JBAT 14(1) (2025) 9 – 14

Jurnal Bahan Alam Terbarukan

Accredited SINTA 3 by Ministry of Research and Technology/National Research and Innovation Agency
SK No. 10/C/C3/DT.05.00/2025
http://journal.unnes.ac.id/nju/index.php/jbat

p-ISSN 2303 0623
e-ISSN 2407 2370

Synthesis of Hydroxyapatite from Green Mussel Shells Using Micro-wave Method Through Ultrasonic Mixing Process and Magnetic Stirrer

Agus Prasetyo[1,✉], Risky Ismail[1], Athanasius Priharyoto Bayuseno[1], Samsudin Anis[2], Deni Fajar Fitriyana[2], Muhammad Afrizal[1], Januar Parlaungan Siregar[3,4]

DOI: https://doi.org/10.15294/jbat.v14i1.25497

[1] Department of Mechanical Engineering, Faculty of Engineering, Universitas Diponegoro, Semarang, Indonesia
[2] Department of Mechanical Engineering, Faculty of Engineering, Universitas Negeri Semarang, Semarang, Indonesia
[3] Faculty of Mechanical and Automotive Engineering Technology, Universiti of Malaysia Pahang Al-Sultan Abdullah (UMPSA), Gambang, Pahang, Malaysia
[4] Centre for Automotive Engineering (Automotive Centre), Universiti Malaysia Pahang Al-Sultan Abdullah (UMPSA), Pekan, Pahang, Malaysia

Article Info

Article history:
Received
19 April 2025
Revised
19 May 2025
Accepted
26 May 2025
Published
June 2025

Keywords:
Biomaterial;
Calcium phosphate;
Hydroxyapatite;
Mixing method;
Synthesis

Abstract

Hydroxyapatite (HAp) is a biomaterial containing calcium phosphate with the chemical formula $Ca_{10}(PO_4)_6(OH)_2$ which is similar to the structure of human bone, so it is good for the purpose of healing human bones and teeth. HAp synthesis has several methods that can affect the results of HAp synthesis, such as the method of mixing CaO with phosphate, namely mixing with ultrasonic machines and magnetic stirrers. However, the discussion about the difference of HAp results with the mixing method of ultrasonic machine and magnetic stirrer is still rarely done. Therefore, it is important to examine and compare the characteristics of HAp synthesized by ultrasonic and magnetic stirrer mixing. The result of XRD test shows that HA1 has a purity of weight percentage (wt.%) of HAp crystal of 99.8%, while HA2 has a weight percentage (wt.%) of HAp crystal of 97.7%. For the FTIR test results of both specimens detected the presence of phosphate groups, and hydroxide, where both are the basic form of hydroxyapatite. Carbonate groups were also detected in the test, but it cannot be said to be bad because carbonate is a natural substitute for phosphate.

INTRODUCTION

Hydroxyapatite (HAp) is a biomaterial containing calcium phosphate with the chemical formula $Ca_{10}(PO_4)_6(OH)_2$ which is similar to the structure of human bone, so it is good for the purpose of healing human bones and teeth (Irfa et al., 2024; Pu'ad et al., 2020; Rachmantio & Irfai, 2023). Due to its biocompatible, bioactive, and osseointegration properties, hydroxyapatite is widely used in orthopedics and dentistry as a bone substitute and implant coating (Mahdi et al., 2024). In addition, HAp has a high specific adsorption capacity, so it is useful for developing treatment methods in the field of orthopedics (Gade et al., 2025).

