2019 IEEE 9th International Conference on Nanomaterials: Applications & Properties (NAP – 2019)
Odesa, Ukraine, September 15-20, 2019

Ministry of Education and Science of Ukraine
Sumy State University
IEEE Nanotechnology Counsil
IEEE Magnetic Society
The International Union of Pure and Applied Physics

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Track: Biomedical Applications

Track: Theory & Modeling

Synthesis and Characterization of Hydroxyapatite Composite Materials Loaded with ZnO Nanoparticles

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Hydroxyapatite (HA) based composite biomaterials are widely used for bone substitution. Incorporating of inorganic metal ions or metal oxide nanoparticles into composite materials could increase antibacterial properties. ZnO nanoparticles show significant antibacterial activity against most pathogenic and nonpathogenic bacteria. Meanwhile, the incorporation of Zn in an implant material promotes the proliferation and differentiation of osteoblast cells, leading to enhanced osteogenesis [1].

a) *Synthesis of hydroxyapatite nanoparticles.* $Ca_{10}(PO_4)_6(OH)_2$ – stoichiometric HA was synthesized by following scheme:

$$10\ Ca(NO_3)_2 + 6\ (NH_4)_2HPO_4 + 8\ NH_3 \cdot H_2O \rightarrow Ca_{10}(PO_4)_6(OH)_2 + 20\ NH_4NO_3 + 6\ H_2O$$

Analytically grade salts $Ca(NO_3)_2$ and (NH4)2HPO4 were taken in concentrations 0.1 M and 0.06 M respectively. First 0.1 M $Ca(NO_3)_2$ solution was mixed with ammonia solution, after that 0.06 M $(NH4)_2HPO_4$ solution was added dropwise to the above mentioned solution. Solution pH was adjusted to 10 by adding 25% ammonia solution. After aging for two days it was rinsed by distilled water until pH = 7. For obtaining of the composite material it was taken as wet slurry.

b) *Synthesis of metal oxide nanoparticles.* ZnO nanoparticles were synthesized by polyol method using $Zn(CH_3COO)_2 \cdot 2H_2O$ and ethylene glycol as a reaction medium. Schematically it can be shown as following:

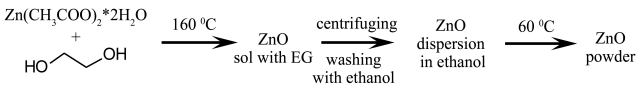

Fig. 1. Scheme of ZnO nanoparticles synthesis.

HA and ZnO nanoparticles with average size 80 and 10-30 nm respectively were mixed as a wet slurry in relation 5:1 by weight, than obtained composite material was centrifuged at 2500 rpm. TEM microscopy of obtained ZnO nanoparticles and XRD that shows presence of zinc oxide (JCPDS 01-089-1397) is shown at the Fig.2.

The mechanism of ZnO antimicrobial activity in obtained HA/ZnO composite material was examined against Gram-positive (*Staphylococcus aureus*) and Gram-negative

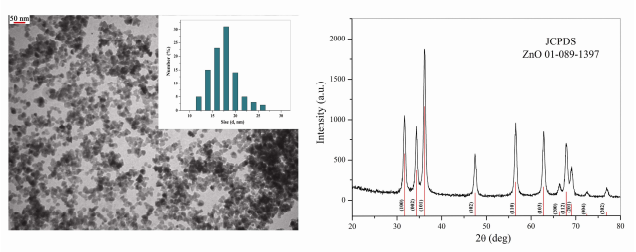

Fig. 2. TEM image and XRD analysis of ZnO nanoparticles.

(*Escherichia coli*) bacteria. The isolates were cultivated overnight in nutrient broth at 37°C. The cultures were diluted with nutrient media to the turbidity equivalent to McFarland 0.5 standard (1.5×10^8 CFU/ml). The MIC of the ZnO was measured by serial dilution method according to the international recommendations provided by the Clinical and Laboratory Standarts Institute (SLCI). Several secondfold dilutions were used to decrease the sample concentration. As controls test-tubes containing growth medium and microorganisms without ZnO were used. Determining the colony count carried out using streak plate technique.

The tested material demonstrated antibacterial activity depending on the concentration of the ZnO in solution against both types of microorganisms. MIC of ZnO in suspension of composite material prevented growth *E. coli* and *S. aureus* was equal 0.0337 g/mL for both of them. At lower concentrations of ZnO in the sample decreasing of bacteriostatic properties was revealed.

Acknowledgments

This research supported by H2020 Marie Skłodowska-Curie Actions Grant NanoSurf 777926 and Ukraine MES Grant “Effectiveness of nanocomposite (chitosan-nanometal) antimicrobial action against multiresistant clinical strains” № 62.20.02-01.18/20.).

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