Conference Paper

The Effect of Calcination Temperature to Mechanical Properties of Sheep Hydroxyapatite

J Triyono, S E Prawira, and T Triyono

Departement of Mechanical Engineering, Universitas Sebelas Maret, Surakarta, Indonesia

Abstract

In this study, we conducted manufacturing processes of bone fillers from Sheep Hydroxyapatite (SHA). The femur bone of sheep was cut into a form of a scaffold with dimensions of 5x5x5 mm. The calcination process was performed at four variations of temperature (700 °C, 900 °C, 1100 °C and 1300 °C). Characterization of scaffold material done before and after calcination process is intended to find out the influence and relationship between calcination of temperature on the mechanical properties of SHA material. The results showed that the higher the calcination temperature would increase SHA material hardness. The optimum hardness occurs at 38.23±0.985 VHN (1100 °C). Meanwhile, high calcination temperature will also decrease the compressive strength of SHA materials. The optimum compressive strength was 2.23±0.249 MPa (1100 °C). The morphology of SHA scaffold was analyzed by Scanning Electronic Microscopy (SEM). The observation of SEM shows that the occurrence of porous interconnections in all temperature variations. SEM analysis results showed that the porous interconnect is formed at all temperature variations with a diameter size of ± 100-500μm. Very high calcination temperature will give the impact of SHA wall is getting thinner, and the porous diameter is getting bigger.

Corresponding Author:
J Triyono
jokotri5528@gmail.com

Received: 23 February 2019
Accepted: 6 March 2019
Published: 25 March 2019

Publishing services provided by Knowledge E

Selection and Peer-review under the responsibility of the ICO-HELICS Conference Committee.

OPEN ACCESS

1. Introduction

The use of bone graft as a bone filler material in Indonesia continues to increase mostly due to the high number of accidents. By using a bone graft, the damaged bone can be reconstructed by giving bone filler material. There are several methods used for bone grafting processes, namely autograft, allograft, and xenograft. The method of autograft and allograft, material is taken from humans while the xenograft method is taken from animals [1].

Allograft and xenograft have weaknesses, namely the tendency of an immune reaction, rejection by body tissues, and the possibility of disease transmission [2]. Autograft and allograft techniques require additional surgical procedures that can cause trauma. In addition, this technique can also extend surgical time and treatment costs.

The solution to overcome the risks arising from several methods of bone reconstruction is by creating a Hydroxyapatite (HAp). Hydroxyapatite is a natural mineral from calcium phosphate apatite compound in the form of crystalline salt with the formula Ca10(PO4)6(OH)2. HAp is the most stable calcium apatite compound compared to other calcium phosphates, namely octacalsium phosphate (OKF), dicalcium phosphate dihydrate (DKFD), and tricalcium phosphate (TKF) [4].

This paper explains the potential use of sheep bones as an ingredient of HAp. In this study, temperature variations were used in the calcination process. The mechanical properties studied were compressive strength and hardness. Observation of microstructure was observed with SEM.

2. Methods

2.1. Preparation of SHA scaffold specimens

The first step in this study was the preparation of the femur of sheep bone. Sheep bone was obtained from Slaughterhouse for Sheep in Tahu Street, Gelung Village, Paron District, Ngawi Regency, East Java. After being cleaned from the sticky meat, it was boiled for 3 times to remove the smell of oil and fish. The next step was to dry the bone by drying it in the sun. This process was carried out for approximately 3 days. After the bones were completely dry, the bones were then cut with a bone saw and then cut to a size of 5x5x5 mm.

2.2. Calcination process

The calcination process was carried out with four temperature variations, namely 700 °C, 900 °C, 1100 °C and 1300 °C. The furnace used is Kejia Furnace KJ-1700X. The calcination process starts from room temperature of 27 °C, with a temperature increase of 10 °C/minute, holding time for 2 hours.

3. Results and Discussion

3.1. The effect of calcination temperature on SHA scaffold color changes

Physically, there is a difference in color at each temperature stage that has been done as shown in Table 1.

From Table 1, it appears that there are color differences between SHA before and after calcination. Before calcination, the bone is still brownish white. This shows that bones still contain organic matter. After calcination to a temperature of 700 °C, significant color changes occur. The color of the bone turns white. Increasing the temperature to 900 °C and 1300 °C increases the color more white.

