 DOI: 10.33892/aph.2021.91.310-311

P-72

Biogenic Iron, Silver and Gold Nanoparticles Against Opportunistic Pathogenic Yeasts and Dermatophytes

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Keywords: *Biological synthesis, toxicity, antifungal activity, dermatophytes, opportunistic pathogenic yeasts*

1. Introduction

Epidemiologic observations indicate that the number of systemic fungal infections has increased significantly during the past decades, however in human mycosis, mainly cutaneous infections predominate, generating major public health concerns and providing much of the impetus for current attempts to develop novel and efficient agents against cutaneous mycosis causing species[1]. Innovative, environmentally benign and economic nanotechnology-based approaches have recently emerged utilizing principally biological sources to produce nanosized structures with unique antimicrobial properties[2]. Due to the obvious advantages, the green synthesis of nanoparticles is a rapidly progressing area of the nanobiotechnology.

In line with this, the aim of this present study was to investigate the suitability of various green materials such as *Parthenocissus quinquefolia* plant extract and *Phaffia rhodozyma* cell-free extract for the preparation of iron nanoparticles (FeNPs), silver nanoparticles (AgNPs) and gold nanoparticles (AuNPs) by biological synthesis and to determine the toxicity of nanoparticles to human keratinocyte cells as well as against various fungal species with a special emphasis on antifungal efficiency against dermatophytes.

2. Materials and methods

For the preparation of iron, silver and gold nanoparticles the *Parthenocissus quinquefolia* plant extract and red yeast *Phaffia rhodozyma* cell-free extract was used as a reducing and stabilizing agent during the synthesis. The morphological features of the synthesized FeNPs, AgNPs and AuNPs were analyzed by transmission electron microscopy. The hydrodynamic particle size distribution of the samples was assessed by dynamic light scattering analysis. The crystal structures were examined by X-ray powder diffraction. The optical properties of the obtained FeNPs, AuNPs and AgNPs were studied by ultraviolet-visible spectroscopy.

MTT mitochondrial activity assay was performed to measure HaCaT immortalized keratinocyte cell viability. Cytotoxicity of the synthesized nanoparticles was assessed by crystal violet staining as well. The antifungal activity of the nanoparticles was tested against pathogenic yeasts as well as against various dermatophytes (Table 1).

Table 1 *List of the tested strains*

Species	Strain number
Candida albicans	ATCC 10231
Candida glabrata	CBS 138
Candida krusei	CBS 573
Candida parapsilosis	CBS 604
Candida tropicalis	CBS 94
Cryptococcus neoformans	IFM 5844
Microsporum gypseum	SZMC 11124
Trichophyton mentagrophytes	SZMC 11102
Trichophyton tonsurans	SZMC 11103

3. Results

Our results indicate that the green synthesis was successful in all three cases. The as-synthesized

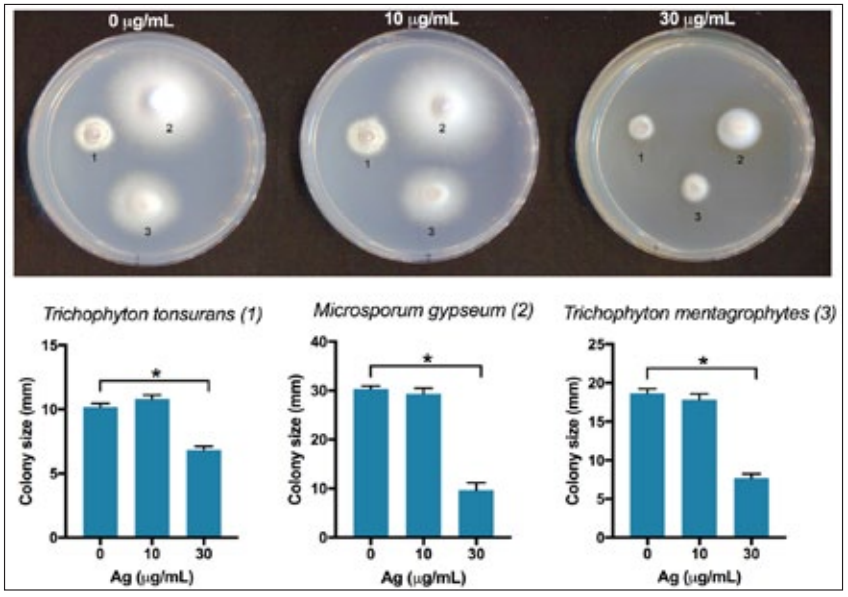

Figure 2 *Anti-dermatophyte activity of silver nanoparticles. Colonies of Trichophyton tonsurans (1), Microsporum gypseum (2) and Trichophyton mentagrophytes (3) on PDA and PDA supplemented with different concentrations of AgNP*

silver and gold nanoparticles were phase pure, well crystalline with a face-centered cubic structure and have the characteristic peak which can be attributed to surface plasmon resonance. AgNPs were quasi-spherical, well separated from each other and minor polydispersity could be observed. The average size of the AgNPs proved to be 4.1±1.44 nm. Gold particles were almost monodispersed, well separated and spherical with a narrow size distribution (2.22±0.7 nm). The FeNPs were around 50.1±2.44 nm and the particles were trapped in the plant matrix. Furthermore, screening of their biological activities confirmed that the iron and silver nanoparticles exhibited good antimicrobial activity against all examined microbial species whereas the gold nanoparticles were toxic only to *Cryptococcus neoformans*. We also assessed the cell viability of human skin-derived keratinocyte HaCaT cells treated with biologically synthesized AgNPs and AuNPs. Both AgNPs and AuNPs, applied in 0–60 μg/mL concentrations, were not toxic to HaCaT human keratinocyte cells. All the three examined dermatophyte species were sensitive to AgNP treatments in growth inhibition assay (***Figure 1***). AgNPs in 30 μg/mL concentration induced significant inhibition of colony growth, indicating an approximately 80% growth inhibition for each strain. On the other hand, none of the dermatophyte species reacted to AuNPs, when applied in 10 or 30 μg/mL concentrations, indicating that AuNPs are not suitable for the growth inhibition of the examined cutaneous mycosis-causing dermatophyte fungi.

4. Conclusions

In this work, we report a simple and eco-friendly method for the synthesis of iron, silver and gold nanoparticles using the *Parthenocissus quinquefolia* plant extract and red yeast *Phaffia rhodozyma*. The as-prepared iron, silver and gold nanoparticles were characterized by transmission electron microscopy, powder X-ray diffraction, dynamic light scattering and ultraviolet–visible spectroscopy. Moreover, a complex biological screening was carried out to determine the biological activity of the nanoparticles. We demonstrated that these nanoparticles are potent against cutaneous mycosis-causing dermatophytes and opportunistic yeast species and are non-toxic to human keratinocytes. Considering our data in the context of the current clinical results, the biocompatibility to skin cells and the outstanding antifungal performance of our biosynthesized nanoparticles might be exploited during topical treatment of cutaneous mycosis or as a prophylactic treatment against the recurrence of the infection.

5. Acknowledgements

This research was supported by the New National Excellence Program of the Ministry for Innovation and Technology from the National Research, Development, and Innovation Fund (ÚNKP-21-5-SZTE-576 for A.R. and ÚNKP-21-5-SZTE-592 for M.K.) and the János Bolyai Research Scholarship of the Hungarian Academy of Sciences (BO/00384/21/7 for A.R. and BO/00878/19/8 for M.K.).

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