Empowering students in science: Phytosynthesis, characterization and biomedical application of metallic (Ag, Si, Cu) and oxide (ZnO, TiO_2, CuO, Fe_2O_3) nanoparticles: microbiology, biokinetics and toxicology aspect

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Abstract:
Four students at Chemistry Department, Faculty of Natural Sciences and Mathematics, University of Banja Luka were involved in national research project "Phytosynthesis, characterization and biomedical application of metallic (Ag, Si, Cu) and oxide (ZnO, TiO_2, CuO, Fe_2O_3) nanoparticles: microbiology, biokinetics and toxicology aspect" led by their professors. Students role was to synthesize metallic oxides: ZnO, TiO_2, CuO, Fe_2O_3 using green chemistry approach and to evaluate metallic oxides physicochemical properties. All the metallic oxide sample were phytosynthesized using methanolic extract of flowers of *Geranium robertianum* L. (Geraniaceae). For characterization students used UV/VIS spectrophotometry, infrared spectroscopy and optical microscope. Additionaly, students evaluated photocatalytic properties of obtained samples, as well as, antimicrobial activity. All the samples were in nanorange, ZnO (530 nm), TiO_2 (460 nm), CuO (710 nm), Fe_2O_3 (980 nm), showed good antimicrobial properties against gramm negative (E.coli) and gramm positive (S. Aureus) bacteria with average MIC value 0.312 mg/mL for S. Aureus and 0.156 mg/mL for E. coli. Photocatalytical efficiency was between 77 - 96% depending on the metallic oxide. The main focus of students involvement in research project was giving importance to scientific work and students' independence in scientific research.

Key words: phytosynthesis, photocatalysis, antimicrobial activity, nanoparticles

Introduction:

Chemical and physical stability, high adsorption capacity and low-toxicological profile are very important features for application of nanoparticles (NPs) in the biomedical field. Physicochemical and biochemical characteristics of nanoparticles are very much dependent on the synthesis method. One of the promising, enivironmentally conscious method is the green synthesis of NPs using plant extracts. In this study methanolic extract of flowers of Geranium robertianum L. (Geraniaceae) was used for phytosynthesis of zinc oxide (ZnO NPs)., copper oxide (CuO NPs), Ttitanium oxide (TiO2 NPs) and iron oxide (Fe2O3). The green method of nanoparticle synthesis using plant extract is a simple alternative to chemical and physical processes. Chemical and physical methods are harmful because the chemicals used are toxic, flammable and not easily disposed of in the environment. The use of biological samples such as plant extracts for the synthesis of nanoparticles has advantages over chemical and physical methods. In recent years, phytosynthesis of nanoparticles has attracted significant attention due to the growing need to develop pure and non-toxic chemicals, environmentally friendly solvents and renewable materials. Research has shown that the size, morphology, stability and physicochemical properties of metal nanoparticles are strongly influenced by experimental conditions and play key roles in controlling the physical, chemical, optical and electronic properties of these nanomaterials. Four nanoparticles systems were synthesized and characterization was performed using UV/VIS spectroscopy, infra red spectroscopy and optical microscope.

Experimental:

Materials: The powder of ZnO, Cuo, TiO_2 and Fe_2O_3 were synthesized by green chemistry technique. The chemicals $Cu(NO_3)_2 \cdot 3H_2O$, $Zn(CH_3COO)_2 \cdot 2H_2O$, $Fe(NO_3)_3 \cdot 9H_2O$ and $TiO(SO_4)_2$ were purchased from Merck. Synthesis of NPs started with preparation of a solution of proper precursor and Geranium robertianum L. solution. pH was adjusted using 2M NaOH. The solutions were heated on in an open glass beaker with constant magnet stirrer for 2h at 550 rpm. After , samples of NPs were centriguated for 5 min at 3000 rpm. The heating was then continued in an oven up for 24h.

UV/VIS spectrophotometry: Photocatalytic study was performed by using sunlight as a light source. In order to determinate photocatalytical power of NPs and with 100 mL of Methylene Blue (10 ppm) and we followed degradation of Methylene Blue in time. For absorption experiment Perkin Elmer Lambda 25 spectrophotometer was used.

IC spectroscopy: Fourier transform infra-red spectroscopy (FT-IR) was carried out by using Tensor 27 instrument with addition of platinum stand on which a very small amount of pure NPs/germanium sample was placed (Bruker, USA).

