SYNTHESIS OF ULTRAFINE HYDROXYAPATITE IN A $\text{Ca}(\text{OH})_2\text{-H}_3\text{PO}_4\text{-H}_2\text{O}$ REACTION SYSTEM USING ULTRASOUND IRRADIATION

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Abstract: This paper reports the application of ultrasound irradiation in the synthesis of ultrafine hydroxyapatite (HAp) with a high specific surface area of $300 \text{ m}^2/\text{g}$. We examined the effects of initial temperature, horn diameter and suspension concentration on the powder properties, especially specific surface area, of ultrafine HAp. Amorphous calcium phosphate (ACP), which is a precursor of HAp, was formed in the early stage of the reaction, and then rapidly crystallized to form HAp. In the case of an initial temperature at 30°C , the largest specific surface areas of $250 \text{ m}^2/\text{g}$ and $300 \text{ m}^2/\text{g}$ were obtained using horn diameters of 19 mm and 6 mm, respectively. Based on particle size distribution measurements, the mode and average diameters of the HAp powders were found to be 5.0 and 6.3 nm, respectively. These values were well-agreed with the average diameter calculated from the specific surface area.

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INTRODUCTION

Hydroxyapatite (HAp) is very insoluble, and is used as, e.g., a biomaterial, ion-exchange material and catalyst. In general, HAp is synthesized by solid-phase reaction at around 1000°C , but synthesis is possible at 100°C or lower by liquid-phase reaction¹⁻⁴. The phosphate used in the synthesis of HAp is important. For example, calcium hydrogen phosphate dihydrate ($\text{CaHPO}_4 \cdot 2\text{H}_2\text{O}$, DCPD) is easily formed through the use of a Na_2HPO_4 solution⁵. Although HAp can be synthesized directly by using Na_3PO_4 , the Na^+ ion ends up in the HAp as an impurity. When the HAp is used as a biomaterial, such impurities may create lattice defects and reduce its performance¹. In addition, there are many reports on the synthesis of fine HAp, most of which synthesized the HAp by adding organic matter⁶⁻⁸. Furthermore, fine HAp has also been synthesized using powder jet deposition or microwave solvothermal synthesis^{9,10}. To our knowledge, the highest reported specific surface area of HAp is $256 \text{ m}^2/\text{g}$ ¹⁰. A sol-gel method and a $\text{Ca}(\text{OH})_2\text{-H}_3\text{PO}_4\text{-H}_2\text{O}$ reaction system are known as methods that avoid contamination of HAp by impurities^{11,12}. Fine HAp is desirable because it has good sintering properties. An ultrasound irradiation has been used to form fine particles^{13,14}.

The authors have reported the synthesis of fine particles of calcium carbonate and HAp by ultrasound irradiation during reaction^{15,16}. Characteristics, such as crystal shape, particle size and its distribution were influenced by the frequency and amplitude of the ultrasound. For example, we obtained calcium carbonate with a specific surface area of $100 \text{ m}^2/\text{g}$, which is approximately three times larger than that typically achieved without ultrasound irradiation, when carbon dioxide was bubbled into the calcium hydroxide suspension¹⁵. In addition, fine HAp with a specific surface area of $150 \text{ m}^2/\text{g}$ was synthesized using a $\text{CaCl}_2\text{-Na}_3\text{PO}_4$ reaction system under various Ca/P atomic ratios, ultrasound amplitudes and frequencies¹. In this study, we examined the effect of temperature, ultrasound amplitude and suspension concentration on the synthesis of impurity-free ultra-fine HAp particles in a $\text{Ca}(\text{OH})_2\text{-H}_3\text{PO}_4\text{-H}_2\text{O}$ reaction system.

EXPERIMENTAL

Calcium hydroxide ($\text{Ca}(\text{OH})_2$; purity 96%) and phosphoric acid (H_3PO_4 ; 60%) were used as raw materials for the HAp syntheses. All of the reagents were obtained from Kanto Kagaku Co., Ltd. (Japan) and were of high grade.

