

# Strengthening of Deformable Bone Cements in the Calcium Phosphates–Chitosan System by Tricalcium Phosphate Granules

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**Abstract**—The possibility of strengthening chitosan-containing calcium phosphate bone cements sintered with tricalcium phosphate granules was studied. The strength was found to depend on the granules content in a nonmonotonic manner. For materials reinforced with 300–500  $\mu\text{m}$  granules in the amount of 20 wt %, the compression strength of about 15 MPa was reached on day 14 of air hardening, which was 50% higher than that of the unreinforced cement matrix. The main crystalline phase of the cement matrix is amorphous calcium phosphate. Dissolution of cement in isotonic saline corresponds to first-order reaction kinetics.

**Keywords:** biomaterials, bone cements, reinforcement, strength

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## INTRODUCTION

Calcium phosphate bone cements (CPCs) are designed for plastic repair of bone tissue defects [1, 2]. They consist of solid (powder) and liquid components upon mixing of which the cement paste undergoes setting and hardening as a result of chemical reactions occurring in the system to form a cement stone. The disadvantage of CPCs is the fragility of the hardened material and, as a consequence, its low strain capacity and operational reliability. To increase the deformability, it has been proposed to add a flexible polymer to the CPC composition where such polymer forms a continuous framework providing the cement with the necessary deformability [3–5]. The use of chitosan as a polymeric constituent provided the cement based on amorphous calcium phosphate (ACP) with flexibility and allowed increasing considerably the material deformability: the compression strain after holding in physiological fluid reached 25–30% [6, 7]. The process of the designed cement is based on the change in solubility as a function of the solvent pH and chitosan molecular weight, namely, on a dramatic decrease in the solubilities of high-molecular-weight chitosans upon pH increase from a neutral to alkaline value [8]. In [6, 7], the pH was changed by addition of tetracalcium phosphate (TeCP) to the charge stock after dissolution of chitosan in cement liquid (CL) and its mixing with ACP.

Another significant disadvantage of the designed cement is its low strength. The increase in the strength of known CPCs was achieved by reinforcement with polymer, metal, and ceramic particles and fibers [9–12]. The problem of strengthening highly strained cements in the calcium phosphates–chitosan system with a con-

tinuous polymeric framework still remains unstudied. In particular, it is not understood how the reinforcement can influence the most interesting characteristic of these composite materials, namely, their high ultimate strain to fracture. It is important to maintain biocompatibility of the material; therefore, it is reasonable to carry out the reinforcement using biocompatible granules of calcium phosphate, in particular, tricalcium phosphate (TCP). The aim of the present work was to study the effect of ceramic TCP granules reinforcement of deformable chitosan-containing CPCs on their strength and deformability.

## EXPERIMENTAL

The powdery component of CPC consisted of ACP and TeCP in the ratio of 1 : 1.3 [6]. Ceramic TCP granules with a size of 100–200 and 300–500  $\mu\text{m}$  were added to the ACP powder in the amount of 10, 20, or 30 wt %. The granules were obtained by dispersing a suspension of powdery TCP in gelatin onto drops followed by thermal treatment at 1300°C [13]. CL was added to the resulting mixture and, then, a TeCP powder was added to change the pH to neutral, which resulted in solidification of chitosan to form a framework. The cement liquid was a 3% solution of high-molecular-weight chitosan (500 kDa) in orthophosphoric acid. The solid/liquid ratio in CPC was 1 : 1, since at a lower CL amount we failed to obtain a homogeneous cement paste and at a higher CL content the cement paste had a very thin consistency and the setting time was more than several hours. Hardening occurred in air and upon immersion in a solution simulating extracellular fluid (SBF). The composition of SBF (g/L): NaCl 6.457;  $\text{NaHCO}_3$  2.268; KCl