The occurrence of the color change in the calcination process can be explained as follows. In this process, there is a material decomposition process. Organic substances such as fat, protein, and collagen will burn. An-organic substances are still left in the material. This is in accordance with the research of Ooi et al. (2006) [5] who reported that bone calcined at a temperature of ≥700 °C, the color of the sample turned white, indicating that the organic material in the bone had disappeared.

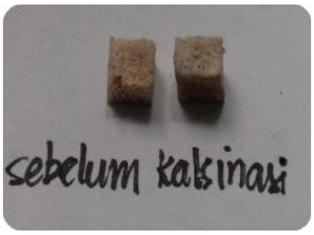
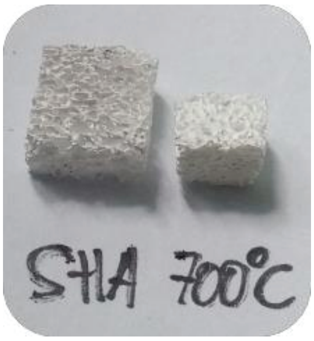
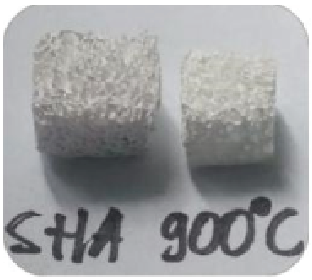
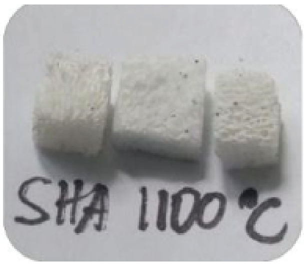
In the calcination process, 1100 °C and the 1300 °C bone color look bright white. At this temperature, the material begins to change to another phase. This analysis is in line with the research conducted by Niakan et al. (2015) [6] who reported that in the calcination process to a temperature of 1000 °C the material would decompose to another phase of calcium phosphate and tri-calcium phosphate (TCP). Ooi et al. (2006) [5] stated that based on the results of the FTIR test, the calcination process above 1100 °C caused the release of OH ions.

From the analysis of the results of the SHA sample calcination and the explanation of Ooi, et al. in 2006 [5], and in 2015 [6], it can be concluded that the calcination temperature plays a vital role in controlling the phase stability of HA. The color changes that occur in the variation of the SHA sample are 700 °C and 900 °C from brownish yellow to white after the calcination process is caused by the decomposition of organic substances (protein, fat, and collagen), while the color changes that occur in the SHA sample are 1100 °C and 1300 °C after calcination becomes rather bright white, it is because SHA decomposes to the tri-calcium phosphate (TCP) phase.

3.2. Hardness test analysis (Vickers hardness tester)

From the tests that have been carried out, it can be seen that the calcination temperature influences the mechanical properties of SHA scaffold material. The test used Vickers

Table 1: Changes in SHA scaffold color before and after calcination.

Temperature	SHA Scaffold	Color
Before calcination		Brownish white
700 °C		White
900 °C		White
1100 °C		Bright white
1300 °C		Bright white

Microhardness test equipment based on ASTM E384-11e1 with a 25 gf and indentor suppression time for 10 seconds. The data that has been obtained is then presented in a graphical form which can be seen in Figure 1.

From the graph in Figure 1, it is generally seen that the value of SHA scaffold hardness has increased, ranging from 700 °C to 1300 °C calcination temperature variations. The test results of SHA scaffold samples at 700 °C have the lowest hardness of 16.73±0.612 VHN, while in the 900 °C calcination temperature variation, the SHA scaffold sample hardness value has increased to 18.98±0.172 VHN. The hardness of the SHA sample continues to increase until it reaches the optimum hardness number of 38.23±0.985 VHN (1100 °C). The phenomenon of the increase in the hardness value of SHA scaffold samples ranging from calcination temperature variations of 700 °C to 1100 °C is caused by the decarbonation process and dehydroxylation of SHA material [7]. The loss of organic matter and the change in SHA scaffold of sheep bone into hydroxyapatite crystalline phase after calcination process is also the cause of the hardness of the control sample lower than the SHA scaffold after calcination.

