

SYNTHESIS AND CHARACTERIZATION OF ZINC OXIDE DOPED HYDROXYAPATITE USED FOR BONE REGENERATION IN ORTHO IMPLANT APPLICATION

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ABSTRACT

Hydroxyapatite (HAp) is one of the bioceramics and has been used for several biomedical applications mainly in dentistry and orthopaedics. Due to its poor mechanical properties and lack of osseointegration gives a substantial demand in the application of HAp for the bone regeneration and as well in bone implant techniques. The present study performs that zinc doped with HAp by using wet chemical method. These synthesized materials were analysed using X-Ray Spectroscopy (XRD), Field Emission Scanning Electron Microscope (FESEM) with Energy Dispersive Spectrometer (EDS) and Fourier Transform Infrared Spectroscopy (FTIR). The morphology, elemental analysis and crystallite particle sizes were calculated. Hence this material can be a promising biomaterial for bone regeneration in ortho implant application.

Keywords: HAp, XRD, FESEM, EDS, FTIR

1. INTRODUCTION

The most commonly used biomaterial in orthopaedic surgeries is hydroxyapatite $[\text{Ca}_{10}(\text{PO}_4)_6(\text{OH})_2]$. The chemical composition of HAp is found in human bones and teeth, being the main mineral constituent of natural hard tissue and also properties such as morphology, stoichiometry, crystallinity and crystal size distribution (CSD) have great influence in the production of materials for biomedical applications, especially in orthopaedic

application owing to its biocompatibility [1-3]. The development of HAp with many additives has been one of the primary aims in the field of biomaterials, in order to higher up the quality materials suitable to use in artificial bone replacement. Hence, it is necessary to optimize how these hydroxyapatite properties are affected by different additives. Obviously, the incorporation of foreign elements can drastically alter the properties of HAp [3]. It has been reported that incorporation of trace ions such as Ag, Zn, Ti and Cu into HAp structures not only provides crystallinity, but also improves their antimicrobial property [4-6]. Among other trace elements Zn is the main reservoir for bone, which accounts for 28% of the total Zn content in the body [7]. In addition, Zinc promotes the bone density and prevents bone loss [8]. The development of zinc oxide (ZnO) nanoparticles has been greatly increased, mainly because of their altered physical, chemical and biological properties [9]. In this work, to improve the mechanical strength and osseointegration, ZnO is doped with the HAp matrix was synthesized by wet chemical synthesis method and was analyzed.

2. MATERIALS AND METHOD

Hydroxyapatite samples were prepared by a wet chemical synthesis technique. The precursor materials were commercially procured $\text{CaCl}_2 \cdot 2\text{H}_2\text{O}$, $(\text{NH}_4)_2\text{HPO}_4$ and NH_4OH (Aldrich). HAp precipitate was prepared by slow addition of 0.2 M ammonium phosphate to a 1.0 M calcium chloride dehydrate powder at 90°C . The pH was monitored and maintained as 10 by adding NH_4OH to the medium. The mixture was thoroughly stirred for 8 hours while maintaining the temperature at 90°C . The precipitate was then dried at room temperature for two days. Finally, the precipitate was washed with water [10]. HAp with zinc oxide was synthesized separately and then mixed together. The mixed solutions were stirred for an hour until it mixed thoroughly. The obtained suspensions were filtered and dried in a hot air oven

at 400°C for 5 hours. The obtained HAp + ZnO were characterized using XRD for phase confirmation and the particle size and crystallinity were studied.

3. CHARACTERIZATION TECHNIQUES

3.1 XRD Characterization

X- Ray diffractometer (XRD) studies were used to determine the structural properties of synthesized HAp + ZnO powders. The investigations were carried out using a Panalytical X'PERT powder diffractometer with a high intensity CuK α radiation ($\lambda=1.540\text{\AA}$). The scans were performed in a 2θ angle ranging from 10-100°. The average crystallite size of HAp + ZnO powder was calculated using the Debye-Scherrer's formula [6],

$$D = 0.9\lambda / \beta \cos\theta$$

Where D is the crystallite size in nanometers, λ is the wavelength of CuK α radiation (1.540 \AA), β is full width of the peak at half of the maximum (FWHM) and θ is the diffraction angle of the corresponding reflection.