Various methods have been developed for the synthesis of hydroxyapatite, such as wet precipitation, sol-gel, and hydrothermal methods. However, these methods often require long process times and strict parameter control to obtain results with suitable morphology and crystal size (Laonapakul, 2015). Therefore, an alternative approach utilizes micro - wave technology with

✉ Corresponding author:
E-mail: agusprasetyo@students.undip.ac.id

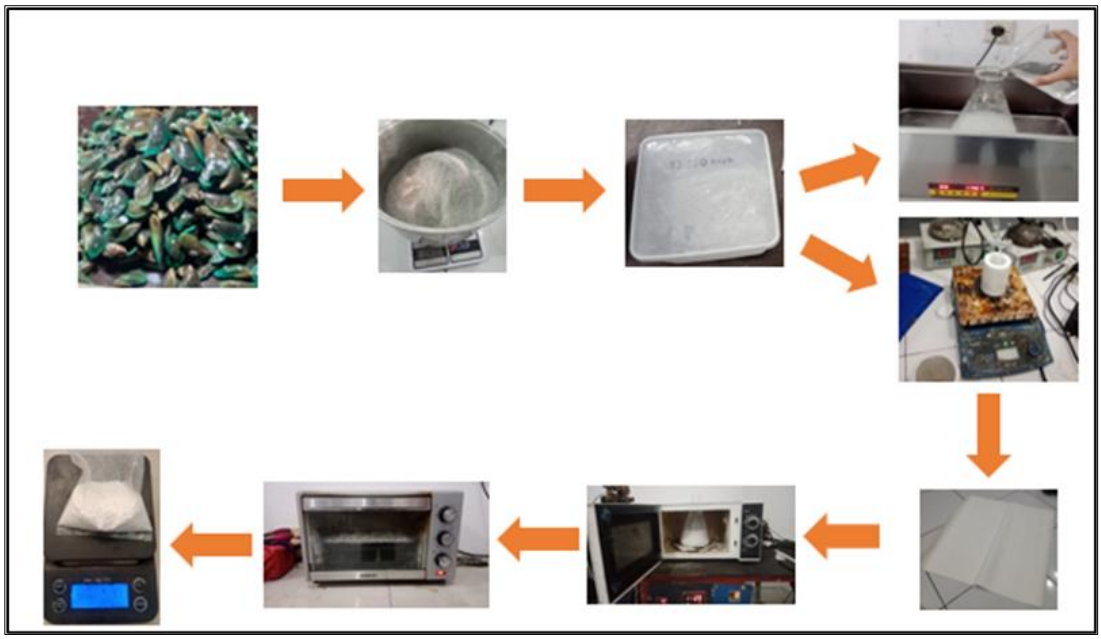


Figure 1. HAp synthesis steps.

ultrasonic solution mixing and magnetic stirrer stirring.

The use of ultrasonics in synthesis processes plays an important role in accelerating chemical reactions through cavitation effects, improving the homogeneity of the mixture (Nikolaev et al., 2018; Poinern et al., 2009). Mixing with a magnetic stirrer maintains the stability of the solution and prevents uneven precipitation during the reaction process (Perwiranegara, 2022). Mixing with magnetic stirrer has been widely used in HAp synthesis, while ultrasonic is still rarely used by researchers.

This study aims to synthesize hydroxyapatite using the micro wave method with ultrasonic mixing and magnetic stirrer and evaluate the characteristics of the synthesized results, such as morphology, crystal size, to see the potential application in biomedicine.

MATERIALS AND METHOD

This research utilized green mussel shells collected from the north coastal area of Semarang City, Central Java, Indonesia. Before use, the shells were cleaned of any remaining meat. Next, the shells were dried in the sun until completely dry. After drying, the shells were crushed using a crusher and sieved to reach a mesh size of 80. The crushed powder was then ready to undergo the calcination process. The calcination process was carried out using a Thermolyne Chamber F6010 Furnace at temperatures of 900°C for five hours.

Figure 1 describes the hydroxyapatite synthesis process using the calcination of green mussel shells, where the calcination results in the form of CaO. The composition of 2.94 grams of CaO was mixed with 20 mL of distilled water. The mixing process was carried out using ultrasonic and magnetic stirrer to compare the hydroxyapatite synthesis results. The mixing was carried out for 20 minutes while the diammonium phosphate mixture that had been dissolved with distilled water was added. After mixing is complete, then synthesize Hydroxyapatite by heating the mixture in the microwave at 450 Watt power for 3 minutes. The synthesized liquid is then soaked using distilled water until its pH reaches a neutral number, namely pH 7. After that, the solution is filtered using filter paper to reduce water content. The last step is the drying process which is carried out using an oven at a temperature of 110°C for one hour, or until completely dry HAp. The specimen code of this research is HA1 for HAp with ultrasonic mixing, and HA2 indicates HAp mixing magnetic stirrer.

