Characterization and fabrication of bio-composite filaments for fused deposition modeling 3D printing

P Phengchan, A Chaijaruwanich*, W Nakkiew and S Pitjamit
Advanced Manufacturing Technology Research Center (AMTech), Department of Industrial Engineering, Faculty of Engineering, Chiang Mai University, Chiang Mai, 50200, Thailand.

Email address: anirut@eng.cmu.ac.th

Abstract. The metal bone screws are generally used in bone fractures. The bio-composite bone screws will be fabricated soon by using hydroxyapatite (HA) to reinforce polylactic acid (PLA) and polycaprolactone (PCL) filament for 3D printing. In this study aimed to characterize morphological and physical properties of bio-composite filaments, HA was synthesized from bovine bone. X-ray diffraction analysis indicated that the obtained HA and HA standard showed a similar chemical composition. The morphological observation of bio-composite filaments by scanning electron microscope (SEM) revealed the distribution of HA in bio-composite filaments. The elemental composition of the filament by energy dispersive spectrometer indicated the consisting of calcium, carbon and oxygen elements. The diameter measurement of all bio-composite filaments indicated the values ranged from 1.66 to 1.86 mm which were non-significantly different value in each composite filament and similar with a filament standard.

Keywords: Poly-lactic acid, Polycaprolactone, Hydroxyapatite, Bio-composites filament, Fused deposition modeling

1. Introduction
Bone fractures are common small animal orthopedic injuries which can be treated by using bone screw. The bone screws development has been performed to increase the treatment abilities especially in canine humerus, femur, and tibia fractures. Nevertheless, the bone screws removal is required due to the complications in short term and long-term implantation. Titanium and stainless steel did not provide the living cells with the biological environment according to the bone screw materials [1]. This research is looking for biomaterials that can eliminate metal bone screws limitations. At present, the use of 3D printing in biomedical technology is rising rapidly. There have been several studies of bone screws applications using 3D printing for orthopedic surgery, according to the rapid process of 3D printing [2]. The most satisfied 3D Printing technique is fused deposition modeling (FDM). Thermoplastic filament will be feed through the heating nozzle. The melted filament will be printed layer by layer until the end. The advantage of FDM technique is production speed, material costs and design flexibility [3]. Various types of biomaterials can be printed using the FDM technique. Previous studies have been focused on a biodegradable polymer and bio-ceramics that activate cell creation includes polylactic acid (PLA) that can be degraded by microorganism and turn into carbon dioxide (CO₂) and vapor (H₂O) and sourced by corns, tapiocas and sugarcanes which has linear thermoplastic fiber and widely used in commercial term [4], also with Polycaprolactone (PCL) is thermoplastic and semi-crystalline polymer that has ester group in its component which synthesized by ring opening polymerization of e-caprolactone with catalysts and heat transfer [5]. Hydroxyapatite (HA) has compound from calcium and Phosphorus combination which can be found in mammals’ bones and synthetic [6]. To increase biological and mechanical properties, it can be fabricated into a structure or combined with other biomaterials. Thus, HA is used to create bio-composite filament for bone screws 3D printing as a reinforcing material for PLA/PCL.
In this study, the filaments were extruded, and their morphological and physical characteristics of the obtained filaments will be investigated before printing. The factors of filaments extrusion consisted of HA ratio, extrusion temperature, and extrusion speed. Then, the bio-composite filament will be tested diameter stability property. Finally, appropriate bio-composite filament will be printed and tested with mechanical and biological properties also with simulated the actual treatment in Finite element analysis in the future study.

2. Materials and Methodology

2.1 Synthesis of Hydroxyapatite (HA)
First, bovine bone was boiled to remove tissue completely. Then, the bones were soaked in oxidizing solution for 72 hrs. Next, bones were calcined by incubation at 950 °C in oven for 12 hrs. After cooling, bones were milled and sieved to a powder [7].

