Evaluating hydrothermal synthesis of fluorapatite nanorods: pH and temperature

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ABSTRACT
In this study, the fluorapatite was synthesised by a hydrothermal technique in different pH and temperature using apricot tree gum surfactant. The fluorapatite was synthesised in different shapes such as spherical, Chrysanthemum flower and rod. The effect of two factors (pH and temperature) on the shape and dimension of synthesised fluorapatite was investigated through the full factorial design. An experimental strategy was developed based on the analysis of variance to create mathematical models for the shape and dimension of synthesised fluorapatite. Findings revealed that the pH of hydrothermal solution is more significant factor than temperature in terms of shape and dimension of the synthesised fluorapatite. It was illustrated that similar nanorods structure to the human tooth enamel can be achieved in pH of 10 and temperature of 70 °C. The transmission electron microscopy (TEM), field emission scanning electron microscopy (FESEM) and energy-dispersive X-ray spectroscopy (EDS) spectroscopy were carried out for characterisation of synthesised fluorapatite.

1. Introduction
Hydroxyapatite (HA) with chemical formula of [Ca10 (PO4)6 (OH)2] is widely used for biomedical applications due to its structural and biological similarity to human bones [1,2]. It is known that HA has the ability to bond to the surrounding tissue [3,4]. Moreover, the hydroxyl group in HA can be substituted with fluoride to fabricate the fluorapatite (FHA). Generally, fluorapatite is a bioceramic and can be found in the tooth and bone. It is known that fluorapatite has more acid resistance than HA. The dissolution process of HA inside the body is related to the crystallinity and its chemical composition. Therefore, addition of fluoride to the structure of HA is a suitable way to decrease the dissolution rate of HA. In addition, fluorapatite has more crystallinity compared to the HA as well as more thermal and chemical stability. Fluorapatite is harder and process slightly...
better thermal stability than the HA. It is demonstrated that the most portion of human teeth and bones [5] are formed of fluorapatite and HA. Great stiffness, significant resistance to acid damage and suitable biocompatibility of fluorapatite make it potential candidates for biomedical applications. The application of fluorapatite has been mentioned for dentistry application such as crowns, inlays, dentin simulators, coatings and cements [5,6]. In addition to dentistry applications, fluorapatite is an important biomaterial in orthopaedic applications as bone regeneration [7,8]. It was reported that desirable length and diameter for synthesised FHA nanorods to be used as enamel of tooth are between 100–1000 nm and 33–65 nm, respectively [9]. Up to now, it has been reported that fluorapatite can be synthesised through different techniques including the sol–gel [10–12], wet-chemical processing [4,13], solid-state reaction [14] and hydrothermal process [13,14]. Among these techniques, hydrothermal process is considered as a promising approach for synthesise of FHA crystals owing to the advantages such as high quality, chemical homogeneity and low cost of production in large-scale [15].

In our previous study, we reported on the hydrothermal synthesis of fluoridated HA nanorods using different surfactants such as ATG, ethylenediaminetetraacetic acid disodium salt dihydrate (EDTA) and sodium dodecyl sulphate (SDS) [16]. The feasibility formation of FHA nanorods in different sizes, crystallinities and shapes at low temperature of hydrothermal conditions was investigated. Findings revealed that synthesised FHA nanorods by ATG surfactant were larger in dimension as compared to the EDTA and SDS. In addition, results indicated that the size and morphology of synthesised FHA nanorods can be controlled by pH value and surfactant. In this study, an experimental strategy based on the full factorial design is performed using analysis of variance (ANOVA) to investigate the effect of pH and temperature on the shape and dimension of fluorapatite in the hydrothermal condition. Finally, a precision mathematical model is created based on the effective parameters and their interactions to predict the desirable shape and dimension of fluorapatite in the hydrothermal conditions.

2. Materials and methods

The HA and sodium fluoride were purchased from sigma Aldrich (i-Chem Solution-Malaysia). The nitric acid and ammonium hydroxide for adjusting the pH were purchased from Merck (Putaka Elite). Apricot tree gum (Prunus Armenia) was achieved from the city of Taleghan in Iran.

To synthesis fluorapatite, three suspensions with different pH were prepared and put in the different hydrothermal conditions similar to our previous work [16]. 105 mg of HA powder with 8 mg of sodium fluoride was mixed in 100 ml of distilled water. The HNO₃ was added to the suspensions until the HA and sodium fluoride powders dissolved (pH was around 2.3). Thereafter, the ATG surfactant was added to the solutions with the amount of 200 mg. Solutions were stirred for two hours and then the NH₃ was added dropwise to each solution until the pH of 6, 8 and 10 is achieved. The solutions were placed in autoclave under 50, 70 and 90 °C at 1 atm hydrothermal condition.