The degree of crystallinity is determined from the XRD sample by using the formula,

$$X_c = (I_{300} - V_{112/300}) / (I_{300})$$

Where, X_c is the fraction of crystalline phase, $V_{112/300}$ is the intensity of trough between (112) & (300) of HAp and I_{300} is the intensity of 300 plane.

The lattice strain is calculated using the Williamson Hall equation,

$$\varepsilon = (\beta \cos\theta - k\lambda/D) / 4\sin\theta$$

The Dislocation density is calculated using Debye-Scherrer's formula,

$$\text{Dislocation density} = 1/D^2$$

3.2 FESEM/EDS Analysis

The topography of the samples were analysed using F E I Quanta FEG 200 - High Resolution Scanning Electron Microscope. The short rod shaped micro structure particle size

on the surface of HAp + ZnO powders was observed. The elemental analysis of ZnO doped HAp powders were performed using Bruker - Energy Dispersive Spectrometer.

3.3 FTIR Analysis

The Fourier Transform Infrared (FTIR) Spectroscopy (PERKIN ELMAR; SPECTRUM RXI) was used to identify the functional group in HAp + ZnO powders. The FTIR spectra within the range of 400-4000 cm^{-1} were obtained from the samples that were made into pellets, having 1 mg of sample mixed with 200 mg of spectroscopic grade KBr.

4. RESULTS AND DISCUSSION

4.1 X-Ray Diffraction (XRD) Characterization

Figure (1) shows the XRD patterns of ZnO incorporated HAp sample. The observed diffraction peaks of prepared sample ensured the emergence of calcium rich phosphate apatite phases by matching well with the peaks in the standard JCPDS data file (02-1350). The intensity of the diffraction peak of plane (211) of HAp increased with the ZnO content, mainly because of the preferential orientation of ZnO in the apatite crystal. The peak intensity of the ZnO incorporated HAp peak, increased with increasing ZnO content. Hence, the added zinc content was involved in the formation of HAp. The crystallite size, lattice strain and dislocation density of HAp + ZnO nanopowder were determined by using the Debye Scherer's formula ($D = k\lambda/\beta\cos\theta$), Williamson Hall equation [$\epsilon = (\beta\cos\theta - k\lambda/D) / 4\sin\theta$] and Dislocation density ($1/D^2$) where $k = 0.9$, $\lambda = 1.54\text{\AA}$, $\beta = 0.0093$ and $\theta = 0.5129$ as 15.06 nm, 0.02764 and 0.04060 respectively. These data confirm the emergence of apatite mineral on the nanometer scale.

