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Nanoscale Apatite Biomaterials for Osteointegration

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The thermal behavior of bone tissue has been investigated in order to assess how it is affected structurally by incineration. The need of the development of such technology is caused by big number of patients with oncology and dystrophic bone tissue diseases which use the artificial bone tissue replacement. The samples like small plates that cut from local area of bone fragment, were exposed to annealing in temperature range 560-720 °C and were characterized by XRD, SEM, EPMA, XPS and method of differential dissolution.

Keywords: Bone Tissue, Thermal Decomposition, SEM, EPMA, XRD, XPS, Differential Dissolution.

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1. INTRODUCTION

Nanoscale crystals of biogenic apatite from bone tissue have complicated structure that due to their functionality. The synthesis such structure in artificial materials for osteoplastics is impossible. This circumstances is stimulated the investigation of natural bone tissue apatite as source of biomaterials. The organic component of donor bone tissue may be removed by pyrolysis or chemically treatment or by their combination for the absence of immune reaction. For today the problem of optimization of treatment process for full organic components removement at minimal changes of biomineral structures is very important. Nanoscale bioapatite crystals with saved natural properties could be the basic material for development of artificial mineral organic composites for osteointegration.

The need of the development of such technology is caused by big number of patients with oncology and dystrophic bone tissue diseases which use the artificial bone tissue replacement.

In this work the thermal behavior of bone tissue has been investigated in order to assess how it is affected structurally by incineration.

As a result, new technology for the production of nanostructured materials based on natural bioapatite were developed.

2. MATERIALS AND METHODS

The objects of investigation were bioapatite samples obtained from cortical (dense) bone from different animals. During preliminary preparation, the samples were mechanically cleaned, washed in distilled water and dried in air. In order to obtain temperature series, the samples (in the form of small plates cut from one local region of the bone fragment) were annealed in temperature range corresponding to recrystalization of apatite (560°C-720°C) in 40°C steps. At each fixed temperature, the samples were held for 1h and then were slowly cooled down to room temperature. The samples were characterized using physical approaches that available as a result of conjunction the next methods: XRD, SEM, EPMA, XPS and differential dissolution.

3. RESULTS AND DISCUSSION

According to the literature [1] bone annealing includes the following processes:

a) ~ 120 °C - the loss of surface-bound water;

b) 400-500 °C - the loss of crystal water. This loss is accompanied by a decrease of the lattice parameter a ~ 0.02 Å for Na-bearing carbonate apatite, and a ~ 0.003 Å - in the absence of ions Na;

c) ~ 600 °C - the loss of water and ions HPO42-;

d) for a well-crystallized carbonate apatite CO2 emissions starting at 550 °C, and usually ends at 900 ... 1000 °C;

e) if the molar ratio of Ca/P < 1.667, the ß-TCP is also presents.

f) In biological apatite carbonate CO2 loss can occur at lower temperatures than in well crystallized apatite.

Bone extracellular matrix has a complex multi-level organization of the uneven distribution of the micro and macro elements in the osteon-lamellar structures [1, 3] (see Fig. 1).

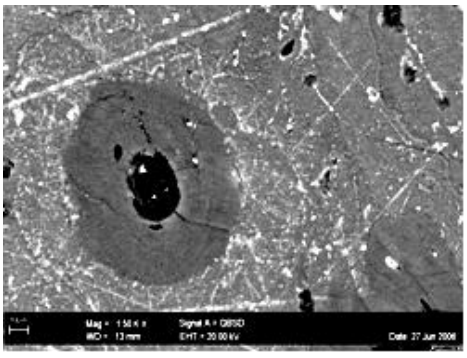

Fig. 1 – SEM image of bovine bone tissue osteon-lamellar structure.