The synthesised fluorapatite at different pH values and temperatures was then characterised by transmission electron microscopy (TEM), field emission scanning electron microscopy (FESEM) and energy-dispersive X-ray spectroscopy (EDS). Moreover, the mathematical analysis was performed to investigate the effect of hydrothermal condition on the synthesis of fluorapatite in terms of shape and dimension.
An experimental investigation of pH and temperature effect on the shape and dimension of synthesised fluorapatite was carried out. In this study, the pH and temperature were considered as two independent variables. The low, middle and high levels of these two factors are given in Table 1. The obtained data were investigated through full factorial design with two replications. There are three centre points with the level of zero for each factor.

### 3. Results and discussion

Figures 1–3 show the synthesis of fluorapatite in different pH and temperature values through the hydrothermal process. The statistical analyses were carried out with ImageJ software to calculate the vertical and horizontal length of synthesised spherical,
Chrysanthemum flower-like and rods structure of fluorapatite. It can be seen in FESEM images that the synthesised fluorapatite is in nano scales but in dissimilar shapes and dimension for different pH and temperature. In addition, it is demonstrated that pH values have a great effect on the shape of synthesised fluorapatite. A summary of qualitative and quantitative data of experimental results can be seen in Table 2. It is revealed that by changing the temperature of hydrothermal procedure, the shape and defined dimension component ($\alpha = \text{horizontal per vertical length}$) are affected. Moreover, the quantitative and qualitative statistical analyses of FESEM images (Table 2) illustrate the dependencies of dimension and shape to the temperature and pH of hydrothermal solution.

The nanorods’ structures of fluorapatite with high amount of $\alpha$ were achieved at 90 °C and the pH of 10. Figures 3 and 4 illustrate the FESEM and TEM images of synthesised fluorapatite. TEM image of synthesised fluorapatite is shown in Figures 4. This image indicates the rode suture of fluorapatite in nano scales. Moreover, the EDS spectrum of synthesised fluorapatite (Figures 5) demonstrates the existence of Ca, P and F elements and the Ca/P ratio value of 1.63 which can confirm the formation of fluorapatite structure.

### 3.1. Analysis of variance (ANOVA) for shape

The ANOVA was applied to test for the significance of the regression model, the test for significance on individual model coefficients. The ANOVA table for the shape is

| No. | pH | Temperature | Shape                      | Average ($\alpha$) |
|-----|----|-------------|----------------------------|-------------------|
| 1   | 6  | 50          | Spherical particles        | 1                 |
| 2   | 6  | 70          | Spherical particles        | 1.2               |
| 3   | 6  | 90          | Chrysanthemum flower-like  | 1.5               |
| 4   | 8  | 50          | Chrysanthemum flower-like  | 1.3               |
| 5   | 8  | 70          | Chrysanthemum flower-like  | 1.8               |
| 6   | 8  | 90          | Rods                       | 2.3               |
| 7   | 10 | 50          | Rods                       | 2.1               |
| 8   | 10 | 70          | Rods                       | 5                 |
| 9   | 10 | 90          | Rods                       | 2.9               |

Note: Horizontal length/vertical length = $\alpha$. 

**Figure 3.** FESEM image of synthesised fluorapatite in pH 10, temperature 70 °C.
presented in Table 3. Values less than 0.050 for ‘Prob $> F$’ indicate that the model is significant, which is desirable as it reveals that the terms in the model have a significant effect on the response [17,18]. The Model $F$-value of 16.50 implies that the model is significant. There is only a 0.26% chance that a ‘Model $F$-Value’ this large could occur due to noise. Values of ‘Prob $> F$’ less than 0.0500 that indicate the model terms are significant. In this case A are significant model terms. Values greater than 0.1000 that indicate the model terms are not significant. If there are many insignificant model terms (not counting those required to support hierarchy), model reduction may improve the model [19].

