Processing and characterization of glass reinforced biogenic hydroxyapatite composites with ferromagnetic additives

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Abstract

Biogenic hydroxyapatite-glass composites, with addition of up to 2 wt.% of Fe or Fe_3O_4, were fabricated from biogenic hydroxyapatite powder and four types of reinforcing glasses by sintering under different conditions. It has been established that the relative density, mechanical and magnetic properties of the prepared composites depend on the composition and sintering conditions. The composites with 1 wt.% Fe or Fe_3O_4 sintered at 500°C in vacuum have a magnetic susceptibility of $2–3\cdot10^{-3}$ cm³/g. This parameter decreases to $0.858\cdot10^{-3}$ cm³/g for the specimens with 1 wt.% additives sintered at 800°C in vacuum and to $0.27–1.3\cdot10^{-3}$ cm³/g for the specimens with 2 wt.% additives sintered at 500°C under the usual atmospheric conditions. The magnetic susceptibility for the specimens with 2 wt.% additives sintered at 780°C under the same atmospheric conditions decreases to $0.24–1.25\cdot10^{-6}$ cm³/g. These ferromagnetic additives influence the degradation rate in vitro within the first 40 min of the composite soaking. Short-term treatment of specimens by the magnetic field leads to an increase in the initial degradation rate, but insignificantly influences it within more prolonged soaking.

Keywords: bioceramics, hydroxyapatite-glass composites, magnetic susceptibility, porosity

I. Introduction

Hydroxyapatite (HAp) is used predominantly to replace bone tissue in many biomedical applications in the form of granules or separate blocks. The works, carried out by Santos et al. [1], have shown that HAp could be reinforced by adding a P_2O_5-CaO glassy phase during its sintering. Other authors have also shown that phosphate glasses added to HAp promote atomic diffusion and development of strong bonds with crystal particles, and densification occurs via liquid formation [2–4]. It has been shown [5] that an average hardness of 383 HV, density of 2.72 g/cm³, and compressive strength of 83 MPa are achieved by sintering at 1200°C with addition of 10 wt.% phosphate bioglass into HAp.

The authors have shown that granules of biogenic hydroxyapatite (BHAp) can be reinforced by the Na-B-Si glasses addition (15–70 wt.%) [6]. Mechanical strength of such glass reinforced BHAp is high enough, and Si-O complexes in their composition activate the interaction of an implant with bone tissue [7].

Development of bioactive materials with ferromagnetic properties is an extremely urgent task for the directed transport of drugs and for magnetic resonance [8–10]. The basic concept in magnetic bioseparation is to selectively bind a biomaterial of interest (e.g., a specific ell. Protein, or DNA sequence) to a magnetic particle and then to separate it from the surrounding matrix using a magnetic field [8]. Nanoparticles of Fe_3O_4 with diameters in the 5–100 nm range are "supermagnetic" and typically used for such separation. They are dispersed in pores of larger microparticles, tagged to the

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biomaterial of interest, and can be removed from the matrix using a magnetic field, but they do not agglomerate after the cutting the field off [8].

In this study, BHAp-glass composites were sintered together with magnetite or iron nanopowders, which are commonly used in medical applications due to their low reactivity. The structure and properties of such composite material are presented and discussed.

II. Experimental

Different BHAp-glass composites, with addition of up to 2 wt.% of Fe or Fe_3O_4, were prepared from BHAp powder and four types of reinforcing glasses by sintering under different conditions. The BHAp powder was produced by treatment of mammal's native bones corresponding to the medical requirements ASTM F1185-88. The compositions of four different reinforcing glasses, used for preparation of the BHAp-glass composites, are presented in Table 1.

The BHAp-glass samples were prepared by classical ceramic technology using two-stage sintering procedure (the first sintering temperature was 1100°C, whereas the second one was selected depending on the type of BHAp-glass composites) [6]. Thus, the OK-50 and OK-50(M3) composites were produced using the second sintering temperature of T_2=780°C. The OK-57 specimens were sintered in vacuum at 500 or 800°C to reduce ferromagnetic additive oxidation. The OK-85 specimens were, in the second stage, sintered at 500°C under the usual atmospheric conditions (Table 2).