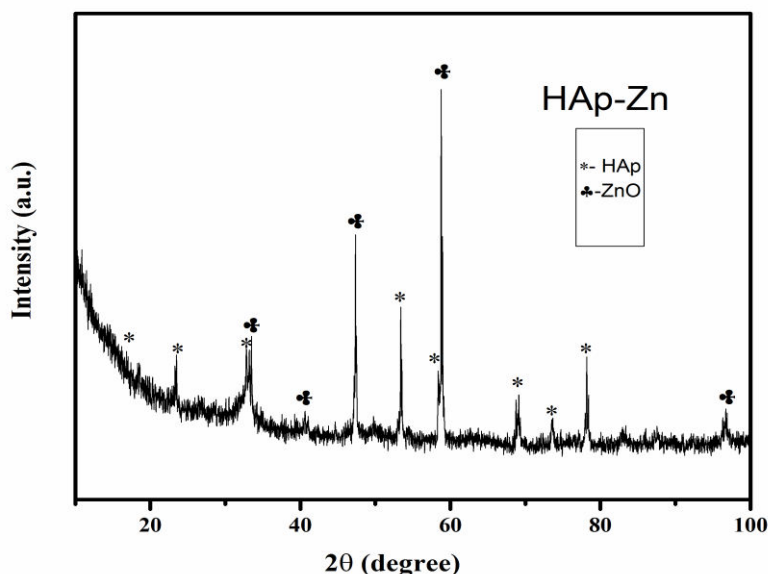


Fig.(1) XRD pattern ZnO doped HAp sample

4.2 Field Emission Scanning Electron Microscopy (FESEM) and Energy Dispersive Spectrometer (EDS) analysis

The morphology and elemental analyses of ZnO doped HAp sample were examined using field emission scanning electron microscopy (FESEM) and energy dispersive spectrometer (EDS) as shown in figure (2). The SEM analysis revealed the emergence of short rod shaped morphology in ZnO doped HAp sample and also confirms that particle sizes ranged from 30 to 50 nm with the incorporation of ZnO. The result of elemental analysis of ZnO doped HAp confirmed the presence of Ca, P, O and Zn as shown in figure (3). The observed elemental concentration confirmed the Ca-P rich apatite crystalline phase. In general, the theoretical value of Ca/P ratio for HAp is 1.67 and the same was considered here in all experimental conditions. However, In reference to the conventional Ca/P ratio, the calculated ratio of the synthesized ZnO doped HAp to be 2.63. Further, the degree of crystallinity for HAp + ZnO is 86.36 due to Zn inclusion in the HAp matrix. This is in consistence with the observed XRD results.

