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# 2. Synthesis of Hydroxiapatite from Muscle Shell Waste Using The Precipitation Method

*by Ika Nawang Puspitawati*

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## Synthesis of Hydroxyapatite from Muscle Shell Waste Using The Precipitation Method

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### Abstract

Hydroxyapatite is a calcium phosphate compound that is the main inorganic component of bones and teeth. Hydroxyapatite has a main role in the medical world because of its identical chemical properties and structure to human bone. Kupang shells have a high CaCO<sub>3</sub> content of 95-99% by weight. So this concurrence, the mussel shells are to be used as good as an ingredient for making Hydroxyapatite. The variables used in this research are Temperature of the Furnace and Phosphoric Acid Concentration, which were 700°C, 750 °C, 800 °C, 850 °C, 900 °C and Phosphate Acid Concentrations 0.4M, 0.6M, 0.8M, 1M, and 1.2M. The product results were tested with XRD analysis to determine the crystal structure contained in the product and the level of Hydroxyapatite in the product. The best research results in this study at a temperature of 900°C at a concentration of 1.2M phosphoric acid with a Hydroxyapatite content of 100% with lattice parameters an (Å)=b (Å) = 9.422, c (Å)=6.8835, and gamma = 120°.

**Keywords:** Hydroxyapatite, Precipitation, Temperature, Concentration

### 1. Introduction

Indonesia is an archipelagic country located between two large oceans, specifically the Pacific Ocean and the Indian Ocean, so it has excellent fishery potential resources [1]. Kupang is one of the marine biota or shellfish with a small size that lives on the bottom of the waters and is a fishery product that is commonly found in the coastal waters of Sidoarjo, East Java. Kupang is processed into a special food, lontong Kupang, favoured by the people of Sidoarjo and Surabaya. Food production is carried out daily, so the mussel shell waste produced is also abundant [2]. That is one of the reasons for using research materials. In addition, the main constituent of mussel shells is CaCO<sub>3</sub>, with 95-99% by weight. Therefore, mussel shells can be a high source of

calcium [3][4]. Through the right technology, mussel shell waste can be a product with high economic value, namely hydroxyapatite or (hydroxyapatite/HAp)[5][6].

Hydroxyapatite (HAp) is a kind of apatite material [M<sub>10</sub>(XO<sub>4</sub>)<sub>6</sub>Z<sub>2</sub>][5] with the chemical formula Ca<sub>10</sub>(PO<sub>4</sub>)<sub>6</sub>OH<sub>2</sub>. The composition of the constituent elements (in ideal weight %) is Ca 39.9%, P 18.5%, H 0.2%, O 41.41%, and the ideal calcium-phosphate (Ca-P) ratio is 1.67. The type of crystal structure of hydroxyapatite can be monoclinic or hexagonal. The monoclinic hydroxyapatite structure was gained only under pristine conditions with stoichiometric composition, while the hexagonal hydroxyapatite structure was generally obtained from 1:1.67-stoichiometric hydroxyapatite synthesis. The lower the (Ca-P) molar ratio value, the more

acidic it is and the more soluble it is [7][8][9]. The monoclinic structure (Fig. 1) is due to the OH<sup>-</sup> arrangement forming the OH-OH-OH-OH<sup>-</sup> sequence, whereas the hexagonal structure (Fig. 2) can also be obtained under stoichiometric conditions if the OH<sup>-</sup> the arrangement is irregular [10].

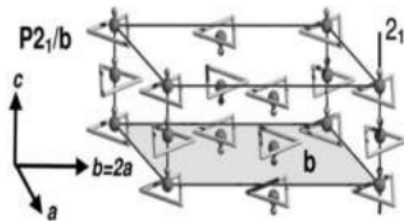


Fig. 1 Monoclinic Hydroxyapatite (HAp) Structure

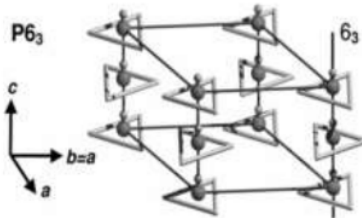


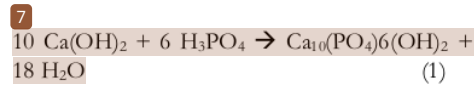
Fig. 2 Hexagonal Hydroxyapatite (HAp) Structure

Hydroxyapatite (HAp) has a density of 3.156 g/cm<sup>3</sup> and a molecular weight of 502.31 g/mol. Hydroxyapatite is a bioceramic compound in solid form. The components of hydroxyapatite are similar to those of bone. In the collagen matrix network in bone, hydroxy ions (OH<sup>-</sup>) can be replaced by F<sup>-</sup>, Cl<sup>-</sup>, and CO<sub>3</sub><sup>2-</sup>[11].