The iron powders with an average particle size of 60 nm were obtained by Kushchevska [10]. The magnetite (Fe_3O_4) powders, produced by Integran Techn., had an average particle size of 9 nm. Ferromagnetic additives (1 or 2 wt.%) were introduced into composite powders by mechanical mixing before second-stage sintering. The primarily sintered composites were ground into granules ≤ 160 μm, mixed with additives and pressed into regular cylindrical specimens for secondary sintering.

The specimen compositions were determined using chemical, X-ray and infrared spectroscopy analyses (specimen/KBr weight ratio was 15/300). To study the composite microstructure, the scanning electron microscope CAMEBAX was used. Magnetic susceptibility was measured by Faraday's method relative to the strong paramagnetic material GdB_4 with the known temperature dependence of magnetic susceptibility [11,12].

Taking into consideration the small cylindrical sizes (15.0 mm diameter and 8.0 mm height) of the tested specimens, the measurements of mechanical properties were limited to the compressive splitting strength and indentation fracture toughness. The compressive splitting strength, R_{cs}, was determined by compression of a disc with flatness 0.02 mm, according to:

$$R_{cs} = 2\,P/\pi\,D\,t \qquad (1)$$

where P is the splitting load, D is the diagonal of the disc and t is the disc thickness.

Table 1. Composition of reinforcing glasses used for preparation of the BHAp-glass composites

Composite notation	Composition [wt.%]						
	SiO_2	Na_2O	K_2O	CaO	MgO	B_2O_3	Total
OK-50	46	26	-	-	-	28	100
OK-50(M3)	66	16	3	1	-	14	100
OK-57	56	21	1.5	0.5	-	21	100
OK-85	70.4	29.6	-	-	-	-	100

Table 2. Composition, sintering condition and some parameters of the BHAp-glass composites

Composite notation	BHAp [wt.%]	Sintering temperature T_2 [°C]	Portion and type of additive [wt.%]	Porosity (±1.3) [%]	Relative density (±0.04) [g/cm³]	Magnetic susceptibility $\chi \cdot 10^6$ [cm³/g]
OK-50	50	780	-	40	1.63	0.25–0.48
			2%, Fe_3O_4	40	1.64	0.25–1.25
OK-50(M3)	50	780	-	45	1.49	0.20–0.41
			2%, Fe_3O_4	41	1.60	0.24–1.18
OK-57	57	500 (vacuum)	-	50	1.34	-
			1%, Fe_3O_4	50	1.34	3000.00
			1%, Fe	53	1.34	2000.00
	57	800 (vacuum)	-	54	1.36	-
			1%, Fe_3O_4	54	1.33	-
			1%, Fe	54	1.36	858.00
OK-85	85	500	-	60	1.21	-
			2%, Fe_3O_4	powder	-	270.00
			2%, Fe	powder	-	1300.00

The fracture (splitting) of the specimen occurs along the diagonal. Strength properties were studied using the Instron system with a rate of 0.2 mm/min. Indentation fracture toughness K_{IC} was determined by means of direct crack measurements. The relationship between the fracture toughness and the size ratio crack/indent with the known Young's modulus exhibited a universal behaviour. The median/radial type of cracks with $c/a \geq 2.5$ can be described according to:

$$(K_{IC} \varphi/H a^{1/2}) (H/E\varphi)^{2/5} = 0.129 (c/a)^{-3/2} \qquad (2)$$

where K_{IC} is the critical stress intensity factor, H is the Vickers hardness, E is the Young modulus, a is the half-diagonal of the indent, c is the sum of the later and the crack length and φ is the constrain factor. The right side of this equation is equal to *0.035 (c/a)$^{-1/2}$* for cracks of Palmqvist type. Young's modulus E, Poisson's ratio and shear modulus G were evaluated using measurements of the propagation rate of longitudinal and crosswise ultrasonic waves. Microhardness was measured in micro-areas at a load of 25 G (HV_{025}).