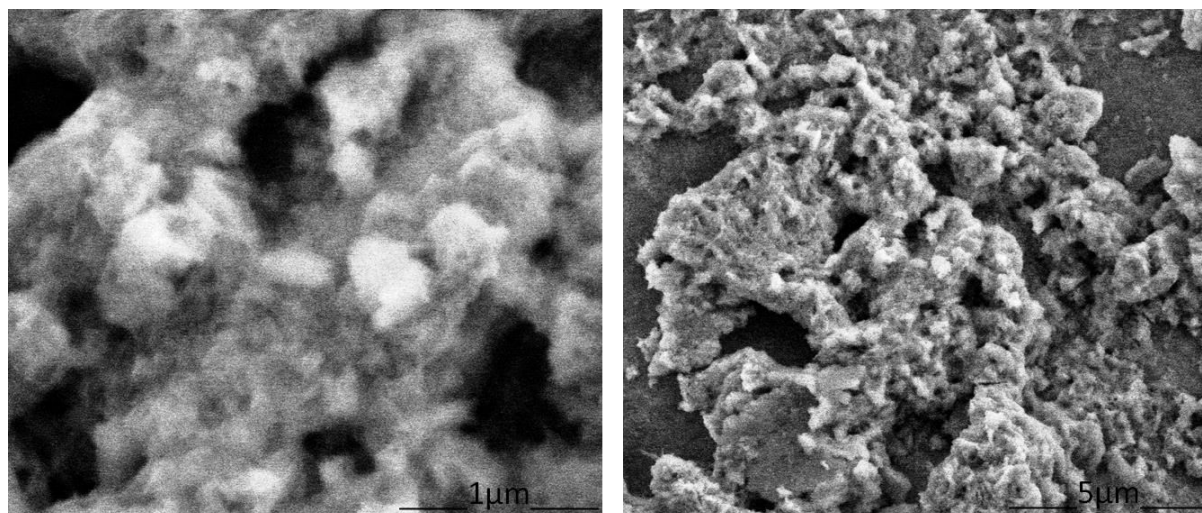


Fig.(2) FESEM images ZnO doped HAp

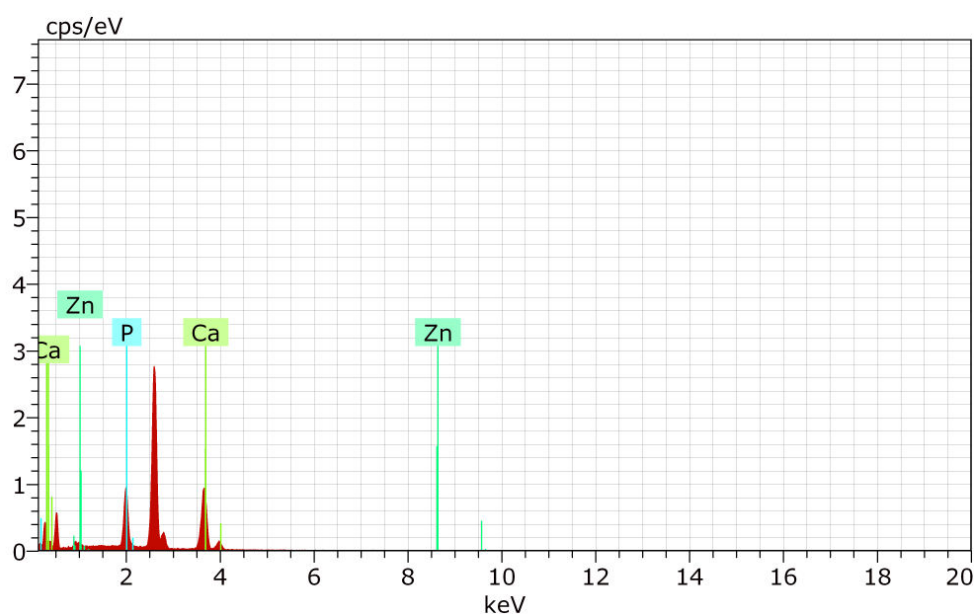


Fig.(3) EDS analysis of ZnO doped HAp

4.3 Fourier Transform Infrared Spectroscopy (FTIR) Analysis

The FTIR spectra of ZnO doped HAp sample is shown in figure (4). The FTIR bands observed at 602 cm^{-1} is assigned to symmetric stretching modes ν_1 and ν_2 phosphate group. The peak which is observed at 561 cm^{-1} was assigned to ν_4 O-P-O bending bands corresponding to HAp. The band at 1042 cm^{-1} was assigned to ν_3 P-O asymmetric stretching mode. The bands observed at 3381 and 3150 cm^{-1} is due to stretching vibration of hydroxyl

group. The band at 1632 cm^{-1} confirms the presence of CO_3^{2-} group and 1402 cm^{-1} decreased with increasing ZnO content.

