

Synthesis, characterization and antibacterial properties of modified calcium phosphates

Grynyuk I.I.¹, Vasyliuk O.M.², Strutynska N.Yu.³, Slobodyanik M.S.³

¹Igor Sikorsky Kyiv Polytechnic Institute, Peremohy av., 37, Kyiv-03056, Ukraine, e-mail: igrnyuk@ukr.net

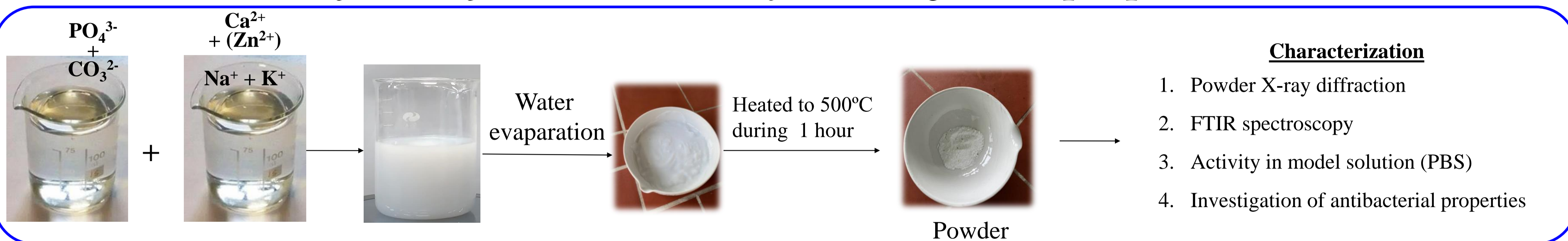
²Zabolotny Institute of Microbiology and Virology, National Academy of Science of Ukraine, Zabolotnogo str. 154, Kyiv-03143, Ukraine

³Taras Shevchenko National University of Kyiv, Volodymyrska Str. 64/13, Kyiv-01601, Ukraine



Apatite ($\text{Ca}_{10}(\text{PO}_4)_6(\text{OH})_2$) and whitlockite ($\beta\text{-Ca}_3(\text{PO}_4)_2$) related calcium phosphates due to their biocompatibility and bioactivity are widely used in orthopedics and dentistry for bone restoration. At the same time, the biphasic calcium phosphates (mixture of apatite and whitlockite-type phases) have also significant interest due to the possibility of regulation of material bioresorption by varying the weight ratios of their components. The main way for influence on functional properties of synthetic calcium phosphates is their chemical modification with cationic and anionic substitutions (mainly carbonate (CO_3^{2-}) and trace metals (Na^+ , K^+ , Zn^{2+})). The foreign ions change chemical and thermal stability, mechanical and biological properties of such synthetic materials. **The aim** of the study focuses on the synthesis of nanoparticles of calcium phosphates which contain of sodium or potassium cations as well as their combination with addition of Zn^{2+} and fixed amount of carbonate-anions, characterization of prepared phosphates using powder X-ray diffraction and FTIR spectroscopy methods and investigation of their antibacterial activity.

