Phases formation in cerium-doped hydroxyapatite

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Abstract. Cerium-substituted hydroxyapatites (Ce-HAs) with an estimated content of Ce$^{3+}$ of 0; 0.1; 0.25; 0.5 mol. % were prepared by two different synthesis methods: precipitation from aqueous solutions of salts and mechanochemical synthesis. The powders after synthesis were heat-treated at 1250 °C to achieve well-crystallized phases. The X-ray diffraction analysis (XRD), scanning electron microscopy (SEM), energy dispersive analysis (EDA), and dilatometry methods revealed that cerium ions didn’t not fully incorporate into the HA structure, but rather were transform to the CeO$_2$ phase. It was attributed to the transition of Ce valency (III)$\rightarrow$(IV) through the oxidation reaction under air conditions during materials heating. Two different synthesis methods provided similar phase composition of the obtained compounds, though different microstructure of the resulted ceramics. It was shown that cerium introduction prevents HA sintering during thermal activation, possibly due to contribution of large atomic mass of cerium ions and formation of cerium (IV) oxide at the crystal boundaries.

1. Introduction

The various pathological diseases caused by injuries, surgery, or pathological changes in the body that lead to bone damage is the serious problem in medicine. In this regard, the development of ceramic biomaterials intended for bone implantation is an urgent area of materials science. To this regards, hydroxyapatite (HA) occupies a special position, since it is the main mineral component of bones and teeth and has good biocompatibility and bioactivity [1-4]. Synthetic HA is widely used in medicine as a bone substitute material, as well as a coating on the metal implants [5-9].

It is known that HA of natural bone contains a large number of impurities, such as Mg, Na, K, etc., and all of them are biologically important. Isomorphic substitutions in the structure have a significant effect on the structural characteristics of HA, which, in turn, affects the behavior of the material in the body. It is known that isomorphic substitutions lead to deformation of the crystal lattice of the base material, reducing the particle size, that affects the phase stability during heat treatment [10-15]. In this regard, in this paper, the preparation of the cation-substituted hydroxyapatite is considered as one of the methods to obtain its highly dispersed powders.

Some promising area of bone treatment is the use of biomaterials with luminescent properties due to possibility to observe of biological processes dynamics [16, 17]. One of the elements that carry these
properties is cerium, which possesses also antibacterial and antitumor properties and can equip HA structure with them [18-20]. However, there are several difficulties in obtaining cerium-substituted HA, primarily the difference in the oxidation state of Ca$^{2+}$ and Ce$^{3+}$ ions, as well as the presence of the Ce$^{4+}$ oxidation state and the high atomic mass of the element. These features can lead to an irregular distribution of dopant in the HA structure and thermal instability of the compounds. Ones of the most common methods for producing HA with high dispersion are the precipitation of salts from aqueous solutions and mechanochemical synthesis [1-3, 21, 22]. The first method is easy enough to use in powder technology of nanoscale materials. Synthesis under conditions of mechanochemical activation is of interest due to the simplicity of the hardware (the use of mills) and the efficiency of the exposure to the material.

The aim of this work was to study the effect of the synthesis method (precipitation from aqueous solutions of salts/mechanochemical synthesis) of cerium-substituted HA on the phase composition and microstructure formation.

2. Materials and methods

2.1. Materials and synthesis methods

Powders of Ce-doped HAs of Ce content of 0; 0.1; 0.25, and 0.5 mol. % were produced via two synthesis methods, the aqueous precipitation and the mechanochemical synthesis. The obtained materials were named as HA-0; HA-0.1; HA-0.25; HA-0.5. The aqueous precipitation method to synthesis Ce-HAs was performed using analytical reagent grade calcium nitrate tetrahydrate (Ca(NO$_3$)$_2$×4H$_2$O) and cerium nitrate hexahydrate (Ce(NO$_3$)$_3$×6H$_2$O) as the source of Ca$^{2+}$ and Ce$^{3+}$ ions. The diammonium phosphate ((NH$_4$)$_2$HPO$_4$) was used as the precipitator. The mixture of 0.5 M calcium nitrate solution and 0.1 M cerium nitrate solution was prepared, and 0.5 M diammonium phosphate solution was added dropwise to the nitrate mixture. The pH value was kept at 11.5±0.5 by regular addition of 25 % solution of aqueous ammonium (NH$_4$OH). Synthesis was carried out under constant intensive stirring for 3 hours at room temperature. The reagents amount was calculated in accordance with the reaction (1):

$$ (10-x)\text{Ca(NO}_3\text{)}_2 + (2x/3)\text{Ce(NO}_3\text{)}_3 + 6(\text{NH}_4\text{)}_2\text{HPO}_4 + 8\text{NH}_4\text{OH}\rightarrow \text{Ca}_{(10-x)}\text{Ce}_{(2x/3)}(\text{PO}_4)_6(\text{OH})_2 + 26\text{H}_2\text{O} + 20 \text{NH}_4\text{OH}. $$

