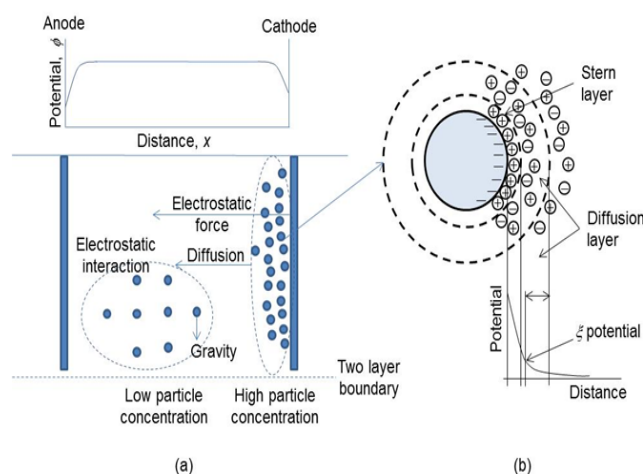

Figure 3. The change in solution pH at three different positions: near the cathode, near the anode and in the bulk solution.

Figure 4. Schematic illustration of the charged-particle behavior in an electric field during electrosynthesis.

edge of the upper layer. In addition, the electrostatic interaction may also push the particles down. As a result, the lower layer gradually became more turbid. The high particle concentration may cause the particles to collide with each other and coagulate to form larger particles. Then, the effect of gravity on the larger particles became significant, and the particles fell down. The densities of the upper layer and the lower layer at that time were, respectively, 1.0774 and 1.0828 g/cm^3. Figure 5 shows the SEM images of particles taken from the upper layer (a) and the lower layer (b). It can be observed that particles in the upper layer are much smaller than those in the lower layer. Particles in the lower layer are large agglomerated particles composed of smaller particles.

Figure 6 shows the XRD patterns of particles sampled from the upper layer, the region near cathode and the lower layer. The pattern for all samples match the standard pattern of HA (JCPDS 03-0747, hexagonal, a = b = 9.4302, c = 6.88911 Å, space group P6$_3$/m).
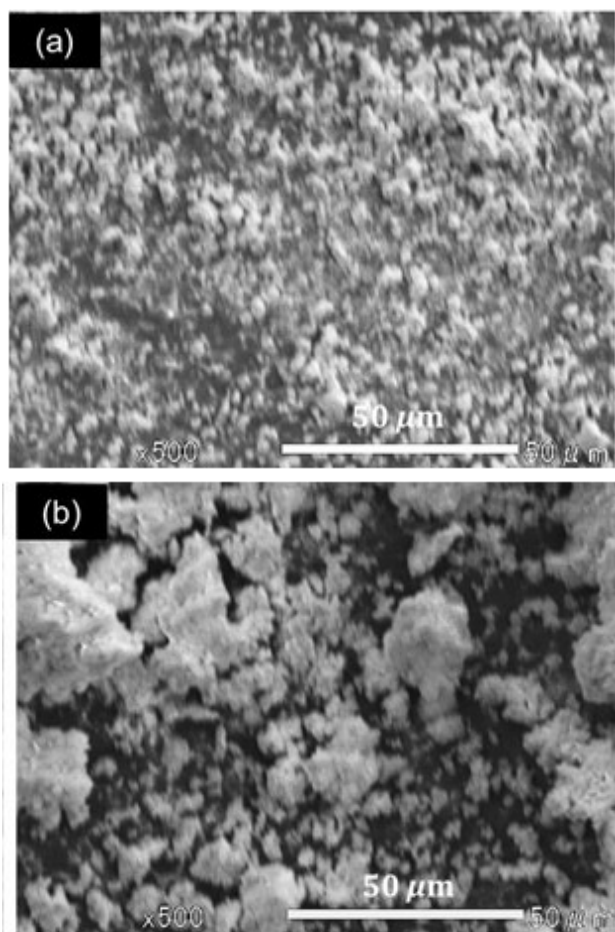
Based on the above observations, the route of the formation of the HA particles during the electrolysis may be described by the following: At the beginning of electrolysis, water was first oxidized at the anode and reduced at the cathode according to reactions (1) and (2). The generation of OH$^-$ ions at the cathode influenced the dissociation of phosphoric acid according to the following equilibrium reactions:

$$H_3PO_4 \leftrightarrow H^+ + H_2PO_4^- \qquad (3)$$

$$H_2PO_4^- \leftrightarrow H^+ + HPO_4^{2-} \qquad (4)$$

$$HPO_4^{2-} \leftrightarrow H^+ + PO_4^{3-} \qquad (5)$$

These equilibrium reactions may be the reason why the pH value near the cathode was relatively constant at the initial time of electrolysis (Figure 3). Conversely, although H$^+$ ions were generated near the anode, the pH was nearly constant. This observation may be caused by the equilibrium reactions of phosphoric acid near the anode (Equations 3-5) that acts like a buffer.