Characterization methods used are X-Ray Diffraction (XRD), and Fourier Transform Infra-red (FTIR). XRD is used to characterize the phase and crystal structure of a material, both crystalline and non-crystalline. Crystalline structure analysis in this study used XRD testing with a Malvern Panalytical X'PERT3 Powder X-Ray diffractometer (Malvern Panalytical, Netherlands). The crystalline

phase contained in the sample was identified with the help of HighScoor Plus version 3.0e. Meanwhile, FTIR was used to analyze the chemical compounds contained in HAp. FTIR testing using FTIR 8201PC Shimadzu brand with the analysis area at wave numbers $(4000 - 400\ cm)^{-1}$. FTIR testing was conducted at the UNNES Physics Laboratory.

RESULTS AND DISCUSSION

Figure 2 displays the results of X-Ray Diffraction (XRD) analysis of HA1 and HA2 specimens. Specimen HA1 shows the presence of Hydroxyapatite (HA), calcite and portlandite crystal phases, while HA2 indicates hydroxyapatite and pirtlandite crystal phases. The presence of HAp crystals in all specimens was validated using the Crystallography Open Database (COD) with reference code 96-900-6838. HA1 shows HAp crystals, as seen in the diffraction peaks at angles 2θ = 22.844°, 25.689°, 28.997°, 31.795°, 39.897°, 44.481°, 46.665°, 49.288°, 53.185°, 64.064°. While HA2 diffraction peaks at angles 2θ = 22.848°, 25.830°, 28.931°, 32.155°, 41.990°, 43.803°, 48.603°, 50.484°, 53.084°, 66.370°. It also shows that the crystalline phase is a hexagonal crystal system with lattice constants a = b = 9.4000 Å, and c = 6.9300 Å in HA1. While HA2 shows a hexagonal crystalline phase with lattice constants a = b = 9.4210 Å and c = 6.8930 Å. According to Asra, Yauma et al., (2018), the crystal structure is formed by a unit cell, which is a collection of atoms arranged periodically in a lattice (Yauma et al., 2018). The phases indicated at the 2θ angle can provide information about the direction of the hkl plane (Ali et al., 2022).

The analysis confirmed in Figure 3 which indicates the weight percentage (wt.%) of HAp of 99.8%, portlandite crystals of 0.1%, and calcite crystals of 0.1% for HA1. While HA2 contained HAp crystals of 97.7%%, and portlandite contained 2.3%, and no calcite crystals were detected.

