The Impact of Mg Concentration, Microwave Irradiation Time, and Sintering on Magnesium-Hydroxyapatite Synthesis

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Abstract. Hydroxyapatite synthesis is an inorganic compound that makes up the hard tissues of the human body such as bones. This study aims to substitute Magnesium (Mg) ions, which have an important role in the structure and function of the human body, in Calcium (Ca) ions from Hydroxyapatite. MgHA crystals are synthesized by mixing a solution of diammonium hydrogen phosphate and magnesium hydroxide into a calcium hydroxide solution which is then irradiated with microwaves, with variations in the concentration of Mg and irradiation time. From the XRD results show that along with the increase in Mg concentration and irradiation time the lattice parameter values a and c are reduced by 0.03 in lattice a and 0.01 in lattice c. The increase in irradiation time is proportional to the increase in crystal size (L) and crystallinity index (CI). At t = 35 it is found L = 19.08 nm and CI = 0.14. The increase in Mg concentration is proportional to the increase in crystal size and crystallinity index, the increase in Mg concentration above 0.6 M shows the presence of saturation in the binding process of Mg in the apatite structure. The sintering process at 900ºC increases the crystal size value from 19.08 nm to 52.09 nm and the crystallinity index from 0.14 to 2.97. Morphology of MgHA produces rod-shaped particles with agglomeration caused by a large amount of Mg content in apatite.

Keywords: Magnesium-Hydroxyapatite, Microwave Irradiation, Biomaterial

1. Introduction

Bone is a connective tissue formed by microscopic phosphate calcium crystals from the matrix of collagen and hydroxyapatite. Bone consists of 65% inorganic components such as hydroxyapatite minerals, calcium phosphate, calcium carbonate, and magnesium. And it contains 35% organic components such as collagen which include collagen and osteogenic [1].

Hydroxyapatite is an inorganic compound making up hard tissue of the human body such as bones and teeth. This material is biocompatibility, bioactivity, and osteoconductivity, thus making hydroxyapatite (HA) suitable as a biomaterial [2]. Calcium and phosphorus are the main elements in the formation of hydroxyapatite so that it has a composition and crystal structure similar to bone and is currently the most widely used material in biomedical applications [3].

Magnesium (Mg) has an important role in the structure and function of the human body. The adult human body contains approximately 25 grams of magnesium [4]. The distribution of magnesium in the body is estimated to be 66% in bone, 33% in muscle and soft tissue, and approximately 1% in the blood. Magnesium in the bone serves to regulate metabolism in the bone. By substituting Mg on HA, it can reduce crystallinity and increase its bioactivity [5].
This study aims to produce magnesium-hydroxyapatite crystal (MgHA) using microwave irradiation method by knowing the effect of variations in Mg concentration, irradiation time, and sintering on the characteristics of MgHA. The samples produced were irradiated by microwaves at 540 W with variations in irradiation time of 15 minutes, 25 minutes, and 35 minutes with variations in the concentration of Mg 0.2 M to 1 M at intervals of 0.2 M and followed by the sintering process.

The difference between synthetic hydroxyapatite and those contained in bone minerals is in the bones with other mineral ion ions such as CO, F, Mg and Na ions, so that to enhance the biological properties of synthetic HA substitution, ion ions can be carried out [4]. It can be substituted with Ca ions contained in HA in the cationic ion. By doing substitution the mineral ion will change the size of the crystal and there will be an inhibitory effect on crystallization and result in a reduction in thermal stability.

The substitution process of other ions into the HA structure causes a change in apatite size, this is due to differences in the size of Ca and Mg ions, where Mg has a smaller atomic radius of 173 pm and Ca at 231 pm. In addition, other ion substitutions will cause inhibition of the crystallization process in HA.

Microwave irradiation processes have several advantages, namely high energy efficiency, high heating rate, volumetric heating from the inside to the outside and diverse heating [1] and relatively short heating times [6].

