



## *In situ* synthesis of hydroxyapatite-grafted titanium nanotube composite

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### ABSTRACT

The present study is an investigation to demonstrate the effectiveness of *in situ* approach in the synthesis of hydroxyapatite-grafted titanium nanotube composite (HA-TNT). This method involves combining the process of HA sol–gel and rapid breakdown anodisation of titanium in a novel solution consisting of NaCl and N<sub>3</sub>PO<sub>4</sub>. This new synthesis approach produced a uniform dispersion of Anatase and Rutile phases of TiO<sub>2</sub> nanotubes with minimal agglomeration in the matrix of crystalline HA. The characterisation of homogenised HA-TNT composite was investigated via field emission scanning electron microscopy (FESEM), energy dispersive spectroscopy (EDS), transmission electron microscope (TEM) and X-ray diffraction (XRD). FESEM and TEM images indicated the nanostructure of composite with TiO<sub>2</sub> nanotube diameter of approximately 10 nm. XRD and EDS analyses confirmed the formation of HA crystalline with the Ca/P ratio of 1.58 and formation of Anatase and Rutile phase of TiO<sub>2</sub> nanotubes.

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## 1. Introduction

Nowadays, considerable attentions have been drawn to the application of hydroxyapatite (HA: Ca<sub>10</sub>(PO<sub>4</sub>)<sub>6</sub>(OH)<sub>2</sub>) in biomedical fields. This is due to the similarity of natural bone to HA in terms of structural and chemical compositions. [1–3] Due to its bioactivity and biocompatibility, HA has been widely used as an alternative material for the damaged teeth or bone over the past three decades. [4–7] HA can be synthesised through various techniques such as hydrothermal, sol–gel, precipitation, electrodeposition and biomimetic deposition. [8–10] Among these techniques, sol–gel is highly preferred due to its homogeneity, low synthesis temperature and possibility of forming nanosized particles. [11] As it has been recorded, the mixtures of nano or submicron particles are efficient in improving osseointegration *in vitro* and *vivo* environments. Usually, sol–gel technique is

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utilised to synthesise the nano-to-micro particles of HA in crystal structures. [12,13] However, HA ceramic suffers relatively poor mechanical properties which impede its usage in long-term load-carrying applications. [14,15] Some attempts have been made to enhance the mechanical properties of HA by implementing fillers such as bio-inert ceramics like alumina, [16] zirconia [17] and titania [18] as well as other metals, polymer fibres and bioglass [19] in the HA matrix.

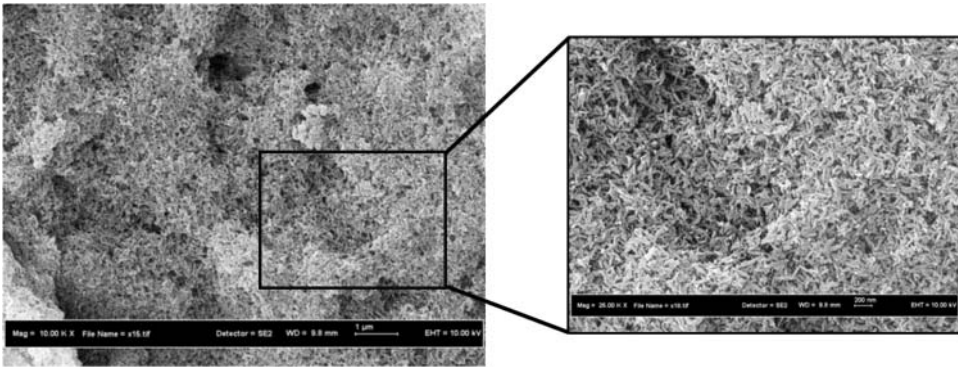
Among the diverse HA-based composites, HA/TiO<sub>2</sub> composites have drawn impressive consideration recently. This is due to the assumption that titania has a great capability to improve osteoblast bond, and it can incite cell development. [20] It has been also demonstrated that cohesive and adhesive strength of HA implant significantly increased by adding titania as the reinforcement. [21] In addition, Xiao et al. have reported that the incorporation of TiO<sub>2</sub> in the matrix of HA improved the corrosion resistance of pure HA. [22] Moreover, several studies have demonstrated that Titanium nanotube is bio-inert and biocompatible in nanoscale; therefore, it could be useful for bioapplications. [23] Furthermore, the presence of the nanotube structure induced a significant acceleration in the growth rate of osteoblast cells. [24] Thus, it can be concluded that the proposed composite coating layer could be a good alternative for bio implant coating applications.

