

INVESTIGATION THE STRUCTURAL INFLUENCES OF SILVER OXIDE ADDITION IN THE BIOACTIVE PHOSPHATE GLASSES[†]

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This research investigates the impact of varying concentrations of silver oxide on the structure and morphology of phosphate bioactive glass (PBG). PBGs are gaining popularity as a potential replacement for traditional silicate glasses in biomedical applications due to their adjustable chemical resistance and exceptional bioactivity. Upon examination of the scanning electron microscope of the composites without Ag₂O, it was observed that the grains tended to merge together, and the surface particles appeared to be larger than those in composites with Ag₂O at concentrations of 0.25, 0.5, and 0.75 wt%. The study found that the diffraction pattern of phosphate bioactive glass composites sintered without Ag₂O showed the presence of Strontium di-phosphate and Calcium di-phosphate. The XRD pattern of these composites without Ag₂O revealed specific planes that corresponded to both types of di-phosphate. However, when Ag₂O was added, a new cubic phase was detected, and the intensity of the calcium and strontium diphosphate increased with higher Ag₂O content. The XRD pattern of the composites with Ag₂O displayed specific planes that corresponded to Ag₂O. In other words, the absence of Ag₂O in the composite material led to larger particle sizes and less distinct boundaries between grains. In addition, it has been found that, as the concentration of Ag₂O increased from 0 to 0.25, 0.5, and 0.75 wt%, the average crystallite size decreased from 36.2 to 31.7, 31.0, and 32.8 nm, respectively. These results suggest that the addition of Ag₂O can effectively reduce the average crystallite size of the composite materials. Also, as the concentration of Ag₂O increased from 0 g to 0.5 wt% within the composite material, the average lattice strain increased from $3.41 \cdot 10^{-3}$ to $4.40 \cdot 10^{-3}$. In simpler terms, adding Ag₂O to the composite material resulted in a slight increase in the average lattice strain.

Keywords: phosphate bioactive glass (PBG); silicate glasses; Calcium di-phosphate; Silver oxide Strontium carbonate (SrCO₃)

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1. INTRODUCTION

Phosphate bioactive glasses (PBGs) have emerged as a promising alternative to conventional silicate glasses, thanks to their adjustable chemical resistance that can be customized to meet specific application requirements[1-3]. PBGs can be designed to degrade completely in a timeframe ranging from hours to years. This property, along with their demonstrated bioactivity, makes them an attractive material option for bone repair and reconstruction. Additionally, PBGs exhibit favorable thermal properties, as evidenced by the numerous studies on bioactive phosphate fibers conducted in recent years, suggesting a wider processing range[4-6].

The dissolution behavior of glasses in the 50P₂O₅-10Na₂O-(40-x)CaO-xSrO system was found to exhibit a minimum when half of the CaO was replaced with SrO [7]. The chemical resistance of the glass was observed to be influenced by its structure, with a reduction in the length of the phosphate chains resulting in increased chemical resistance. Additionally, the presence of SrO was found to enhance the proliferation and growth of gingival fibroblast cells, in contrast to the cell death observed with SrO-free glasses within 24 hours. The CaO-free glass displayed similar cell counts to those measured on the surface of the reference glass S53P4. The improved cell viability with SrO-containing glasses is partly attributed to the decreased initial dissolution rate, resulting from the replacement of CaO with SrO. The effect of strontium ions, both in the surrounding media and within the reactive layer of the glass, was also found to play a significant role [8, 9].

Metaphosphate glasses have shown promising results as potential biomaterials for bone repair and reconstruction. However, they exhibit a rapid initial dissolution rate, which can result in the late formation of a reactive layer. This can hinder the attachment and proliferation of cells during the first few days of culture, which is a crucial step in the healing process. The reactive layer may contain precipitates or other species that can interfere with cell adhesion and growth. This issue has been observed in previous studies, where fast-dissolving glasses were found to be unsuitable for promoting proper cell proliferation. Therefore, further research is required to address these challenges and optimize the performance of metaphosphate glasses as biomaterials [5].

The addition of metal ions, such as silver (Ag), copper (Cu), or cobalt (Co), to glass compositions can modify their physical and chemical properties and provide unique functionality for clinical applications. These metal ions are known to possess antimicrobial properties that can be useful in preventing or controlling infections [13-19]. For instance, silver ions are highly effective against a broad range of microorganisms, including bacteria, viruses, and fungi, by disrupting their cellular processes. Similarly, copper ions can damage the cell membranes of microorganisms, while cobalt ions not only have antibacterial properties but can also promote bone growth and enhance the mechanical strength of the glass.