2. Materials and Methods

2.1 Synthesis of MgHA

This study mixed three chemicals, namely calcium hydroxide, diammonium phosphate, and magnesium hydroxide to obtain magnesium-hydroxyapatite with variations in Mg concentration (0.2 M, 0.4 M, 0.6 M, 0.8 M, 1 M) and microwave irradiation time (15, 25, 35 minutes) with a power of 540 Watts. The number of precursors was calculated based on the Ca/P molar ratio of 1,67. The tools used in this study include digital scales used to measure the mass of chemicals, spatulas are used to extract chemicals, burette and static are used for mixing methods, which are inserted into the burette is a solution of diammonium phosphate and magnesium hydroxide at a rate of 5 ml/minute, petri dishes are used to place samples, magnetic stirrers are used as magnetic stirrers, beaker glass is used for solution containers, ovens as drying processes and microwaves are used as microwave irradiation. The material used in the synthesis of this research is calcium hydroxide as a calcium precursor, diammonium hydrogen phosphate as precursor phosphate, magnesium hydroxide as a magnesium precursor, and aquades as a solvent. The results of this mixture will then be irradiated by microwaves and then put into the oven to carry out the drying process until the sample mass is constant. The dried sample is ready to carry out the characterization test process.

Figure 1. Schematic of synthesis MgHA using microwave irradiation.
2.2 Characterizations of MgHA
From the XRD pattern that has been obtained for MgHA, the calculation of the crystallinity index (CI) and crystallite size (L) is carried out in the Miller (002) field, this is because the measurements in fields (300) have more than one adjacent peak, however, smoothing from the tool consider some of these peaks as one peak, then, the use of the FWHM value from the field (300) is less accurate than the field (002).

The crystal size (L) of MgHA powder was determined using the Scherrer formula [7], as follows:

$$L = \frac{K\lambda}{\beta \cos \theta}$$

with K is a constant associated with a crystal form that has a value of 0.9, β is the peak width of the diffraction peak profile at half its maximum height (rad), λ is the X-ray wavelength (nm), and θ is the Bragg angle. The crystal size is measured in the Miller (002) field. The crystallinity index (CI) of MgHA powder was determined using the following equation [8]:

$$CI = \frac{0.24}{\beta}\frac{1}{3}$$

with β being the peak width at half maximum height or full width at half maximum (FWHM) (o) for the Miller (002) field, a value of 0.24 is a constant for various types of calcium phosphate apatite.

3. Result and discussion
3.1 X-Ray Diffraction
Based on the results of the characterization, the synthesis results performed have a high HA purity. From the XRD pattern with 35 minutes irradiation time in Figure 2 it can be seen that at each increase in Mg concentration the peak intensity in the plane (310) with the diffraction angle 38 which is the main angle of the Mg increases significantly. The peak at this angle is the peak arising from the Mg bond with the hydroxyl group, this shows that the higher the concentration of Mg the higher the intensity of the Mg contained in the synthesis results.

The deviation results for the average lattice parameters a and c for all variations of Mg concentration and irradiation time showed a reduction in the a and c axes when compared with the hydroxyapatite (HA) lattice parameter values with a = 9.417 Å and c = 6.880 Å. Similarly, the results of the deviation of the lattice parameter values of MgHA with lattice a = 9.4272 Å and c = 6.8614 Å which also shows the reduction of the a and c axis.

The change in deviation value, when viewed from the increase in Mg concentration, shows that the synthesis carried out results in a smaller axis change than the axis in the HA crystal. This is caused by Mg ions which have a smaller size than Ca ions. The lattice parameters tend to shrink on all three axes as Mg increases, so the apatite size is denser than pure HA, which means that Mg has been successfully substituted in the crystal side. The decrease in lattice value is caused by the nature of Mg as an inhibitor of HA growth. The decrease in lattice parameter values a and b against HA lattice parameter values tends to be greater for each increase in Mg concentration [10].

![Figure 2. XRD MgHA pattern with variation of Mg concentration at 35 minutes irradiation](image-url)
Table 1. Average lattice parameters a and c and MgHA deviation at 35 minutes irradiation

| Mg Concentration | $a = b$ (Å) | $c$ (Å) | Deviation from HA [14] | Deviation from MgHA |
|------------------|-------------|---------|------------------------|---------------------|
| 0.2 M            | 9.38        | 6.85    | -0.04                  | -0.03               |
| 0.4 M            | 9.38        | 6.84    | -0.04                  | -0.04               |
| 0.6 M            | 9.39        | 6.86    | -0.03                  | -0.02               |
| 0.8 M            | 9.38        | 6.86    | -0.04                  | -0.03               |
| 1 M              | 9.39        | 6.85    | -0.03                  | -0.04               |

Increased irradiation time tends to form more dense crystals with the lattice stretching towards the a-axis due to compressive stress along the c-axis. Reduction of the a-axis as a result of an increase in magnesium content was also reported by other studies [11]. This occurs because the size of the Mg ion is smaller than the Ca ion, which is 1.28 Å compared to 1.37 Å [12], thus forming a tight apatite structure. The length of the c-axis, which tends to shrink shows that most of the Mg ions in the initial mixture enter the crystal lattice. From the table, the lattice parameter values with a higher irradiation time produce deviation values that are close to the literature.