To evaluate the bioactivity of the investigated composites, they were immersed at 36–37°C in medical isotonic physiological solution (0.9% NaCl) and human blood plasma with a solid/liquid ratio of 1 g/30 ml. A change in pH of physiological solution was measured over soaking time from 5 to 120 min. To evaluate the influence of the additional magnetization of some composites on their bioactivity, they were exposed for 30 sec to the magnetostatic field (600 Oe).

III. Results and Discussion

In accordance with the chemical analysis of composites, the calcium and silicon contents corresponded to the calculated ones with an error of ±0.5 wt.%. The XRD pattern showed a crystalline structure, in which the major phase was $Ca_{10}(PO_4)_6(OH)_2$.

An example of SEM micrograph of the investigated specimen surfaces and distribution of Ca, P, Si, and Fe is presented in Fig. 1. It indicates on uniform distribution of calcium, phosphorus and silicon, but not-uniform distribution of iron ions, which are partly in the form of large agglomerates (up to ~20 μm).

A typical IR transmission spectrum of the investigated composites is presented in Fig. 2. The spectral positions of the basic absorption bands for the BHAp and some composites are given in Table 3. The basic IR absorption bands are related to the vibrations of $(PO_4)^{3-}$ and $(OH)^-$ complexes. The absorption band 6 near 1050 cm^{-1} changes most of all. It is related to $(PO_4)^{3-}$ complexes and to intense absorption of Si-O complexes in the glass phase. The presence of iron oxides in the investigated specimens is determined by the band 10 near 1629.3 cm^{-1}. It was shown that the displacement of the IR-absorption bands of the composites with Fe and Fe_3O_4 additives in relation to ones without them is in the limits for the other specimens. The presence of some other absorption bands, for example the bands 8 and 9 (Table 3), indicate on the presence of admixtures remained in the initial BHAp after the removal of the bone organic constituent.
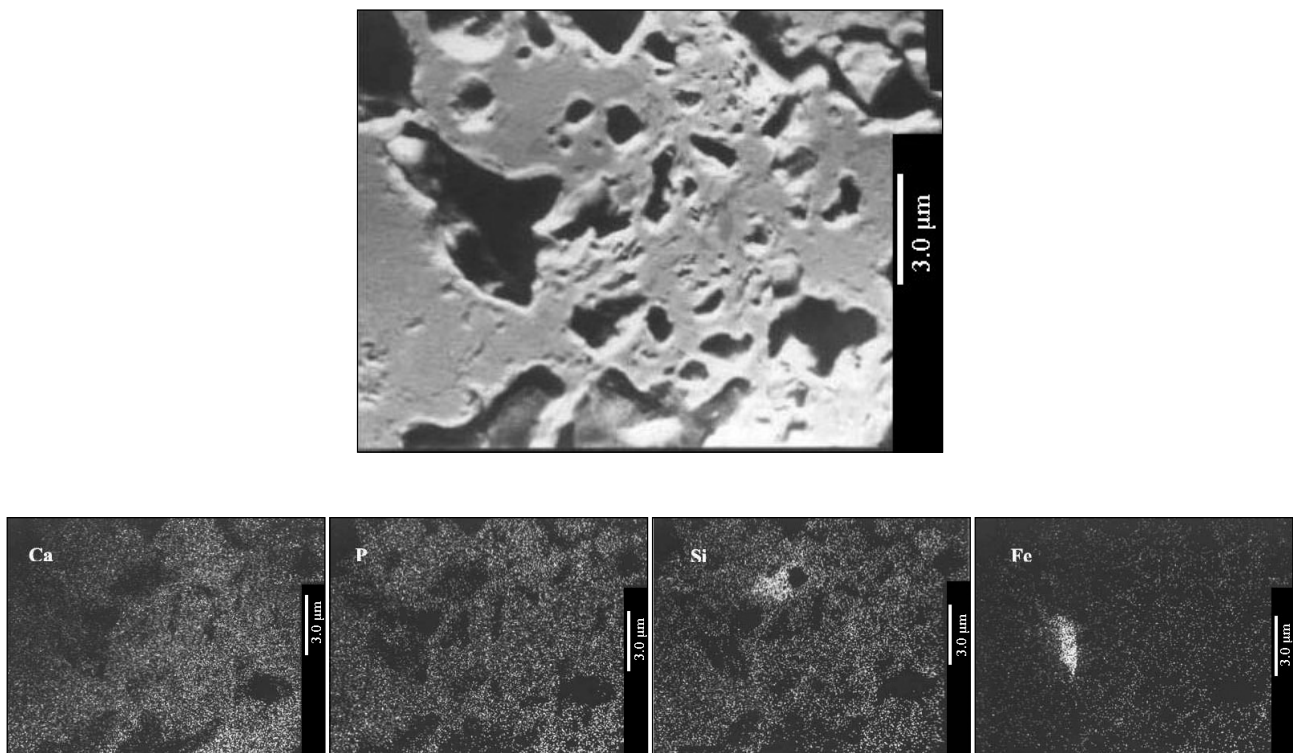