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Therefore, incorporating metal ions into glasses can expand their potential applications in the biomedical field and improve their functionality [17, 19].

Silver (Ag) is a widely used metal ion dopant in biomedical devices due to its effectiveness against various microbial growth and low toxicity to human cells, even at low concentrations. Ag ions in the form of nitrate or oxide are commonly employed in healthcare products such as Ag-coated catheters or wound dressings to prevent or control infections caused by microorganisms. Ag ions interact with the cellular structures of microbes, disrupt their metabolic processes, and eventually cause cell death, giving rise to their antimicrobial properties. Furthermore, Ag ions can promote tissue regeneration and wound healing, making them an excellent choice for biomedical applications[20]. Hence, the incorporation of Ag as a dopant in glass compositions can offer antimicrobial functionality, enhance the safety and efficacy of different medical devices, and improve patient outcomes[21]. Ag-based ointments have been traditionally used for treating wounds that are susceptible to bacterial infections. In the case of phosphate-based glasses (PBGs) doped with Ag, the metal ions are thought to be incorporated within the glass structure. Unlike silicate glasses, PBGs tend to dissolve congruently, which allows for the controlled release of Ag ions as the glass dissolves. Ahmed et al. conducted a study on silver-doped bioactive glasses by substituting Na_2O with Ag_2O up to 15 mol%, and found that the maximum antimicrobial effect was achieved in glasses containing 1 to 5 mol% of Ag_2O . The authors also examined the structure and properties of the silver-doped glasses. More recently, the antimicrobial properties of silver-doped phosphate glasses were investigated for glasses with 65 and 70 mol% of P_2O_5 [22].

Previous studies have shown that an increase in the Ag content in phosphate glasses results in an increased antimicrobial effect, while increasing the phosphate content in Ag-free glasses has a similar effect. However, these studies were conducted with glasses having a phosphate content of 50 mol% or greater, which was found to inhibit growth and bone antigen expression due to the fast dissolution rate of the glass in the P_2O_5 -CaO- Na_2O family[9]. On the other hand, glasses with slow solubility were found to promote cell proliferation. Despite a P_2O_5 content of 50%, previous research on Sr-containing bioactive glasses has demonstrated that gingival fibroblast cells can attach and proliferate. Therefore, it is important to investigate the impact of Ag doping on this glass composition and determine whether it is possible to obtain Ag-doped strontium-containing phosphate glasses with antimicrobial properties[8].

The objective of this study was to examine the impact of varying amounts of silver oxide on the structure and properties of a bioactive glass composed of metal-phosphate. The glass was synthesized using phosphor oxide, strontium carbonate, and calcium carbonate and subjected to characterization via X-ray diffraction (XRD), scanning electron microscopy (SEM), and atomic force microscopy (AFM). By analyzing the data obtained from XRD, AFM, and SEM, the researchers aimed to gain a better understanding of the physical and chemical characteristics of the glass, including its stability, reactivity, and biocompatibility. This would enable the optimization of its performance for diverse biomedical applications.

2. EXPERIMENTAL

2.1. Materials

Sodium carbonate was purchased from Shandong Yifengtuo Chemical Co., Ltd. Silver oxide Strontium carbonate (SrCO_3), and Calcium carbonate (CaCO_3) was purchased from Jinan Future Chemical Co., Ltd. Phosphor oxide was purchased from Hebei Ruisite Technology Co., Ltd.

2.2. Metal-Phosphate Bio-Active Glass

In order to prepare Metal-Phosphate Bio-Active Glass, the following steps were carried out: Firstly, the required amounts of different materials, including sodium carbonate (0.75g), phosphor oxide (1.5g), strontium carbonate (0.75g), calcium carbonate (0.427g), and varying amounts of Ag_2O (0g, 0.25g, 0.5g, or 0.75g), were accurately measured and mixed thoroughly using a mortar and pestle. This resulting mixture was then transferred to a mold, which had a diameter of 2 cm and a height of 0.5 cm. A pressure of 1000 psi (0.000445Ton) was applied to the mold using a hydraulic press for 2 minutes, following which the mold was sintered in a furnace at 780°C for a duration of 2 hours. Once the sintering process was completed, the mold was allowed to cool down to room temperature, following which the solidified glass was removed. These steps were undertaken to prepare the Metal-Phosphate Bio-Active Glass sample for further analysis.