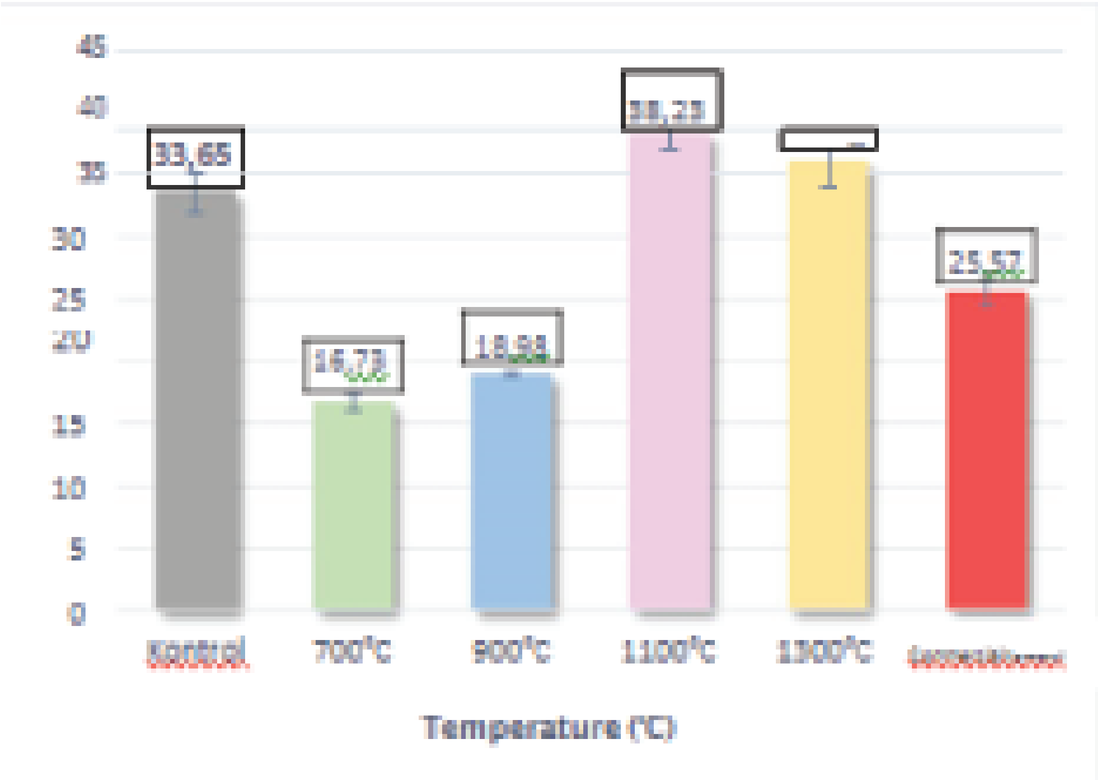


Figure 1: Result of SHA hardness test.

From the graph in Figure 1 it is shown that the hardness of the SHA control scaffold sample is only 33.65 VHN. This value is lower than the hardness value of the SHA scaffold sample after calcined on a temperature variation of 1100 °C which is 38.23±0.985

VHN, with a difference in the hardness value of 4.58 VHN. Meanwhile, the value of SHA scaffold hardness in the variation of calcination temperature 1100 °C is higher than the value of commercial HAp hardness. This is because the commercial HAp is produced by calcining the bones of cattle at 900 °C, while in this study the hardness value of sheep bone HAp at 900 °C is 18.98±0.172 VHN. The difference was 6.59 VHN lower than the commercial HAp hardness value, which was 25.57±1.069 VHN.

The hardness of SHA scaffold samples on temperature variations of 1300 °C tends to decrease the hardness value, which is 36.06±1.716 VHN. The decrease in the value of this hardness can occur because the temperature of 1300 °C is too high if it is used for the calcination process of Sheep HAp scaffold which aims to produce pure HAp. As a result, several grains of HAp compounds become damaged and begin to form another phase, namely tricalcium phosphate (TCP). Damage to HAp content at calcination temperatures above 1100 °C has also been reported in previous studies, namely the synthesis of HAp made from mandibular bone strong squid [8]. In this study, HAp scaffolds were made in powder form using the ball mill method and then calcined at variations of temperatures of 900 °C, 1000 °C, and 1100 °C. From FTIR testing and SEM observation, it was explained that the elements of Ca and $PO4^{-3}$ at a calcination temperature of 1100 °C are semi-crystalline or amorphous in the form of agglomerates or clumps so that the temperature of 1100 °C is not suitable for calcination which aims to produce pure HAp.