Figure 1. SEM micrograph and element distribution for the OK-50 with 2 wt.% Fe₃O₄ composite
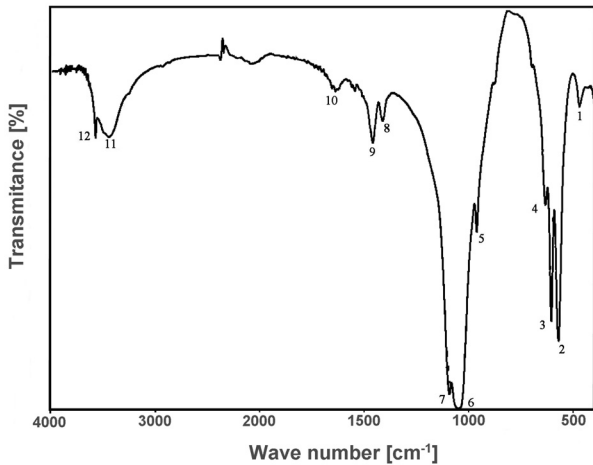
Figure 2. IR transmission spectrum of the OK 57 (1 wt.% Fe) composite

Selected mechanical properties of the tested materials are presented in Table 4. It can be seen that microhardness is in the range of 150–542 HV. The addition of 2 wt.% Fe_3O_4 to the OK 50(M3) resulted in increase of microhardness. Within the precipitates of the Fe_3O_4 phase observed in that sample the microhardness reaches 542 HV. Compressive splitting strength of the OK-50 and OK-50(M3) specimens (measured along the sample diameter) is within a range of 3.6–5.0 MPa. The OK-

57 composites sintered in vacuum showed low compression splitting strength (<1 MPa). Modulus of rigidity and Poisson's ratio are in ranges of 7.9–9.7 GPa and 0.252–0.307, respectively. The composite compressive strength is in the range 63–126 MPa and depends on a specimen porosity and treatment with the physiological solution (Table 4). The ferromagnetic additions lead to increase in compressive strength of the untreated samples (before soaking), but its decrease for the samples soaked in the physiological solution.

For all the specimens, the indentation fracture toughness K_{IC}, is within the range 0.6–1.0 MPa m$^{1/2}$. Mechanical properties of the sample OK-50 with 2 wt.% Fe_3O_4 are somewhat different from the other tested materials. This specimen exhibits the highest Young's modulus and relative density (Table 2), but its compressive splitting strength (R_{cs}=3.6 MPa) and Poisson's ratio (v=0.25) are lower. This is apparently related to its higher porosity, which makes walls of the pores thinner and proner to the fracture.