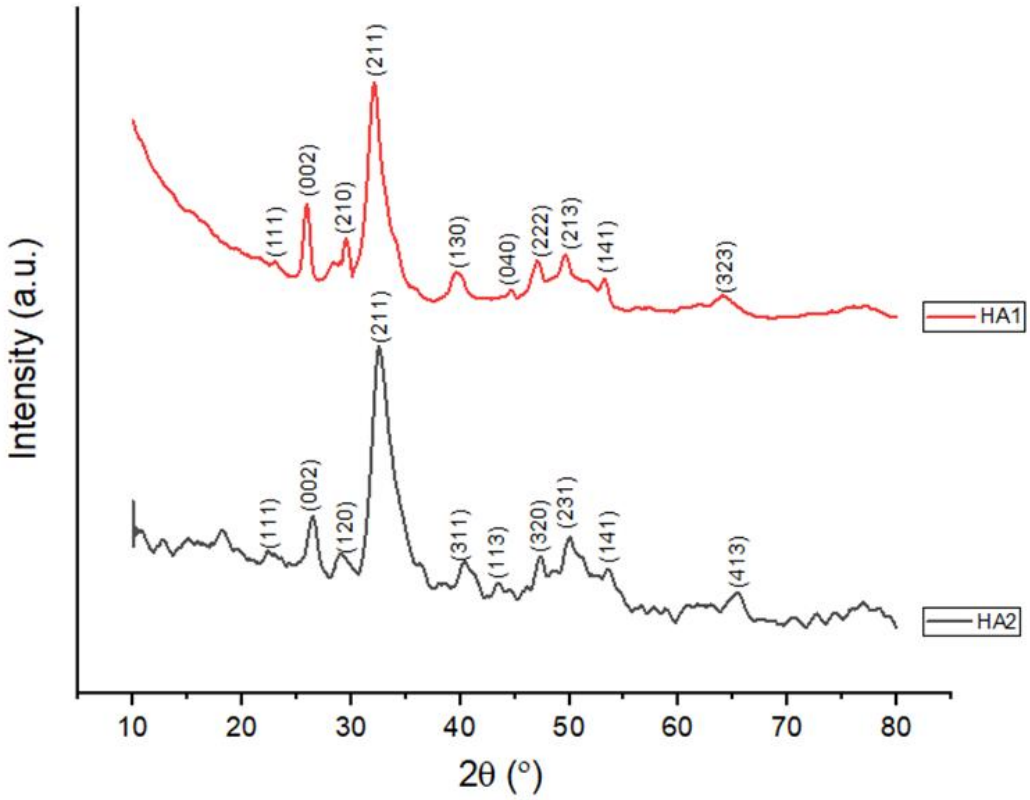
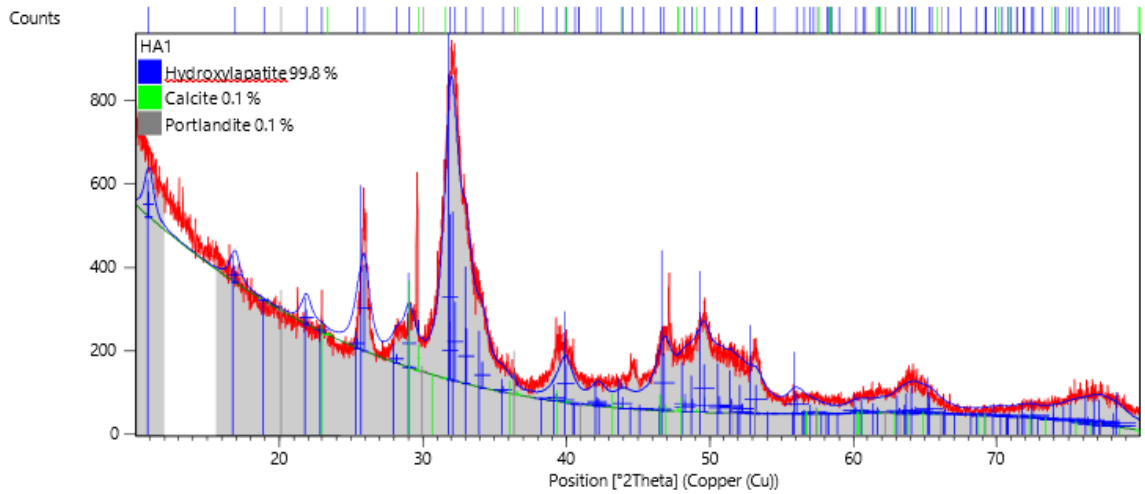
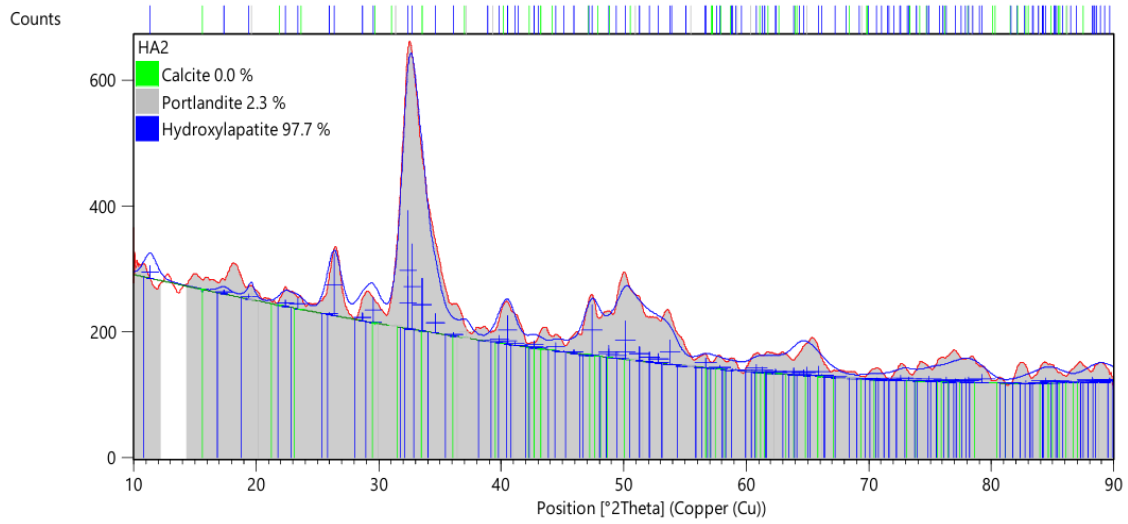