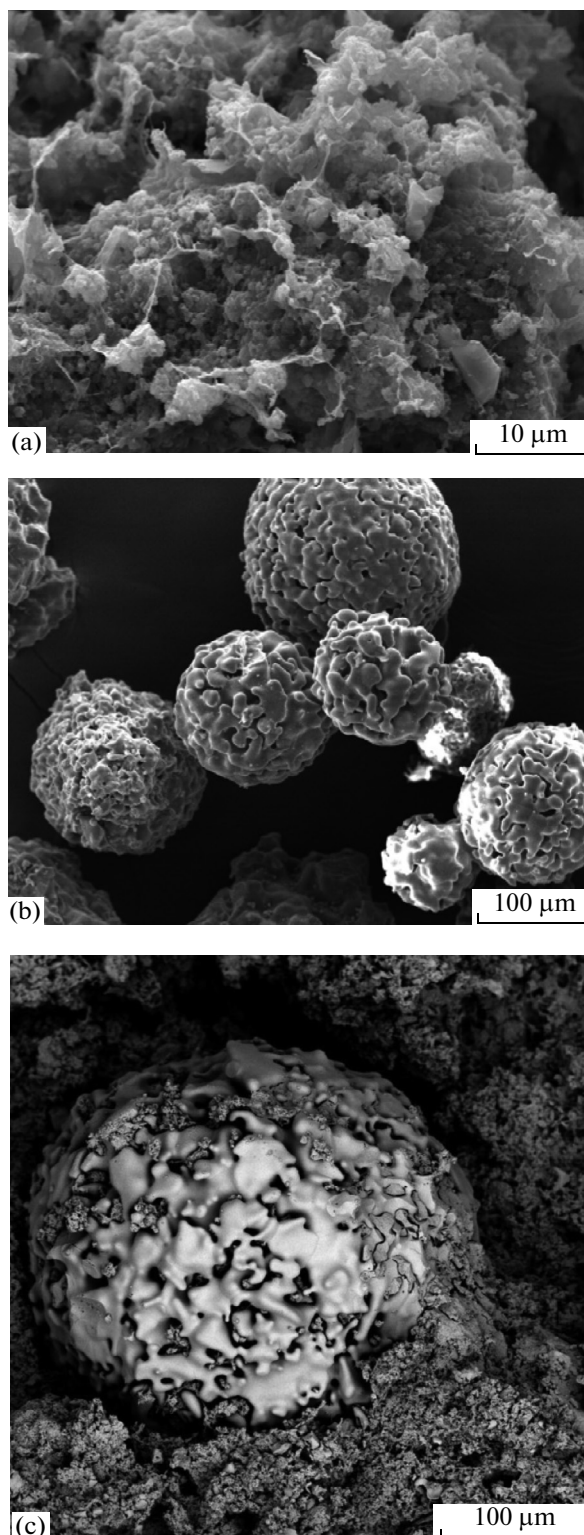
0.373;  $\text{Na}_2\text{HPO}_4 \cdot 12\text{H}_2\text{O}$  0.178;  $\text{MgCl}_2 \cdot 6\text{H}_2\text{O}$  0.305;  $\text{Na}_2\text{SO}_4$  0.071;  $\text{CaCl}_2 \cdot 2\text{H}_2\text{O}$  0.368; Tris buffer 6.057.

The samples were studied by scanning electron microscopy (SEM, TeScan Vega II microscope) and X-ray diffraction (XRD, Shimadzu XRD-6000 diffractometer). Mechanical tests were performed upon axial compression of cylindrical test pieces on an Instron 5581 testing machine. Solubility tests were performed according to GOST R ISO 10993-14—2001 measuring the calcium ion content in solution. The samples were kept in normal saline (0.1 M NaCl) for 28 days at constant volume of the liquid phase (closed system), pH 7.4, and 37°C. The calcium concentration in the liquid phase was measured using an Optima inductively coupled plasma atomic emission spectrometer. The measurement error was 0.01 mg/L. The setting time was determined on a Vicat apparatus at the load of 100 g on needle.