To prepare Metal-Phosphate Bio-Active Glass, we followed the following steps: First, we measured out the required amounts of sodium carbonate (0.75g), phosphor oxide (1.5g), strontium carbonate (0.75g), calcium carbonate (0.427g), and Ag_2O (0g, 0.25g, 0.5g, or 0.75g). Mix these materials thoroughly in a mortar and pestle, and transfer the resulting mixture to a mold (2 cm in diameter and 0.5 cm with height). Apply a pressure of 1000 psi (0.000445 Ton) for 2 minutes using a hydraulic press, and then sinter the mold in a furnace at 780°C for 2 hours. After the sintering process is complete, allow the mold to cool to room temperature and then remove the solidified glass.

2.3. Characterization

Images of the current samples were captured by a JEOL/EO SEM (version 1.0). Platinum (Pt) coating was performed using an Auto fine coater-JEOL (JEC-3000 FC) for 60 seconds at 20 mA of operating current. The accompanying INCA x-act was used for the EDS analysis (Oxford instruments). Samples' EDS spectra were acquired at acceleration voltages of up to 10 keV.

To perform X-ray diffraction (XRD) analysis, the sintered compacted discs were first ground into powder and then analyzed using a Philips PW 1800 instrument. The instrument was set to a voltage of 40 kV and a current of 30 mA, and it used Cu-K α radiation with a wavelength of 1.54 Å. XRD patterns were recorded in the range of $10^\circ \leq 2\theta \leq 90^\circ$ at a scanning rate of $2^\circ/\text{min}$ for qualitative analysis.

Due to the limitations of SEM in revealing fine topological details, atomic force microscopy (AFM) was used to analyze the morphology of Metal-Phosphate Bio-Active Glass. Specifically, an NTEGRA Probe Nano Laboratory System manufactured by NT-MDT in Moscow, Russian Federation was used. The system was equipped with an NSG01 cantilever that had a tip with a resolution of 10 nm.

3. RESULTS AND DISCUSSION

3.1. Morphological Study

The morphology of powder samples sintered at 780°C for 2h with and without varying concentrations of Ag_2O (0, 0.25, 0.5 and 0.75g) is shown in Figure 1, evaluated using a field emission scanning electron microscopy image. Sintered sample powders are spherical with aggregation of particles, as can be seen plainly (Figure 1a).