To evaluate magnetic susceptibility of the composites with Fe and Fe_3O_4 additives, hysteresis loops were determined (Fig. 3). The OK-57 composites with 1 wt.% Fe or Fe_3O_4 powders sintered at 500°C in vacuum have a magnetic susceptibility of 2–3·10^{-3} cm^3/g (Table 2). This parameter decreases to 0.858·10^{-3} cm^3/g for the OK-57 specimens with 1 wt.% additives sintered at 800°C in vacuum

Table 3. Spectral positions of basic IR absorption bands of BHAp composites

Band No.*	BHAp	OK-50 OK-57	OK-50(M3) +2% Fe_3O_4	OK-57 +1% Fe_3O_4	OK-57 +1% Fe	Fe_3O_4	Complex
	Wave number of IR absorption band [cm^{-1}] (±0.5 cm^{-1})						
1	473.2	473	469.1	472.3	472.3	-	$[PO_4]^{3-}$
2	569.2	570	570.1	569.5	569.9	567.6	$[PO_4]^{3-}$, Fe–O
3	604.0	604	603.1	603.8	603.6	-	$[PO_4]^{3-}$
4	632.4	633	~630	632.6	632.4	-	$[OH]^-$
5	961.8	961	~960	961.2	961.4	-	$[PO_4]^{3-}$
6	1050	1047	1048.4	1045.9	1047	-	$[PO_4]^{3-}$, Si-O
7	1090	1090	~1090	1090.1	1090.4	-	$[PO_4]^{3-}$
8	1411.8	-	~1411	1412.1	1411.5	-	C–O ,C–N
9	1456.9	1457.2	~1460	1458.2	1459.0	-	C–O ,C–N
10	1634.3	1633.8	1631.9	1637.6	1629.9	1629.3	Fe–O
11	3421.8	3433	3422.0	3437.4	3431.7	3427.3	H_2O
12	3571.7	3570	~3570	3570.2	3570.5	-	$[OH]^-$

* See numbers in Fig. 2

Table 4. Mechanical properties of BHAp composites

Composite notation	Portion and type of additive [wt.%]	Compressive splitting strength (±0.5) [MPa]	Young modulus E (±0.5) [GPa]	Microhardness HV$_{.025}$	Compressive strength (±1) [MPa]	
					Before soaking	After soaking
OK-50	-	5.0	23	240–445	70	121
OK-50	2%, Fe_3O_4	3.6	25	150–460	115	63
OK-50(M3)	-	-	-	220–508	112	126
OK-50(M3)	2%, Fe_3O_4	-	-	260–542	122	104

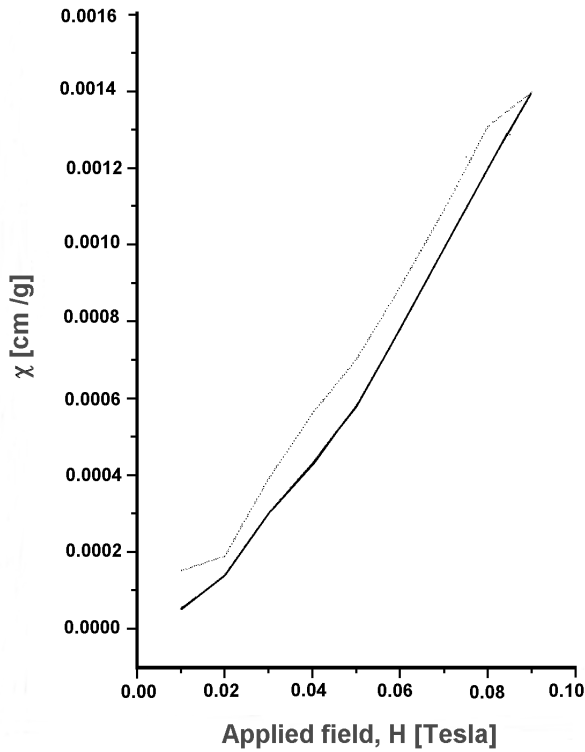**Figure 3. Hysteresis loop for specimen OK-85 with 2% Fe**