Figure 2. XRD pattern of HA specimen.



(a)



(b)

Figure 3. Results of rietveld analysis on (a) HA1, and (b) HA2.

This research shows that HA1 is better than HA2. This is due to the less than optimal synthesis method and processing conditions (Charlena et al., 2022). Both specimens can be said to be in accordance with medical use standards, where the medical standard set is 95% for HAp content (Charlena et al., 2022; Fitriyana et al., 2024; Rimus et al., 2024).

Fourier Transform Infra-red (FTIR) provides details on the carbonate substitution in the phase. FTIR spectrophotometer is a tool that can be used for the identification of functional groups. The FTIR spectra of HA1 and HA2 specimens are shown in Figure 4. In general, HA is characterized by various vibrational modes of hydroxyl (OH-) and phosphate (PO_4^{3-}).

All specimens showed vibration modes corresponding to phosphate, hydroxyl and carbonate groups. Groups PO_4^{3-} have four vibration modes, namely symmetrical stretching (υ1) at a wave number of about 956 cm^{-1}, symmetric bending (υ2) at 430–460 cm^{-1}, asymmetric stretching (υ3) at 1040–1090 cm^{-1}, and asymmetric bending (υ4) at 575–610 cm^{-1} (Yauma et al., 2018). he results of FTIR analysis show that phosphate vibrates at υ2 470.68 cm^{-1}, 464.09 cm^{-1}. υ4 transmission band at wave numbers 471.14 cm^{-1}, 557,97 cm^{-1}. The υ3 and υ4 bands of the phosphate are asymmetric bands indicating that the specimen is not completely amorphous. The hydroxyl bond strain was observed in the range of 3645.92 cm^{-1}, 3498,52 cm^{-1} in all samples. Carbonate substitution in apatite was identified by the characteristic peaks of carbonate ion around 866.76 cm^{-1} (asymmetric strain) and 1416.82 cm^{-1} (symmetric bending). Carbonate groups are thought to replace phosphate groups and cause contraction in lattice parameters (Fitriyana et al., 2024; Gnanasekaran et al., 2024).

