

Effect of Aging Time and Sintering Temperatures on Structural, Morphological and Thermal Properties of Coralline Hydroxyapatite

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Abstract

Apatite is the major constituent of vertebrate bones and teeth. Biphasic Calcium Phosphate (BCP) nanoparticles comprise of Hydroxyapatite (HAP) and Beta-Tricalcium Phosphate (β -TCP) can significantly increase the biocompatibility and bioactivity of synthetic biomaterials. In present study Coralline HAP was synthesized using Goniopora corals using wet precipitation method. Corals were preheated and converted to powder to use as a calcium ion source throughout followed by the successful synthesis of BCP, effect of aging time (24 and 48 hours) at an aging temperature of 25°C was observed on its structural, morphological and thermal properties. X-Ray Diffraction (XRD) confirmed the presence of HAP and β -TCP in major proportions along with Calcium carbonate (CaCO_3) and Calcium Oxide (CaO) in traces revealing the BCP nature of synthetic powders. Crystalline size, lattice parameters, percentage weight fractions, calcium deficiency and resultant Ca/P ratio were calculated and compared. Fourier Transform Infrared (FTIR) spectroscopy confirmed the presence of various PO₄ ion groups and supported the XRD results. Thermo gravimetric Analysis (TGA) disclosed the thermal stability of synthesized powders beyond 600°C. Scanning Electron Microscopy (SEM) observed the agglomerated powder having irregular particle shapes and vivid particle sizes. With the rise of sintering temperature, weight fraction of HAP increased at both the aging times respectively. Aging time of 24 hours at a selected aging temperature yielded the maximum HAP percentage revealing the more bioactive nature of coralline HAP. In addition, weight fraction of trace impurities was also observed to be least at the aging time of 24 hours, thus confirming the optimum parameters for BCP powder synthesis.

Keywords

Hydroxyapatite, Corals, Aging, Sintering Temperature.

I. Introduction

Hydroxyapatite [$\text{Ca}_{10}(\text{PO}_4)_6(\text{OH})_2$; HAP] is chemically similar to the inorganic component of bone matrix. A study by Taniguchi et al. show that sintered HAP exhibits excellent biocompatibility with soft tissues such as skin muscle and gums. Such capabilities have made HAP an ideal candidate for orthopedic and dental implants. Synthetic HAP has been widely used to repair hard tissues. Common uses include bone repair, bone augmentation, as well as coating of implants or acting as fillers in bone or teeth. The material factors affecting the biological response to the implant include the property of HAP powder, Sintering property, Structural configuration and the presence of secondary phases.

In present study, Wet precipitation method is used in the process; the calcium carbonate skeleton of marine coral is converted into hydroxyapatite. The resultant material is called coralline hydroxyapatite (CHAP). In this study the synthesis and characterization of CHAP powder, effect of aging time and sintering temperature are reported. Systematic characterization of the coral before conversion to CHAP has also been carried out.

II. Materials and Methods

A. Coralline Hydroxyapatite Preparation

The corals of genus "GONIOPORA" were obtained which are easily available in coastal areas of India. They exhibit a porous structure with all pores interconnected throughout the skeleton and the skeleton resembles that of a bone. The corals were washed repeatedly with warm water followed by sintering at 900°C for 2 hours to remove organic matter and impurities from the raw corals. The sintered coral were again washed with deionized water followed by overnight drying in air oven at 100°C. On heat treatment the corals become very brittle and were easily crushed to powder. The obtained powder samples were characterized using XRD, FTIR and SEM analysis.

The preheated coral powder is converted into CHAP by using wet precipitation method. Chemical precipitation is performed by first preparing a solution of calcium hydrogen phosphate ($\text{CaHPO}_4 \cdot 2\text{H}_2\text{O}$), 10.29 grams in 100ml of distilled water and Coral solution, 10.11 grams of coral powder in 100 ml of distilled water. In order to obtain CHAP Phosphate solution was added drop wise into coral solution under vigorous stirring at room temperature for 2 hours. Aging was done at 25°C for 24 hours and 48 hours respectively to obtain different test samples. Each sample was centrifuged at 3500 rpm for 5 minutes and gel was extracted in crucible. A systematic series of test samples were prepared by doing Sintering at different temperatures which were selected after preliminary TGA.