Initially composed of intergrown collagen and hydroxyapatite (HAP), combustion of the organic component is complete by 560 °C, with most mass loss (50-55%) occurring by 500 °C. Combustion of collagen is accompanied by an increase in HAP mean crystallite

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size at temperatures greater than 600 °C, from 28 nm to a constant value of 190 nm at 700 °C [2]. The temperature range of 600-700 °C is characterized by significant structural and micro-structural rearrangements of bioapatite.

Energy Dispersive X-ray Analysis (EDX) along and across the osteon of bone tissue samples and EDX compositional mapping were perfomed.

It was found that *Mg* in biopatite can be both on the surface of the nanocrystals and embedded in-crystal lattice, replacing calcium. That is in good agreement with literature data [4, 5].

It has been found that the source of systematic error may be an option of a reference sample.

Surface chemical composition was studied by XPS technique. The XPS measurements indicate that the samples have lines typical for phosphorous, carbon, calcium, oxygen, sodium and magnesium. Intensities of photoelectron lines from other elements were on the noise level and within the bounds of XPS method sensitivity are not detected. The survey photoelectron spectrums of investigated samples are represented on Fig. 2.

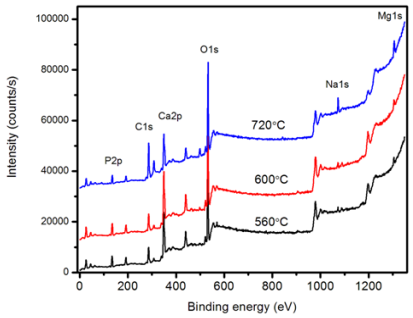

Fig. 2 – XPS survey spectrums from annealed at different temperatures (560 °C, 600 °C, 720 °C) bone samples.

Analysis of recorded with high resolution lines of calcium - BE(Ca2p)=347.3 eV and phosphorous - BE(P2p)=133.3 eV indicate that calcium is in state of Ca2+ and phosphorous is in state of PO4-, that corresponds to the next chemical compounds, for example, calcium phosphat Ca3(PO4)2 (CaP) or hydroxyapatite Ca10(PO4)6(OH)2 [6, 7].

According to differential dissolution analysis the composition of main phase of bone tissue can be fragmentary represented (without oxygen and hydrogen) by the formula:

$$Ca_{1.66}Mg_{0.054}Sr_{0.001}Na_{0.06}P_{1.00}$$

The content of the phase in the sample is 99.9%. Sample also contains small amounts of phase - probably phosphates - *Mg*, *Sr*, *Na* and *K*. Ratios of these elements in the small phases are *Mg* 1.4 rel.%, *Sr* 4.4 rel.%, *Na* 3.3 rel.% - from the total in the sample content of each of these elements. These elements are almost completely covered in the main phase. At the same time in the main phase is not potassium, which either forms its own phase or concentrated in the surface layers of the main phase.

The received results accomplish a modern performances about ultrastructural organization of nanoscaled apatite of bone tissue biominerals.

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REFERENCES

1. J.C. Elliott, *Structure and Chemistry of the Apatites and Other Calcium Orthophosphates. Studies in Inorganic Chemistry* (Amsterdam. Elsevier. 1994).
2. F. Betts, N.C. Blumenthal, A.S. Posner, *J.Cryst.Growth* **53**, 63 (1981).
3. S.N. Danilchenko, A.V. Koropov, I.Yu. Protsenko et al., *Cryst. Res. Technol.* **41** No3, 268 (2006).
4. A. Bigi, E. Foresti, R. Gregorini, *Calcif. Tissue Int.* **50**, 439 (1992).
5. A. Bigi, G. Falini, E. Foresti, *Acta Cryst.* **52**, 87 (1996).
6. C.C. Chusuei, D.W. Goodman, M.J. Van Stipdonk, D.R. Justers, Schweikert, *Anal. Chem.* **71**, 149 (1999).
7. S. Kaciulis, G. Mattogno, L. Pandolfi, L. Cavalli, G. Gnappi, *Appl. Surf. Sci.* **151**, 1 (1999).