One of the applications of hydroxyapatite (HAp) can be used as bioceramics in the medical and dental fields. Hydroxyapatite has bioactive, osteoconductive properties and low electrical and thermal conductivity. In addition, hydroxyapatite has excellent biocompatibility properties and a high affinity for biopolymers. Hydroxyapatite can be synthesised by mixing calcium precursors with phosphate precursors[12][13][14].

Various methods have been developed to make hydroxyapatite by previous researchers, including the hydrothermal method; the sol-gel method and the precipitation method

[15][16][17]. The precipitation method is the leading wet chemical widely used to synthesise hydroxyapatite. This technique can synthesise large amounts of HA without using organic solvents and is inexpensive. This method uses sundry precursors that have contents of calcium and phosphate, such as calcium hydroxide (Ca(OH)<sub>2</sub>) and phosphoric acid (H<sub>3</sub>PO<sub>4</sub>). Water is the only by-product, and the reaction does not involve foreign elements [18][19].



The precipitation process is an acid-base reaction that produces crystalline solids (reaction salts) and water. The advantages of this method are the process with cheap raw materials, relatively simple chemical reactions, and the size and homogeneity of the particle sizes obtained tend to be good. A simple process with yields >87% makes it very suitable for large-scale (industrial) production, the absence of foreign contaminants and the only byproduct produced is water, requiring inexpensive reagents. Also, Ca/P products with the composition of varying phases can be obtained [16].

Previous researchers have synthesized hydroxyapatite from cow bone waste Yenti et al. The materials used for Ca and P precursors are Ca(OH)<sub>2</sub> and H<sub>3</sub>PO<sub>4</sub>. Then it was found that hydroxyapatite was more dominant at a (Ca-P) ratio of 1,67. As well as variations in the concentration of H<sub>3</sub>PO<sub>4</sub>, HAp was more dominant at concentrations of 0.8 and 1.2 M H<sub>3</sub>PO<sub>4</sub> [16]. This research will use the precipitation method with different raw materials from mussel shell waste. The resource calcium comes from the shell of mussels and phosphoric acid by controlling the temperature or even the concentration of the solvent.

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## 2. Material and Method

### 2.1 Material

The primary material of mussel shells used in this study was mussel shells in Balungdowo Village, Candi sub-district, Sidoarjo district, East Java. Other ingredients include phosphoric acid and NaOH purchased at the Ngagel Jaya Kimia shop, Surabaya.

## 2.2 Production of Hydroxyapatite Synthesis

This research uses a series of calcination tools consisting of a furnace and a furnace pot for synthesising Hydroxyapatite using a magnetic stirrer, glass beaker, and stir bar. The series of tools for the hydroxyapatite synthesis process below with the following descriptions of numbers 1 to 4 sequentially, namely aluminium cover, glass beaker, stirrer bar, magnetic stirrer

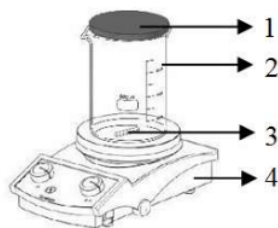


Fig. 3 The Set Up of Hydroxyapatite Synthesis

The variable used is the stirring speed of 300 rpm, and the pH is set to 10. The variable changes in this study are the furnace temperature of 700°C, 750°C, 800°C, 850°C, and 900°C and the concentration of phosphoric acid is 0.4M, 0.6M, 0.8M, 1M and 1.2M

## 2.3 Preparation of Primary Material

Preparing primary materials begins with cleaning the mussel shell with a brush and running water. Then it is dried in the open air and under the sun. Then it was pulverized with a wooden pulverizer to a powder size of about 100 mesh.

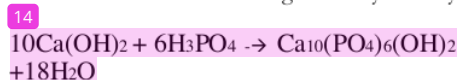
## 2.4 Calcination Process

After the mashed mussel shells, it calcined in a furnace at 900 °C for 5 hours. After it was calcined, the mussel's shell was then sieved with a mesh sieve according to the provisions of 100 Mesh.

## 2.5 Hydroxyapatite Synthesis Process

The calcium hydroxide suspension was stirred at 300 rpm for 1 hour while the temperature was maintained at 37°C, and the pH was adjusted to 10. Then the mixture was aged

at room temperature for 24 hours. After that, the precipitate formed was filtered using Whatman 42 filter paper. Then the precipitate was dried using an oven at 100°C for 5 hours. The dry precipitate obtained was then heated at a temperature corresponding to the changing variables (700°C, 750°C, 800°C, 850°C, 900°C) for 3 hours to increase the degree of crystallinity.