So far, various techniques such as sol–gel, hydrothermal and microwave hydrothermal have been employed to synthesise HA/TiO<sub>2</sub> nanocomposites. As a drawback, all these techniques require high-temperature heat treatment to obtain crystalline powder. In all these techniques, heat treatment is the only step that increased the expense and the other steps are simple and cheap to be prepared.

In contrast, *in situ* precipitation technique can produce HA/TiO<sub>2</sub> crystalline nanocomposites through low-cost and low-temperature (room temperature) procedures. In this study, HA/TiO<sub>2</sub> nanocomposite was synthesised through *in situ* precipitation technique from HA and titania precursors. In this novel technique, HA-TiO<sub>2</sub> nanocomposite was synthesised by employing sol–gel and rapid breakdown adonisation (RBA) simultaneously. Morphology and microstructure of hydroxyapatite-grafted titanium nanotube composite (HA-TNT) were evaluated by scanning electron microscope (SEM) and transmission electron microscope (TEM). X-ray diffraction (XRD) was also utilised to characterise the composition and structure of the samples.

## 2. Materials and methods

Titanium sheet (5 cm × 5 cm), platinum foil, CaCl<sub>2</sub>, Na<sub>3</sub>PO<sub>4</sub> and ammonium hydroxide were purchased from Sigma Aldrich (Malaysia). Two precursors with 0.41 and 0.25 M concentration of CaCl<sub>2</sub> and Na<sub>3</sub>PO<sub>4</sub> were prepared using distilled water. The pH of Na<sub>3</sub>PO<sub>4</sub> solution was adjusted to 4.3 using NaOH. The CaCl<sub>2</sub> precursor was added to the Na<sub>3</sub>PO<sub>4</sub> solution at the rate of 5 mm/min while differential potential of 20 V was applied to cathode (platinum foil) and anode (titanium sheet) with the distance of 2 cm. During one hour of RBA process, the solution was stirred at the rate of 450 rpm. This process was conducted for less than 1 hour until the initial foil was completely transformed into TiO<sub>2</sub> nanotubes powder to produce HA-TNT. Then, several centrifugal washing processes (10,000 rpm) were conducted in order to remove the Na and Cl residues which are soluble in the water solution. Finally, the composite was dried for 24 hours at 60 °C in an oven. In order to evaluate and compare the hardness of HA-TiO<sub>2</sub> nanotubes composite, the pellets