3.3. Compressive strength analysis

Calcination temperature influences the compressive strength of SHA scaffold material. This is evidenced by the compressive strength testing that has been carried out using Universal Testing Machine (JTM Technology Machine, 0.5 T Capacity) based on ASTM F451-95 with load loading of 100 kg and with a suppression speed of 5 mm/minute. Data from the test results of the compressive strength of several SHA scaffold samples from several calcination temperature variations are shown in Figure 2 below.

In the graph shown that SHA sheep bone scaffold for temperature variations 700 °C has the lowest compressive strength value of 0.87±0.053 MPa, at a calcination temperature of 900 °C the strength value increases to 1.29±0.081 MPa. The optimum compressive strength value on the SHA sample press test was obtained at 1100 °C calcination temperature variation of 2.23±0.249 MPa. The significant increase of SHA compressive strength value from 700-1100 °C calcination temperature occurs because the SHA scaffold of sheep bone has begun to form pure HAp. In addition to the formation of pure hydroxyapatite, the increase in compressive strength is also due to an increase in

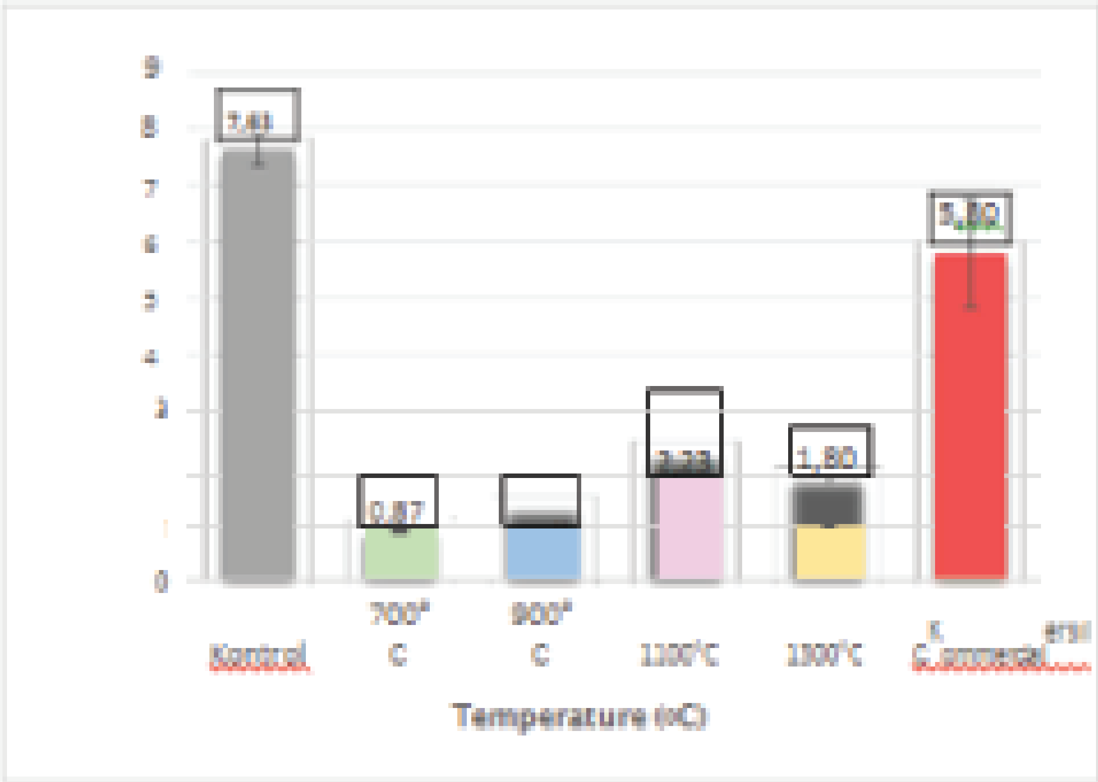


Figure 2: Result of SHA compressive strength.

crystallinity when the sheep bone is calcined at a temperature of 700 to 1100 °C. The phenomenon of increasing crystallinity of HAp has also been found in the study of Ooi (2007) [5] who reported an increase in hydroxyapatite crystallinity in BHA samples when the calcination temperature reached 600 to 1000 °C.