The ultrafine HAp was synthesized by liquid-phase reaction in a $\text{Ca}(\text{OH})_2\text{-H}_3\text{PO}_4\text{-H}_2\text{O}$ reaction system. An equivalent volume of 0.10 mol/dm^3 H_3PO_4 solution with a Ca/P atomic ratio of 1.67 was added to a 0.167 mol/dm^3 $\text{Ca}(\text{OH})_2$ suspension (60 cm^3) at $15 - 40^\circ\text{C}$. At this stage, the suspension concentration was 8.75 mass%, and the suspension concentration was changed from 7 to 12.25 mass%. The suspension was either mixed by mechanical stirring (200 rpm) or ultrasound irradiation for 20 min. Approximately 1 g of ultrafine HAp was obtained per reaction. The ultrasound homogenizer apparatus (250D Branson Sonifier, Branson, USA; 200 W, 20 kHz) had 19 mm and 6 mm diameter horns. The unit of ultrasound amplitude is watt density (W/cm^2), and it depends on the insertion depth of the horn, as well as on the volume and size of the container. However, it is not possible to simply compare the watt density at different frequencies. In the present study, the unit of amplitude is given in micrometers rather than watt density because the experimental design is such that the amplitude in micrometers can always be fixed. The actual energy input rate was approximately 95-97%¹. The ultrasound amplitude using the 19 mm diameter horn was 70%. The maximum elastic distance (amplitude 100%) of this horn is $135 \mu\text{m}$, and at 10% it is $19 \mu\text{m}$. Therefore, the elastic distance at an amplitude of 70% is $93 \mu\text{m}$. On the other hand, the maximum stretch distance (amplitude 100%) of the 6 mm diameter horn is $247 \mu\text{m}$, and at 10% it is $60 \mu\text{m}$. Therefore, the elastic distance at an amplitude of 30% is $122 \mu\text{m}$. The pH and temperature were also measured during the reaction. The tip of the horn was placed at a depth of 1 cm below the solution surface. The obtained HAp suspension was centrifuged (10000 rpm) and then filtered using a membrane filter to obtain ultrafine HAp. The ultrafine HAp particles were then stored in a vacuum dryer.

After the reaction, the resulting suspension was filtered, washed with pure water, and then dried under vacuum conditions. The obtained sample was characterized by powder X-ray diffractometry (XRD; Rigaku MultiFlex, Japan), infrared (IR) spectrometry (JASCO FT/IR-4600, Japan), field-emission electron microscopy (FE-SEM; Hitachi D-4500, Japan), scanning transmission electron microscopy (STEM, JEOL JEM-F200, Japan), nano particle size distribution measurement (TSI LiquiScan-ES 3980, USA) and BET specific surface area measurement (micromeritics GeminiVII, USA). The XRD was conducted at a scan rate of $8^\circ/\text{min}$, and the IR measurements were conducted using the KBr disk method.

RESULTS AND DISCUSSION

Effect of initial temperature on specific surface area of HAp

In these experiments, we used the horn with a diameter of 19 mm. Diffraction peaks of DCPD and unreacted $\text{Ca}(\text{OH})_2$ were observed when the reaction was carried out with stirring in the $\text{Ca}(\text{OH})_2\text{-H}_3\text{PO}_4\text{-H}_2\text{O}$ system reaction. It is thought that neutralization of H_3PO_4 resulted in the formation of HPO_4^{2-} ions, which reacted with Ca^{2+} ions to generate DCPD. It was confirmed that HAp was only generated when the system was irradiated with ultrasound. However, the temperature of the suspension rose to near 80°C as the irradiation continued, and the specific surface area of the HAp began to decrease when the temperature became too high. Thus, we cooled the reaction vessel with ice from the outside to prevent too much of a rise in temperature during ultrasound irradiation.

FIGURE 1 Change in suspension temperature during ultrasound irradiation.

Frequency: 20 kHz, Amplitude: 70 %, Ca/P atomic ratio: 1.67, Initial temperature: ● 15 °C, ● 25 °C, ● 30 °C, ● 40 °C

Figure 1 shows the change in suspension temperature during ultrasound irradiation. The temperature of the suspension increased regardless of the initial temperature. That is, at an initial temperature of 15°C , the temperature rose linearly and reached up to 38°C after 8 min. At an initial temperature of 25°C , the temperature of the suspension reached to 38°C after 4 min and then rose gradually. When the initial temperature was at 30°C , it reached to 41°C after 4 min and then equilibrated. Finally, in the case of an initial temperature at 40°C , the temperature only increased by 1°C .

FIGURE 2 X-ray diffraction patterns of the product synthesized at various reaction times (initial temperature: 40 °C).