When particles started appearing near the cathode after approximately 90 min, the pH near the cathode also started increasing (Figure 3). The increase in pH may be caused by the generation of OH$^-$ due to the formation of HA particles, in addition to the generation of OH$^-$ ions by water reduction produced during the following reaction [12]:
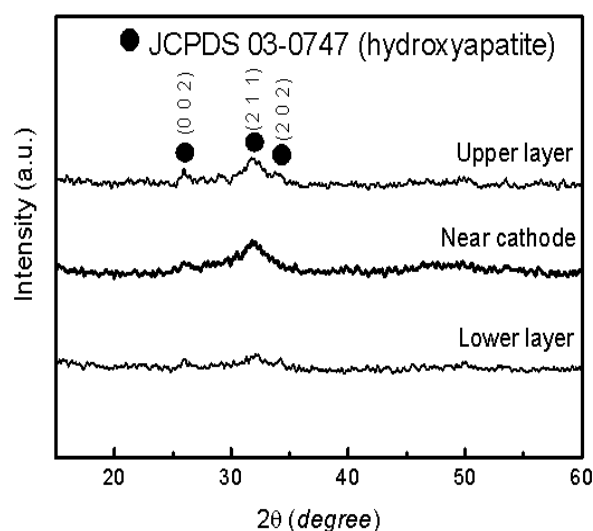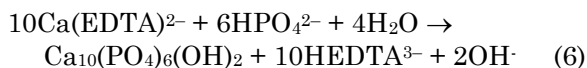

Figure 5. SEM images of the particles sampled from (a) the upper layer and (b) the lower layer

Figure 6. XRD patterns of the particles sampled from the upper layer, the region near cathode and the lower layer.

$$10Ca(EDTA)^{2-} + 6HPO_4^{2-} + 4H_2O \rightarrow$$
$$Ca_{10}(PO_4)_6(OH)_2 + 10HEDTA^{3-} + 2OH^- \quad (6)$$

The pH continued to increase until an elapsed time of approximately 240 min and was then constant. It seems that the system has reached steady state as indicated by the similar color of suspension in the upper layer that differs from the color before an elapsed time of 240 min (brighter).

The mechanism of HA formation as described above is different from that proposed by Montero *et al.* [8] and Djošić *et al.* [9]. They proposed that the precipitation of HA occurred by consuming the OH⁻ ions generated by water reduction according to the reaction:

$$10Ca^{2+} + 6PO_4^{3-} + 2OH^- \rightarrow Ca_{10}(PO_4)_6(OH)_2 \quad (7)$$
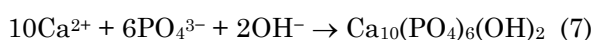
However, if this reaction occurs, the pH near the cathode should have decreased in our experiments, not increased as our observations showed. In Montero and Djošić's experiments, the solution with an initially high pH was stirred during electrolysis, and they did not monitor the solution pH as a function of time. Of course, as they suggested, the first step in order for reaction (6) to occur should be the liberation of Ca²⁺ ions from EDTA:

$$Ca(EDTA)^{2-} \leftrightarrow Ca^{2+} + EDTA^{4-} \quad (8)$$

The equilibrium constant of this reaction is pH dependent. The equilibrium constant increases as the pH increases. Thus, in our case, when the concentration of OH⁻ increased due to water reduction, i.e., the pH increased, the reaction shifted to the right, i.e., liberated more Ca²⁺ ions. These Ca²⁺ ions then reacted with HPO_4^{2-} and H_2O to form HA according to
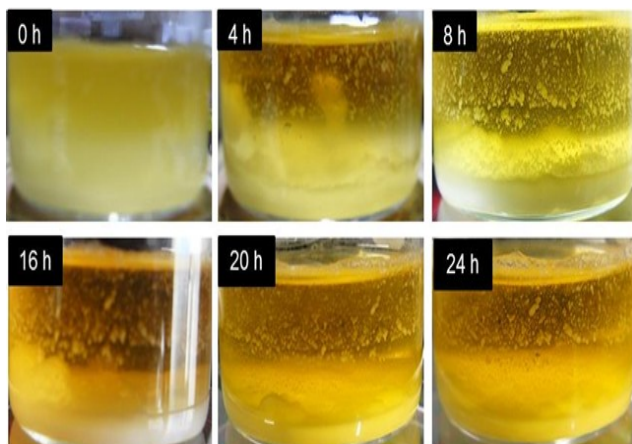
Equation (6).