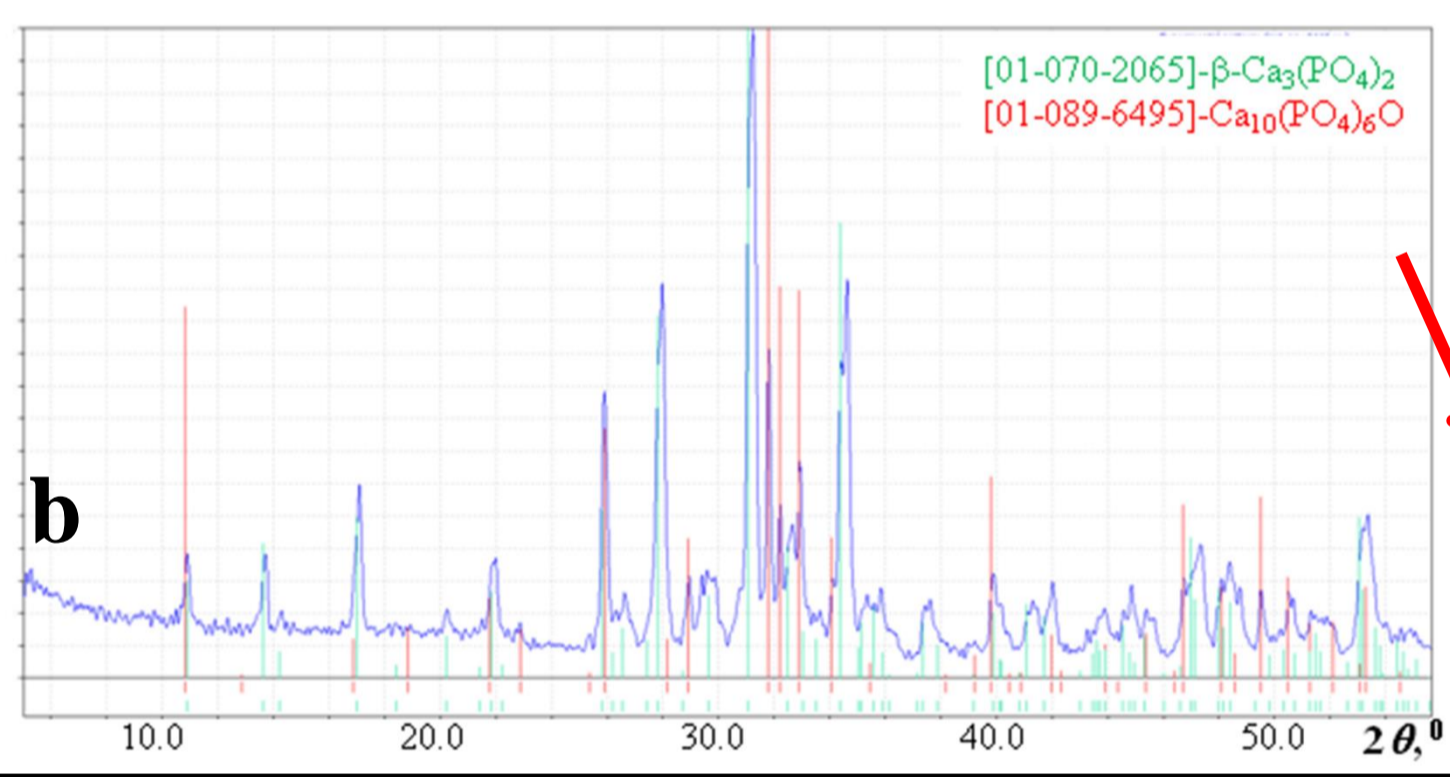
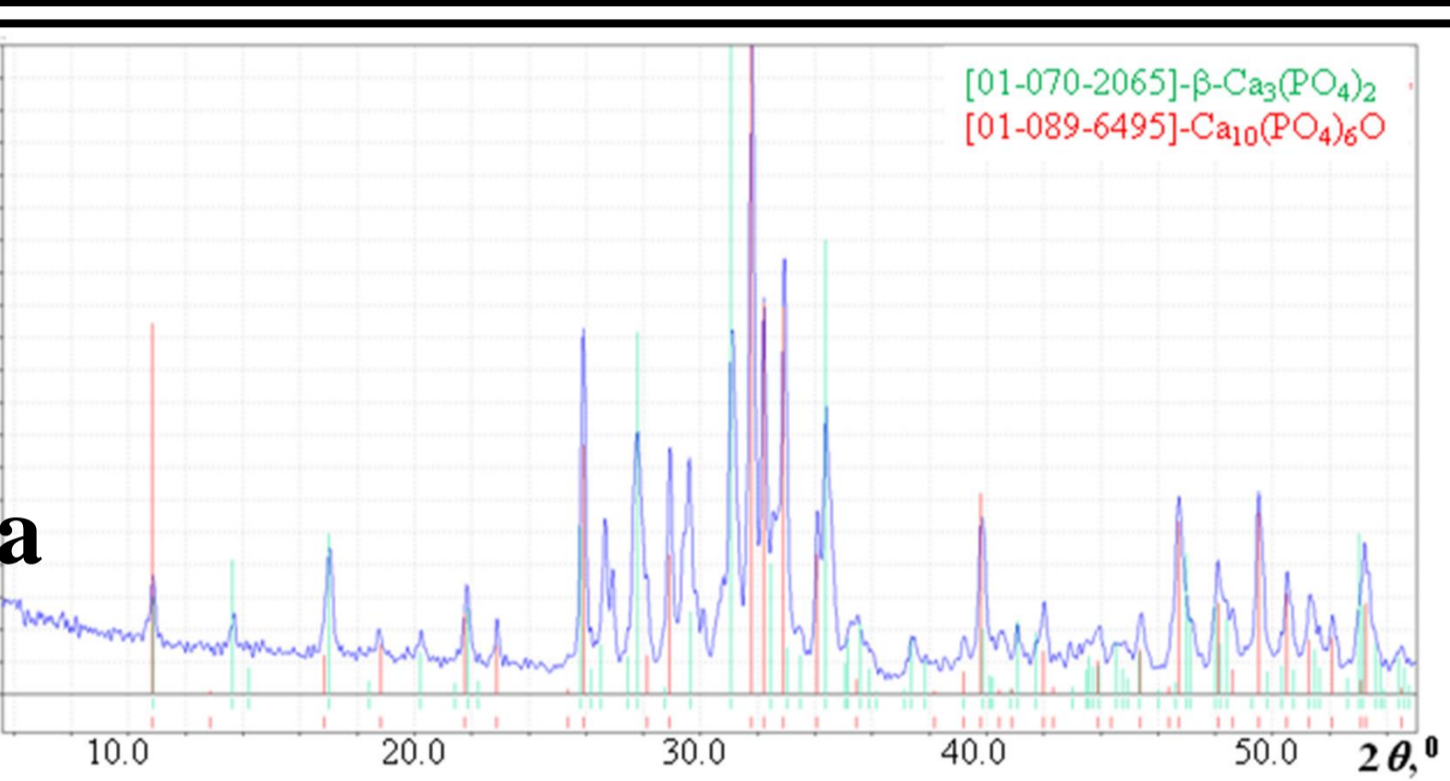
Synthesis of Na^+ , K^+ , Zn^{2+} , CO_3^{2-} -containing calcium phosphates