and to $0.27-1.3 \cdot 10^{-3}$ cm³/g for the OK-85 specimens with 2 wt.% additives sintered at 500°C under the usual atmospheric conditions. The magnetic susceptibility for the OK-50 and OK-50(M3) specimens sintered at 780°C under the same atmospheric conditions is only $0.20-1.25 \cdot 10^{-6}$ cm³/g (Table 2). The OK-50 and OK-50(M3) composites revealed rather low susceptibility, probably due to the small volume fraction of the magnetic powder and to the partial oxidation of Fe_3O_4 (Table 2). The magnetite susceptibility values of the OK-50 and OK-50(M3) specimens with 2 wt.% Fe_3O_4 are correlated. A nonuniform distribution of iron ions in the investigated specimens leads to a certain spread of their magnetic susceptibility values. In the composites without additives, there is also certain amount of iron (0.02–0.20 wt.%) originated from BHAp, which conditions their magnetic susceptibility (Table 2).

The comparison of mass losses in the composites with or without additives in both blood plasma and physiological solution indicates the influence of the iron ion presence on the degradation rate (Figs. 4 and 5, Table 5). It follows from Fig. 4 that the presence of iron ions in the OK-57 specimens increases their degradation rate in the initial stage (for 40 min) of soaking in the physiological solution. The comparison of the OK-50 and OK-50 (M3) specimens with or without addi-
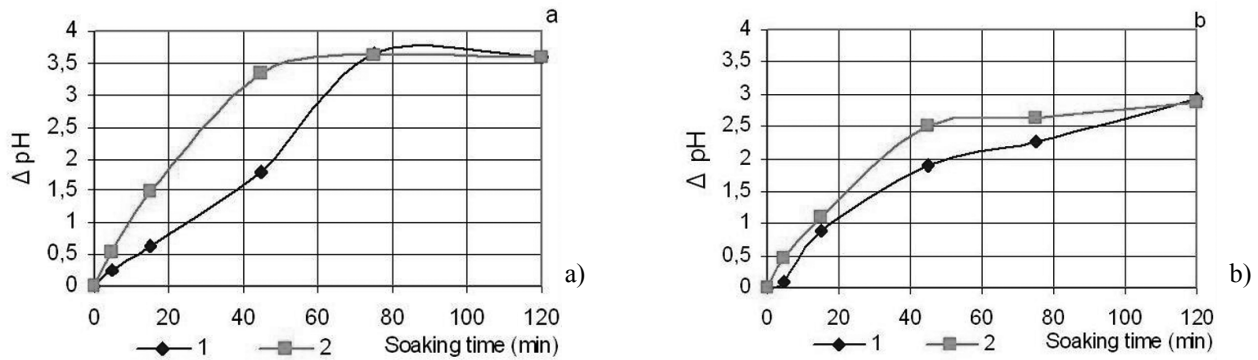

Figure 4. Dependence of the physiological solution pH change on the soaking time for OK-57 (a) and OK-57 with 1 wt.% Fe_3O_4 (b) composites without (1) and with (2) magnetic processing
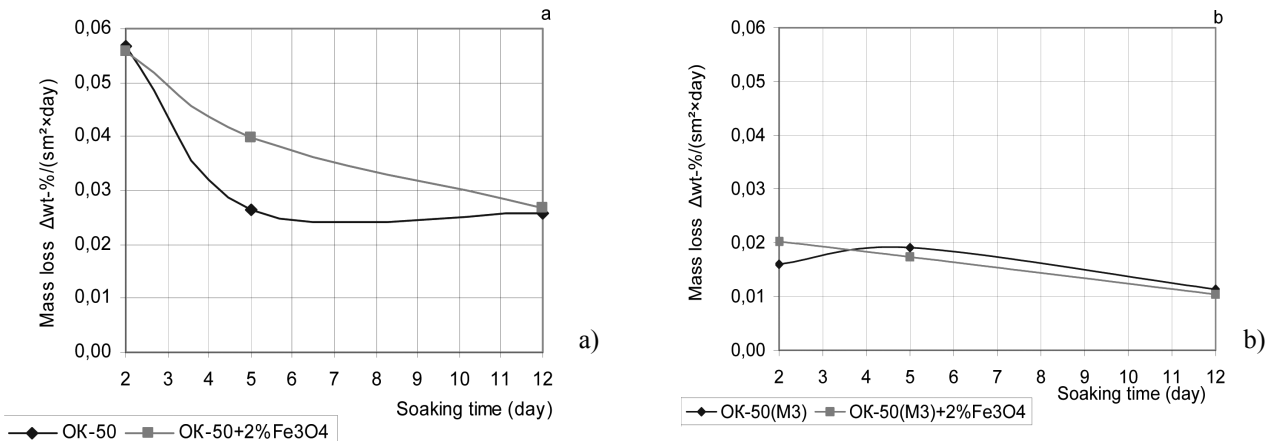

