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Synthesis Nano Zinc Oxide Materials and Their Activity on Fungus Growth

Abstract- In this study Zinc oxide Nano particles was synthesized by employing sol-gel technique to explore novel antifungal agents to beat the developed of fungi resistance to many conventional fungicides. Prepared of Zinc oxide Nano particles were investigated via using Scanning Electron Microscopy (SEM, the VEGA Easy Probe), X ray powder diffraction (XRD), FTIR spectrum, The antifungal properties of ZnO nanoparticles were tested against *Fussarium spp.*, *Alternaria spp.*, *Penicillium spp.*, and *Aspergillus spp.* by using agar diffusion method. The percentage of growth was lowest comparison with control for all types of fungi when used zinc oxide Nano in different concentration.

Keywords- ZnO nanoparticles, sol-gel method, antifungal agents, percentage of growth.

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1. Introduction

The increasing resistance of microbial organisms to multiple antibiotics, and continuing emphasis on healthcare costs, has led researchers to develop new antimicrobial substances that are effective against drug resistance [1].

Whereas the diseases of Fungi are among the most major issues woodland recovery is confronting and they can now and again cause substantial misfortunes because of high death rates. A great parcel of the contagious fungi is seed-borne and is moved to woods nurseries through seeds and that move toward becoming on youthful plants. Aside from their seed-borne nature, soil-borne contagious pathogens are annihilating when attaching youthful plants with timberland plantation [2]. *Fusarium spp.* is ordinarily present on coniferous seedling establishes and in the rhizosphere, and also, can in like manner be accountable for pre-and post-rise damping-off upon seed germination [3], and can carry on as fungal pathogens bringing on damping-off and root mold of youthful coniferous plants, That leads to serious yield and financial misfortunes in backwoods nurseries. This fungal genus is pervasive in many compartments and bare root youthful plantation on healthy and unhealthy coniferous plants, in incubation soils, and on coniferous seeds of various types [4]. Additionally, Species of the variety *Alternaria spp.* Considered regular area growths, as both

saprotrophic and types of plant pathogenic that also impact grain, vegetables, and crops of fruit in the region or source decay postharvest. Moreover, a lot of *Alternaria spp.* possess a high toxigenic potential as It has the ability to produce secondary metabolism toxins called mycotoxins [5] Which is not dealt with by humans and animals. The important mycotoxins are alternariol's, altenuene, adjust poisons and tenuazonic corrosive [6] that is the cause of different types of cancers [7].

While *Penicillium* is outstanding and one of the most widely recognized organisms happening in an assorted scope of habitats, from soil to vegetation to air, indoor environments and different food products. It has an overall dissemination and an expansive financial effect on human life. Its fundamental capacity in nature is the disintegration of natural materials, where species cause destroying decays as pre-and postharvest pathogens on sustenance crops [8]. As well as producing a different range of mycotoxins [9].

Aspergillus niger is one of the known important fungus species belonging to the class *Aspergillus*. And is the main reason of the disease of black mold on specific vegetables and fruits like peanuts, grapes and onions, Which is also the major cause of food contamination. *Aspergillus niger* has the ability to produce certain kinds of fungal toxins that may cause hepatocarcinogen,

nephrogenic immunological for the human. Besides, this fungus is also responsible for decay maladies in some fruits and vegetables [10]. It is hard to control fungal growth since this organism consists of great resistance to a lot of conventional fungicides such as benzimidazoles and dicarboximides [11].

Developing resistance microbial strains against antibiotics is one of the important challenges in treatment of diseases. The limited choice of antifungal agents is also one of the most challengeable problems about fungal diseases [12, 13].

To conquer this protecting, it's imperative to investigate different antifungal factors, which may supplant immediate control systems. In last year's, Nanoparticle (NP) materials have gotten expanding consideration because of their special chemical and physical characterization which contrast essentially from their ordinary examples [14]. In comparison with organic antibiotics, inorganic antibacterial metal elements

(e.g., silver (Ag), copper (Cu) and zinc (Zn) have attracted great attention due to their perfect stabilities, superior broad-spectrum antibacterial properties, relatively low toxicity to human cells and low risk of producing resistant strains [15]. Zinc oxide (ZnO) has generated considerable attention because of its a unique material that exhibits semiconducting and piezoelectric dual properties. magnetic, antibacterial and Its nanostructures exhibit interesting properties very promising photocatalyst for photocatalytic degradation of water pollutants: high catalytic efficiency and strong adsorption capacity, owing to its high activity, low cost and environmental friendly feature. [16, 17]

Furthermore, zinc is a metal component basic to the health of living organisms and also its one of the important supplement for zinc. Also ZnO NPs have great biocompatibility to living organism's cells [18]. The antibacterial and antifungal activity of bulk ZnO powders has been demonstrated already [19]. In farming, zinc mixes are essentially utilized as fungicides [20]. There are several ways to prepare Zinc Oxide nanoparticles approaches including sol-gel processing homogeneous precipitation mechanical milling, microwave method, spray pyrolysis, thermal evaporation and chemical synthesis [21].