In Figure 2, it is also shown that the compressive strength of SHA of sheep bone has decreased after calcined above 1100 °C. The compressive strength of SHA sheep at a temperature of 1300 °C is 1.80±0.058 MPa. The difference in the decrease in compressive strength is 0.43 MPa. This happens because the SHA scaffold is increasingly porous. The higher the temperature used for SHA calcination, the greater the porous interconnection that occurs in sheep bone scaffold samples. This amount of porous causes the compressive strength of a sheep bone scaffold sample to decrease at a temperature of 1300 °C. According to Solechan's research (2014) [8], the high calcination temperature makes the scaffold more fragile and porous, besides that the strength of the bond between particles becomes weaker. Weak particle bonds result in a very brittle scaffold and low compressive strength.

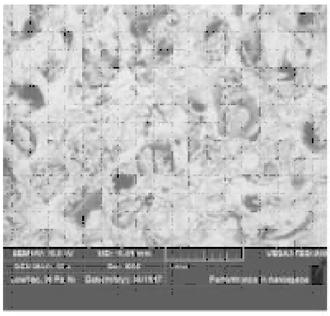
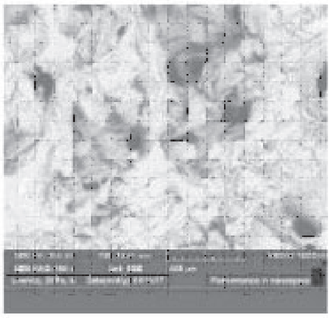
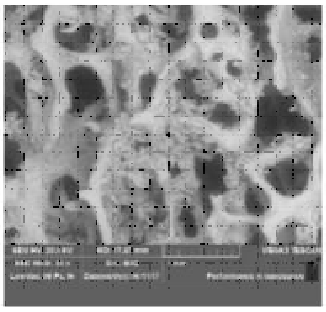
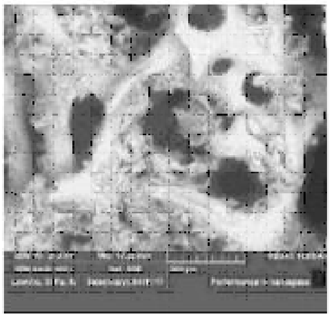
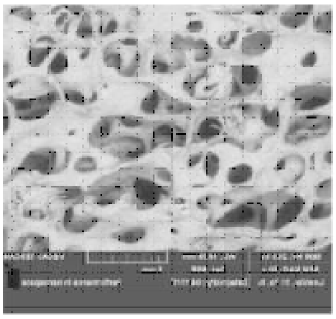
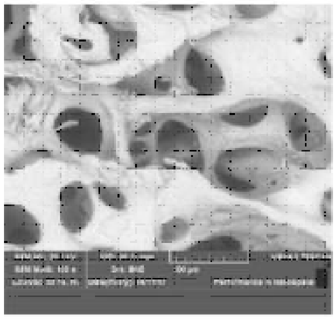
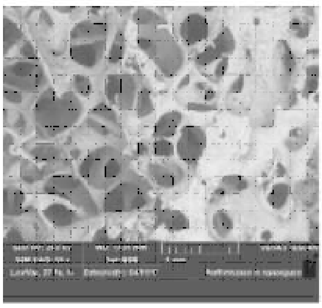
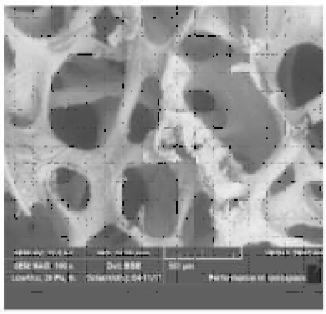
Based on the data of this compressive strength test, it can be concluded that in general, the compressive strength of SHA scaffold made from sheep bone material is strongly influenced by the calcination temperature used. High calcination temperature will reduce the compressive strength properties of SHA scaffold of sheep bones. This was evidenced by compressive strength testing and it was seen that the SHA scaffold compressive strength before calcined (control SHA) was 7.63±0.062 MPa which decreased the compressive strength after calcined at several high-temperature variations (700 °C, 900 °C, 1100 °C, 1300 °C). The decrease in compressive strength is due to the reduction of organic binder and is increasingly porous (Tontowi et al., 2012) [9]. This conclusion is also strengthened by Solechan's (2014) [8] study which states that high calcination temperatures cause the scaffold to become more fragile and porous, in addition to the weaker bond strength between particles. Weak particle bonds result in the very brittle scaffold and low compressive strength.