Figure 5. Degradation of OK-50 (a) and OK-50 (M3) (b) composites with (▪) and without (♦) additives over the soaking time in the physiological solution

Table 5. Some results of experiments *in vitro*

Composite notation	Sintering temperature T_2 [°C]	Portion and type of additive [wt.%]	Mass loss, ±0.005 Δ mass [%/(cm²·day)]	
			2 days	5 days
OK-50	780	-	0.07*(3 days)	0.10*
OK-50	780	2%, Fe_3O_4	0.10*(3 days)	0.13*
OK-57	500 (vacuum)	-	0.19	-
		1%, Fe_3O_4	0.25	-
		1%, Fe	0.23	-
	500 (vacuum)*	-	0.20	-
		1%, Fe_3O_4	0.25	-
		1%, Fe	0,09	-
	800 (vacuum)	-	0.07	0.08
		1%, Fe_3O_4	0.06	0.09
		1%, Fe	0.04	0.06
	800 (vacuum)*	-	0.10	0.08
		1%, Fe_3O_4	0.05	0.08
		1%, Fe	0.05	0.05

*magnetic processing

tives indicates that degradation after 2–3 day soaking of the OK-50 composite with additives is different from that of the composite without Fe_3O_4 (Fig. 5).

It was established that the degradation rate of the OK-57 composites without additives processed by magnetic field before their immersion in the physiological solution increased within the first 40 min of soaking (Fig. 4a). Similar changes in the degradation rate were observed for the OK-57 specimens with 1 wt.% Fe_3O_4 (Fig. 4b). Analogous results were also obtained for the OK-50 specimens with 2 wt.% Fe_3O_4. The composite degradation in the physiological solution during more prolonged soaking weakly depends on treatment by magnetic field (Table 5). The above features may be related to the effect of the local magnetic field of composite ferromagnetic additives on their bioactivity *in vitro*. Short-term treatment by external magnetic field of composites containing ferromagnetic ions strengthens their internal magnetic field.

Addition of ferromagnetic ions to an implant based on the BHAp-glass biomaterials can increase the bone regeneration rate *in vivo*. The short-term processing of patient's operated section with a weak magnetic field can strengthen this effect both at the initial stage after implantation and over prolonged time.

IV. Conclusions

Biogenic hydroxyapatite-glass composites, with addition of up to 2 wt.% of Fe or Fe_3O_4, were fabricated from biogenic hydroxyapatite powder and different reinforcing glasses by sintering under different conditions. Sintering of BHAp-glass composites with ferromagnetic additives at ≤ 500°C under the usual atmospheric conditions preserves high enough values of the

magnetic susceptibility up to $1.3 \cdot 10^{-3}$ cm³/g. Sintering them in vacuum makes it possible to increase magnetic susceptibility, but their mechanical strength is considerably decreased. The presence of ferromagnetic additives leads to an increase in the degradation rate *in vitro* of composites with both high and low magnetic susceptibility within the first 40 min of soaking. Short-term processing of the composites (with addition of Fe or Fe_3O_4) with a magnetic field leads to increase in their initial degradation rate.

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