2. Material and method of work

1. Preparation of Zinc oxide Nano particles

Zinc oxide nanoparticles were prepared via utilizing the sol-gel technique (Sol-gel technology

is a chemical solution process in which the dispersed phase is so small (1 to 1000nm) that gravitational force are negligible and interactions are dominated by short range force, such as van der Waals attraction and surface charge and too it is based on the hydrolysis of metal oxide precursors and allows mixing of elements into an atomic level without the need for very high processing temperatures, with a high degree of porosity) [22, 23]. When prepared ZnO NPs, we dissolved 8 g of Sodium Hydroxide in 10 ml of (DW) distilled water and 2 g of Zinc acetate dehydrate were dissolved in 10 ml of distilled water after weighted this materials utilizing a weighting balance and measured distilled water via a measuring cylinder. The solution was blended with a continuous stirring for around five minutes. After mixed the solution well, sodium hydroxide solution was mixed with the solution containing zinc acetate by addition its drop after drop with a continuous stirring via a magnetic stirrer for five minutes. White precipitate formed after the reaction completed, then filtered the solution and dry the matter in the oven at 70°C for 24h and later grind to become white powdered. As shown in figure (1). As shown in figure (1).

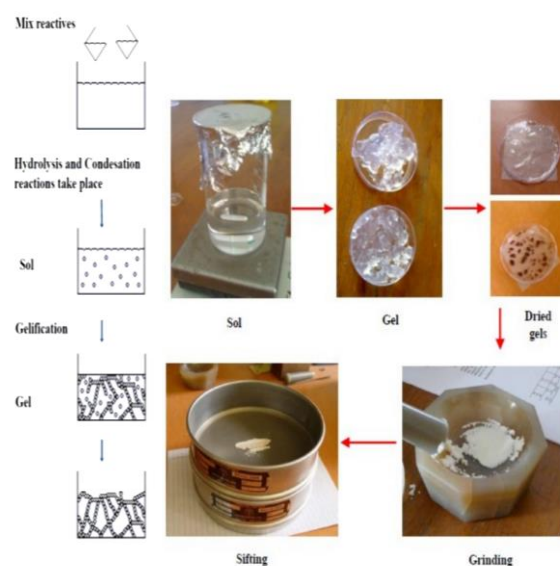


Figure (1): Show Diagram Process to the Synthesis of ZnO Nano Particles

II. Preparation of Fungal Culture

Four types of fungi were used in this research its *penicillium spp.*, *Fusarium spp.*, *Alternaria spp.* and *Aspergillus niger* that's culture in PDA from (3-7) days obtain from Nanotechnology and Advanced Materials Research Center (NAMRC).

III. Characterization and Bioassay

Scanning electron microscopy (SEM, the VEGA Easy Probe), X ray powder diffraction, FTIR spectrum and anti-fungal activity was tested for identification of zinc oxide and clarification of important in inhibition of fungal growth. Three concentration of zinc oxide Nano particles (2.5µg/ml , 5µg/ml and 10µg/ml) added to potato dextrose agar separately and sterilization in autoclave at 121 C for 15 min. ,then pour in petri dishes and a well put in middle of agar at 6mm in diameter for culture of fungi and incubated at 24 °C for (3,7,12,15) days.

3. Results and Discussion

I. Results of Zinc Oxide Nano Particles

The FTIR spectrum of zinc oxide nanostructures are shown in Figure (1) that was possessed in a chain absorption summit from 400 to 4000 cm⁻¹. Bands of 417, 437, and 740 cm⁻¹ spectra of ZnO and 1540.1, 1492.9, 1357, 1043.49, 833.25 a companion with expansion oscillation of crystalline hexagonal ZnO expansion vacillation. The broad absorption peaks at the range (3200-3600) C-1 belong to the presence of hydroxyl group of vibration at the surface of ZnO samples. The FTIR observation backing the XRD outcome [24].

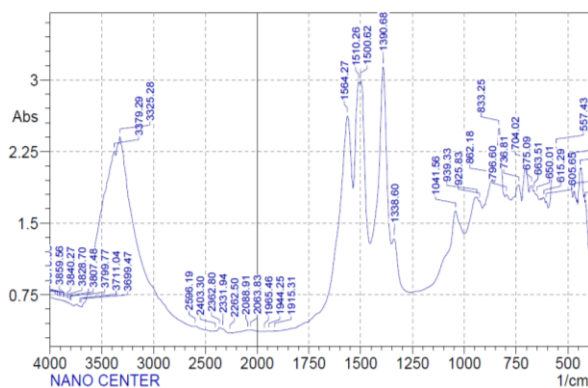


Figure 2: FTIR spectrum of zinc oxide nanostructures

Figure (3) shows XRD pattern of synthesized ZnO. The pattern shows that all the diffraction peaks indexed to the hexagonal phase of pure ZnO with a quartzite structure. Also, there is no diffraction peaks belong to free Zn or some other debasements can be seen in the spectrum, this in result demonstrates to the excellent quality of the synthesized products. Moreover, the clear and sharp peaks additionally uncovered that the ZnO Nano structure has a high crystalline quality [24].