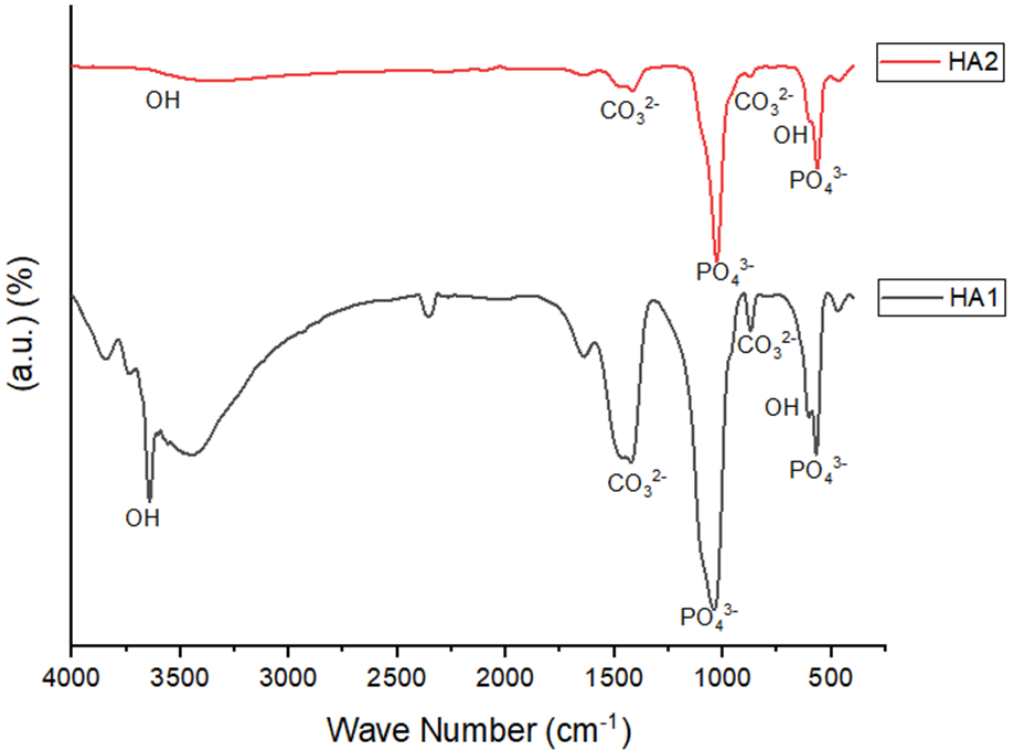


Figure 4. FTIR spectra of HA1 and HA2.

CONCLUSION

This research shows that HA1 has better purity than HA2 which is known through XRD test. HA1 has 99.8% purity of HAp crystal, while HA2 has 97.7% purity of HAp crystal. The purity of both specimens meets the standard of medical use, where in its application the purity of HAp used is at least 95%. The crystalline phase obtained is hexagonal, HA1 with lattice constants a = b = 9.4000 Å, and c = 6.9300 Å. While HA2 shows a hexagonal crystalline phase with lattice constants a = b = 9.4210 Å and c = 6.8930 Å. FTIR test results detected the presence of PO_4^{3-} (phosphate) groups, where phosphate is an important component in the formation of HAp. Hydroxide was also detected in both specimens. In addition to these two contents, both specimens also detected the presence of carbonate groups. This cannot be said to be bad, because carbonate can be a natural substitute for phosphate in bone. The results show that the ultrasonic mixing method is more homogeneous than the magnetic stirrer. Ultrasonic mixing can break particle agglomeration more effectively, resulting in a better and more homogeneous mixture. The use of ultrasonic mixing is a better method for hydroxyapatite synthesis applications.

ACKNOWLEDGMENTS

Thanks to the supervisor who has helped make this article and CBIOM3S for helping fund this research.

REFERENCES

Ali, A., Chiang, Y. W., Santos, R. M. 2022. X-ray Diffraction Techniques for Mineral Characterization : A Review for Engineers of the Fundamentals , Applications , and Research Directions. Minerals. 12(2): 1–25.

Charlena, Maddu, A., Hidayat, T. 2022. Synthesis and Characterization of Hydroxyapatite from Green Mussel Shell with Sol-Gel Method. Jurnal Kimia Valensi. 8(2): 269–279.