## 2.6 Hydroxyapatite Analysis with XRD

The characterization of the synthesized hydroxyapatite was carried out using X-ray Diffraction (XRD). Before being used, the tool is calibrated first and the control XG (in the form of current), water flow, shutter and door open are set. While waiting for the calibration of the instrument, as much as 2 mg of the sample was placed in the holder of the diffractometer. The voltage used is 40 Kv and the generator current is 30 mA with a wavelength source of 1.5406 Å. The results obtained in the form of a diffractogram identified based on the intensity and angle of 2θ. termination of the emerging phase refers to the Joint Committee on Powder Diffraction Standard (JCPDS).

## 3. Results and Discussion

### 3.1. Kupang Shell Calcination

The calcination process in mussel shell waste aims to eliminate organic components and convert the initial compound calcium carbonate (CaCO<sub>3</sub>) into Calcium Oxide (CaO) compounds which will later be used as Calcium (Ca) precursors. The calcination process at a temperature of 250°C. The water will evaporate then all organic components will be oxidized below a temperature of 450°C. At a temperature of 540°C, calcium carbonate (CaCO<sub>3</sub>) decomposition into CaO at a temperature of 750°C [5].



HAp was synthesized using the precipitation method with variable furnace temperatures of 700°C, 750°C, 800°C, 850°C, and 900°C. The hydroxyapatite was tested using XRD analysis, and the results were compared to standard hydroxyapatite.



### 3.2. Effect of Temperature on HAp Concentration

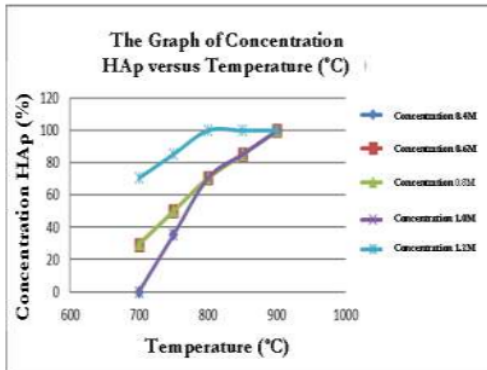


Fig. 4. The Graph of Concentration HAp versus Temperature (°C)

Based on Figure 4, shows that the higher the temperature equivalent with the yield of hydroxyapatite obtained, and at a concentration of 0.4 M phosphoric acid with the lowest value at a temperature of 700°C and the good results at a temperature of 900°C, the following graphs of XRD analysis results at a concentration of phosphoric acid at 900°C.

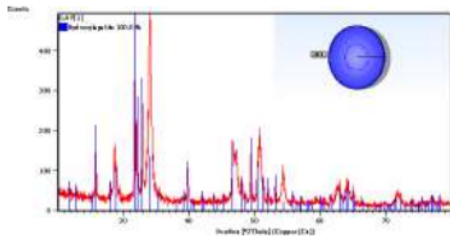


Fig. 5 The Graph of result Analisa XRD with Poshpate Acid Concentration 0.4M and Temperature 900 °C

In the graph, a temperature of 900°C and concentration of 0.4 M peaks the highest intensity at an angle of 2θ: 34.0818 hydroxyapatite 100% obtained with 455.87cts height.

Table 1. Hydroxyapatite kisi parameter value 0.4M

Temperature	900°C	850°C	800°C	750°C	700°C
a (Å)	9,41	9,413 3	9,41 66	9,38 33	9,35
c (Å)	6,879	6,876 7	6,87 45	6,87 82	6,88 2
Gamma (°)	120	120	120	120	120

Table 1 shows that at a temperature of 900°C the best value got 100% hydroxyapatite acid, lattice parameters an (Å) = b (Å) = 9.41, c (Å) = 6.879, and gamma = 120°. That shows that the crystal structure of the product is close to the literature, which shows the lattice parameters a = b = 9.432 Å, c = 6.881 Å, and gamma = 120°.

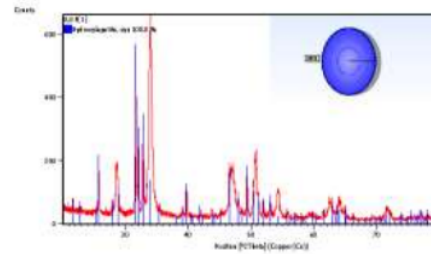


Fig 6. The Graph of result Analisa XRD with Poshpate Acid Concentration 0.8M and Temperature 900 °C

The graph above shows the position of the angle 2θ: 34.0467 also obtained a hydroxyapatite intensity of 100% with a height of 606.71 cts.