B. Sample Characterization

Morphological and micro structural analysis of the coral sample was carried out using Scanning electron microscopy (SEM) Information from thermogravimetric analysis (TGA) was used to select the calcination temperatures more appropriate for the study here reported

Fourier Transform Infrared Spectroscopy (FTIR) was used to investigate the chemical composition of the controls and of the samples calcined at different temperatures.

X-ray Diffraction (XRD) was applied to monitor the phase composition features of the samples before and after calcination at different temperatures ranging from 80°C to 800°C. The structural properties of samples were calculated using Scherrer Formula. The Lattice parameters were calculated by using relation:

$$1/d^2 = 4/3(h^2+hk+k^2)/a^2+1^2/c^2 \quad (1)$$

Where, d = d spacing i.e. distance between adjacent planes in set of miller indices.

a & c = lattice parameters.

Scherrer equation [Eq (2)] was used to calculate crystalline size (t).

$$t = (0.9 \cdot 0.154 \cdot 180) / (\text{FWHM} \cdot \pi \cdot \cos 2\theta / 2) \quad (2)$$

Calcium deficiency Factor (x) was evaluated using relation:

$$(1004)/(310) \cdot \text{wt. ratio} = 3x/1-x \quad (3)$$

Calcium Phosphorous ratio was calculated as under

$$\text{Ca/P} = (10-x)/6 \quad (4)$$

III. Results and Discussion

A. Coral Samples

1. SEM Analysis

Fig. 1 shows SEM micrographs of Raw Coralline powder at various magnifications. SEM micrographs of raw coral powder revealed the loose, unagglomerated mixture of particles having vivid particle sizes ranging from very small to large having 1.669-72.488 μm respectively. Aspect ratio of particles was averaged to be 1.558 with mean roundness of 0.889.

2. FTIR Analysis

Small and narrow peak at 3640 cm^{-1} as shown in fig. 2 belongs to freely available OH functional group, thus revealed the presence of water adsorption deformation in the raw coral powder.

B. Coralline Hydroxyapatite Samples

Various CaP powders were synthesized and categorized using different aging times (24 and 48 hours) and aging temperature 25°C.

Clustered peaks present between 400-4000 cm^{-1} may be due to the vibrations caused by the various molecules present and making the compound.

3. XRD Analysis

XRD diffractogram of raw coral powder as shown in fig. 3 shows that the pretreated coral powder was mainly constituted of Calcium Carbonate (CaCO_3) and Calcium Oxide (CaO) phases as was expected and reported by various authors. Its color was fine white to off white and was observed to be micro-crystalline in nature. Concluding the results earmarked by XRD, SEM and FTIR of raw coral powder; it has been finalized that coral powders were mainly comprised of CaCO_3 and CaO phases. Particles of raw coral powder were observed to be irregular in shape having vivid particle sizes ranges from 1.669 -72.488 μm .