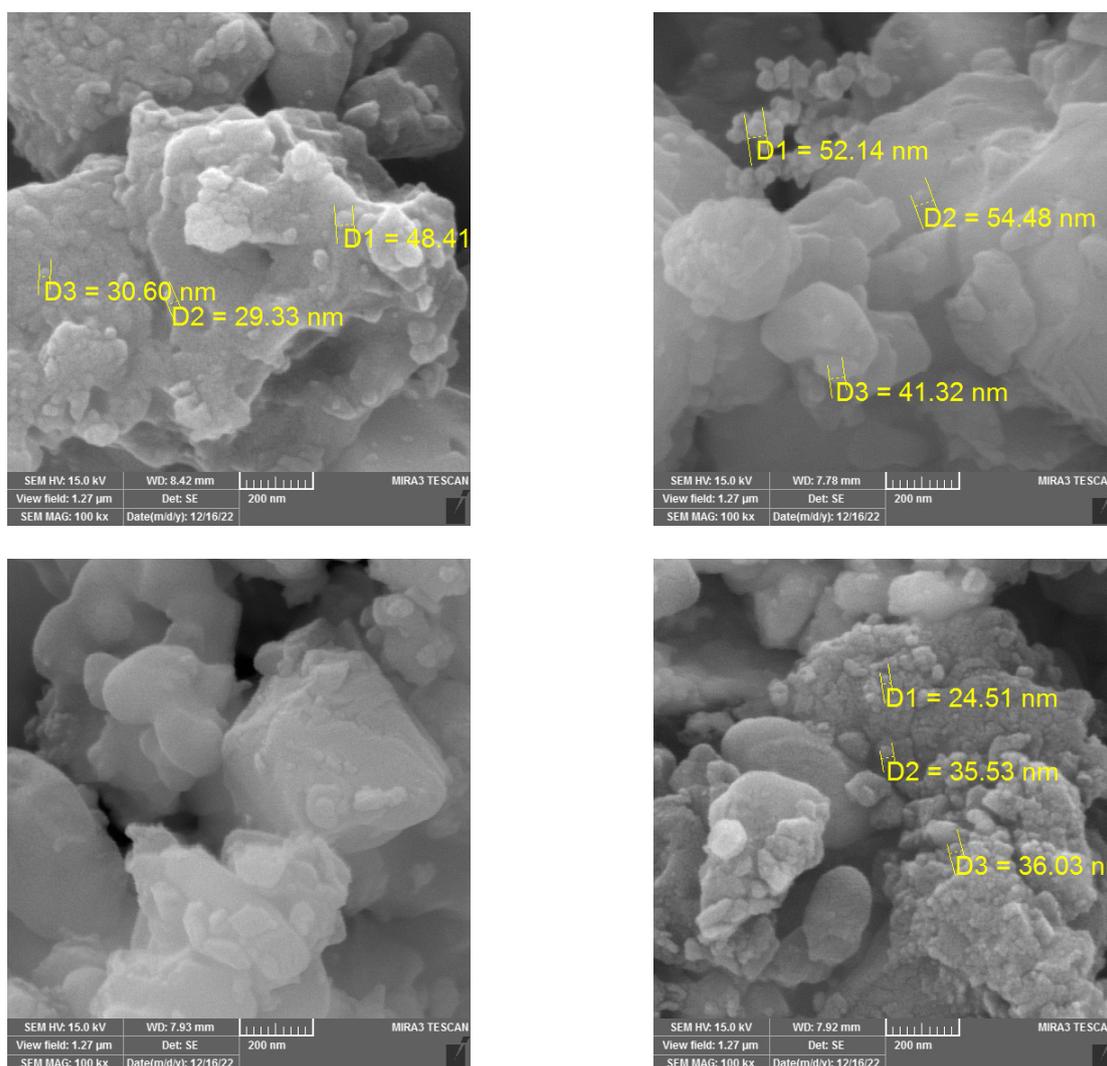


Figure 1. SEM Surface characteristics of calcium phosphate ceramic with (a) - 0 wt% Ag_2O , (b) - 0.25 wt% Ag_2O , (c) - 0.5 wt% Ag_2O , and (d) - 0.75 wt% Ag_2O

Micrographs of Ag_2O -free composites show the grains coalescing and Surface particles are larger than Ag_2O (0.25, 0.5, and 0.75g) particles (Figure 1b-d). The results show that Ag_2O nucleation occurred on the surfaces of all the ceramics, and the Ag_2O -rich ceramics exhibited more particle growth. SEM examination leads us to the conclusion that adjusting the Ag_2O concentration in the bioactive glass can influence the rate at which the bioactive layer forms on its surface. In other words, the addition of Ag_2O leads to the depolymerization of $(\text{PO}_4)_3$ and the formation of a compact phosphate network.

In Figure 2a, it can be seen that, the composite without Ag_2O particles were formed as irregular dense block shapes with dispersion of agglomerates particles over the composite surface. Figures 2a-d manifest the interaction between the

ingredients including Ag₂O loaded on the surface of metal-phosphate bioactive glass which confirmed by EDS elemental analysis. Carbon, Oxygen, Nitrogen, Sodium, Phosphorus, Strontium, and Silver were all added to form the metal-phosphate composite bioactive glass that provided experimentally. Also, the SEM photos revealed that the addition of Ag₂O particles leads to form homogenous composites with effective attraction between the components to produce identity agglomeration shape. Increasing of Ag₂O to the composites contents leads to formation of smaller particles size as it appears in the external area of the SEM photography's. The surface pictures demonstrated that the P₂O₅ substrates with the Ag₂O/CaCO₃/SrCO₃/Na₂CO₃ had been successfully manufactured.