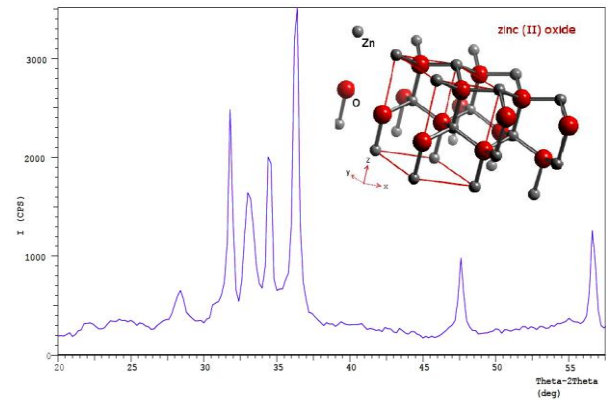


Figure. 3: XRD patterns of ZnO nanostructures.

SEM images at different magnification where SEM micrograph of the ZnO nanostructure shown in Figure (4). The ZnO structures have an average diameter of 100 nm and have found different in size, length and shape, in the Nano rods. And in another region, we have notice that pack of Nano rods is ranged with each other in a various direction [24].

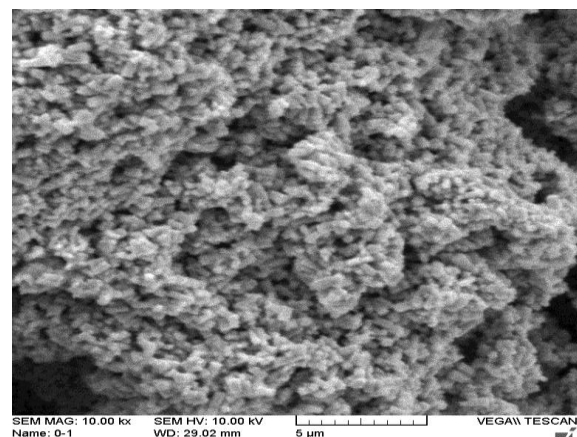
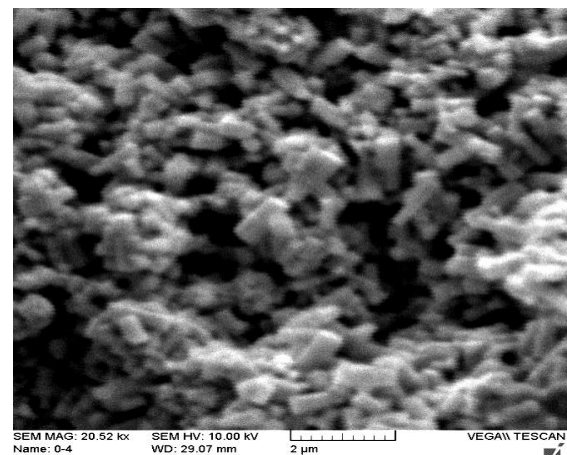


Figure. 4: SEM of ZnO nanostructures.

II. Results of Anti Fungi Activity

The effect of zinc oxide Nano particles on *Fusarium spp.* Show in Fig.5 and Fig.6 and the results explain ability of zinc oxide to inhibition of fungi growth in different period (3, 7, 12, 15) day for all types of fungi. The Percentage growth of fungi *Fusarium spp.* in concentration (0.25 , 0.5 , 1.0) $\mu\text{g/ml}$ of Zinc oxide Nano particles compared with normal growth of fungi for (3 days) was (100, 75,75) % , for (7 days) was (100, 75,75) % , for (12 days) was (89, 56,33) % and for (15 days) was (100, 44,25) % .

While Percentage growth of fungi *penicillium spp.* show in Fig.7 and Fig.8 was in concentration (0.25 , 0.5 , 1.0) $\mu\text{g/ml}$ of Zinc oxide Nano particles compared with normal growth of fungi for (3 days) was (100, 86,86) % , for (7 days) was (44, 22,20) % , for (12 days) was (60, 30,24) % and for (15 days) was (63, 31,25) %.

This results agree with [25] that suggest ZnO Nanoparticles may damage and harm the conidia of fungi. Subsequently, the growth of fungi was mightily prevented and give *Fusarium oxysporum* and *Penicilium expansum* hypha with ideal net form and soft surface would lost their softness and seemed swollen and disintegrated when treated with ZnO NPs .