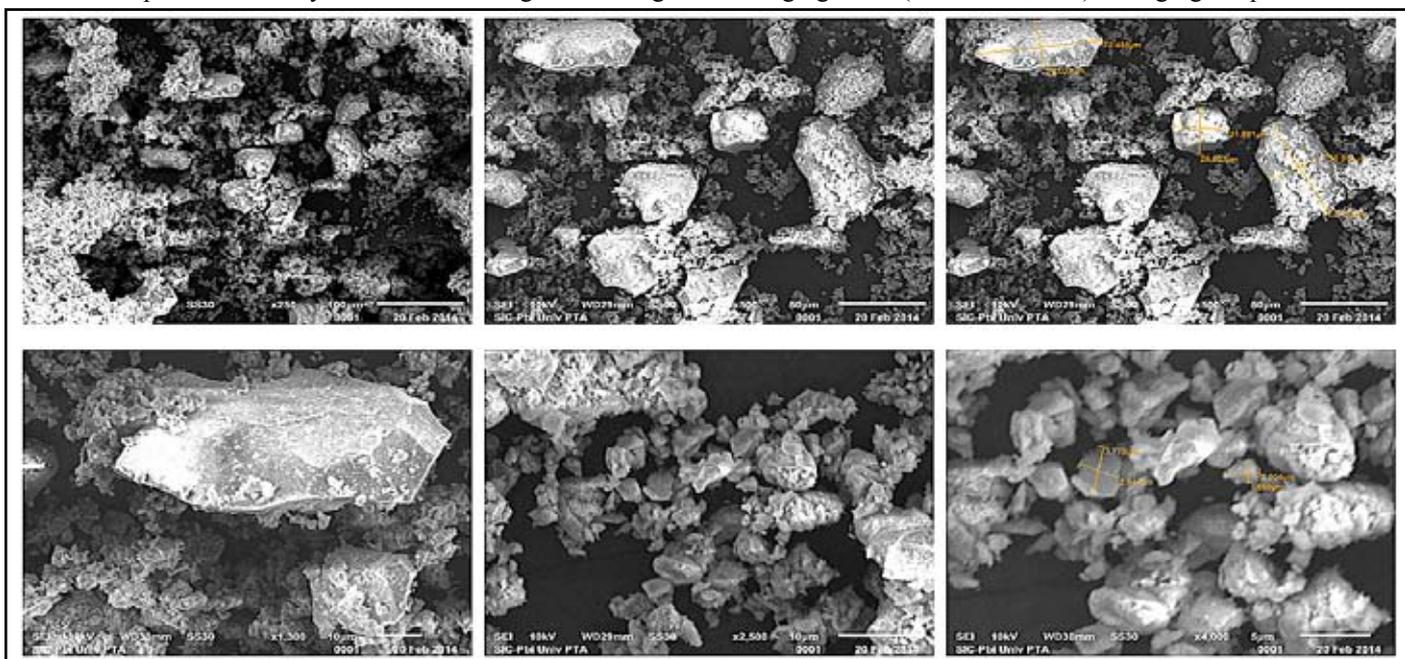


Fig. 1: SEM of Raw Coral Powder

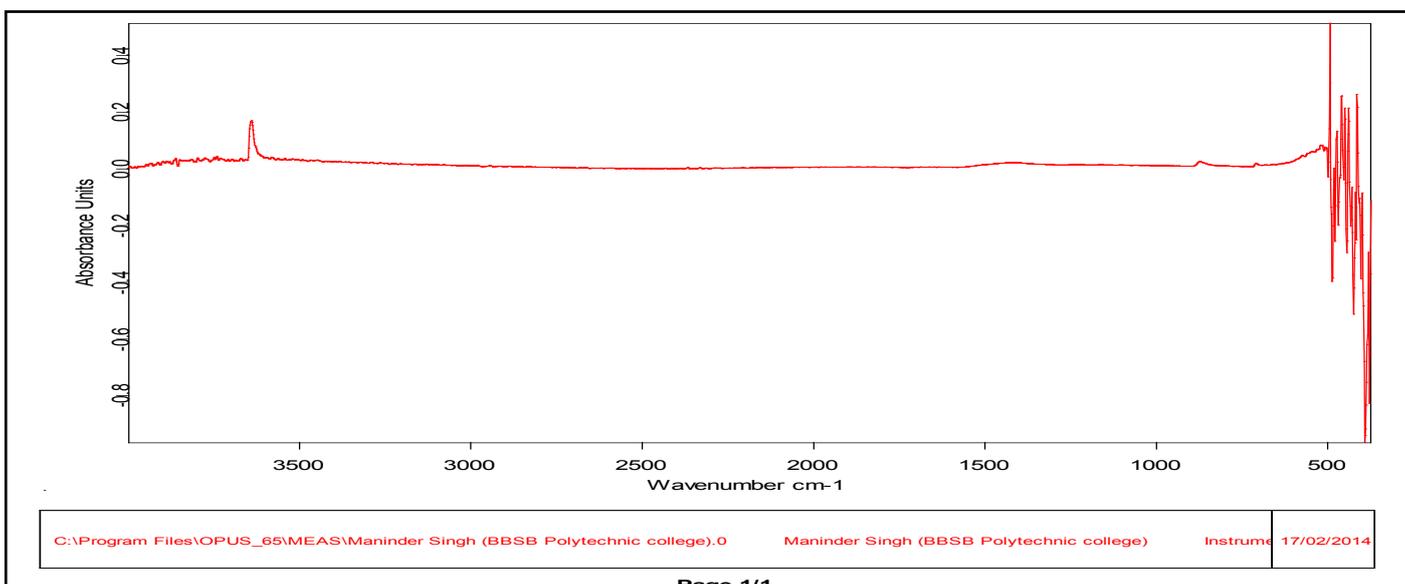


Fig. 2: FTIR of Raw Coral Powder

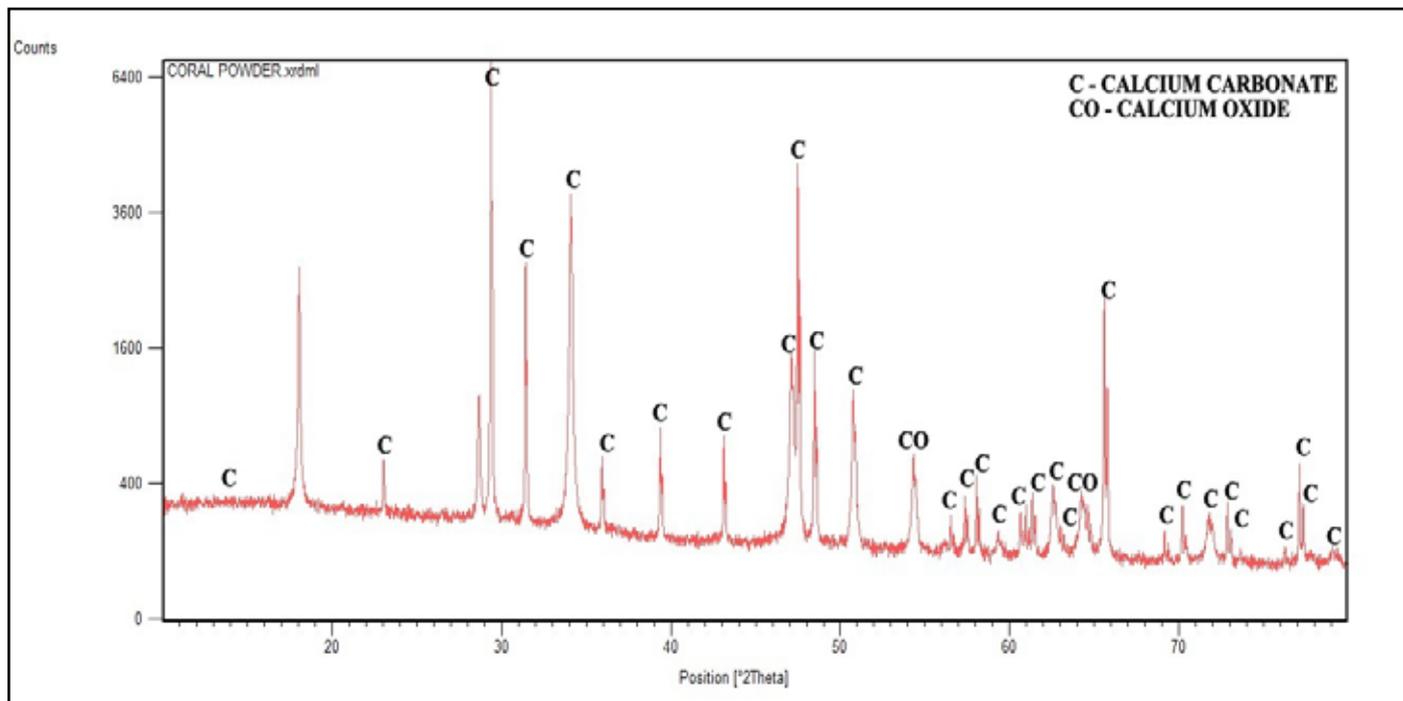


Fig. 3: XRD of Raw Coral Powder

Various synthesized powders were denoted with the different names on the basis of aging times have been mentioned in Table 1 respectively. Six different powder samples were prepared taking care of aging times and sintering temperatures respectively.