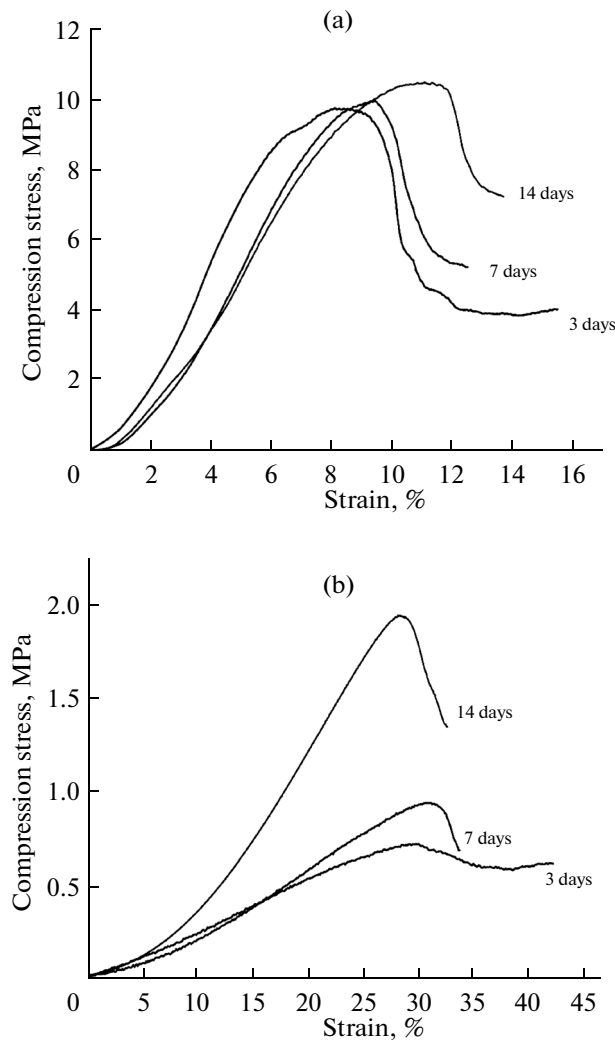
## RESULTS AND DISCUSSION

The setting time of the cement paste was from 30 min to 2 h. The initial pH of the system was 7.1 and increased to 7.9 by day 7. According the XRD data, the single crystalline phase in the product of reaction in SBF on day 14 was the  $\text{Ca}_3(\text{PO}_4)_2 \cdot n\text{H}_2\text{O}$  phase, whereas, after exposure to air, the reaction product contained a considerable amount of unreacted TeCP. Figure 1 shows the SEM images for cement matrix, TCP granules, and fracture surface of CPC reinforced with TCP granules. The matrix is porous and has a pore size up to 20  $\mu\text{m}$ . One can see chitosan as films interlinking the phosphate particles (see Fig. 1a). The TCP granules mainly have a nearly spherical shape (see Fig. 1b), although some granules have an arbitrary shape. The granules are also porous, but the particles within granules are sintered to each other. Figure 1c shows the fracture surface of the material reinforced with 300–500  $\mu\text{m}$  granules. The fracture occurs along the porous matrix and at the interface between the matrix and granules.

Figure 2 shows the stress-strain diagrams upon compression of CPC samples containing 20 wt % of TCP granules with a size of 100–200  $\mu\text{m}$  hardened in air (see Fig. 2a) or in SBF (see Fig. 2b) for the time up to 14 days. The strength increases with increasing hardening time. The air-hardened material containing no TCP granules has a compression strength of about 8 MPa on day 14 of hardening [7]. The addition of ceramic TCP granules increases the cement strength. The dependence of strength on the reinforcing TCP granules is nonuniform: the maximum values are reached at the granule content of 20 wt % (Fig. 3). The value of the achieved strengthening effect is influenced by the granule size: reinforcement with 300–500  $\mu\text{m}$  granules results in a higher strength (up to 15 MPa on day 14 of air hardening, see Fig. 3c) compared to reinforcement with 100–200  $\mu\text{m}$  granules (up to 11 MPa, see Fig. 3a).



**Fig. 1.** SEM images for cement matrix (a), TCP granules (b), and fracture surface of composite materials reinforced with TCP granules (c).