And the results of Percentage growth of fungi *A.niger* in concentration (0.25 , 0.5 , 1.0) $\mu\text{g/ml}$ of Zinc oxide Nano particles compared with normal growth of fungi appear in Fig.9 and Fig.10 for (3 days) was (100, 86,86) % , for (7 days) was (75, 50,38) % , for (12 days) was (86, 43,29) % and for (15 days) was (94, 59,35) % (

G. Baskar et al. (2013) explain The various functional groups related were observed to be C=O, C-N, N-H, O-H, N=O makes the integrated zinc oxide Nano particles as the powerful antimicrobial factors. Prepared zinc oxide Nano particles its can utilized as powerful antifungal

factors. This Nano particles were added to the sustenance to decrease the food toxicity impact by the different *Aspergillus spp.* , that is lawfully confirmed [26].

While Percentage growth of fungi *Alternaria spp.* show in Fig.11 and Fig12 in concentration (0.25 , 0.5 , 1.0) $\mu\text{g/ml}$ of zinc oxide nanoparticles compared with normal growth of fungi for (3 days) was (80, 47,47) % , for (7 days) was (100, 56,25) % , for (12 days) was (100, 59 ,29) % and for (15 days) was (100, 62, 34) % and Antifungal impact of ZnO Nano particles on some pathogenic microorganisms like *Alternaria spp.* was clarified via [27] That watched a noteworthy diminishment in mycelia increase and spore up growth incubated with ZnO nanoparticles.

The inhibitory impact of nanoparticles may be because of the arrival of metabolites and extracellular enzymes this worked as a factor for survival when exposed to stress from harmful compounds and in differences temperature that shown in the situation of fungus *Trichoderma reesei* and a lot of microorganisms [28].

Antimicrobial affectivity of several nano particles of silver has additionally been mentioned in a few fungi like fungi that decayed wood, a lot of phytopathogenic fungi and *Fusarium spp.* It's likewise watched that antifungal effectiveness that responsible of pathogenesis might is because of concealment of Poisons and enzymes utilized by the fungal pathogenic species [29].