(a): Commercial HAp, (b) – (f): Synthesized HAp
Reaction times: (b) 0.5 min, (c) 1 min, (d) 2 min, (e) 3 min, (f) 4 min

The ○ symbols indicate HAp peaks.

X-ray diffraction patterns recorded at various irradiation times (reaction time) for an initial temperature of 40 °C are shown in Figure 2. The peak was hardly observed after 0.5 min of ultrasound irradiation, and it appeared to be a halo observed in the X-ray diffraction of glass. This is considered to be due to the generation of amorphous calcium phosphate (ACP), which is a precursor of HAp. As ACP is generated at an initial temperature of 40 °C, it is inferred that ACP is also generated at the initial stage of the reaction for all lower initial temperatures. Diffraction peaks attributable to HAp were observed around $2\theta = 26^\circ$ and 32° at a reaction time of 1 minute. It was confirmed that the $2\theta = 32^\circ$ peak was not separated as compared with the reagent HAp, and the crystallinity was considerably low. There was almost no change in the diffraction peak even as the reaction time was increased. It was expected that decreasing the initial temperature would delay crystallization from ACP to HAp.

As X-ray diffraction does not reveal the presence of ACP, IR spectra were measured. Figure 3 shows IR spectra of the sample at various reaction times for an initial temperature of 40 °C. At a reaction time of 0.5 min, an absorption attributable to phosphate ions (PO_4^{3-}) near $1000 - 1100 \text{ cm}^{-1}$ was noticeably rounded^{17,18}. This occurs when the PO_4^{3-} ions are not regularly arranged, suggesting the formation of ACP. However, after 1 minute of reaction time, the PO_4^{3-} ion was split into two, which indicates the disappearance of ACP. That is, when the reaction is carried out at an initial temperature of 40 °C, ACP is considered to crystallize to form HAp

FIGURE 3 IR spectra of the product synthesized at various reaction times.

Initial temperature: 40 °C, Reaction times: (a) 0.5 min, (b) 1 min, (c) 2 min, (d) 3 min, (e) 4 min

within 1 minute. Almost no further change was observed in the IR spectrum when the reaction time was 2 min or more. The adsorption near 1450 cm^{-1} observed in all cases corresponds to CO_3^{2-} ions but is insignificant in the context of this study. Furthermore, no OH absorption was observed in the IR spectra of the reagent and ultrafine HAp.

FIGURE 4 Effect of initial temperature on specific surface area of HAp.

Initial temperature: ● 15 °C, ● 25 °C, ● 30 °C, ● 40 °C

Figure 4 shows the effect of initial temperature on the specific surface area of HAp. The specific surface area of the product at an initial temperature of 40 °C was about $105 \text{ m}^2/\text{g}$ after 0.5 min of reaction, which is because the product is primarily still ACP at this stage. The specific surface area of ACP varies depending on the synthesis method, and a range of $60 - 155 \text{ m}^2/\text{g}$ has been reported^{19,20}. The specific surface area increased as it crystallized. On the other hand, as the value of the specific surface area after 2 min of reaction at an initial temperature of 30 °C exceeds

200 m²/g, most of the ACP is expected to crystallize to HAp. The maximum specific surface area of 250 m²/g was achieved after 3 min of reaction and an initial temperature of 30 °C. ACP crystallization was delayed due to the lower initial temperature. However, too much ultrasound irradiation (5 min) lead to a decrease in the specific surface area of HAp to 150 m²/g, which is considered to be due to crystal growth of HAp. For an initial temperature of 25 °C, the specific surface area reached a maximum by 5 min, whereas a maximum was reached after 7 min for an initial temperature of 15 °C. The time required to reach a maximum specific surface area decreased as the initial temperature increased. The reason is considered to be that ACP becomes more unstable at higher temperatures, and crystallization into HAp is promoted.

FIGURE 5 FE-SEM (a) and TEM (b) micrographs of HAp with a specific surface area of 250 m²/g. Initial temperature: 30 °C, Reaction time: 3 min

Figure 5 shows FE-SEM and TEM micrographs of HAp with a specific surface area of 250 m²/g. Based on the FE-SEM image, the HAp particles were estimated to be 10 – 50 nm in size, and it was observed that they were aggregated in an irregular shape. In addition, thin plate-like particles were observed in the TEM image.

Effect of horn diameter and suspension concentration on specific surface area of HAp

An initial temperature of 30 °C was found to provide the largest specific surface area for synthesizing ultrafine HAp when using a horn diameter of 19 mm. Next, horn diameter and suspension concentration were varied to attempt to increase the specific surface area of HAp.