Table 1. Various Coralline hydroxyapatite Powder samples and Their Nomenclature

S. No.	Powder Name	Aging Time	Sintering Temperature
1	CHAP -25-24-80	24 Hours	80°C
2	CHAP -25-24-600	24 Hours	600°C
3	CHAP -25-24-800	24 Hours	800°C
4	CHAP -25-48-400	48 Hours	400°C
5	CHAP -25-48-520	48 Hours	520°C
6	CHAP -25-48-800	48 Hours	800°C

1. TGA Analysis

TGA test of CHAP gel prepared at 24 Hours and 25°C of Aging Time and Aging Temperature respectively as shown in Figure 4 suggested 80°C, 600°C and 800°C as the temperatures at which change of mass of respective CHAP powder was observed to be affected maximally. Hence, such were the temperatures at which structural properties i.e. phases, lattice parameters etc. changed critically. Therefore, taking care of TGA interpretation; respective CHAP gel was further sintered at 80°C, 600°C and 800°C respectively and characterized for their structural and morphological properties.

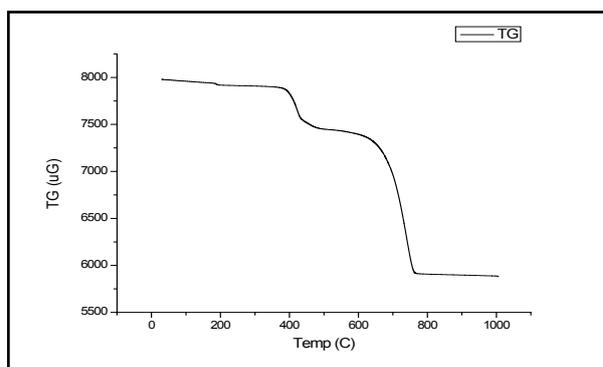


Fig. 4: TGA Behavior of CHAP Powder Prepared at 24 Hours and 25°C of Aging Time and Aging Temperature Respectively

TGA test of CHAP gel (aging time of 48 hours and aging temperature of 25°C) as shown in Figure 5 suggested 400°C, 520°C and 800°C as the temperatures at which change of mass of respective CHAP powder was observed to be affected maximally. Hence, such were the temperatures at which structural properties i.e. phases, lattice parameters etc. changed critically. Therefore, taking care of TGA interpretation; respective CHAP gel was further sintered at 400°C, 520°C and 800°C respectively and characterized for their structural and morphological properties.

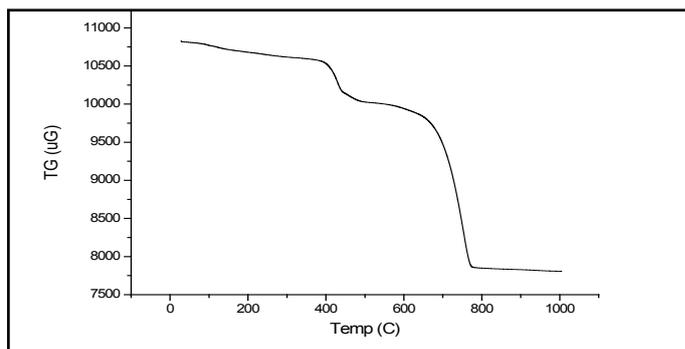


Fig. 5: TGA Behavior of CHAP Powder Prepared at 48 Hours and 25°C of Aging Time and Aging Temperature Respectively

2. XRD Analysis

Fig. 6 shows the XRD diffractograms of raw coral and synthesized CHAP powders prepared at an aging time of 24 hours, aging temperature of 25°C followed by sintering at 80°C, 600°C and 800°C respectively. From the XRD patterns, diffraction peaks have been identified as the hexagonal phase of the HAP powder (JCPDS card no. 09-0432). The observed sharp peaks in the XRD patterns in the samples confirm that the samples are crystalline. On the basis of XRD results, various nano-structural properties viz. phases, lattice parameters, mean crystallite size, weight fractions, calcium deficiency and resultant Ca/P ratio were calculated and have been shown in Table 2. The standard lattice constants $a = 9.418 \text{ \AA}$ and $c = 6.884 \text{ \AA}$ (JCPDS card no. 09-0432) for the HAP has been matched approximately with the calculated values in the present study. It reveals the chemical constitution of synthesized powders primarily comprised of HAP, β -TCP followed by impurities viz. CaCO_3 and CaO in varied percentages. Lattice distortion of HAP phase was remained near to the normal value of 0.730. As expected, crystallite size of HAP phase was observed to increase with the rise of sintering temperature as shown in Table 2. Increase

in sintering temperature caused the percentage of HAP phase increased along with the variation in allied phases consequently; Calcium deficiency and Ca/P ratio of respective synthetic powders were calculated and have been shown in Table 2.