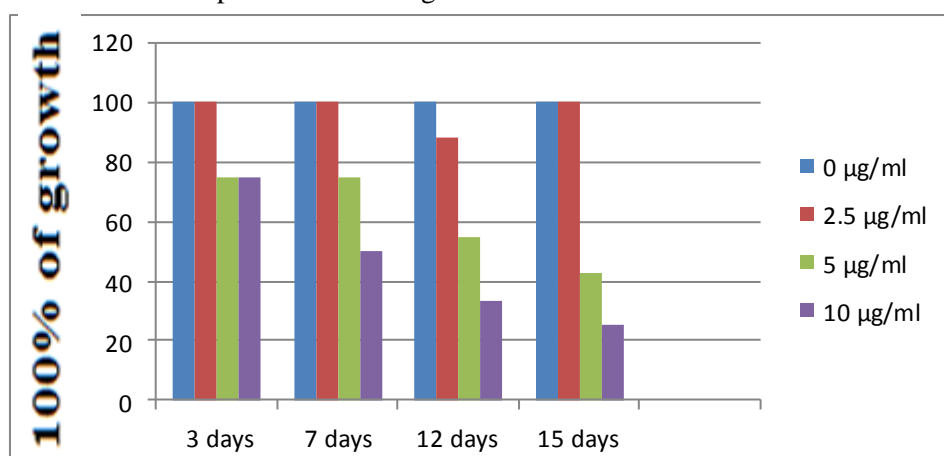


Figure.(5) :percentage growth of *Fusarium spp.* Comparison with control growth

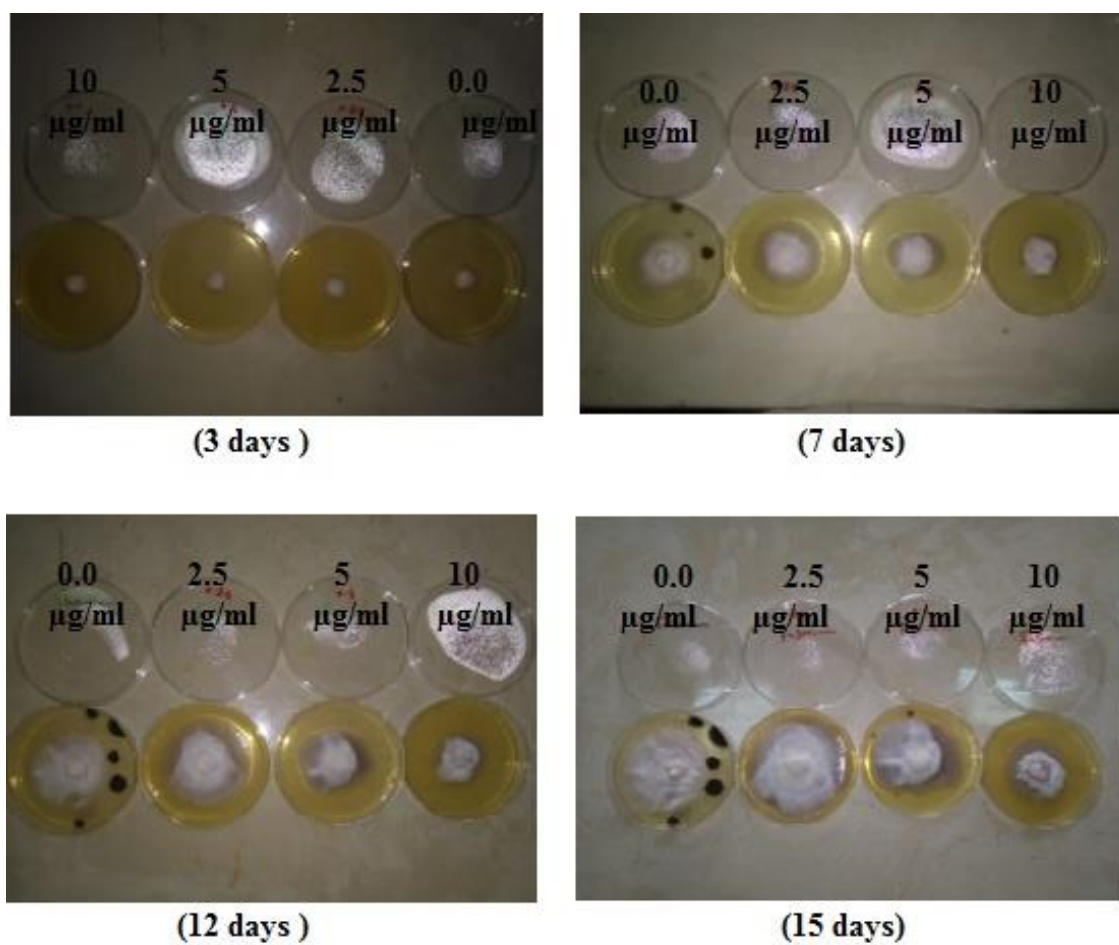


Figure.(6) : growth of *Fusarium* spp. In different period and different concentration

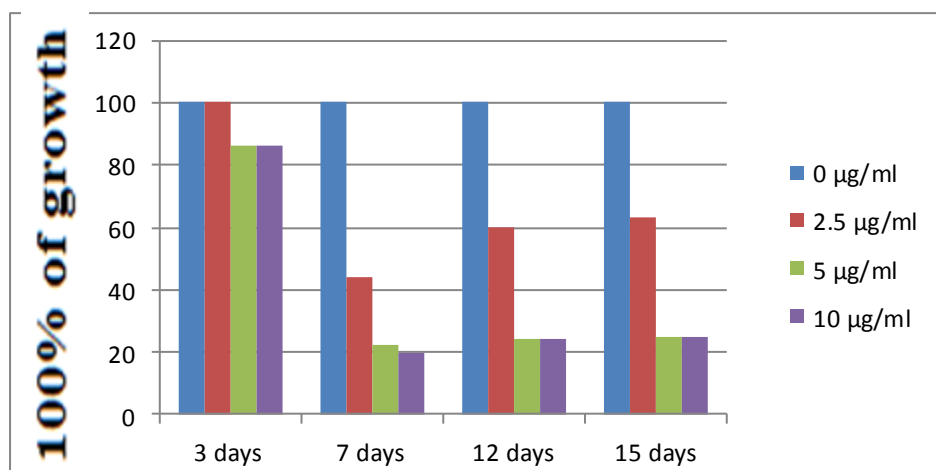


Figure (7) percentage growth of *Penicillium* spp. comparison with control growth