Firstly, the horn diameter was changed from 19 mm to 6 mm. The elastic distance can be increased by shortening the horn diameter, so it is considered possible to accelerate the reaction locally. Figure 6 shows the effect of reaction time on the specific surface area of HAp. The specific surface area of the product at a reaction time of 1 minute was about 120 m²/g, which indicates that ACP is the product at this stage. ACP gradually crystallized into ultrafine HAp

FIGURE 6 Effect of reaction time on specific surface area of HAp synthesized using a horn diameter of 6 mm. Frequency: 20 kHz, Amplitude: 30%, Initial temperature: 30 °C

FIGURE 7 Particle size distribution of HAp with a specific surface area of 300 m²/g. Horn diameter: 6 mm, Initial temperature: 30 °C, Reaction time: 3 min

with increasing reaction time, which is reflected in the increase in specific surface area. The specific surface area at 2 min increased compared to the results in Figure 3. It was possible to synthesize ultrafine HAp with a specific surface area of 300 m²/g in 3 min; however, the specific surface area of HAp decreased after 4 min of reaction time. The mean particle size of the HAp with specific surface area of 300 m²/g was calculated to be 6.3 nm.

We measured the particle size of HAp using different methods, particle size distribution measurement and BET specific surface area measurement. The measured particle size distribution of HAp with a specific surface area of 300 m²/g using LiquiScan-ES 3980 is shown in Figure 7. The particle size was distributed in the range of 3 – 20 nm and the mode diameter was 5.0 nm. Furthermore, the mean particle size was 6.3 nm, which was slightly lower but in good agreement with the value calculated

based on the specific surface area. The value was slightly lower than the calculated value.

FIGURE 8 Effect of suspension concentration on specific surface area of HAp.

Amplitude: 30%, Horn diameter: 6 mm, Initial temperature: 30 °C, Reaction time: 3 min

FIGURE 9 STEM micrographs of HAp with a specific surface area of 300 m²/g.

Amplitude: 30%, Initial temperature: 30 °C, Time: 3 min, (a): BF image, (b): DF image

Next, the effect of the suspension concentration on the specific surface area of HAp was examined, and the results are shown in Figure 8. The specific surface area of HAp exhibited a maximum of 300 m²/g at a suspension concentration of 8.75 mass% when the initial temperature was 30 °C and the reaction time was 3 min. The specific surface area slightly decreased with increasing suspension concentration, but by less than 10%. It was confirmed that a large amount of fine HAp could be synthesized at a suspension concentration of 12.25 mass%. Although the specific surface area decreased when the suspension concentration was too low, this is thought to be because the fine HAp particles dissolve.

Figure 9 shows a bright-field (BF) and a dark-field (DF) STEM micrograph of the synthesized HAp with a specific surface area of 300 m²/g. Based on the BF image, the particles aggregated into ellipses of 5 – 10 nm in size. Thus, it was confirmed that the synthesized HAp particles were not uniform.

Even in Figure 5, the smallest particle was 5 nm, the largest particle was about 10 nm, and the average was about 6.7 nm. In the dark-field (DF) image, the elliptical or elongated particles are rounded, as in the BF image.

CONCLUSION

Ultrafine HAp with a high specific surface area was synthesized by ultrasound irradiation in a $\text{Ca}(\text{OH})_2\text{-H}_3\text{PO}_4\text{-H}_2\text{O}$ system reaction. In this reaction system, ACP was formed immediately after the addition of phosphoric acid, and ultrafine HAp was obtained upon crystallization of the HAp. When ultrasound was irradiated for 3 min using a horn diameter of 19 mm and an initial temperature of 30 °C, HAp with a specific surface area of 250 m²/g was obtained. Furthermore, when the horn diameter of the ultrasonic wave was 6 mm, ultrafine HAp with a specific surface area of 300 m²/g was obtained, which corresponds to the highest specific surface area for HAp reported to date. From our particle size distribution measurements, the mode diameter of this HAp was found to be 5.0 nm, and the mean diameter was 6.3 nm. The maximum concentration was obtained at a suspension concentration of 8.75 mass%, but HAp with a relatively high specific surface area was obtained even at 12.25 mass%. In future work, we plan to study the sinterability and catalytic properties of this ultrafine HAp.

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