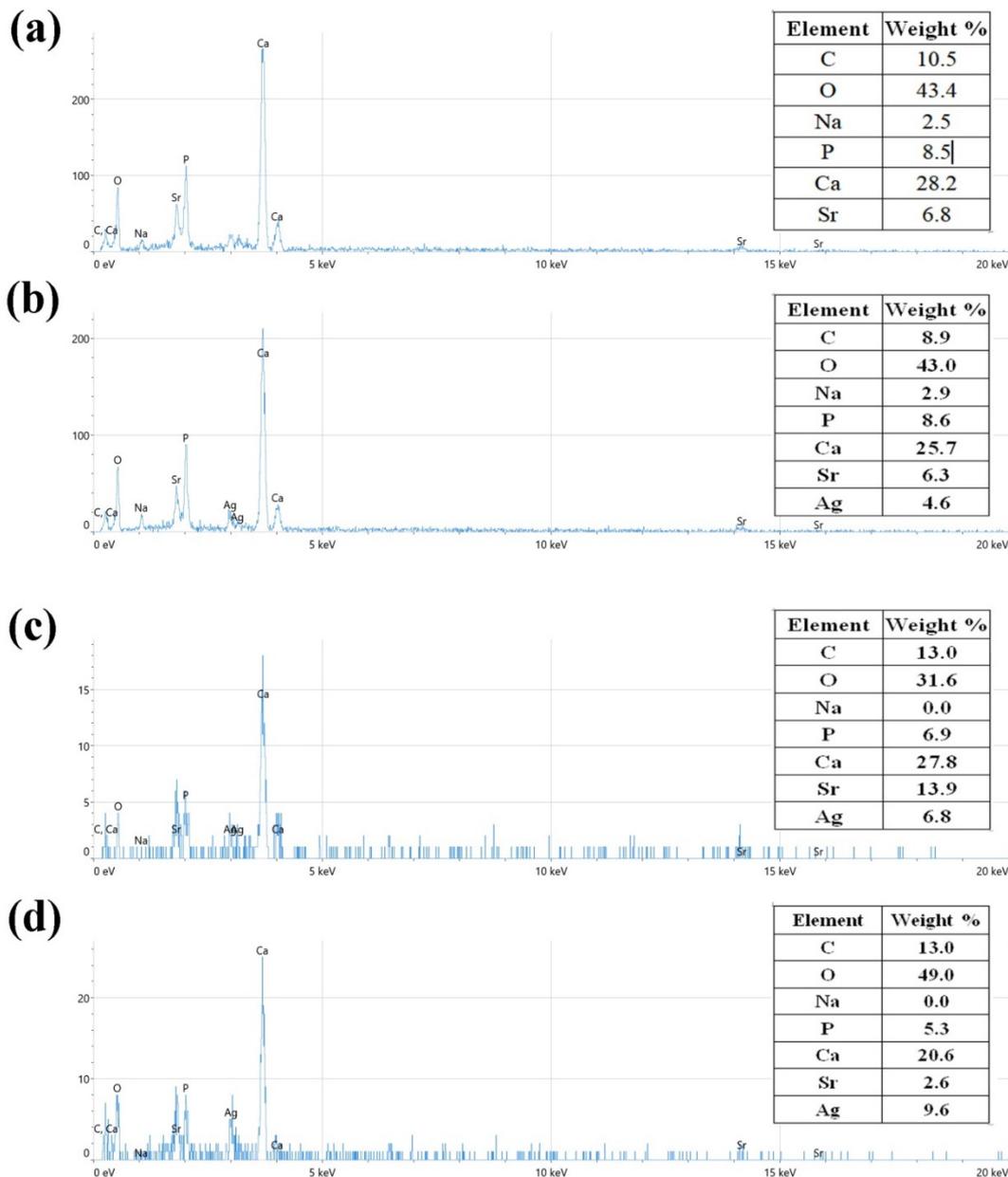


Figure 2. EDX analysis of calcium phosphate ceramic with (a) - 0 g Ag₂O, (b) - 0.25 g Ag₂O, (c) - 0.5 g Ag₂O, and (d) - 0.75 g Ag₂O

3.2. X-ray diffraction

Figure 3 shows the XRD patterns of the samples sintered at 780 °C for 2h without and with different contents of Ag₂O (0.025, 0.5 and 0.75g). The diffraction pattern of the phosphate bio active glass composites sintered without Ag₂O displays the Strontium di-phosphate (Sr₂O₇P₂) ((orthorhombic - a=8.9459 Å b=5.4133 Å c=13.2114 Å) and the Calcium di-phosphate ((tetragonal - a=6.6858 Å c=24.1470 Å)) (Ca₂O₇P₂) (Figure 3). The reflections for phosphate bioactive glass composites without Ag₂O at 25.74°, 26.02°, 33.6947°, 36.3317°, 47.53°, 48.74° in the XRD pattern can be indexed to (210), (013), (204), (303), (322), and (413) planes of Strontium diphosphate, respectively. While the reflections at 26.94,