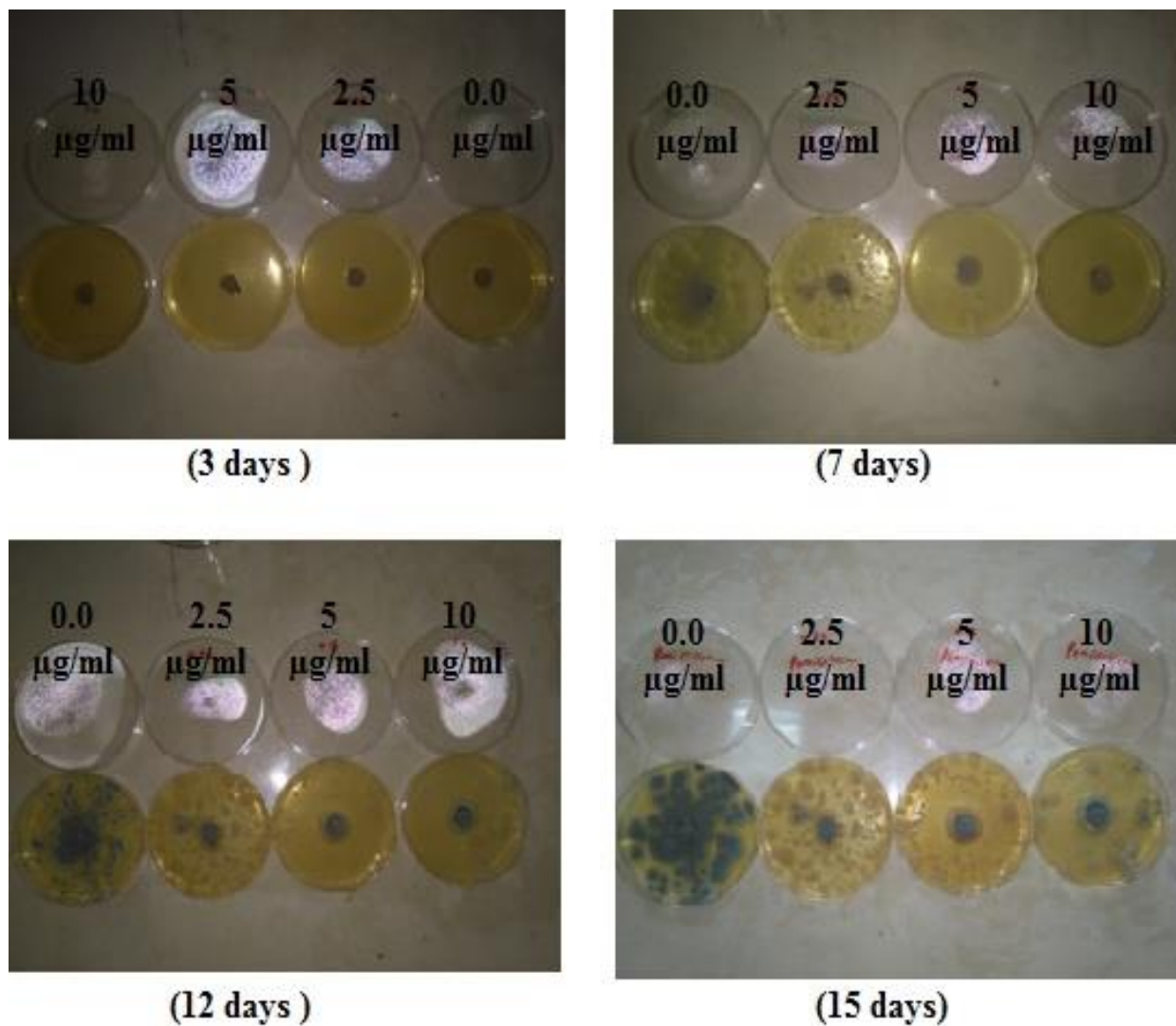


Figure.(8) : growth of *Penicillium* spp. in different period and different concentration

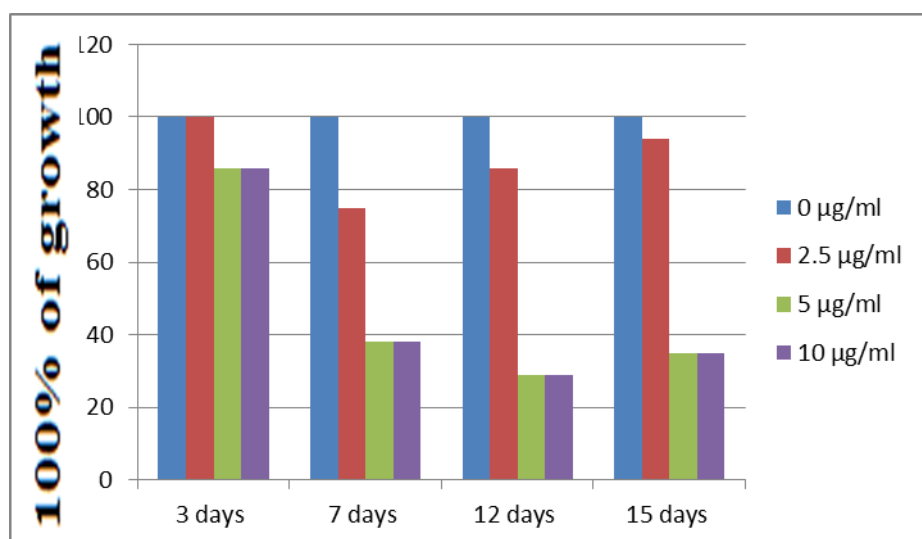


Figure (9) percentage growth of *Aspergillus niger* comparison with control growth