Table 2. Hydroxyapatite kisi parameter value 0.8M

Temperature	900°C	850°C	800°C	750°C	700°C
a (Å)	9,418	9,412	9,424 4	9,423 1	9,421 8
c (Å)	6,8835	6,884 2	6,885	6,883 1	6,881 3
Gamma (°)	120	120	120	120	120

Table 2. it can be seen that the excellent value was had at a temperature of 900°C, which is 100% hydroxyapatite acid, with lattice

parameters  $a$  ( $\text{Å}$ ) =  $b$  ( $\text{Å}$ ) = 9.418,  $c$  ( $\text{Å}$ ) = 6.8355, and  $\gamma$  =  $120^\circ$ .

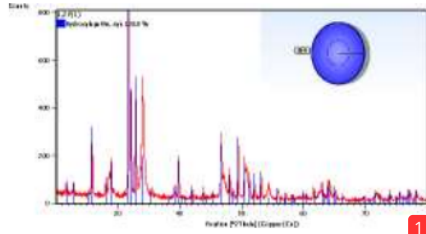


Figure 7. Graph of XRD analysis results at a concentration of 1.2M phosphoric acid at a temperature of 900 °C

The perfect result where the concentration variable 1.2M with a temperature of 900°C shows an angle position of  $2\theta$ : 31.7092 with a 100% Hydroxyapatite presentation with a height of 784 cts.

Table 3. Hydroxyapatite kisi parameter value 1.2M

Temperature	900°C	850°C	800°C	750°C	700°C
a (Å)	9,422	9,42	9,418	9,414	9,41
c (Å)	6,8835	6,8867	6,89	6,869	6,848
Gamma (°)	120	120	120	120	120

Based on table 3. it showed that at a temperature of 900°C the finest value was obtained, which is hydroxyapatite acid of 100%, with lattice parameters  $a$  ( $\text{Å}$ ) =  $b$  ( $\text{Å}$ ) = 9.422,  $c$  ( $\text{Å}$ ) = 6.8835, and  $\gamma$  =  $120^\circ$ . The product's crystal structure is close to the literature, which shows  $a = b = 9.432 \text{ Å}$ ,  $c = 6.881 \text{ Å}$ , and  $\gamma = 120^\circ$  of the lattice parameters.

According to the picture below, the hydroxyapatite of the diffractogram shows a similar characterisation result pattern from the XRD analysis of hydroxyapatite from the JCPDS Committee on Powder Diffraction Standards Data. According to de Wolf (JCPDS 09- 0432), the three significant peaks of hydroxyapatite are at  $2\theta$ ; 31.77°, 32.90° and 32.19° respectively. The concentration of 1.2M is close to the current value at  $2\theta$ ; 31.71°; 32.11°; and 32.86°. The value

of the hydroxyapatite intensity is 100% at the peak altitude of 784 cts.

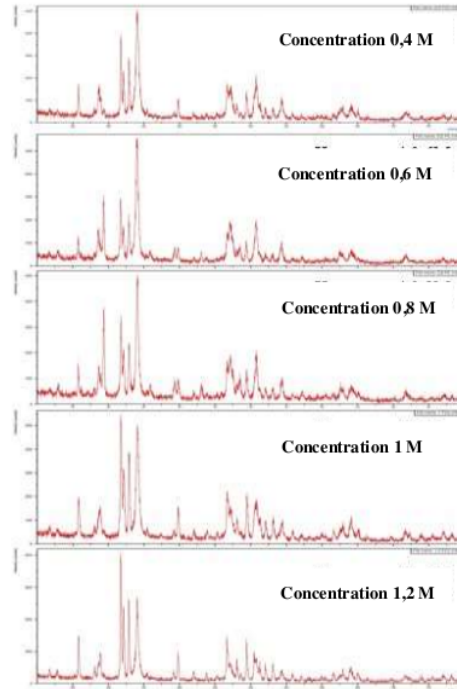


Fig 8. Graph of the comparison of all concentrations at a temperature of 900°C

#### 4. Conclusions

The shell waste of mussel shells can be used as the primary material for the manufacture of hydroxyapatite. Increasing the furnace's temperature would make the intensity of hydroxyapatite that would be produced more significant. The concentration of phosphoric acid does not affect the product's concentration but affects the product's lattice parameters. The best results were at a temperature of 900°C and at a concentration of 1.2 M phosphoric acid, which obtained 100% hydroxyapatite acid, with lattice parameters  $a$  ( $\text{Å}$ ) =  $b$  ( $\text{Å}$ ) = 9.422,  $c$  ( $\text{Å}$ ) = 6.8835, and  $\gamma$  =  $120^\circ$ .

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## 2. Synthesis of Hydroxiapatite from Muscle Shell Waste Using The Precipitation Method

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