Fig. 6 shows the XRD diffractograms of raw coral and synthesized CHAP powders prepared at an aging time of 48 hours, aging temperature of 25°C followed by sintering at 400°C, 520°C and 800°C respectively. In this sample observed sharp peaks in the XRD patterns are formed at 400°C, 520°C and 800°C. When the sintering temperature increases percentage of HAP also increases. Crystalline sizes vary according to the sintering temperature. Ratio of two elements calcium and phosphorus slightly differ according to sintering temperature. Very close value of Ca/P ratio is 1.62, when compared with the standard value 1.67. These results show the influence of synthesis parameters which play important role in synthesis process. Calcium deficiency decreases with the increasing sintering temperature. Lattice parameters of HAP are shown in Table 3. Lattice distortion of HAP phase was remained near to the normal value of 0.730.

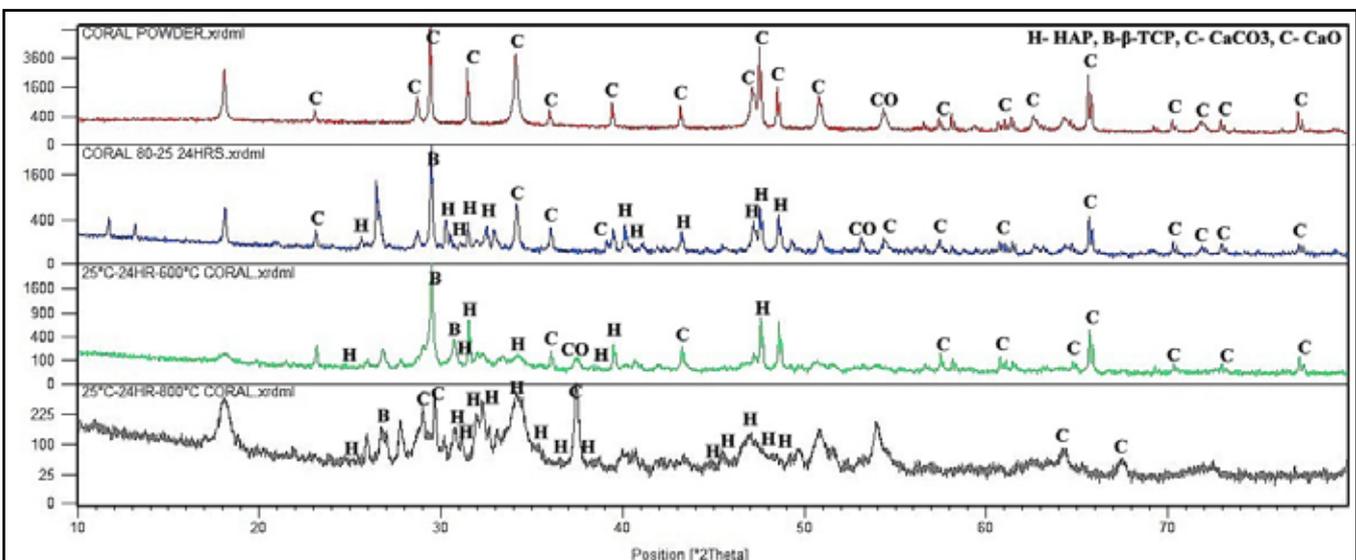


Fig. 6: XRD Diffractograms of Raw Coral and Synthesized CHAP Powders at 80°C, 600°C and 800°C shown From Top to Bottom