The increase in crystal size (L) and the crystallinity index (CI) when viewed by the increase in irradiation time, are associated with microwave irradiation energy that is transferred to the molecules that react. The longer the time of irradiation, the acceleration of atomic motion lasts longer so that collisions that occur more and more so that it causes a decrease in viscosity of solvents and the possibility of atoms binding to each other the higher [13]. The reduction in solvent viscosity triggers the diffusion barrier to decrease, so that with more irradiation time the atoms have a longer time to fill the crystal lattice with increased polarization and aggregation of apatite nuclei to form larger crystals followed by increased crystallinity [14].

Increases in L and CI values are shown in Figure 3 obtained in the following three variations of irradiation time. Based on the results of sintered MgHA characterization, it was seen that the peaks in the sintered MgHA were higher and pointed compared to the non-sintered MgHA. The formation of peak intensity and sharpness which increased significantly as well as not forming additional phases compared to those not sintered showed that crystals from HA had been formed. This proves that the results of the synthesis carried out have a high HA purity crystal [15].

The crystal size (L) of the field (002) obtained from sintered MgHA has a much higher value than the L value in the non-sintered MgHA. A significant increase in L and CI indicates that the sintering process is capable of producing a much larger form of apatite crystals. This change is influenced by the increase in temperature and time in the heating process of the sample. Crystallinity will be higher in proportion to the temperature rise in the sintering process. Very high temperatures of 900°C can cause atomic movements to accelerate. The longer time is also able to fill the crystal lattice by increasing the process of polarization and aggregation of apatite nuclei to form a larger crystal size followed by an increase in crystallinity [15].
Figure 3. The pattern of increasing the size of MgHA crystallites and crystallinity index by varying the concentration of Mg.

| Mg Concentration | (hkl) | 2 Theta | L (nm) | CI   |
|------------------|------|---------|--------|------|
| 0.2 M            | 25.95| 45.44   | 1.93   |
| 0.4 M            | 25.96| 47.1    | 2.28   |
| 0.6 M            | 25.97| 52.09   | 2.98   |
| 0.8 M            | 25.96| 43.33   | 1.68   |
| 1 M              | 25.99| 41.41   | 1.49   |

Figure 4. XRD MgHA pattern with variations of Mg with sintering 900°C for 5 hours and Crystallite size (L) and crystallinity index (CI) for MgHA at irradiation time of 35 minutes and sintered for 5 hours at 900°C.

3.2 Fourier Transform-IR

Figure 5. FTIR MgHA pattern with variation of Mg concentration with 35 minutes irradiation time.
Table 2. Functional groups obtained for MgHA with 35 minutes irradiation time and 900°C sintering for 5 hours.

| Function Group | 0.2 M | 0.4 M | 0.6 M | 0.8 M | 1 M |
|----------------|-------|-------|-------|-------|-----|
| Mg(OH)$_2$ (stretch) | -     | -     | 3690  | -     | -   |
| H$_2$O (stretch)       | -     | -     | -     | -     | 3420|
| CO$_2$ (bend)          | -     | -     | -     | 2362  | 2362|
| H$_2$O (bend)          | -     | -     | -     | 1648  | 1648|
| PO$_4$ (stretch)       | 1032  | 1032  | 1032  | 1032  | 1032|
| PO$_4$ (bend)          | 563   | 563   | 563   | 563   | 563 |

In FTIR spectra with 35 minutes of irradiation time without sintering, the peaks showed an Mg in the apatite structure. This peak arises at wavenumber 3690 in all spectra with different Mg concentrations due to a meeting of magnesium and hydroxyl groups, which causes stretching mode \[16\]. In the graph, it can be seen that MgHA with a concentration of 0.6 M has a very deep peak which means a very high stretch of Mg.