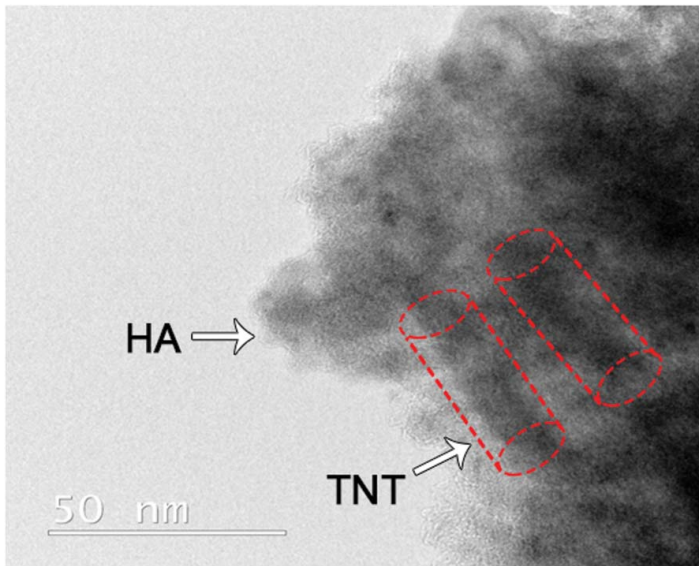


**Figure 1** FESEM image of synthesised HA-TNT composite.

of the composite and pure HA were prepared as control sample. Based on ASTM E92-82, a Vickers micro-hardness machine (SHIMADZU HMV 2T) was used to investigate the hardness of pellets. A diamond indenter with a standard geometry was indented under the load of 0.3 kgf into the pellet for 5 seconds. After the removal of the indenter, the contact area of the resulting indent was calculated (number of reputation is three) based on the following equation:

$$HV = 1.854 \frac{F}{d^2}$$

where  $F$  is load (kg force) and  $d$  is the total length of the diagonal indentation.



**Figure 2.** TEM image of synthesised HA-TNT composite.

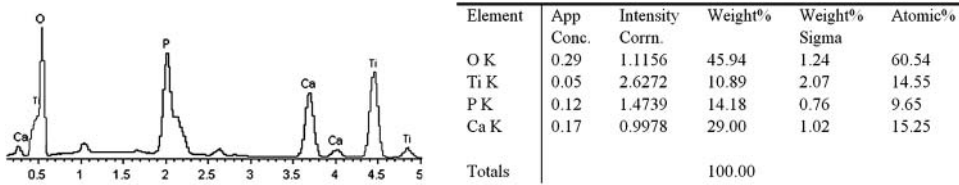


Figure 3. The EDS spectrum and elemental analysis of synthesised HA-TNT composite.

### 3. Results and discussions

Figure 1 shows the morphology of HA-TNT composite. The overall evaluation of the morphological feature indicates a uniform distribution of TiO<sub>2</sub> nanotubes in the HA matrix without agglomeration. As can be seen in Figure 1, TiO<sub>2</sub> of nanotubes structure are well synthesised and consequently well dispersed in the HA matrix. Moreover, TEM characterisation was conducted to confirm the structure of the synthesised TiO<sub>2</sub>. In Figure 2, TEM image manifests that the HA-TNT composite consists of TNT with the diameter of approximately 10 nm. As shown in Figure 3, energy dispersive spectroscopy (EDS) analysis illustrated the presence of O (60.54 at. %), Ti (14.55 at. %), Ca (15.25 at. %) and p (9.65 at. %) with the Ca/P ratio of 1.58.

Figure 2 illustrates that the nanotube structures of TiO<sub>2</sub> have been surrounded by HA particles. This may be due to the *in situ* formation of HA when TNT was synthesised through RBA technique. The homogeneity of TNT into the HA matrix also can be inferred through the one-step procedure of the novel method used in this study.

Figure 4 shows the XRD pattern of HA-TNT composite recorded in the 2θ range of 20–70. The peak indexed at 2θ = 27, 57 and 65 is attributed to Rutile phase of TiO<sub>2</sub> nanotubes and the peaks at 2θ = 27, 57 and 65 corresponded to the Anatas phase of TiO<sub>2</sub> nanotubes. That 2θ = 23, 25, 28, 31, 32, 33, 35.5, 47, 49.5, 51, 52, 53, 54 and 56 confirms the formation of HA crystalline in the synthesised composite. This indicates that the calcium phosphate paste was formed in crystalline phase (HA). Moreover, it has been reported in several studies that HA is stable in simulated body fluid and is also desirable for bioapplications. [24] Furthermore, EDS spectrum in Figure 3 illustrates that the ratio of Ca/P is around 1.58 which confirms the theoretical value of HA in its chemical formula (Ca<sub>10</sub>(PO<sub>4</sub>)<sub>6</sub>(OH)<sub>2</sub>). [25]

