**Continuous synthesis and characterization of hydroxyapatite nanoparticles modified with lecithin**

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**Highlights**

* Y-shaped millireactor application to the continuous synthesis of hydroxyapatite
* Lecithin used as a surfactant to control the morphology of particles
* Synthesis provides carbonated hydroxyapatite with a spherical shape
* The aging temperature affects the particles size

**1. Introduction**

Hydroxyapatite (HAp) is the main inorganic part of human hard tissue (bones and teeth) [1]. Therefore synthetic HAp is a common material in regenerative medicine, particularly in bone tissue regeneration [2]. Those applications create a demand for new methods of HAp synthesis. Among all the methods for producing hydroxyapatite powders, synthesis in a continuous reactor seems to be a promising way to obtain better control over the reaction conditions. Production of hydroxyapatite nanoparticles using ultrasonic tubular microreactor, meso-oscillatory flow reactor, tube-in-tube microchannel reactor, among the others, has been reported in the literature [3]. We present results of HAp nanoparticles synthesis using a Y-type millireactor. For this purpose, we use 3D printing technology to produce a continuous reactor with 1 mm inner hydraulic diameter and 10 mm in length of the outlet channel. We investigated the influence of the lecithin concentration used in the synthesis and the aging conditions on the properties of the powders.

**2. Methods**

We synthesized hydroxyapatite in the presence of lecithin (HAp-LE) by chemical precipitation method using a solution of Ca(NO3)2·4H2O and solution of (NH4)2HPO4 in ultra-pure water as starting media [4]. Ca/P molar ratio was maintained at 1.67 as in a stoichiometric HAp. The synthesis was conducted in Y-shaped continuous reactor (1 mm inner hydraulic diameter and 10 mm in length of the outlet channel) under atmospheric pressure with each starting media flow rate of 500 ml/h. The resulting suspension of hydroxyapatite was aged in an aging reactor for 24 h at room temperature (RT) or at 60 °C, or directly centrifuged without aging (wa). After the aging process, the suspensions were centrifuged, washed with fresh water and then dried in an oven at 50°C for 24 h. Four different concentrations of lecithin were tested: 1, 3, 5 and 15 g/L in the reaction mixture. Resulting powders were characterized by X-ray diffraction (XRD), Fourier transformed infrared spectroscopy (FTIR), scanning electron microscopy (SEM), nanoparticle tracking analysis (NTA), dynamic light scattering (DLS) and zeta potential measurement. Moreover, we implemented the method of Chemical Oxygen Demand (COD) analysis to the quantitative determination of lecithin concentration in powders.

**3. Results and discussion**

Results of investigation of phase purity (XRD) and chemical composition (FTIR) show that we obtained carbonated hydroxyapatite regardless of the aging conditions and the amount of lecithin used. COD measurements confirm predicted increase in the concentration of lecithin in the final product with an increasing concentration of lecithin used in synthesis. SEM images show structure consisting of spherical particles with size 10-25 nm. The sizing techniques used (NTA and DLS) demonstrate broad particle size distribution indicating particles agglomeration with sizes to about 500 nm in NTA and about 1 000 nm in DLS. The values of the zeta potential around –5 mV explain instability and tendency of particles to agglomerate. Of the parameters tested here, the aging temperature has the greatest influence on the final properties of the HAp-LE. An increase in aging temperature causes higher crystallinity, larger crystal size, and larger particle sizes. What is more, we obtain a minimum particle size depending on the lecithin concentration - for samples aged at room temperature with a lecithin concentration of 5 g/L (around 10 nm), and for samples aged at 60 °C and without aging with a concentration of 3 g/L (around 15 nm).

**4. Conclusions**

The continuous process of hydroxyapatite precipitation in a 3D-printed millireactor was achieved and the final products were characterized. We present a simple and low-cost method to produce spherical nanoparticles modified with lecithin. Among tested parameters, the aging temperature has the most significant influence on powders properties (size, crystallinity). The concentration of lecithin also has an influence on the size of the resulting HAp-LE particles. Therefore, we conclude that lecithin is an addition that permits control over the size of the obtained particles. Summarizing, when planning the production of hydroxyapatite using the proposed approach, it is essential to control the lecithin concentration and the temperature in the post-processing reactor.

**Acknowledgements**

Authors acknowledge funding within the project: “Innovative polymer composites for filling bone defects”-INPOLYBOND. NCBR/EC, Smart Growth Operational Program for 2014-2020 of European Regional Development Fund, (POIR.04.01.04.00-0133/15).

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