26.94, 27.89, 29.12, 29.68, 30.09, 32.11, 35.47, 38.51, 39.77, 43.48, 44.59, 45.43, 64.81, and 77.71 can be indexed to (201), (202), (023), (008), (121), (117), (125), (119), (223), (034), (035), (129), (337), and (1316) planes of calcium diphosphate, respectively. The appearance of a new phase relative to Ag_2O (cubic – $a = 4.7600 \text{ \AA}$) was detected after the addition of 0.25, 0.5 and 0.75 g Ag_2O (Figure 3b–d). At higher contents of Ag_2O (after 0.5 g Ag_2O), the intensity of calcium diphosphate and strontium diphosphate increased (Figure 3c–d). The reflections at 32.79° , 37.73° and 54.29° can be indexed to (111), (200) and (220) of Ag_2O , respectively.

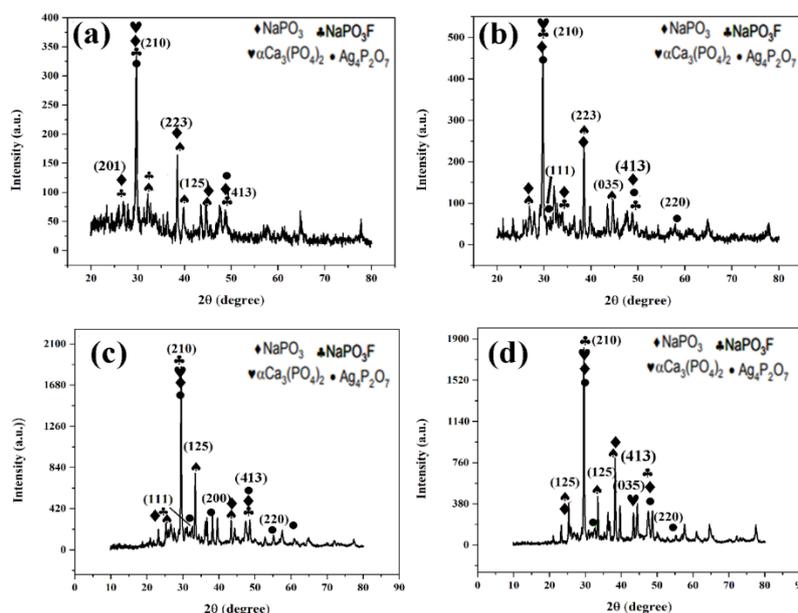


Figure 3. X-ray diffraction patterns of mixture of raw materials with different content of Ag_2O , (a) – 0 g Ag_2O , (b) - 0.25 g Ag_2O , (c) - 0.5 g Ag_2O , and (d) - 0.75 g Ag_2O .

In general, the increase in peak width in the X-ray diffraction and in the peak profile analysis as a result of dislocation growth is by reason of an increase in lattice strain and crystallite size, as a result of dislocation growth.

Equation (1) shows how to determine the size of a crystallite using the Scherrer formula, which is given below[23, 24]:

$$D = K\lambda / \beta \cdot \cos\theta \quad (1)$$

where k is the shape factor (0.9), d is the crystallite size in nm, λ is the wavelength of X-ray ($\text{Cu K}\alpha = 0.15406 \text{ nm}$), and β_{hkl} is the full width at half maxima (FWHM) of an individual peak at 2θ .

Figure 4a and b show the average crystallite size and micro strain measurements for the phosphate bioactive glass composites with and without Ag_2O , respectively. The average crystallite size was decreased from 36.2 nm to 31.7, 31.0, and 32.8 nm with increased Ag_2O from 0 g to 0.25, 0.5, and 0.75 g within the composites, respectively.

The average lattice strain was increased from $4.41 \cdot 10^{-3}$ to $4.40 \cdot 10^{-3}$ with increased Ag_2O from 0 g to 0.5g within the composites (Figure 4b). Lattice strain is important because it can effect on the ceramic performance and full width at half maximum (FWHM: β_{hkl}) of X-ray diffraction peaks; in other word, lattice strain is a measure of the distribution of lattice constants arising from crystal imperfections, such as lattice disruptions and dislocations, in which crystallite size reduction can induces a large amount of strain.