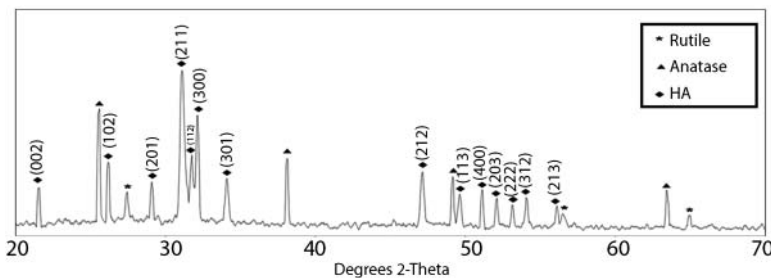
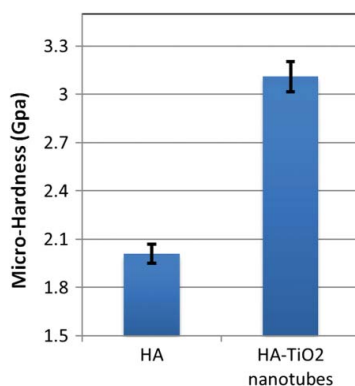


Figure 4. XRD pattern of synthesised HA-TNT composite.



**Figure 5.** Hardness of HA-TiO<sub>2</sub> nanotubes composite and pure HA.

The hardness of HA-TiO<sub>2</sub> nanotubes composite and pure HA was measured using Vickers micro-hardness device (Figure 5). The average hardness of the pellets (number of reputation is three) were 2.01 and 3.11 Gpa, respectively. The results revealed that there was an increase of 54% in the hardness of the composite pellet as compared to the pure HA pellet. Thus, it can be concluded that the incorporation of TiO<sub>2</sub> nanotubes into the matrix of HA possibly increased the micro-hardness due to the nature of TiO<sub>2</sub> in nano-scales. This result is in close agreement with the addition of TiO<sub>2</sub> nanoparticles in the matrix of HA reported by Que et al. and Okta et al. [26,27]

Due to the excellent mechanical and electrochemical properties of TiO<sub>2</sub> nanotubes, HA-TNT composite possibly has higher mechanical properties and corrosion resistance compared to the pure HA. Therefore, this composite can be considered an alternative bio-composite to the current HA composite coating layer on the metallic implant. This is of great consideration particularly in load-bearing applications where high mechanical properties of coated layer are required. In addition, the ion release from the coated implant can be reduced due to the fact that HA-TNT composite may have less porosity compared to the pure HA.

#### 4. Conclusion

In this study, we attempted to develop a novel method of synthesising HA-TNT composite by combining sol-gel and RBA in a novel solution consisting NaCl and N<sub>3</sub>PO<sub>4</sub>. field emission scanning electron microscopy (FESEM) and TEM images from the synthesised composite illustrate the nanostructure of the composite with the TiO<sub>2</sub> nanotube diameter of approximately 10 nm. XRD and EDS analyses confirm the formation of HA crystalline, Anatas and Rutile phases. This novel one-step synthesis method is fast and economical and can be applied for fabrication of HA-TNT composites. Extraordinary mechanical and electrochemical properties of TiO<sub>2</sub> nanotubes make this composite an alternative candidate to the current HA for the coating layer of metallic implant in biomedical applications. Therefore, it can be recommended that this proposed *in situ* approach for synthesising the HA-TNT can be combined with the current commercial plasma spraying or electrospinning techniques which can provide simultaneous spray of HA and TNT on the implant surface and offer concurrent fabrication of HA-TNT scaffold with higher mechanical and electrochemical properties.

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## Disclosure statement


No potential conflict of interest was reported by the authors.

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