Characterization

1. Powder X-ray diffraction
2. FTIR spectroscopy
3. Activity in model solution (PBS)
4. Investigation of antibacterial properties

Characterization of obtained modified calcium phosphates



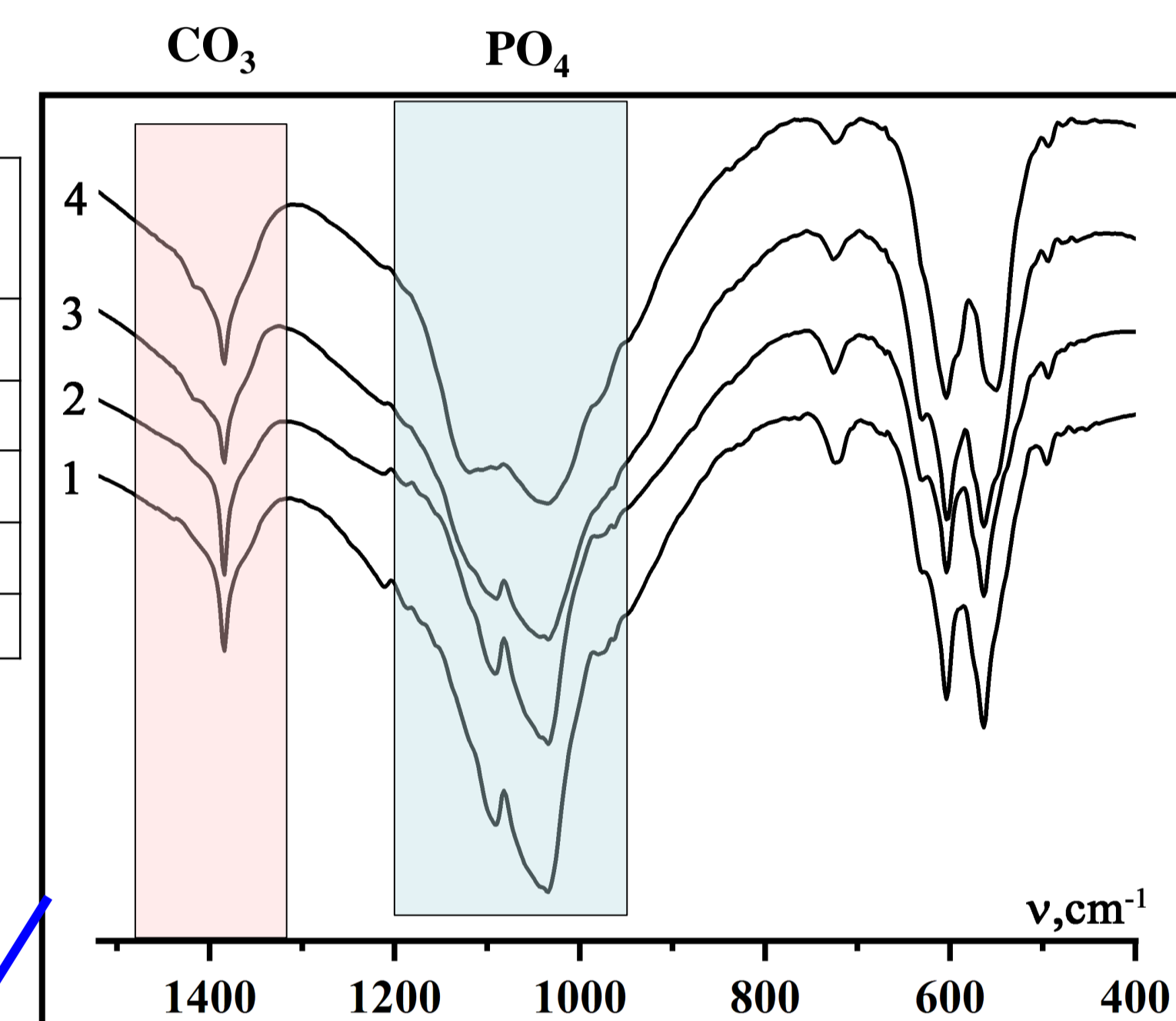
XRD patterns for prepared calcium phosphates: Sample 2 (a) and Sample 4 (b)