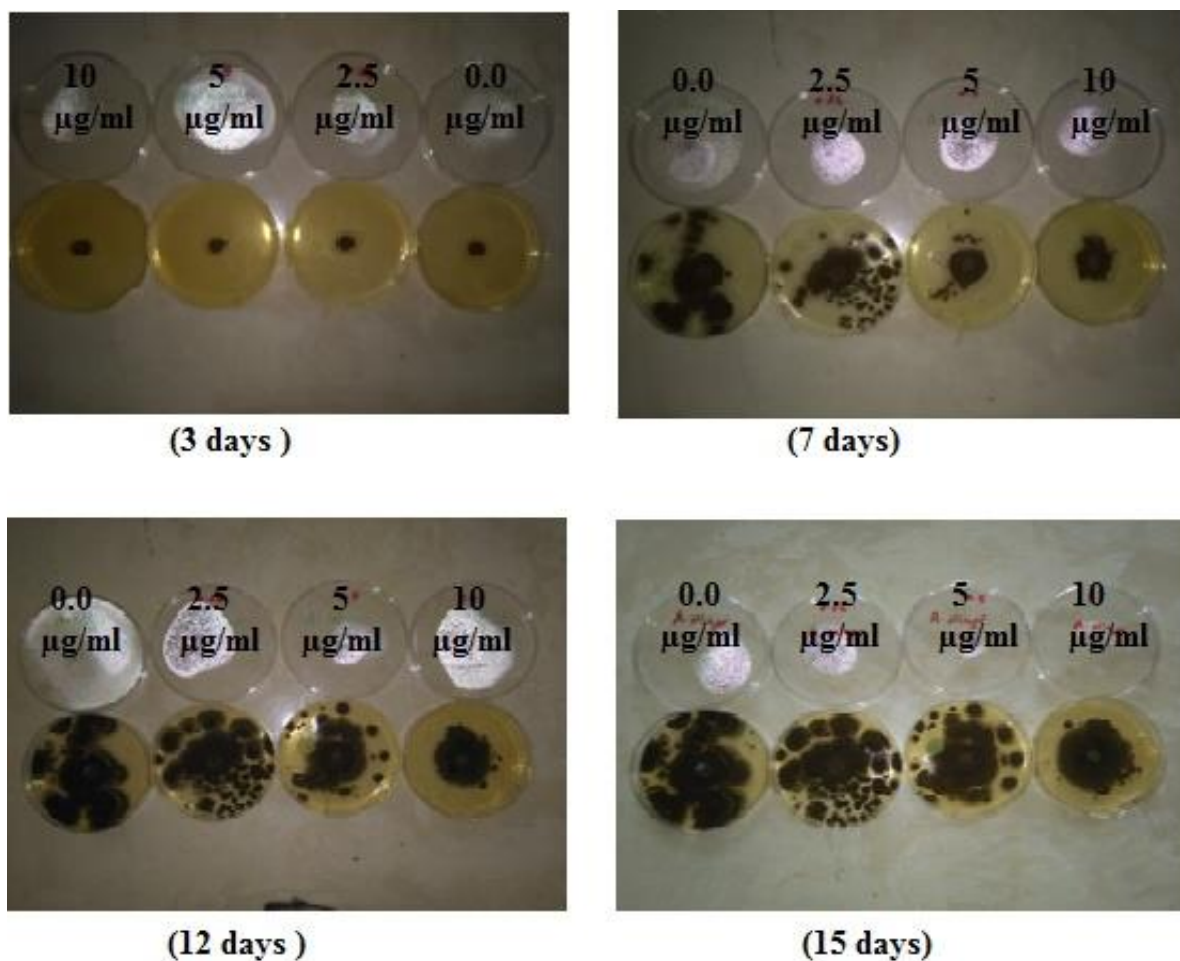


Figure.(10) : growth of *Aspergillus niger* in different period and different concentration