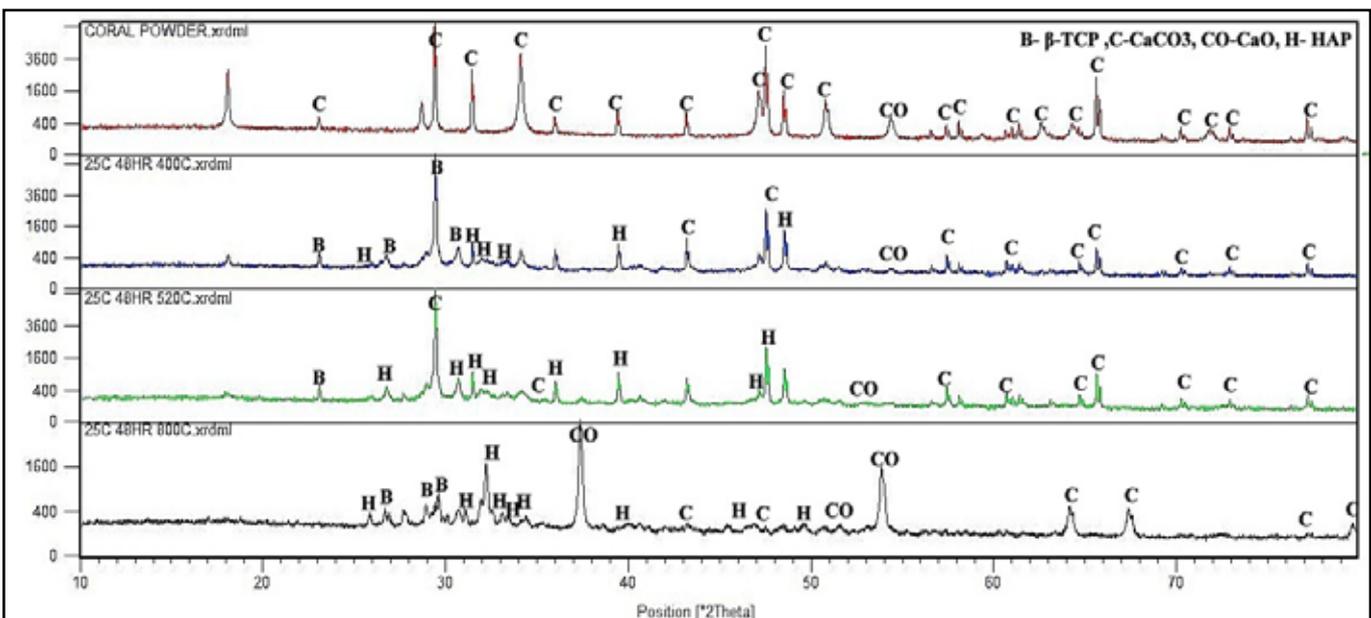


Fig. 7: XRD Diffractograms of Raw Coral and Synthesized CHAP Powders at 400°C, 520°C and 800°C shown From Top to Bottom

Table 2: Calculated XRD Structural Properties of Synthetic CHAP Powders at 25°C, 24 hours

CHAP	Aging Temperature, Aging Time, Sintering Temperature		
	25°C 24Hours, 80°C	25°C 24Hours, 600°C	25°C 24Hours, 800°C
Phases	HAP, β TCP, CaCO_3 , CaO	HAP, β TCP, CaCO_3 , CaO	HAP, β TCP, CaCO_3 , CaO
Lattice Parameter of HAP	a=9.357nm c=6.958nm c/a=0.743	a=9.495nm c=4.865nm c/a=0.512	a= 9.403nm c=6.877nm c/a=0.731
Mean Crystalline Size	111.41nm	32.73nm	30.83nm
Weight Percentage	HAP=23.0 β TCP=44.7 CaCO_3 =28.1 CaO=4	HAP=28.89 β TCP=11.82 CaCO_3 =58.20 CaO=1	HAP=54.24 β TCP=24.44 CaCO_3 =21.32 CaO=Nil
Calcium Deficiency (X)	0.68	0.31	0.33
Ca/P	1.55	1.61	1.61

Table 3: Calculated XRD Structural Properties of Synthetic CHAP Powders At 25°C, 48 hours

CHAP	Aging Temperature, Aging Time, Sintering Temperature		
	25°C 48Hours, 400°C	25°C, 48Hours, 520°C	25°C, 48 Hours, 800°C
Phases	HAP, β TCP, CaCO_3 , CaO	HAP, β TCP, CaCO_3 , CaO	HAP, β TCP, CaCO_3 , CaO
Lattice Parameter of HAP	a=9.503nm c=6.874nm c/a = 0.723	a = 9.503nm c = 6.88nm c/a = 0.724	a=9.431nm c=6.894nm c/a=0.730
Mean Crystalline Size	70.96nm	82.84nm	72.52nm
Weight Percentage	HAP=18.03 β TCP=50.64 CaCO_3 =28 CaO=4	HAP=31.21 β TCP=50.29 CaCO_3 =14.5 CaO=4	HAP=43.94 β TCP=11.49 CaCO_3 =26 CaO=18
Calcium Deficiency (X)	0.75	0.63	0.22
Ca/P	1.54	1.56	1.62