$F$-value of the curvature is 0.14 which implies that the curvature (as measured by difference between the average of the centre points and the average of the factorial points) in the design space is not significant relative to the noise. There is a 72.46% chance that a

![Figure 4. TEM image of synthesised fluorapatite in pH 10, temperature 70 °C.](image)

![Figure 5. The EDS spectrum and elemental analysis of fluorapatite in pH 10, temperature 70 °C hydrothermal condition.](image)
‘Curvature $F$-value’ this large could occur due to noise. The regression model in term of coded factor obtained from data is presented in Equation 1:

$$\text{Shape} = +0.13 + 0.88 A 0.13 B - 0.13 AB$$ (1)

The half normal plots of the shape are shown in Figure 6. A check on the plots revealed that the factors A and B and also the interaction AB are significant and affect the model. Figure 7 illustrates the effect of factor pH and temperature and also their interaction on the shape. This figure indicates that the effect of pH is more significant than the effect of temperature on the shape. The counter plot and three-dimensional (3D) surface graphs are shown in Figure 8. These graphs reveal that by increasing the pH and temperature, the shape of synthesised fluorapatite has been modified from $-1$ (spherical) to 1 (rods). This means that the synthesised conditions (pH and temperature) are the effective factors on the morphology of fluorapatite crystals. These factors have a great effect on the crystal

![Half-Normal Plot](image)

Figure 6. Half normal plot of shape.
growth of the synthesised fluorapatite. Findings demonstrate that the pH is more effective than temperature in this study. It may be due to the release profile of surfactant that can be manipulated by changing the pH of solution. It is interpreted that the release amount of calcium ions can be adjusted and consequently influenced the growth of fluorapatite crystals.

Figure 7. Effects of factors on shape and their interaction.

Figure 8. Counter plot and 3D surface graph for shape.
3.2. Analysis of Variance (ANOVA) for dimension

The ANOVA table for the dimension is presented in Table 4. The Model $F$-value of 84.55 that implies the model is significant. There is only a 0.01% chance that a ‘Model $F$-Value’ this large could occur due to noise. Values of ‘Prob $> F$’ less than 0.0500 that indicate the model terms are significant. In this case, A and B are significant model terms. Values greater than 0.1000 that indicate the model terms are not significant [20]. If there are many insignificant model terms (not counting those required to support hierarchy), model reduction may improve your model. $F$-value of the curvature is 1.19 which implies that the curvature (as measured by difference between the average of the centre points and the average of the factorial points) in the design space is not significant relative to the noise. There is a 31.72% chance that a ‘Curvature $F$-value’ this large could occur due to noise. The regression model in term of coded factor obtained from data is presented in Equation 2:

$$\text{Dimension} = + 1.94 + 0.61 \ A + 0.36 \ B - 0.012 \ AB$$ \hspace{1cm} (2)$$

The half normal plots of the shape are shown in Figure 9. A check on the plots revealed that the factors A and B and also the interaction AB are significant and affect the model. Figure 10 illustrates the effect of factor pH and temperature and also their interaction on the shape. This figure indicates that the effect of pH is more significant than the effect of temperature on the dimension. The counter plot and 3D surface graphs are shown in Figure 11. These graphs reveal that by increasing the pH and temperature, the dimension of synthesised fluorapatite has been modified from $\alpha = 0.9$ to $\alpha = 5$. Increasing the amount of $\alpha$ means the dimension of synthesised fluorapatite changed from symmetric shape (spherical) to the asymmetric shape (rod). It can be seen in Figures 1-3 that the shape of synthesised crystal was changed from the spherical to the Chrysanthemum flower-like and then rod shape. This is related to the release profile of the calcium ions by ATG surfactant as well as the degree of temperature. It is illustrated by increasing the temperatures of solutions from 50 to 90 °C, the shape of crystals tends to the rods structure. Moreover, when the pH values increased, the same process occurred.
Figure 9. Half normal plot of dimension.

Figure 10. Effects of factors on dimension and their interaction.
4. Conclusion

In this study, the effect of pH and temperature on the dimension and shape of fluorapatite through a hydrothermal process was investigated. It was revealed that the presence of ATG as surfactant in different pH and temperature values has remarkable effect on the shape and dimension of synthesised fluorapatite. The similar nanorods’ structure to the human tooth enamel was achieved in pH of 10 and temperature of 70 °C through the hydrothermal process. It was demonstrated that with the increase of pH, the shape of synthesised fluorapatite can be changed from the spherical to Chrysanthemum flower-like and then rod shape. Moreover, the raising of temperature in a constant pH values was found to change the shape and dimension of fluorapatite. The full factorial design mathematical models illustrate that pH is more effective than the temperature in terms of shape and dimension of synthesised fluorapatite.