Elemental and phase compositions of the calcium phosphates prepared from aqueous solution at room temperature and then heated to 500 °C

Sample number	Elemental composition weight %				Phase type weight %	
	Na ⁺	K ⁺	Zn ²⁺	CO ₃ ²⁻	Ca ₁₀ (PO ₄) ₆ O	β-Ca ₃ (PO ₄) ₂
Sample 1	1.2	-	-	3.2	95	5
Sample 2	1.1	0.2	-		85	15
Sample 3	-	2	-		93	7
Sample 4	1.1	0.2	2.1		15	85

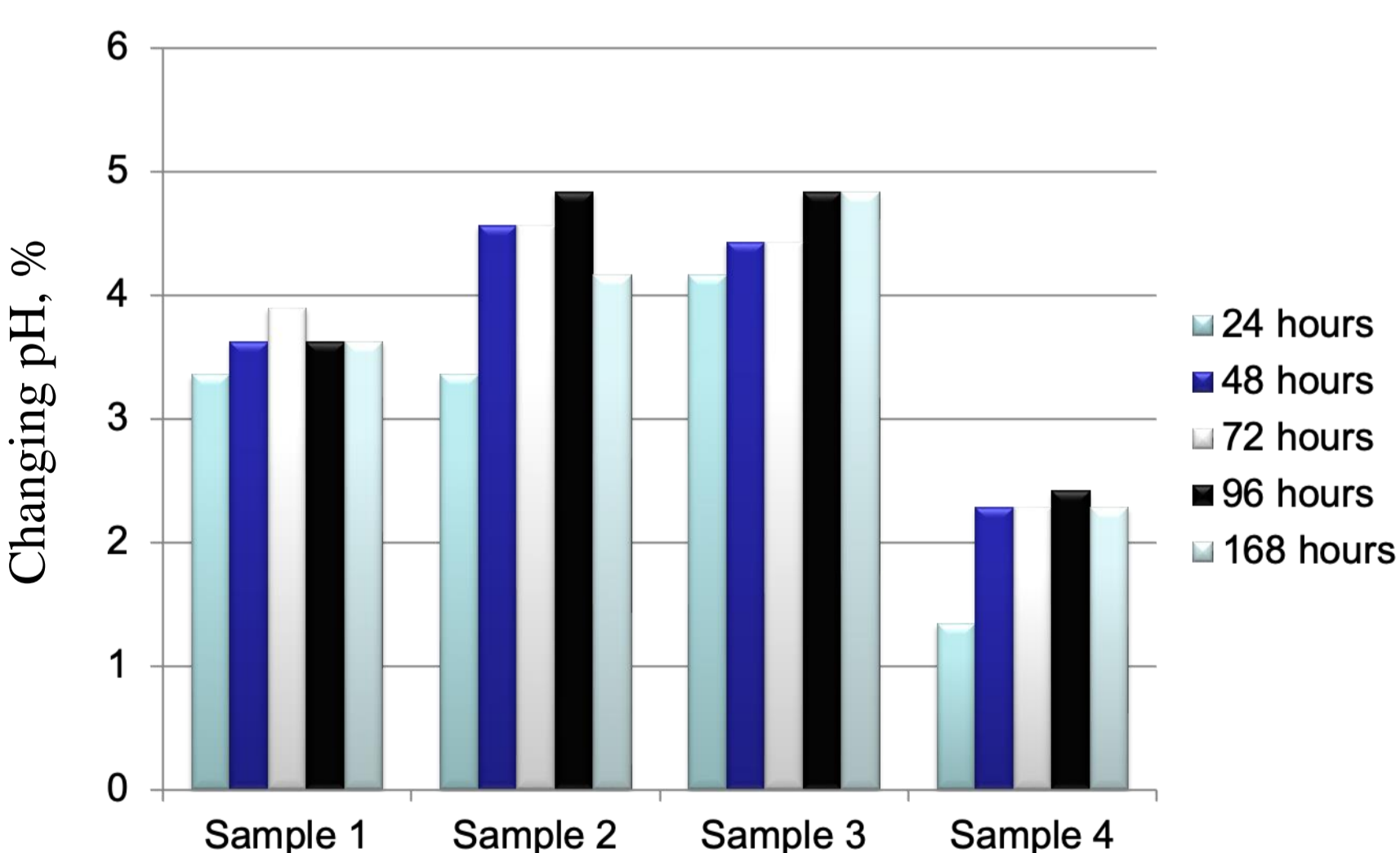
According to powder X-ray diffraction results the biphasic calcium phosphates with different ratios of the main components were obtained.

FTIR spectroscopy data confirm the presence of phosphate and carbonate anions in the composition of prepared modified calcium phosphates.