Fitriyana, D. F., Ismail, R., Bayuseno, A. P. 2024. Characterization of Hydroxyapatite Extracted from Crab Shell Using the Hydrothermal Method with Varying Holding Times. Journal of Renewable Material. 1–19.

Gade, S. D., Lopes, G., Neto, D. O., Queiroz, M. N., Oliveira, A., Steimacher, A. 2025. Bioactive Borate Glass-Hydroxyapatite

Composites : Influence of the Sintering Temperature on Structural Properties and In vitro Bioactivity. Next Materials. 8: 100589.

Gnanasekaran, R., Yuvaraj, D., Muthu, C. M. M., Ashwin, R., Kaarthikeyan, K., Kumar, V. V., Ramalingam, R. J., Al-lohedan, H., Reddy, K. 2024. Extraction and Characterization of Biocompatible Hydroxyapatite (HAp) from Red Big Eye Fish bone : Potential for Biomedical Applications and Reducing Biowastes. Sustainable Chemistry for the Environment. Sustainable Chemistry for the Environment. 7(June): 100142.

Irfa, M. A., Muryanto, S., Prihanto, A., Pusparizkita, Y. M., Ismail, R., Jamari, J., Bayuseno, A. P., Loke, P. 2024. Microwave-assisted hydrothermal synthesis of carbonated apatite with calcium and phosphate resources derived from green mussel shell and bovine bone wastes. Environmental Advances Journal. 17(October): 100582.

Laonapakul, T. 2015. Synthesis of Hydroxyapatite from Biogenic Wastes. KKU Engineering Journal. 42(September): 269–275.

Mahdi, A., Abdul-rasool, A. A., Al-sharify, Z. T., Zaidan, K., Mohammed, D., Hashim, S. 2024. Nano bioceramics : Properties , applications , hydroxyapatite , nanohydroxyapatite and drug delivery. Case Studies in Chemical and Environmental Engineering. 10(December): 100869.

Nikolaev, A. L., Gopin, A. V, Severin, A. V, Rudin, V. N., Mironov, M. A., Dezhkunov, N. V. 2018. Ultrasonic Synthesis of Hydroxyapatite in Non-cavitation and Cavitation Modes. Ultrasonics – Sonochemistry. 44(February): 390–397.

Perwiranegara, S. A., Bayuseno, A. P., Ismail, R. 2022. Pengaruh Daya Microwave Terhadap Karakterisasi Hidroksiapatit Berbahan Cangkang Rajungan. Jurnal Teknik Mesin. 9(4): 559 - 564.

Poinern, G. E., Brundavanam, R. K., Mondinos, N., Jiang, Z. 2009. Synthesis and characterisation of nanohydroxyapatite using an ultrasound assisted method. Ultrasonics Sonochemistry. 16(April): 469–474.

Pu'ad, N. A. S. M., Haq, R. H. A., Noh, H. M., Abdullah, H. Z., Idris, M. I., Lee, T. C. 2020. Synthesis method of hydroxyapatite : A review. Materials Today : Proceedings. 29(Part 1): 233–239.

Rachmantio, C., & Irfai, M. A. 2023. Pengaruh Suhu dan Waktu Kalsinasi Terhadap Kemurnian Hidroksiapatit Berbasis Cangkang Kerang Hijau untuk Aplikasi Pada Bone Tissue Engineering. Jurnal Teknik Mesin. 11(1): 1–6.

Rimus, A., Hartati, W., Herry, A., Fathoni, A., Wendari, P. 2024. Transforming seafood waste : Green mussel shell-derived hydroxyapatite as a catalyst for spirooxindole synthesis. Bioresource Technology Reports. 25 (February): 101796.

Yauma, D. A., Widya, Y. S., Dahlan, K. 2018. Effect of Microwave Irradiation on the Synthesis of Carbonated Hydroxyapatite (CHA) from Chicken Eggshell. IOP Conference Series: Earth and Environmental Science. 187: 012016.