The mechanochemical synthesis of HAs was carried out using analytical reagent grade calcium oxide (CaO), diammonium phosphate ((NH$_4$)$_2$HPO$_4$), and cerium nitrate hexahydrate (Ce(NO$_3$)$_3$×6H$_2$O) in a planetary mill with alumina balls. Calcium oxide was previously calcinated at 1000 °C during 1 h. The synthesis was implemented in two stages. At first stage the solid reagents were intensively mixed in grinding vials during 30 min at rotation speed of 150-200 rpm. Then the distilled water was added to the vials with reagents mixture to perform the second synthesis stage at the same milling conditions. The reagents amount was calculated in accordance with the reaction (2):

$$ 10\text{CaO} + 6(\text{NH}_4)_2\text{HPO}_4 + \text{H}_2\text{O} \rightarrow \text{Ca}_{10}(\text{PO}_4)_6(\text{OH})_2 + 12\text{NH}_4\text{OH}. $$

At the end of the synthesis, the reaction product was subjected to aging for 24 h at room conditions. Then it was filtered, washed by distilled water, and dried at 80 °C. The obtained dry cake was crushed in agate mortar and wiped through the mesh of 100 µm cell size. To obtain well-crystallized phases for further XRD analysis the resulting powders were heat-treated at 1250 °C. The evaluation of the effect of cerium dopant on the microstructure formation of the ceramics was performed. For this purpose, the cylindrical samples of 5 mm diameter were fabricated by uniaxial double-sided pressing of the resulted powders in a steel mold on a hydraulic manual press at 5 MPa. The pressed samples were sintered in the furnace with Si-C heaters at 1250 °C with the rate of 10 °C/min.

2.2. Sample characterization

X-ray diffraction analysis (XRD, Shimadzu) was implemented using the diffractometer with CuKα radiation (average wavelength λ = 1.54183 Å). Data were collected in 20 range of 10–60 deg., step 20 of
0.02 deg. and counting time of 2 s/step. XRD were performed on the powered samples calcinated at 1250 °C. Sintering of the materials was studied by measuring continuous shrinkage on a horizontal dilatometer (DIL 402C Netzsch, Germany), with corundum rigging, equipped with a high-precision displacement sensor (500-5000 µm, accuracy up to 1×10^{-5}) in an argon atmosphere. The linear dimensions of the samples were measured at a continuous temperature increase up to 1400 °C at a constant rate of 10 °/min. Microstructure of the ceramics was investigated by scanning electron microscope (SEM, Tescan Vega II SBU) with integrated Energy dispersive X-ray microanalyzer (EDX, INCA Energy).

3. Results and discussion

According to XRD, the powders obtained by aqueous precipitation corresponded to the apatite structure (Fig. 1). The introduction of cerium of 0.1 and 0.25 mol. % led to calcium oxide presence up to 1-2 wt.% in the main HA phase. All Ce-HAs contained a phase of cerium oxide (IV), CeO_{2}. The diffraction peak at 28.6 deg. characterized this compound (Fig. 1). With Ce amount rising the peak intensity increased, which indicated an increase in the CeO_{2} content.

XRD patterns of the powders obtained by mechanochemical synthesis are presented on Fig. 2. The similar patterns were observed. The small amount of CaO presented in 0.1 and 0.25 Ce-HA and it lacked in 0.5 Ce-HA. All the Ce-HAs contained CeO_{2}, its content increased in the concentration range.

![Figure 1. XRD patterns of Ce-HAs obtained by aqueous precipitation method.](image-url)
Figure 2. XRD patterns of Ce-HAs obtained by mechanochemical synthesis.

The shrinkage measuring of the Ce-HAs obtained by two synthesis methods are presented on Fig. 3. For the powders obtained by precipitation technique it was revealed that the first sintering stage started at 600 °C. The course of the shrinkage curves was similar for all compositions up to 900 °C. The active sintering for all materials began at 870 °C. The HA-0 sample showed more intense sintering with greater shrinkage value of 25 % at 1040 °C. For HA-0.1 and HA-0.25, the sintering was completed earlier (at 1000 °C) with a lower shrinkage value of 22 %. The active sintering interval for HA-0.5 was significantly shorter and completed at 1000 °C with shrinkage value of 20 %. The continuous shrinkage curves for pure and Ce-containing HAs had a traditional general course and showed a decrease in the sintering intensity with an increase in the cerium amount.

For the specimens of Ce-HAs obtained by mechanochemical synthesis the course of the continuous shrinkage curves was slightly different from the previous ones. Although the general tendency to slow down the HA sintering with Ce entry remained. The specimen of HA-0 showed a wide sintering interval from 680-1310 °C with a shrinkage value of 28 %. The shrinkage curve for HA-1 was gentler, with the shrinkage reaching 23 % at the end of intensive sintering at 1290 °C. The course of the shrinkage curves for HA-0.25 and HA-0.5 was the same and shorter, which was explained by the incomplete entry of the dopant into the structure of these materials.

