



The Influence of Different Nanomaterials on the Growth of Fungi



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Introduction

All paintings especially stored in unsuitable conditions suffered from microbial deterioration appeared as stains and discoloration (Figure 1). Bio deterioration is an undesirable process triggered by living organisms such as bacteria, fungi, algae and lichens which can affect cultural heritage and economically important materials. [1]. The physical factors which affect the growth of microorganisms are humidity, temperature and light. Some authors suggest bacteria as the first agents in the colonization of the mural paintings thus providing organic matter to the

next colonizers fungi and lichens [2]. The metabolism of these microorganisms secretes oxalic acid ($H_2C_2O_4$) which can react with calcite $CaCO_3$ present in the paintings giving rise to calcium oxalate (CaC_2O_4) [3]. The growth of biological agents such as fungi is identified as a determinant factor in the degradation of the murals can cause vital deterioration factors such as discoloration of the mural surfaces, detachment of the fragments and biofilms formation. On other hand the bacilli generate energy from the oxidation of reduced sulphur compounds producing sulphuric acid as the end product [3] (Figure 1).



Figure 1: Detail of the painting with places of swabs.

Choosing different nano materials

Currently in this paper nano materials which probably used for cleaning such as nano titanium dioxide, which is common in self-cleaning material, nano calcium hydroxide which consider the most successful consolidation nano materials, and nano silica, nano calcium carbonate which are probably used as filler in consolidation process. Those nano materials are already used for Cleaning and consolidation mural paintings.

Materials and Methods

Sampling

Samples were taken from the colored surface of the current mural painting under aseptic (free from contamination) conditions with sterile swabs.

Isolation and characterization of microorganisms

The samples recovered with sterile swabs were mechanically shaken for IH and inoculated under aseptic condition in different culture media specific to each microorganisms nutrient agar for bacteria , malt extract agar for filamentous fungi and yeast Extract peptone Dextrose agar for yeasts the cultures incubated at 30 °C for bacteria and fungi respectively the identification of the microbial isolates was performed based on the observation and macroscopic features such as texture and color of the colonies , hyphae morphology and observed with an optical microscope (OM) (Leica DM2500P) whose images were acquired in a camera Leica DFC290HD [4].

Current mural painting entails six fungi *A. Fumigatus*, *Aspergillus Flavus*, *Aspergillus Niger*, *Aspergillus Terrus*, *Fuzarium* and

Penicillium specie. The aim of this work is not only the isolation and characterization of the microbial organism affecting the mural painting of the Coptic museum storage backs to saint Jeremiah monastery 5th century but also identify the role of nano material especially the nano materials will be used in cleaning and consolidation the undertaken mural painting which restored using nano materials [5] .

The Disk diffusion test

Disc-diffusion test is a test of the material sensitivity for organisms. Currently nano materials are used to test the effectiveness on the diagnosis microorganisms. In this test, wafers containing nanomaterials are placed on an agar plate where fungi have been placed, and the plate is left to incubate. If the material stops the fungi from growing or kills them, there will be an area around the wafer where the fungi have not grown enough to be visible. Currently in this study the nano-materials effect the growth of fungi through measuring the diameter of clear zone or measuring the diameter of fungal colony then comparing the results before and after the test , even the diameter of the fungal colony shorter than the diameter of clear zone after adding the anti-material that is mean the material is effected the fungi.

Nano- Materials Preparation (Antifungal)

All nano materials purchased from nano tech com. LTD (nano silica, nano calcium carbonate, nano calcium hydroxide, and nano titanium dioxide) prepared in conc. 2% in ethanol (purity >99%). Preparation of the mixed nanomaterial / system. The nanomaterial was sonicated in ultrasonic power 100 for 1 hour to make a homogenous suspension.