When the applied potential was stopped by turning off the DC power supply, the electric field between the electrodes suddenly disappeared, and the particles settled rapidly due to gravity (Figure 7). Then, the suspension was aged at room temperature for 24 h. Figure 8 shows SEM images of the particles before and after aging for 24 h. It can be observed that the particles become smaller after aging. The dissolution may occur during the aging process. To further understand this process, both particles were characterized using XRD. Figure 9 shows the XRD patterns of the particles before and after aging. The patterns for both cases have characteristic peaks (indicated by circles) that match the standard pattern of HA (JCPDS 03-0747). There are diffraction peaks, indicated by square symbols, from substances other than HA in the XRD patterns of the particles after aging. The diffraction peaks match the standard pattern of brushite (AMCSD 0008884, monoclinic, a = 5.812 Å, b = 15.18 Å, c = 6.239 Å). It seems that during ag-
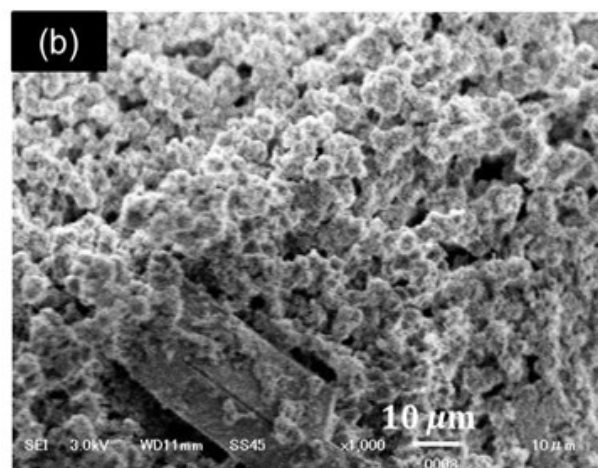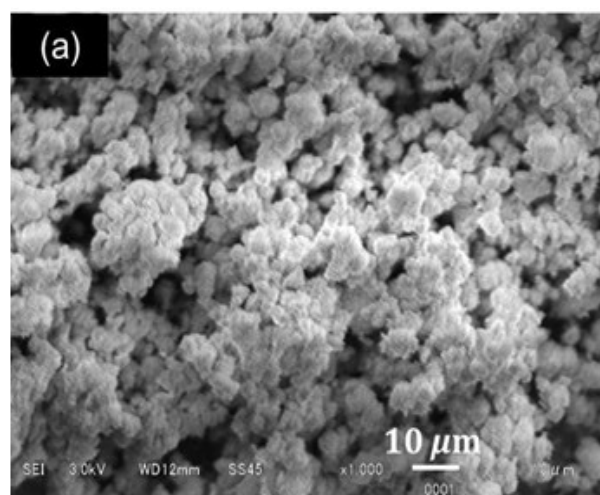
Figure 8. SEM images of the particles (a) before and (b) after aging for 24 h.

Figure 7. Time-resolved pictures of particles during the aging process.

ing, HA particles dissolve and then the reversible reaction between brushite (CaHPO$_4$) and HA occurs:

$$10CaHPO_4 + 2OH^- \leftrightarrow Ca_{10}(PO_4)_6(OH)_2 + 4PO_4^{3-} + 10H^+ \tag{9}$$
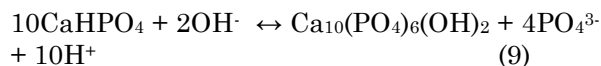
This finding suggests that if HA is desired, the particles should be removed immediately from the system once they form to avoid the formation of brushite.

We conclude that the electro-generation of OH$^-$ ions appears to play an important role in the formation of HA using the electrochemical method. The concentration of OH$^-$ must be sufficiently high to liberate Ca^{2+} from EDTA for the formation of HA to occur. It should be noted that the mechanism of HA formation is completely different from that described by Montero *et al.* [8] and Djošić *et al.* [9]. In their studies, OH$^-$ ions generated by water reduction were consumed to form HA, which contradicts our finding that OH$^-$ ions were produced during the reaction.

4. Conclusions

We have demonstrated that HA particles can be prepared through the electrolysis of a solution containing Ca^{2+}, EDTA and PO$_4^{3-}$. During electrolysis, two layers with a clear boundary occur mainly due to the electrostatic effects on the charged particles. When the particle concentration is sufficiently high, coagulation occurs, and the particles increase in size. The larger particles fall down due to gravity. SEM observations show that the HA particles consist of spherical agglomerates. It appears that the electro-generation of OH$^-$ ions by water reduction at the cathode plays an important role in

the formation of HA by the electrochemical method. The OH$^-$ ions induce the liberation of Ca^{2+} ions and the dissociation of phosphoric acid, which serve as the reactants for the formation of hydroxyapatite. Aging the produced particle suspension for 24 h tends to transform HA to brushite. Thus, if HA is desired, the particles should be rapidly removed from the system to avoid the formation of brushite.

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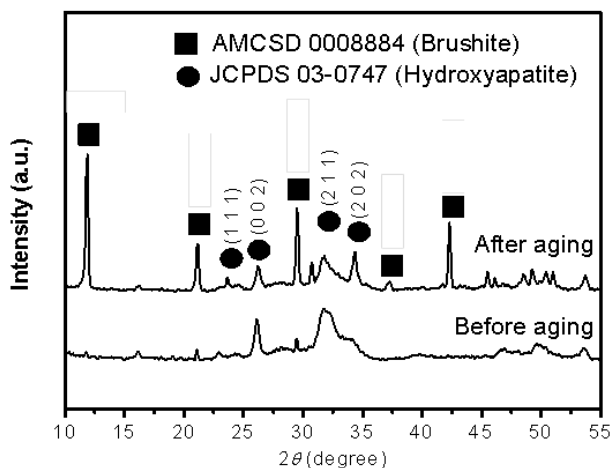
Figure 9. XRD patterns of the particles before and after aging for 24 h.

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