3.4. Scanning electron microscopy observations (SEM)

Table 2 shows the results of SEM from the SHA scaffold sample for variations in calcination temperature 700 °C, 900 °C, 1100 °C and 1300 °C. From Table 2, the scaffold at a calcination temperature of 700 °C is porous, but the diameter of the interconnection is not so apparent that observations of porous interconnect sizes for 700 °C samples cannot be done in both photo magnifications. Meanwhile, the wall of HAp on SHA scaffold temperature of 700 °C has begun to appear and can be observed. Different from the temperature of 700 °C, porous interconnect diameter at 900 °C temperature variation can be observed clearly with porous diameter ±100-200 μm at 100x magnification. The HAp wall is also increasingly clearly visible at 900 °C temperature variations. This calcination process removes organic material and leaves inorganic material, namely hydroxyapatite. At a temperature of 1100 °C, the porous interconnection is perfect and observed at 100x magnification with a porous diameter of ±100-400 μm.

Meanwhile, the SHA at a temperature variation of 1300 °C, porous interconnects tend to enlarge and begin to experience damage. In Table 2, there are several porous interconnection faults which indicate that calcination at a temperature of 1300 °C results in porous interconnection of the SHA sheep scaffold. The porous interconnects which suffered damage were ± 200-500 m in diameter. Meanwhile, the HAp wall at a 1300 °C variation also appears to be thinner when compared to the HAp wall at 1100 °C. The increasing porous size of the SHA and the thinner HAp wall is what causes the SHA scaffold to become more brittle, thereby reducing the compressive strength of the SHA scaffold.

Table 2: SEM results from the SHA scaffold.

(°C)	Magnification 50x	Magnification 100x
700		
900		
1100		
1300		

4. Conclusions

The higher calcination temperature will increase the hardness value of SHA scaffold material. From the results of the Vickers Microhardness test on SHA scaffold material, the highest hardness value was achieved at 1100 °C, temperature variation of 38.23 ±0.985 VHN. SHA scaffold material decreased the compressive strength after calcined at temperatures of 700°C, 900°C, 1100°C and 1300°C. the highest compressive strength of SHA material was reached at a calcination temperature of 1100oC with compressive strength value of 2.23±0.249 MPa. SEM observations of SHA scaffold material at a calcination temperature of 700 °C, 900 °C, 1100 °C and 1300 °C showed porous interconnections with a diameter of ±100-500 µm. The high calcination temperature caused the hydroxyapatite (HAp) wall in the SHA scaffold material to become thinner, and the porous interconnection diameter became more extensive to reduce the mechanical properties of the SHA scaffold material.

References

[1] Dahlan K, Dewi S U, Nurlaila S and Soejoko D 2012 *Synthesis and Characterization of Calcium Phosphate/Chitosan Composites* p 12

[2] Bacáková L and Filová E 2009 Osteogenic cells on bio-inspired materials for bone tissue engineering osteogenic cells on bio-inspired materials for bone tissue engineering *Physiol. Res.* **59** 309-88

[3] Jebahi S *et al*. 2012 Biologic response to carbonated hydroxyapatite associated with the orthopedic device: an experimental study in a rabbit model *Korean J. Pathol* **46(**1) 48-54

[4] Gnanasundaram S, Pal S, Rose C and Sastry T 2001 A novel bio-inorganic bone implant containing deglued bone *Chitosan and Gelatin* **24**

[5] Ooi C Y, Hamdi M and Ramesh S 2007 Properties of hydroxyapatite produced by annealing of bovine bone *Ceram. Int.* **33** 1171-7

[6] Niakan A 2014 Synthesis and sintering of hydroxyapatite derived from eggshells as a calcium precursor *Ceram. Int.* February 2016

[7] Livingstone T J 2008 *Optimisation of Plasma Sprayed Hydroxyapatite Coatings Optimisation of Plasma Sprayed Hydroxyapatite Coatings* (Ireland: Dublin City University)

[8] Solechan 2015 *Making Nano Hydroxyapatite Synthesis Materials for Application of Mandibular Bone Scaffolds From Sontong Squid Bone Using Calcination Method*

5(1) pp 1-12

[9] Tontowi A E, Dewo P, Wahyuni E T and Triyono J 2012 Scaffold bovine hydroxyapatite made by polyvynialchohol coating *Teknosains* **1(**2) 71–143