3. FTIR Analysis

Peaks representing various functional groups present in respective synthesized CHAP powder were revealed to be similar in all the FTIRs as shown in fig. 8 and fig. 9. With the rise of sintering temperature from 80-800° C, peaks were observed to be sharper and narrower, showing the increasing crystallinity of various phases present in the powder; thus supported the XRD results as enumerated in Table 2 and Table 3. On the other hand, various vibrational groups belonging to PO_4 ions were observed and remained intact with the rise of sintering temperature. Presence of PO_4 functional group at the lowest temperature of 80° C and 400° C revealed the presence of HAP phase in it which is one of the achievements of present synthesis methodology and also supports the XRD results.

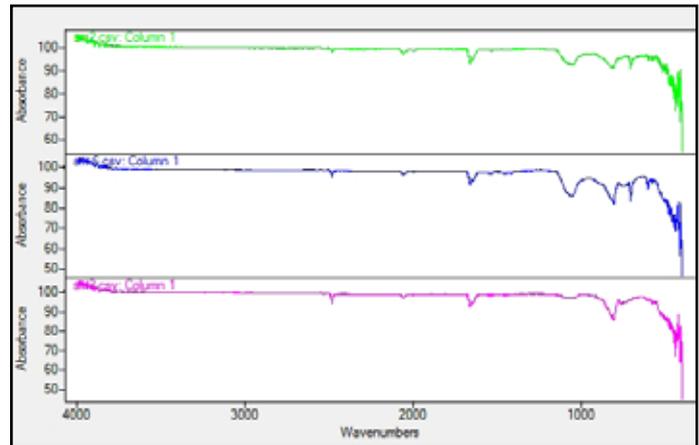


Fig. 8: FTIR Analysis of CHAP Powders Synthesized at Aging Time of 24 Hours, Aging Temperature of 25°C Followed by Sintering at 80°C, 600°C and 800°C shown from Top to Bottom Respectively

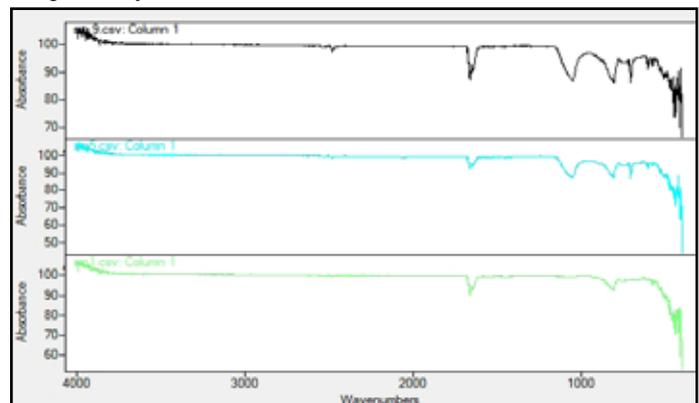


Fig. 9: FTIR Analysis of CHAP Powders Synthesized at Aging Time of 48 Hours, Aging Temperature of 25°C Followed by Sintering at 400°C, 520°C and 800°C shown from Top to Bottom Respectively

IV. Conclusion

The results show that the aging time and aging temperature highly conditions the properties of coralline HAP. On the basis of above results, we can conclude that with the rise in sintering temperature, weight ratio of HAP rises. Lattice parameter of HAP a and c have standard values 9.418nm and 6.884nm for hexagonal structure, c/a ratio is 0.730, when compared with sample's distortion ratio then we can say that low aging temperature samples have distortion ratio very close enough to standard value, it means lattice structure made fine at low aging temperature. When aging was done at 25°C and 24 hours CaO was no longer detected but traces of calcium carbonate was found but the percentage of HAP was found more as compared to aging time 48 hours.

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