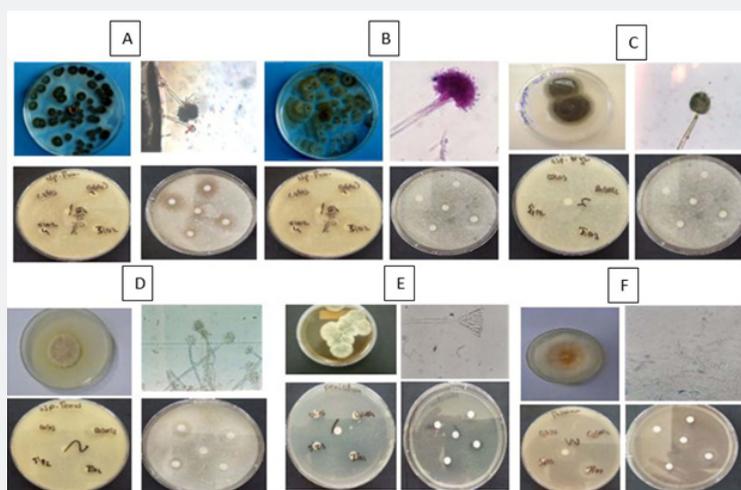


Figure 2:

- A. The growth of *A. Fumigatus* in Petry plate, The fungi under optical microscope, the disk diffusion test.
- B. The growth of *Aspergillus Flavus* in petry plate, the fungi under optical microscope, the disk diffusion test.
- C. The growth of *Aspergillus Niger* in petry plate, the fungi under optical microscope, the disk diffusion test.
- D. The growth of *Aspergillus terrus* in Petry plate, The fungi under optical microscope, the disk diffusion test.
- E. The growth of *Penicillium* sp. in Petry plate, The fungi under optical microscope, the disk diffusion test.
- F. The growth of *Fuzarium* sp. in Petry plate, The fungi under optical microscope, the disk diffusion test.

Nano-materials Effect

Nano SiO₂: There are microorganisms colonizing on Si-O so nano silica has affected some microorganisms as *A. Fumigatus* (the diameter of clear zone) was 2cm that assured the success of nano material in stop the growth of *A. Fumigatus* Figure 2 Nano Ca(OH)₂: assured it is success in consolidation and creating membrane helps in fumigation currently in this study with its circumstances the nano calcium hydroxide affected *A. Fumigatus* Fig.2 as the diameter of clear zone was 1.5cm *A. Fumigatus* and according to terrus with the diameter of clear zone was 2cm the nano material stopped the growth of *Terrus*. Figure 2 Nano CaCO₃: there are microorganisms colonizing CO₃ as microorganisms attacking calcined stone. so nano calcium carbonate hasn't affected any microorganisms. It is well known that the mixture of Titanium dioxide nanoparticles or zinc oxide nanoparticles with calcium hydroxide nanoparticles giving a big protection from the microorganisms specially the calcined stones but currently in this study with its circumstances the nano calcium carbonate affected fungi as *A. Fumigatus* Figure 2 with the diameter of clear zone 2.5cm and *Niger* Figure 2 with the diameter of clear zone 2.5cm that means the nano calcium carbonate stopped the growth of them.

Nano TiO₂: Using Titanium dioxide 50% with calcium hydroxide in nature light or inside artificial light they affected the *Penicillium* and *Niger* [6]. Currently in this study the nano titanium dioxide affected all founded fungi as *A. Fumigatus* with the diameter of clear zone 1.9cm, *Aspergillus Flavus* with the diameter of clear zone 2cm, *Aspergillus Niger* with the diameter of clear zone 1.9cm, *Aspergillus Terrus* with the diameter of clear zone 1.3cm, *Fusarium* with the diameter of clear zone 2cm and *Penicillium* specie with the diameter of clear zone 1.5cm. Titanium dioxide is the most successful nano material in this study as it affects *A. Fumigatus*, *Aspergillus Flavus*, *Aspergillus Niger*, *Aspergillus Terrus*, *Fusarium* and *Penicillium* specie. So nano titanium dioxide must be mixed with material as follows:

a. The calcium hydroxide which is popular used for consolidation must be mixed with titanium dioxide which proved its success in fumigation with all founded kind of fungi in the current mural painting,

b. And according to use nano silica and calcium carbonate as filler must be also mixed with titanium dioxide which proved its success in fumigation.

Conclusion

This study indicates that the paintings of saint Jeremiah monastery have been colonized by fungi which seem to be responsible for the biofilm formation in the painting there seems to be a strong relationship between the most deteriorated areas with structural damage such as cracking and detachment of the paint layer and a higher microbial contamination. The fumigation using different nano materials especially the most successful material such as nano titanium dioxide which must be mixed with calcium hydroxide, or with nano fillers silica and nano calcium carbonate to prevent any growth of microorganisms.

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