2.2 Fabrication composite filaments
First, PLA granules (4043D NatureWorks, LCC) was incubated at 60 °C in oven (PROTRONICS INTERTRADE CO.,LTD. BPG—9070A oven) for 4 hrs, then PLA and PCL were mixed with 70:30 ratio with 0% and 5% wt HA using alumina balls with shaking for 15 min in the cylindrical Teflon mold to homogeneous material [8]. Filaments were fabricated in different factor values of the extrusion process (Table 1). A total 4 conditions consisted of 0% wt HA, 180 °C, 10 rpm, 0% wt HA, 190 °C, 50 rpm, 5% wt HA, 180 °C, 10 rpm and 0% wt HA, 190 °C, 50 rpm. by Wellzoom extrusion machine.

| Factors                     | Value              |
|-----------------------------|--------------------|
| % wt HA                     | Low 0, High 5      |
| Extrusion temperature (°C)  | 180, 190           |
| Extrusion speed (rpm)       | 10, 50             |

3. Testing

3.1 Scanning electron microscope and energy dispersive spectrometer
The top, middle and bottom sites of each sample were cut into 5 mm lengths. HA powder distribution was observed on 1000x magnification by Scanning Electron Microscope (SEM) (EFI Quanta 200 3D) as shown in Figure 2. Energy Dispersive Spectrometer (EDS) analysis of elements in the composite filaments was determined by JSM-5910LV.

3.2 Diameter measurement
Filament prototypes were sampled and measured the diameter stability by analog micrometer (Mitutoyo 0.25 mm – 0.01 mm). A total 10 points of each filament was measured. The obtained diameter values were analyzed on Minitab version 16 program and T-test analysis was performed.
4. Result

4.1 X-ray Diffraction
X-ray Diffraction: XRD of HA powder referred by the recent research [9] for checking the component phase with X-ray diffraction techniques. The XRD result of the synthesized material corresponded to hydroxyapatite (HA) when comparing with HA standard component phase $\text{Ca}_{10}^{}(\text{PO}_4^{})_6^{}(\text{OH})_2^{}$ as shown in Figure 1.

![Figure 1. XRD pattern of synthesized HA powder.](image)

4.2 Scanning Electron Microscope and Energy Dispersive Spectrometer
In morphological testing, use SEM with condition that specific contains HA powder. In some conditions with low temperature (180 °C) non-melting plastic pellets, as shown in Figure 2. And the condition at 190 °C was a suitable condition for PLA melting. Moreover, HA powder was found to be successfully integrated into a composite matrix. The elemental composition of the composite filament has been given in Figure 3. According to the EDS spectrum, the sample mainly consisted of C, O and Ca components. C, O and Ca are primarily derived from the carbonated HA [10] which at high extrusion temperature and speed found that weight percent and atomic of Calcium is higher.
Figure 2. SEM images composite filament at 5% wt HA, 180 °C, 10 rpm (A-D) and 5% wt HA, 190 °C, 50 rpm (E-H). Each images showed top (A,E), middle (B,F), bottom (C,G) and overall (D,H) of filament.

Figure 3. EDS spectrum (A) and (B) of the composite filament at 5% wt HA, 180 °C, 10 rpm and EDS spectrum (C) and (D) of the composite filament at 5% wt HA, 190 °C, 50 rpm.
4.3 Diameter measurement analysis

The diameter values of each filament measurement are shown in Table 2. The obtained diameter values of all composite filaments ranged from 1.66 to 1.86 mm which is non-significantly different by T-test (Table 3).

Table 2. Measurement of filament diameter (in mm) for various conditions

| Sample conditions       | Replicates |
|-------------------------|------------|
|                         | 1  | 2  | 3  | 4  | 5  | 6  | 7  | 8  | 9  | 10 |
| 0%HA, 180°C, 10 rpm     | 1.73 | 1.66 | 1.68 | 1.71 | 1.68 | 1.77 | 1.77 | 1.76 | 1.81 | 1.76 |
| 5%HA, 180°C, 10 rpm     | 1.86 | 1.81 | 1.7  | 1.76 | 1.86 | 1.74 | 1.77 | 1.75 | 1.73 | 1.71 |
| 0%HA, 190°C, 50 rpm     | 1.78 | 1.81 | 1.83 | 1.81 | 1.75 | 1.76 | 1.75 | 1.73 | 1.74 | 1.76 |
| 5%HA, 190°C, 50 rpm     | 1.74 | 1.78 | 1.81 | 1.71 | 1.64 | 1.79 | 1.82 | 1.74 | 1.79 | 1.7 |

Table 3. Results of 1 sample t-test.