The decrease in the relative shrinkage of the material at the end of active sintering was associated with two possible reasons. On the one hand, the partial entry of cerium ions with a higher atomic mass than calcium ions into the HA structure led to the inhibition of diffusion in the processes of mass transfer during thermal activation. On the other hand, in high-temperature processes, the formation of an impurity phase of cerium (IV) oxide at the crystal boundaries or in aggregates could also prevent the HA sintering.
Figure 3. Linear shrinkage of obtained HAs vs the temperature: aqueous precipitation method (a) and mechanochemical synthesis (b).

SEM observation of Ce-HA ceramics obtained from the powders synthesized by aqueous precipitation indicated that cerium introduction resulted in reducing of crystal size and increasing of total porosity (Fig. 4). The ceramics surface of HA-0 was composed of the crystals of average size of 2.5-5 µm. Both dense areas and porous parts with a short particle contact and large pores up to 4 µm in size were viewed. Ce-HAs ceramics was composed of uniform in shape and size particles, of 0.5-1.5 µm. Also, the porous areas were presented in the structure, smaller in size, comparing with HA-0. When regarding the SEM micrographs of HA-0.5, the regions containing particles of a different morphology and size compared to the main phase were observed. A detailed examination and the results of the energy-dispersion analysis gave reason to believe that the phase of CeO₂ was out in the separate particles agglomerates of 0.1 µm in size. According to the EDA results, the Ce content in these areas was 30 times higher than its content in the main material (Fig. 5).

SEM micrographs of the Ce-HAs ceramics obtained from the powders synthesized by mechanochemical synthesis are shown on Fig. 6. The structure of HA-0 ceramics was homogeneous and dense, composed of crystals of 2-3 µm in size, with a small intercrystallite pores up to 0.5 µm. It was revealed that the Ce introduction resulted in slight decreasing of crystal size of the HA ceramics and porosity rising. HA-0.1 ceramics was composed of crystals of 1 to 2 µm, HA-0.25 was consisted of crystals of 3 µm. The structure of HA-0.5 was composed of crystals up to 4 µm, and fine-crystalline highly porous inhomogeneous regions with a particle size of 0.3 µm were observed. According to the EDA results, the cerium content in these areas was 5 times higher than the cerium content in the main material (Fig. 7).

Figure 4. SEM micrographs of the Ce-HAs ceramics obtained from the powders synthesized by aqueous precipitation: a – HA-0, b – HA-0.1, c –HA-0.25, d – HA-0.5 (magnification ×5000).
Table 1

EDA results for HA-0.5 ceramics obtained from the powders synthesized by aqueous precipitation.

| Spectrum | Element content, at. % |
|----------|------------------------|
|          | O  | P  | Ca | Ce |
| 1        | 64,00 | 13,34 | 22,39 | 0,26 |
| 2        | 69,03 | 8,09  | 12,59 | 10,29 |
| 3        | 53,34 | 16,73 | 29,89 | 0,03 |

Figure 5. SEM micrograph (a, magnification ×5000) and EDA results for HA-0.5 ceramics obtained from the powders synthesized by aqueous precipitation.

Table 2

EDA results for HA-0.5 ceramics obtained from the powders produced by mechanochemical synthesis.

| Spectrum | Element content, at. % |
|----------|------------------------|
|          | O  | P  | Ca | Ce |
| 1        | 63,22 | 14,64 | 22,06 | 0,08 |
| 2        | 56,15 | 16,20 | 22,51 | 5,14 |
| 3        | 59,90 | 15,58 | 22,73 | 1,80 |

Figure 6. SEM micrographs of the Ce-HAs ceramics obtained from the powders produced by mechanochemical synthesis: a – HA-0, b – HA-0.1, c – HA-0.25 (magnification ×5000).

Figure 7. SEM micrograph (a, magnification ×5000) and EDA results for HA-0.5 ceramics obtained from the powders produced by mechanochemical synthesis.
4. Conclusion

Thus, the aqueous precipitation and mechanochemical synthesis failed in obtaining of thermally stable HA with calcium ions replaced with cerium ions in case of subsequent temperature treatment in an oxidation environment. It is explained by the incomplete entry of the dopant into the HA structure during synthesis, as well as by the transition of the cerium valency from III to IV under oxidation conditions with temperature increasing. It was revealed that cerium introduction prevents the HA sintering due to inhibition of ion diffusion in the processes of mass transfer during thermal activation and due to formation of cerium (IV) oxide at the crystal boundaries. In this regard, in order to obtain the thermally stable hydroxyapatite with the substitution of calcium ions with cerium ions, it is advisable to study in detail the effect of the temperature and environment of heat-treating on the phase formation of the obtained powders.

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