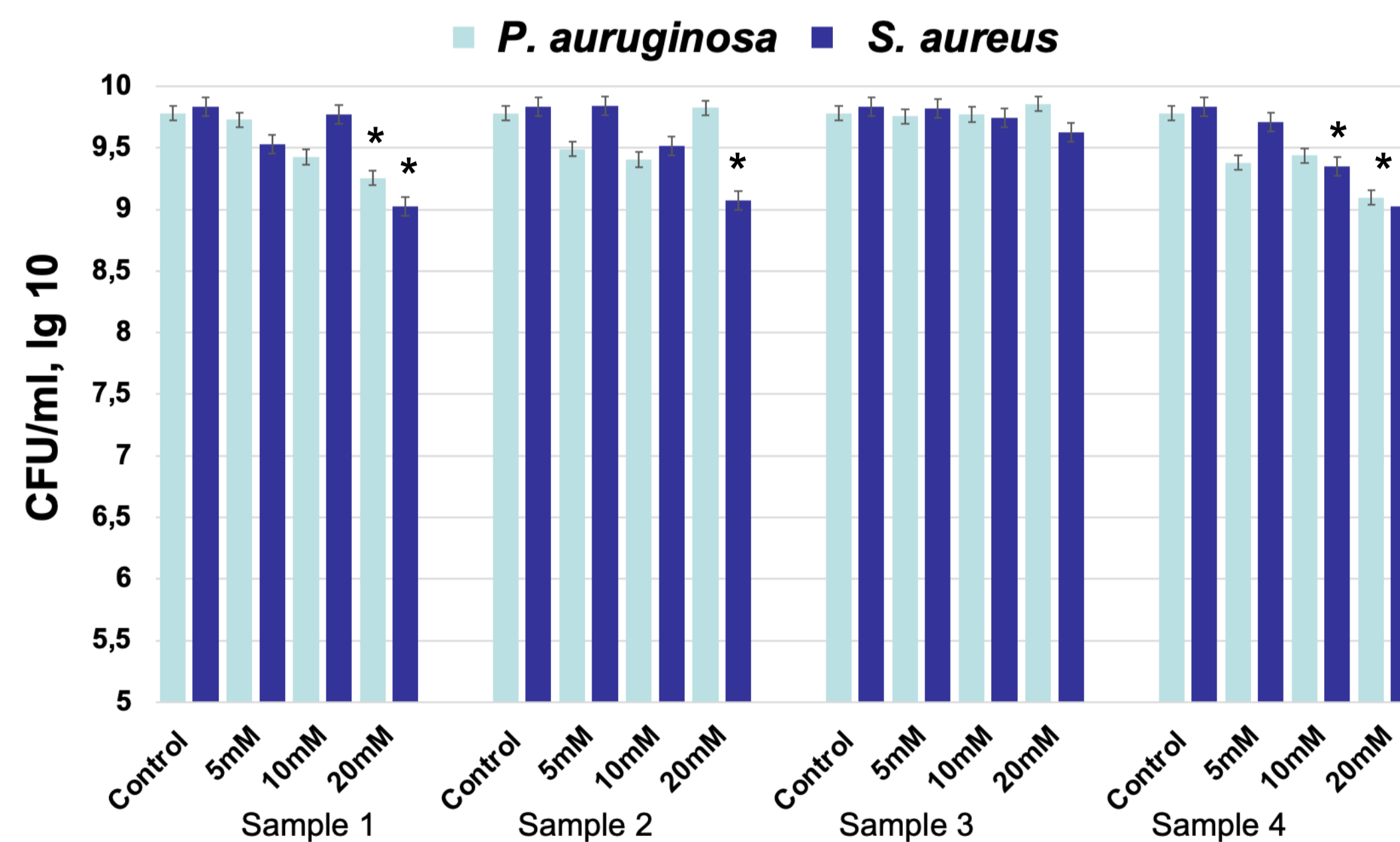
FTIR-spectra of prepared calcium phosphates: Sample 1 (curve 1), Sample 2 (curve 2), Sample 3 (curve 3), Sample 4 (curve 4)

Activity in model solution and antibacterial properties of modified calcium phosphates



Activity of prepared samples with different elemental and phase composition was determined in buffer solution at temperature 37 °C and pH = 7.45.

The changing of pH values was measurement after 24, 48, 72, 96 and 168 hours. The increase of pH values of solutions was observed for all samples. The most growing of pH to 7.8 was observed after 48 h for Sample 2 (Na^+ (1.1wt%), K^+ (0.2wt%), CO_3^{2-} (3.2wt%)-containing calcium phosphate).



The antibacterial activity of prepared samples with different elemental and phase composition against *Staphylococcus aureus* UCM IMV B-904 and *Pseudomonas aeruginosa* UCM IMV B-900 ($M \pm m$, n 4).

The antimicrobial effect of samples 1, 2 and 4 (20 mM) was established after 24 h, while sample 3 did not affect the growth of microorganisms. Samples 1 and 4 had inhibitory activity against the growth of both gram-positive (*S. aureus*) and gram-negative (*P. aeruginosa*) microorganisms in the amount of 20 mM. Sample 4 containing Zn^{2+} had the highest antimicrobial activity. Strain *S. aureus* were more sensitive to the effect of sample 4 in concentration 10 and 20 mM, while its noticeable effect on gram-negative bacteria *P. aeruginosa* was observed only at 20 mM of the sample.

Conclusions. Chemically modified biphasic calcium phosphates with different ratio of the main phases-type ($\text{Ca}_{10}(\text{PO}_4)_6\text{O}$ and $\beta\text{-Ca}_3(\text{PO}_4)_2$) have been prepared from an aqueous solution. FTIR spectroscopy data confirm the presence of phosphate and carbonate anions in the composition of prepared modified calcium phosphates. The antibacterial activity of obtained phosphates against *Staphylococcus aureus* UCM IMV B-904 and *Pseudomonas aeruginosa* UCM IMV B-900 have been evaluated. It was found that samples 1, 2 and 4 with concentration 20 mM have inhibitory effect on both strains *S. aureus* and *P. aeruginosa*. Sample 4 containing zinc had the highest antimicrobial effect against *Staphylococcus aureus*.