Optical miscroscopy: Optical microscope Omax A35140U was used

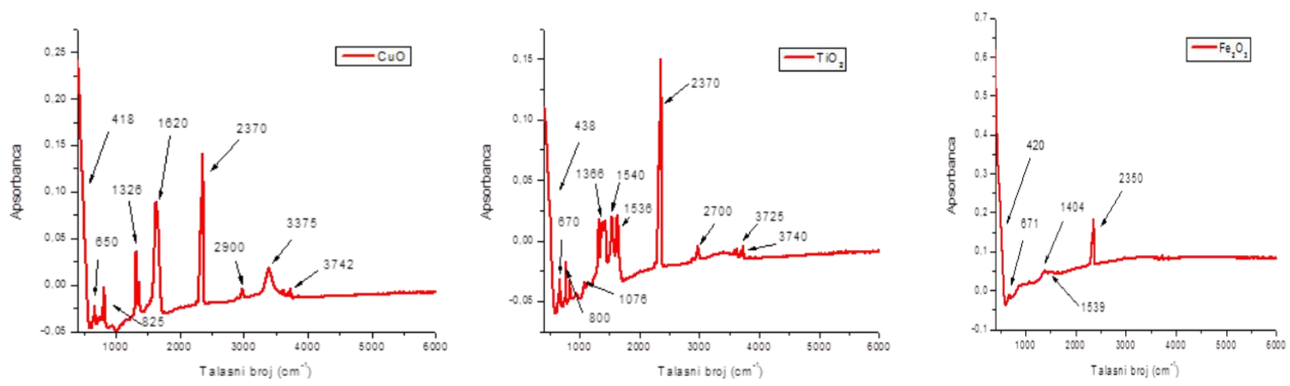

Table 1. Kinetic and photocatalysis parameters

NPs system	K, 1/min	$t_{1/2}$	Photocatalysis efficiency
ZnO	0.0241	28.76	96.0
TiO_2	0.021	33.0	95.4
Fe_2O_3	0.011	63.0	80.43
CuO	0.0087	79.6	77.29

Table 2. Antimicrobial analysis

Acinetobacter

Noncentrifugation	Centrifugation
Fe 10 mm	Fe 10 mm
Zn 10 mm	Zn 15 mm
Ti 9 mm	Ti 9 mm
Cu 8 mm	Cu 8 mm

MRSA

Noncentrifugation	Centrifugation
Fe 11 mm	Fe 14 mm
Zn 11 mm	Zn 14 mm
Ti 10 mm	Ti 11 mm
Cu 11 mm	Cu 12 mm

Pseudomonas

Noncentrifugation	Centrifugation
Fe 11 mm	Fe 12 mm
Zn 12 mm	Zn 11 mm
Ti 13 mm	Ti 11 mm
Cu 10 mm	Cu 10 mm

E coli ATCC

Noncentrifugation	Centrifugation
Fe 11 mm	Fe 12 mm
Zn 12 mm	Zn 10 mm
Ti 11 mm	Ti 10 mm
Cu 11 mm	Cu 12 mm

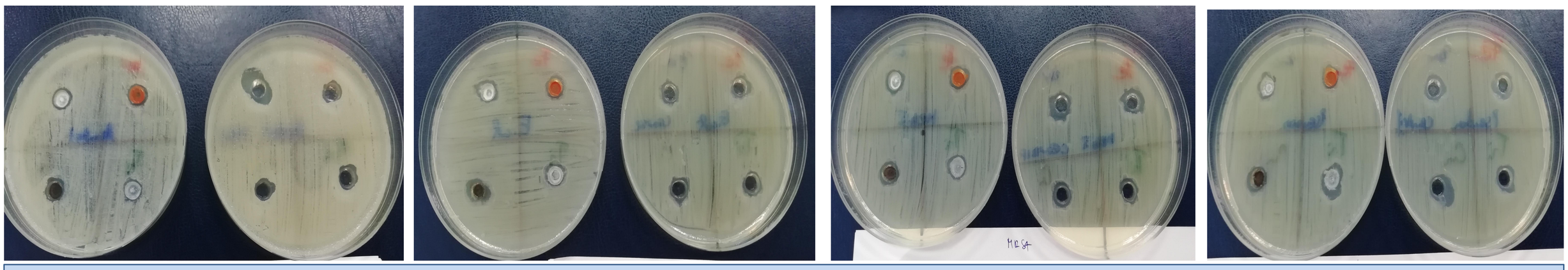

Figure 2. Antimicrobial analysis: Acinetobacter (first), E.coli (second), MRSA (third) and Pseudomonas (fouth)

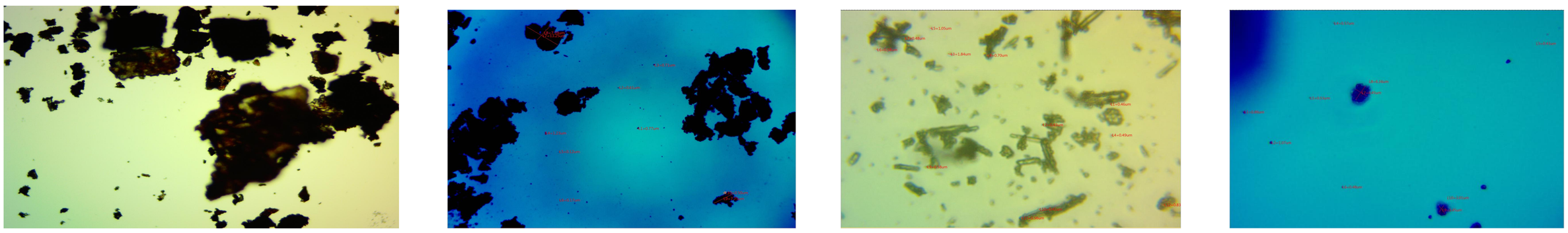

Figure 3: Optical microscope images of green synthesized nanoparticles: iron oxide (first), copper oxide (second), titanium oxide (third) and zinc oxide (fourth)

Conclusions:

Conclusions: Two main goals of presented research were accomplished. First one, accounting green chemistry for NPs synthesis and their characterization and second one, enagagement of chemistry students in scientific project and empowering students to do the science.

Four different systems of nanoparticles were successfully synthesized using green chemistry method. Obtained samples of NPs systems were characterized by three instrumenatal technique: UV/VIS spectroscopy for photocatalysis evaluation, IC spectroscopy and optical miscroscopy. Additionaly antimicrobial analysis of NPs system was performed against gramm-negative and gramm-positive bacteria. NPs showed photocatalytical performances in following order: zinc oxide>titanium oxide>iron oxide>copper oxide. All synthesized NPs samples showed antibacterial activity.

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