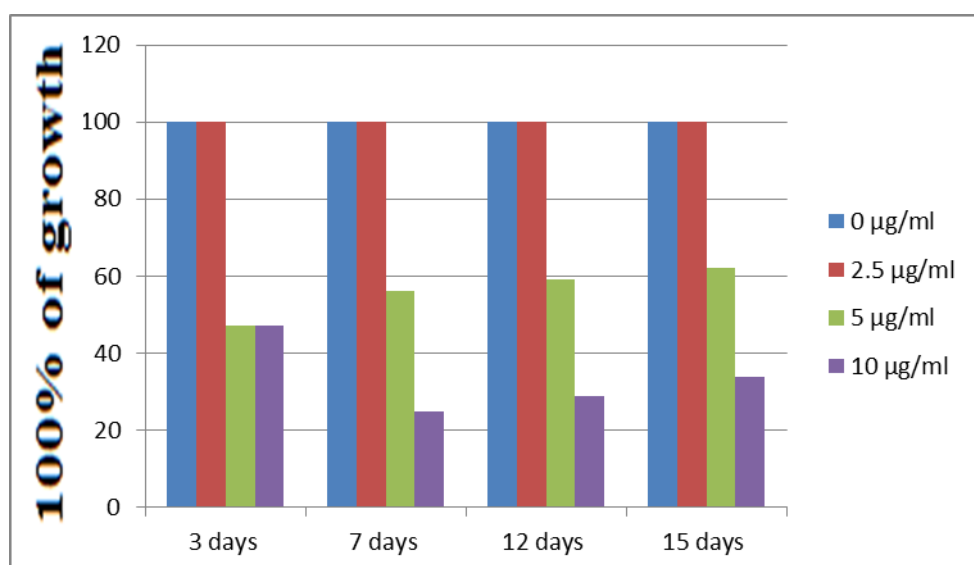


Figure.(11) :percentage growth of *Alternaria spp.* Comparison with control growth

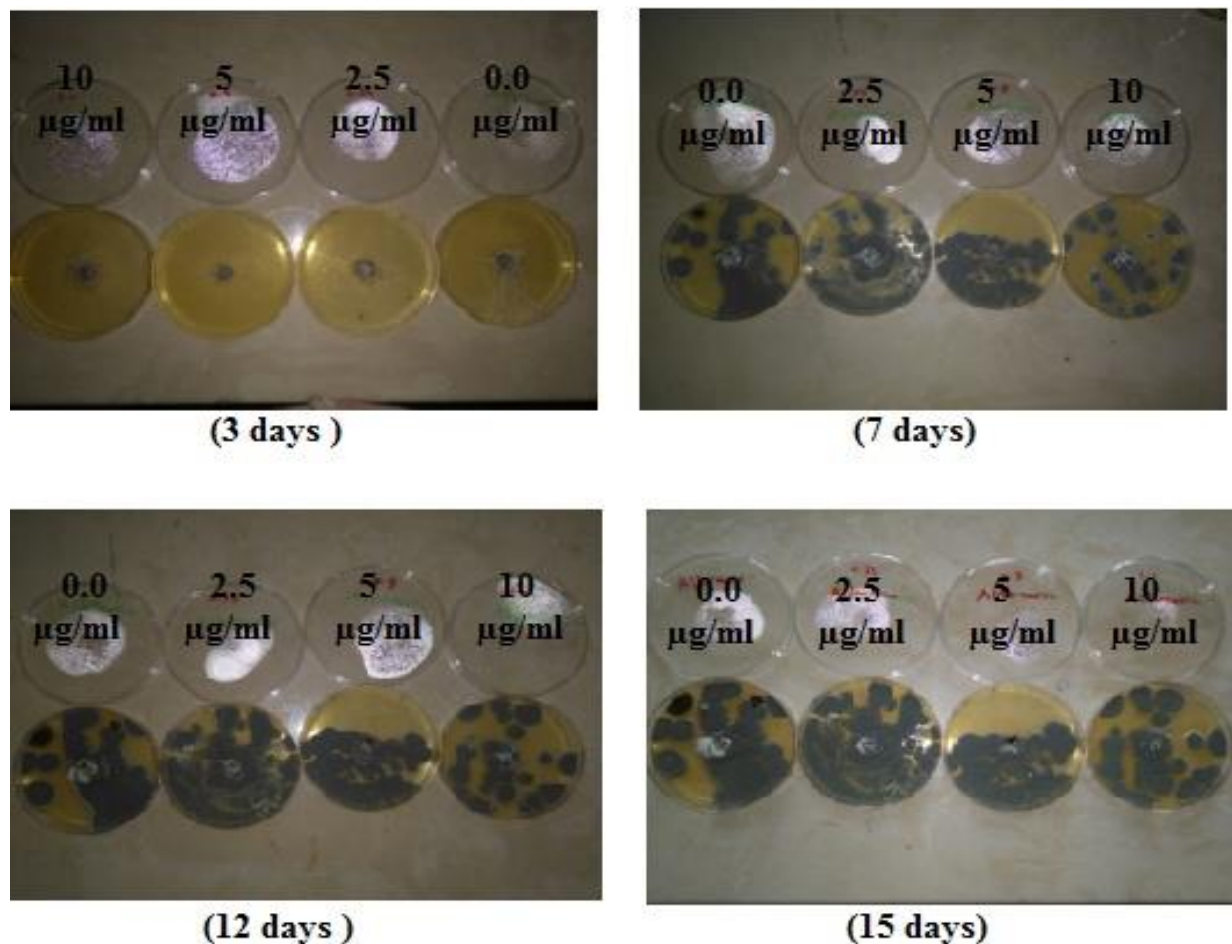


Figure.(12) : growth of *Alternaria* spp. In different period and different concentration

4. Conclusion

The results of this research appeared ability of ZnO Nano particles prepared by sol-gel method to effect on a lot of fungi when used in different concentration.

5. Acknowledgement

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