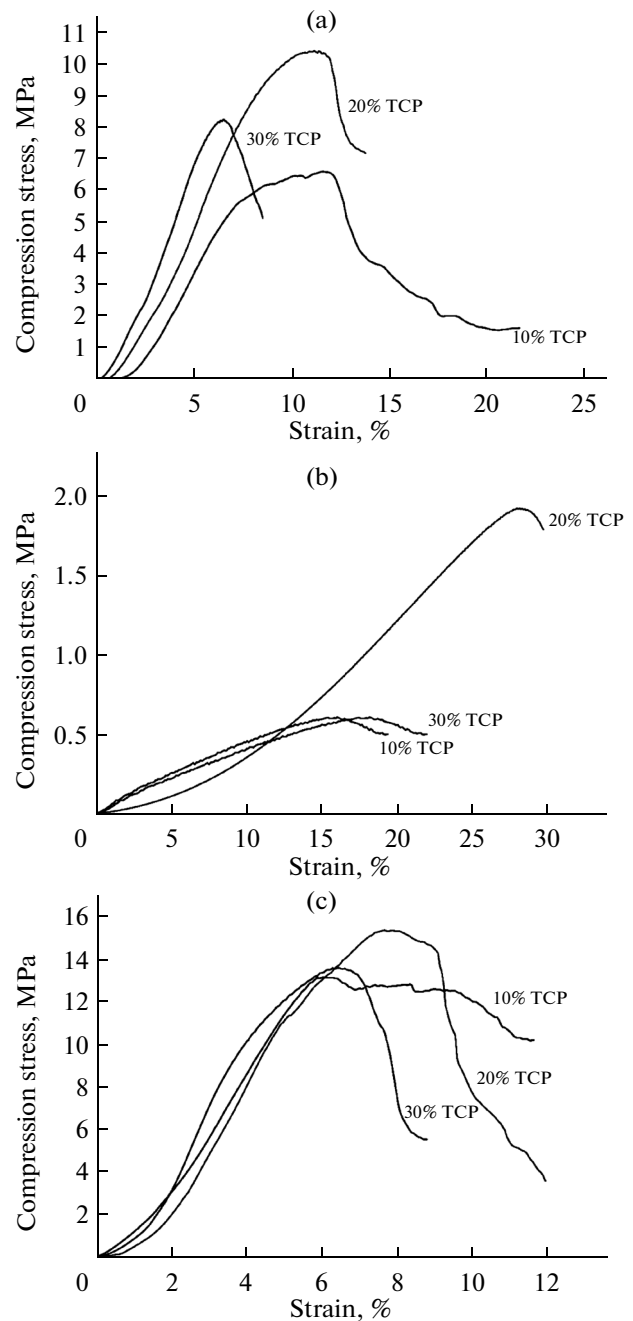


**Fig. 2.** Stress-strain diagrams for CPCs reinforced with 100–200  $\mu\text{m}$  TCP granules in the amount of 20 wt %: (a) air hardening and (b) hardening in SBF.

The magnitudes of strength after hardening in SBF were significantly less (see Figs. 2b and 3b), but the relative effect of strengthening was as high as after air hardening. The maximum strength for unreinforced cements after hardening in SBF was 1.2 MPa [7]. For the same cements containing 20 wt % of 300–500  $\mu\text{m}$  granules, the strength reached 1.8 MPa.

The strain-to-fracture for all reinforced air-hardened cements was 8–12% and that for the cements hardened in SBF was 15–30%, which corresponded to the ultimate strain of unreinforced cements [7]. No unambiguous relationship between the strain to fracture and the sample strength was revealed. A possible reason for the increase in deformability in SBF can be a less fragile state of chitosan after holding in the liquid.

Ceramic materials are known to be stronger than the cement ones of the same composition. This is due to the fact that ceramic materials have stronger bonds between particles along the boundaries formed upon



**Fig. 3.** Stress-strain diagrams for CPCs reinforced with TCP granules on day 14 of hardening at different granule content: (a) in air, granule size of 100–200  $\mu\text{m}$ ; (b) in SBF, granule size of 100–200  $\mu\text{m}$ ; and (c) in air, granule size of 300–500  $\mu\text{m}$ .

sintering. Taking into account the concepts of brittle fracture mechanics, one can assume that the increase in the strength of composite cements as a result of reinforcement is mainly caused by a higher fracture resistance of added ceramic TCP granules compared to that of cement matrix. Therefore, a crack propagating upon fracture envelops the granules (see Fig. 1c), which results in a change in the fracture mode from normal frac-

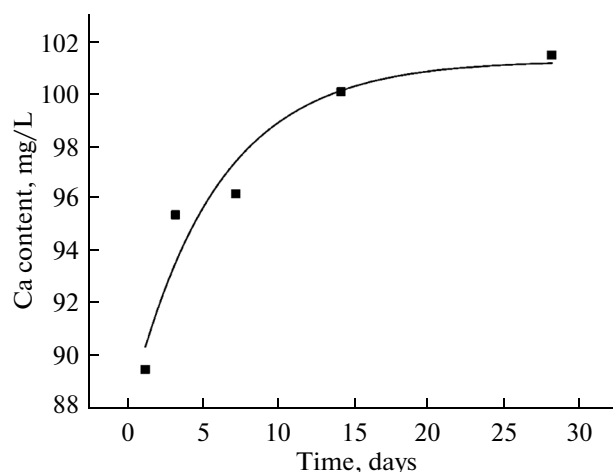


Fig. 4. Kinetic curve for the change in the calcium ion concentration in isotonic saline upon holding therein of CPCs reinforced with TCP granules.

ture to shear requiring higher forces owing to the increase in the crack tip critical stress intensity factor [14].

Figure 4 shows the kinetic curves for the change in the calcium ion concentration in solution upon holding of CPCs reinforced with TCP granules in isotonic saline. The kinetic law can be described by an exponential function, which corresponds to first-order reaction kinetics (the rate of concentration change is proportional to its current value). The comparison with the kinetic curve for the calcium concentration change upon holding of unreinforced cement in isotonic saline results in the conclusion of that the rate of reinforced cement dissolution is defined by the rate of matrix dissolution.

### CONCLUSIONS

Thus, the studies performed showed that the strength of highly strained CPC based on  $\text{Ca}_3(\text{PO}_4)_2 \cdot n\text{H}_2\text{O}$  and chitosan increases upon addition of TCP granules. At the optimum 20 wt % content of granules with a size of 300–500  $\mu\text{m}$ , the strength of 15 MPa was achieved after air hardening for 14 days. We assume that an additional reserve of the increase in the strength of reinforced CPCs consists in a decrease in the porosity of the cement matrix, which can be achieved by optimization of the solid/liquid ratio and particle sizes of components. Cement materials possessing a high deformability are promising for application in bone tissue engineering as matrices providing the vital activity of osteogenic cells.

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