| variable                | Mean(mm) | StDev | SE Mean | 95% CI      | T    | P   |
|-------------------------|----------|-------|---------|-------------|------|-----|
| 0%wtHA, 180°C, 10 rpm   | 1.73     | 0.0490 | 0.0155  | (1.6979, 1.7681) | -1.10 | 0.301 |
| 5%wtHA, 180°C, 10 rpm   | 1.77     | 0.0570 | 0.0180  | (1.7282, 1.8098) | 1.05 | 0.320 |
| 0%wtHA, 190°C, 50 rpm   | 1.77     | 0.0339 | 0.0107  | (1.7477, 1.7963) | 2.05 | 0.071 |
| 5%wtHA, 190°C, 50 rpm   | 1.75     | 0.0567 | 0.0179  | (1.7114, 1.7926) | 0.11 | 0.914 |

5. Conclusion

According to the purpose of the end study, HA will be used to reinforce PLA/PCL bone screws which will be fabricated by 3D printing process. HA should provide the fracture area with a good biological environment to speed up the bone healing process by providing a good surface for proliferation, cell adhesion, and migration. In addition, biological enzymes can degrade HA and PLA/PCL. Thus, the 3D printing HA bone screws reinforcing PLA/PCL eliminates the removal process and provides a good biological environment for bone regeneration. HA synthesized in this study could use for a composition material. The bio-composite filament from 5% wt of HA, extrusion 190°C temperature and 50 rpm extrusion speed condition shows the highest HA distribution after observation by SEM. The bio-composite filaments consisted of calcium, carbon and oxygen elements. The diameter value of bio-composite filament obtained in this study was similar to the filament standard for fused deposition modeling 3D printing. Bio-composite filament for 3D printing will be tested for future work and other properties such as mechanical properties, cytotoxicity, biodegradable and finite element analysis will be observed.

6. Acknowledgment

Authors would like to acknowledge Advanced Manufacturing Technology Research Center (AMTech), Department of Industrial Engineering, Faculty of Engineering, Chiang Mai University for their facility support.

7. References

[1] Navarro, M., Michiardi, A., Castano, O., & Planell, J. A. (2008). Biomaterials in orthopaedics. Journal of the Royal Society Interface, 5(27), 1137-1158.
[2] Rengier, F., Mehndirattu, A., Von Tengg-Kobligk, H., Zechmann, C. M., Unterhinninghofen, R., Kauczor, H. U., & Giesel, F. L. (2010). 3D printing based on imaging data: review of medical applications. International journal of computer assisted radiology and surgery, 5(4), 335-341.

[3] Mohamed, O. A., Masood, S. H., & Bhowmik, J. L. (2016). Mathematical modeling and FDM process parameters optimization using response surface methodology based on Q-optimal design. Applied Mathematical Modelling, 40(23), 10052-10073.

[4] Oksman, K., Skrifvars, M., & Selin, J. F. (2003). Natural fibres as reinforcement in polylactic acid (PLA) composites. Composites science and technology, 63(9), 1317-1324.

[5] Wright, D. D. (2004). Degradable polymer composites. In Encyclopedia of Biomaterials and Biomedical Engineering (pp. 423-432).

[6] Elkayar, A., Elshazly, Y., & Assaad, M. (2009). Properties of hydroxyapatite from bovine teeth. Bone and Tissue Regeneration Insights, 2, 31.

[7] Pitjamit, S., Sriprapha, P., & Nakkiew, W. (2016). Suitable forming condition of hydroxyapatite and bioactive glass composites for a bone fixation plate using Taguchi experimental design. Engineering and Applied Science Research, 43, 466-469.

[8] Russias, J., Saiz, E., Nalla, R. K., Gryn, K., Ritchie, R. O., & Tomsia, A. P. (2006). Fabrication and mechanical properties of PLA/HA composites: a study of in vitro degradation. Materials Science and Engineering: C, 26(8), 1289-1295.

[9] Watcharaprapapong, P., Nakkiew, W., Wattanuchariya, W., & Pitjamit, S. (2018). Effect of forming conditions of poly-lactic acid/hydroxyapatite to tensile strength of canine bone fixation plate using full factorial experimental design. In MATEC Web of Conferences (Vol. 192, p. 01049). EDP Sciences.

[10] Huang, Y., Song, G., Chang, X., Wang, Z., Zhang, X., Han, S., ... & Zhang, X. (2018). Nanostructured Ag+ substituted fluorhydroxyapatite-TiO2 coatings for enhanced bactericidal effects and osteoinductivity of Ti for biomedical applications. International journal of nanomedicine, 13, 2665.