The \( \text{(PO}_4\text{)}^3^- \) function group is at wavenumber 1032 (stretch) and 563 (bend) \[14\]. The H$_2$O function group is shown at 3420 (stretch) and 1648 (bend), this functional group is related to water absorbed by MgHA \[17\]. A wavenumber 2362 there is a functional group for CO$_2$ dissolved by ceramic material \[18\], the peak is only found in MgHA with concentrations of Mg 0.6 M and 0.8 M only. In the functional group 1384 there is a peak that indicates the carbonation process \[6\], which is a reaction process between metal hydroxide and CO$_2$ contained in the atmosphere. In this study, there are two metal hydroxides, namely Ca(OH)$_2$ and Mg(OH)$_2$ which can react with CO$_2$ from the atmosphere, so that the functional group 1384, indicates the presence of Ca(HCO$_3$) or Mg(HCO$_3$).

In general, the resulting FTIR sintering spectrum has the same shape as the FTIR pattern that is not sintered because it has peaks that tend to be the same. Seen in the sintered MgHA, the Mg peak appears only at 0.6 Mg concentration.

In Table 2 it can be seen that many wavenumbers disappear. This occurs due to the sintering process. The \( \text{(PO}_4\text{)}^3^- \) function group is found at the same wavenumber 1032 (stretch) and 563 (bend). The H$_2$O function group shown at 3420 (stretch) only appears at concentrations of 1 M and 1648 (bend) only appear at a concentration of 0.8 M and 0.6 M. At wavenumber 2362, CO$_2$ only appears in the concentration of Mg 1 M.

3.3 SEM-EDS

The morphology of MgHA at Mg 0.6 concentration with a magnification of 20,000 times can be seen in Figure 6. The choice of variation in irradiation time and Mg concentration for SEM evaluation refers to FTIR results containing the most Mg. The results of SEM images indicate rod-shaped particles with an average particle size calculated using Fiji ImageJ software.

The shape produced in the non-sintered sample is seen as an agglomerated rod caused by the presence of Mg on apatite which is still amorphous. This shows that the Mg substitution results in agglomeration of the particles, the more the amount of Mg in the molecule, the more agglomeration that results in inhibiting apatite growth \[11\]. Amorphous properties are also evidenced by the very small crystal size of 19,083 nm.

It is different from the sintered sample. The morphological shapes obtained are shaped rods that are larger than those that are not sintered. This result proves that by doing sintering at very high temperatures it can produce larger crystals. The agglomeration in sintered sample is still visible due to the content of Mg in the apatite structure. By increasing the heating temperature by 900°C resulting in a
decrease in the particle agglomeration rate followed by an increase in the rate of fragmentation or separation of particles. So that with reduced agglomeration, the content of Mg on apatite decreases.

Figure 6. SEM image at irradiation time of 35 minutes with 0.6 Mg concentration and sintering for 5 hours

Table 3. Composition of MgHA elements

| Element | Not Sintered | Sintered |
|---------|--------------|----------|
| Ca      | 22.13 %      | 20.27 %  |
| P       | 11.37 %      | 10.70 %  |
| Mg      | 4.80 %       | 4.87 %   |
| Ca/P    | 1.51         | 1.47     |

This proves the results of XRD and FTIR where the peak Mg decreases after the sintering process is carried out but produces a larger crystal size of 152,098 nm. From the composition obtained through EDX characterization, it can be seen that Ca/P obtained is smaller than Ca/P from pure HA which is equal to 1.67. Reducing the Ca/P ratio shows the composition of Ca which becomes less due to the successful substitution of Mg in HA.

4. Conclusion

In general, based on the analysis of the results of the characterization of Magnesium Hydroxyapatite (MgHA) successfully synthesized using microwave irradiation method with a power of 540 W. The deviation value of the lattice parameters at each increase in Mg concentration and irradiation time shows a and c axes smaller than the HA lattice parameter values. Increase in irradiation time is proportional to the increase in crystal size (L) and crystallinity index (CI). At t = 35 it is found L = 19.08 nm and CI = 0.14. The increase in Mg concentration is proportional to the increase in crystal size and the crystallinity index, the increase in Mg concentration above 0.6 M shows saturation in the binding process of Mg in the apatite structure. The sintering process at 900ºC increases the crystal size value from 19.08 nm to 52.09 nm and the crystallinity index from 0.14 to 2.97. Morphology of MgHA produces rod-shaped particles with agglomeration caused by a large amount of Mg content in apatite. At the concentration of Mg 0.6 M, a lot of agglomeration is formed resulting from the buildup of Mg on the apatite surface. SEM results from sintering MgHA prove that the sintering process at 900ºC can produce larger rod-shaped crystals.
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