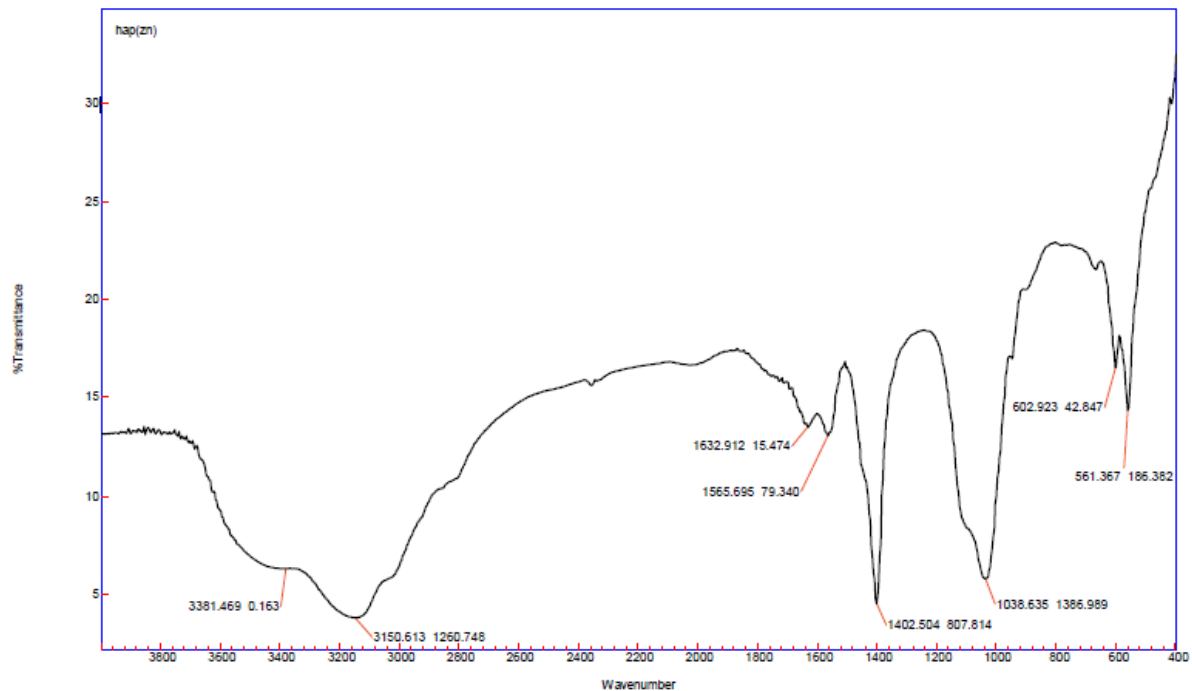


Fig.(4) FTIR spectrum of ZnO doped HAp

5. CONCLUSION

ZnO incorporated HAp sample was prepared using wet chemical synthesis method. The obtained XRD results confirmed the crystallite size, lattice strain and dislocation density of HAp + ZnO as 15.06 nm, 0.02764 and 0.04060 respectively. The incorporation of ZnO was used to increase the average crystallite size of the Ca-P apatite crystal. The morphology of HAp + ZnO is in short rod shaped like structure. The elemental analyses confirmed the emergence of calcium rich apatite crystalline phases. Further, successful integration of ZnO into HAp matrix suggests the possibility of enhancement in mechanical adhesion, osseointegration and antibacterial activity that are primarily considered for bone regeneration in ortho implant applications.

6. REFERENCE

- [1] M. Supova, Substituted hydroxyapatites for biomedical applications: a review, *Ceramics International*, 41 (8), (2015) 9203-9231.
- [2] W. Suchanek and M. Yoshimura, Processing and properties of Hydroxyapatite-based biomaterials for use as hard tissue replacement implants, *Journal of Materials Research*, 13(1), (1998) 94-117.
- [3] D.G. Guo, Y. Wang, K. Han, W. Xu, Characterization, physico-chemical properties and biocompatibility of La-incorporated apatites, *Acta Biomaterialia*, 5 (2009) 3512-3523.
- [4] F. Bir, H. Khireddine, A. Touati, D. Sidane, S. Yala, H. Oudadesse, Electrochemical depositions of fluoro hydroxyapatite doped by Cu²⁺, Zn²⁺, Ag²⁺ on stainless steel substrates, *Applied Surface Science*, 258 (2012), 7021-7030.
- [5] S. Saidin, P. Chevallier, M. R. Abdul Kadir, H. Hermawan, D. Mantovani, Polydopamine as an intermediate layer for silver and hydroxyapatite immobilisation on metallic biomaterials surface, *Materials Science and Engineering C* (2013).
- [6] E. Boanini, M. Gazzano, A. Bigi, Ionic substitutions in calcium phosphates synthesized at low temperature, *Acta Biomaterialia*, 6 (2010), 1882-1894.
- [7] G.S. Kumar, A. Thamizhavel, Y. Yokogawa, S.N. Kalkura, E.K. Girija, Synthesis characterization and in vitro studies of zinc and carbonate co-substituted nano-hydroxyapatite for biomedical applications, *Materials Chemistry and Physics*, 134 (2012), 1127-1135.
- [8] R.Z. Legeros, Calcium phosphate-based materials containing Zinc, Magnesium, Fluoride and carbonate. US Patent 7 (2008), 419,680.
- [9] C. Deepa, A. Nishara Begam, S. Aravindan, Preparation and antimicrobial observations of zinc doped nanohydroxyapatite, *Nanosystems*, 4 (3), (2013) 370-377.
- [10] L. Mohan, D. Durgalakshmi, M. Geetha, T.S.N. Sankara Narayanan, R. Asokamani, Electrophoretic Deposition of nanocomposites (Hap + TiO₂) on titanium alloy for biomedical application, 38, (2011) 3435-3443.