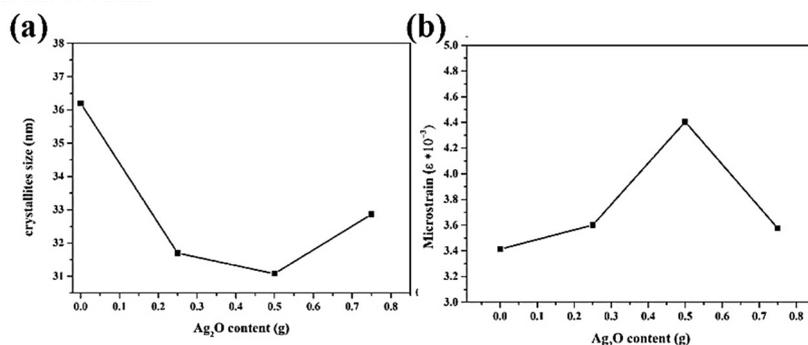


Figure 4. Average crystallites size and microstrain of phosphate bio active glass composites with different content of Ag_2O

3.3. Surface topography

AFM is used to measure the surface topography and roughness of materials [25, 26]. In this study, Metal-Phosphate bio-active glass has been prepared with different concentrations of silver oxide and tested its surface roughness using AFM. By analyzing the AFM data, the results show that the surface roughness decreased as the concentration of silver oxide increased up to a certain point, and then increased again at higher concentrations. This information can be useful in optimizing the composition of prepared glass material for specific applications where surface roughness is an important factor. Therefore, AFM is an important tool for characterizing and optimizing the properties of metal-phosphate Bio-active glass material.

The addition of silver oxide to Phosphate bio-active Glass can potentially affect the surface roughness of the resulting material, but the direction of this effect may depend on various factors, such as the concentration of silver oxide and the processing conditions.

In general, the addition of a foreign material to a glass matrix can introduce variations in the surface topography and roughness. Silver oxide is known to be a powerful nucleating agent, which can promote the formation of crystals or nanoparticles within the glass matrix [27]. These crystals or nanoparticles may alter the surface morphology of the glass, potentially increasing or decreasing its roughness, depending on their size, shape, and distribution. The decrease in surface roughness (Sq) observed in Figure 5 and Table 1 as the amount of silver oxide increased from 0g to 0.5g in the metal-phosphate Bio-active glass can be attributed to several factors. Firstly, silver oxide can act as a smoothing agent, filling in small surface irregularities and voids to reduce surface roughness. Secondly, the addition of silver oxide can inhibit crystal growth in the glass by interacting with the glass matrix and preventing the growth of crystalline phases that contribute to surface roughness [28]. Lastly, the presence of silver ions can promote the nucleation and growth of smaller particles during sintering, leading to a reduction in particle size and contributing to a smoother surface. In contrast, the increase in Sq observed as the concentration of silver oxide increased from 0.5g to 0.75g in the metal-phosphate Bio-active glass can be attributed to several factors. Firstly, there is a possibility of agglomeration or clustering of silver oxide particles during the sintering process at higher concentrations, leading to the formation of larger surface irregularities and voids. Secondly, the addition of a higher concentration of silver oxide may result in the formation of new phases in the glass, such as silver phosphate or silver strontium phosphate, which could contribute to an increase in surface roughness. Lastly, the higher concentration of silver oxide may lead to an increase in the crystallinity of the glass, which can also contribute to an increase in surface roughness.

Table 1. Surface roughness (Sq), average roughness (Sa), and maximum height of the surface (Sz) of metal-phosphate Bio-Active glass samples with varying silver oxide content.

Surface roughness (nm)	Ag ₂ O content (g)			
	0	0.25	0.5	0.75
Sq	42.42	12.67	3.104	89.02
Sz	399.6	107.3	42.9	854.0
Sa	31.28	9.9	1.974	62.77

