1410
doi:10.1017/S1431927622005748
Microsc. Microanal. 28 (Suppl 1), 2022

Morphology of *Coniophora eremophila* Exposed to Silver and Zinc Oxide Nanoparticles in Dextrose Sabouraud Agar Analyzed by Scanning Electron Microscopy

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Introduction

Wood decay is an economic problem linked to agriculture, industry [Hiltunen et al, 2020] and ancient art preservation [Lucejko *et al*, 2018], being fungi the main cause of wood infection and deterioration. *Coniophora eremophila* is a wood pathogen that has been studied as a model for antifungal components such as zinc oxide (ZnONP) and silver (AgNP) nanoparticles [Arzate *et al*, 2017]. Zinc oxide is a well known antifungal agent used in ointments [Fedota *et al*, 2019], and silver is a wide range antimicrobial [Sim *et al*, 2018] with high toxicity that had been been tested against bacteria and fungi of clinical importance, however, not much is known about their effect on *Coniophora* species.

Materials and Methods

Nanoparticles were kindly provided by the Mexican mining company Peñoles and characterized by X-ray diffraction (DRX, Panalytical, X'Pert PRO, Lelyweg, Netherlands) and Scanning electron microscopy (SEM, JEOL equipment model JSM-7401f). Antifungal tests were performed on Petri dishes containing Sabouraud agar, using ZnONP and AgNP as treatments and negative control consisted on Sabouraud agar without nanoparticles. Both treatments were added in concentrations of 120μg/ml and complemented to Sabouraud media before solidification. All plates were inoculated with *C. eremophila* placed in the center of the agar. Incubation was performed for 7 days at room temperature. Samples of the fungus were then drilled from the Petri dish and fixated in acetaldehyde, followed by dehydration on increasing concentrations of alcohol and ketone. Osmium tetroxide was added to the mix in order to stabilize the structure of the cells. The sample was prepared for SEM analysis by the critical point method and covered in gold before observation.

Results

Chemical composition of ZnONP and Ag nanoparticles was confirmed by DRX and EDAX analysis. ZnONP had an average size of 78 nm (Fig. 1A) and AgNP (Fig. 1B) of 25 nm. Even though ZnONP and AgNP partially inhibited the growth of *C. eremophila*, no observable changes in morphology were observed. As presented in figure 2A, morphology in fungus not exposed to treatments are no different than those exposed to ZnONP (Fig 2B) and AgNP (Fig 2C).

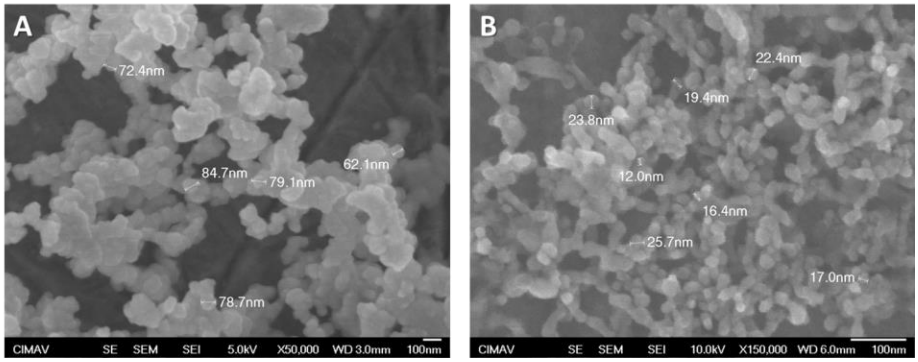

Figure 1. SEM analysis of Zinc oxide (Fig 1A) and Silver (Figure 1B) nanoparticles.

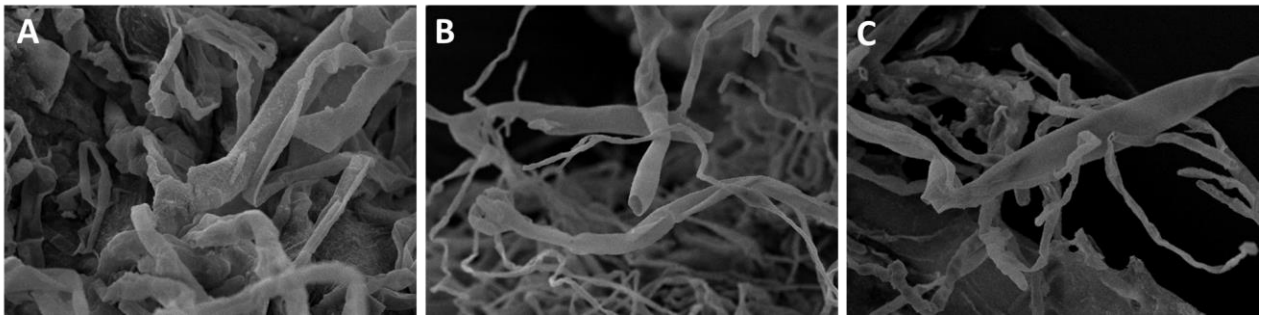

Figure 2. Comparison of morphology of *C. eremophila* hyphae not exposed to nanoparticles (2A), exposed to ZnONP (2B) and exposed to AgNP (2C).

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https://doi.org/10.1017/S1431927622005748 Published online by Cambridge University Press