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Disclosure statement

No potential conflict of interest was reported by the authors.

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References

[1] Rezazadeh Shirdar M, Sudin I, Taheri MM, et al. A novel hydroxyapatite composite reinforced with titanium nanotubes coated on Co–Cr-based alloy. Vacuum. 2015;122:82–89.
[2] Taheri MM, Abdul Kadir MR, Shokuhfar T, et al. Fluoridated hydroxyapatite nanorods as novel fillers for improving mechanical properties of dental composite: synthesis and application. Mater Des. 2015;82:119–125.

[3] Kheimehsari H, Izman S, Shirdar MR. Effects of HA-coating on the surface morphology and corrosion behavior of a Co-Cr-based implant in different conditions. J Mater Eng Perform. 2015;24:2294–2302.

[4] Padial-Molina M, Galindo-Moreno P, Fernández-Barbero JE, et al. Role of wettability and nanoroughness on interactions between osteoblast and modified silicon surfaces. Acta Biomater Acta Materialia Inc. 2011;7:771–778.

[5] Chen Y, Miao X. Thermal and chemical stability of fluorohydroxyapatite ceramics with different fluorine contents. Biomaterials. 2005;26:1205–1210.

[6] Wei J, Wang J, Shan W, et al. Development of fluorapatite cement for dental enamel defects repair. J Mater Sci Mater Med. 2011;22:1607–1614.

[7] Yoon B-H, Kim H-W, Lee S-H, et al. Stability and cellular responses to fluorapatite-collagen composites. Biomaterials. 2005;26:2957–2963.

[8] Dhert WJA, Thomsen P, Klein CPAT, et al. Fluoroapatite-coated implants in experimental arthritis: the response of rabbit trabecular bone. J Mater Sci Mater Med. 1994;5:59–66.

[9] Chen HF, Clarkson BH, Sun K, et al. Self-assembly of synthetic hydroxyapatite nanorods into an enamel prism-like structure. J Colloid Interface Sci. 2005;288:97–103.

[10] Montazeri N, Jahandideh R, Biazar E. Synthesis of fluorapatite-hydroxyapatite nanoparticles and toxicity investigations. Int J Nanomedicine. 2011;6:197–201.

[11] Khattech I, Jemal M. Thermochemistry of phosphate products. Part II: standard enthalpies of formation and mixing of calcium and strontium fluorapatites. Thermochim Acta. 1997;298:23–30.

[12] Zimehl R, Willigeroth SF, Hannig M, et al. Mesophases, polymers, and particles. Prog Colloid Polym Sci Berlin. Heidelberg: Springer Berlin Heidelberg; 2004.

[13] Chen H, Sun K, Tang Z, et al. Synthesis of fluorapatite nanorods and nanowires by direct precipitation from solution. Cryst Growth Des. 2006;6:1504–1508.

[14] Wei M, Evans JH, Bostrom T, et al. Synthesis and characterization of hydroxyapatite, fluoride-substituted hydroxyapatite and fluorapatite. J Mater Sci Mater Med. 2003;14:311–320.

[15] Byrappa K, Yoshimura M. Handbook of hydrothermal technology. Norwich: William Andrew Publishing, LLC; 2013.

[16] Taheri MM, Abdul Kadir MR, Shokuhfar T, et al. Surfactant-assisted hydrothermal synthesis of fluoridated hydroxyapatite nanorods. Ceram Int. 2015;41:9867–9872.

[17] Shirdar MR, Izman S, Taheri MM, et al. Effect of electrophoretic deposition parameters on the corrosion behavior of hydroxyapatite-coated cobalt-chromium using response surface methodology. Arab J Sci Eng. 2016;40(2):591–598. Available from: http://link.springer.com/article/10.1007/s13369-015-1700-3

[18] Shirdar MR, Izman S, Taheri MM, et al. Effect of post-treatment techniques on corrosion and wettability of hydroxyapatite-coated Co–Cr–Mo alloy. Arab J Sci Eng. 2015;40:1197–1203.

[19] Rezazadeh Shirdar M, Taheri MM, Moradifard H, et al. Hydroxyapatite–titania nanotube composite as a coating layer on Co–Cr-based implants: mechanical and electrochemical optimization. Ceram Int. 2016;42:6942–6954.

[20] Noordin MY, Venkatesh VC, Sharif S, et al. Application of response surface methodology in describing the performance of coated carbide tools when turning AISI 1045 steel. J Mater Process Technol. 2004;145:46–58.