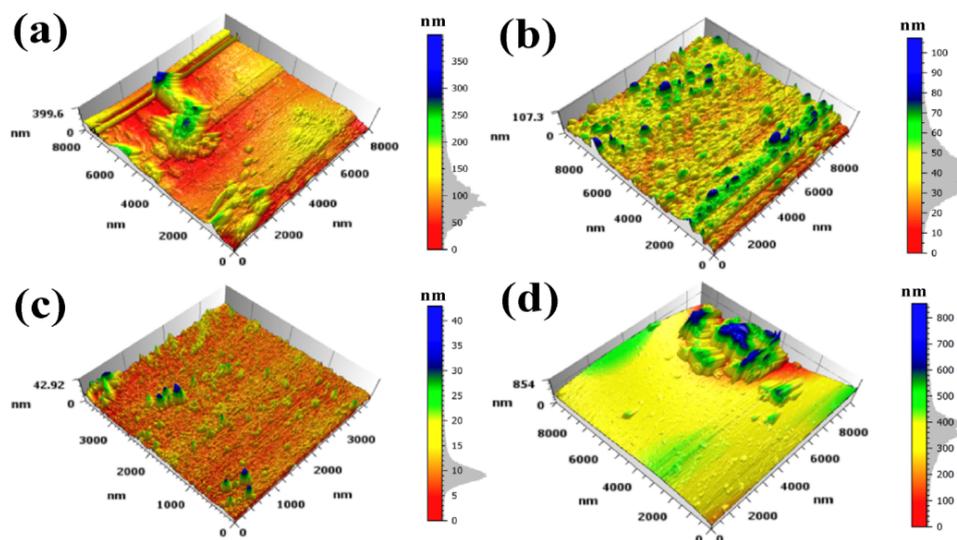


Figure 5. 3D topography of metal-phosphate Bio-Active glass samples with varying silver oxide content. (a) – 0 g Ag₂O, (b) - 0.25 g Ag₂O, (c) - 0.5 g Ag₂O, and (d) - 0.75 g Ag₂O

4. CONCLUSION

In this research, it has demonstrated that the concentration of silver oxide has a significant impact on the physical and structural properties of phosphate Bio-active glass. The addition of Ag₂O to the composite material led to smaller particle sizes and more distinct boundaries between grains, as well as a reduction in the average crystallite size. This

suggests that the incorporation of Ag₂O can improve the performance of the glass in various applications. Moreover, results showed a slight increase in the average lattice strain upon the addition of Ag₂O. These findings have important implications for the design and optimization of phosphate Bio-active glass for various biomedical and industrial applications. Further research is necessary to explore the specific mechanisms underlying these effects and to investigate the potential of these materials for specific applications.

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ДОСЛІДЖЕННЯ СТРУКТУРНОГО ВПЛИВУ ОКСИДУ СРІБЛА В БІОАКТИВНОМУ ФОСФАТНОМУ СКЛІ

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Досліджено вплив різних концентрацій оксиду срібла на структуру та морфологію фосфатного біоактивного скла (PBG). PBG набувають популярності як потенційна заміна традиційному силікатному склу у біомедичних застосуваннях завдяки їх регульованій хімічній стійкості та винятковій біоактивності. При дослідженні за допомогою скануючого електронного мікроскопу композитів без Ag₂O було виявлено тенденцію до злиття зерен, а поверхневі частинки виявилися більшими, ніж у композитах з Ag₂O при концентраціях 0,25, 0,5 і 0,75 мас.%. Дослідження показало, що дифракційна картина фосфатних біоактивних скляних композитів, спечених без Ag₂O, показала присутність дифосфату стронцію та дифосфату кальцію. Рентгенограма цих композитів без Ag₂O виявила специфічні площини, які відповідали обом типам дифосфату. Однак, коли додавали Ag₂O, була виявлена нова кубічна фаза, і інтенсивність дифосфату кальцію та стронцію зростала з вищим вмістом Ag₂O. Рентгенограма композитів з Ag₂O відображала специфічні площини, які відповідали Ag₂O. Іншими словами, відсутність Ag₂O в композиційному матеріалі призвела до більших розмірів частинок і менш чітких меж між зернами. Крім того, було виявлено, що при збільшенні концентрації Ag₂O від 0 до 0,25, 0,5 і 0,75 мас.% середній розмір кристалітів зменшився з 36,2 до 31,7, 31,0 і 32,8 нм відповідно. Ці результати свідчать про те, що додавання Ag₂O може ефективно зменшити середній розмір кристалітів композитних матеріалів. Крім того, у міру збільшення концентрації Ag₂O від 0 г до 0,5 мас.% у композиційному матеріалі середня деформація решітки збільшилася з $3,41 \cdot 10^{-3}$ до $4,40 \cdot 10^{-3}$. Простіше кажучи, додавання Ag₂O до композитного матеріалу призвело до незначного збільшення середньої деформації решітки.

Ключові слова: фосфатне біоактивне скло (ФБС); силікатне скло; дифосфат кальцію; оксид срібла, карбонат стронцію (SrCo₃)