INFLUENCE OF ACTUAL ACIDITY (PH) AND TEMPERATURE ON THE FORMATION OF SILVER NANOPARTICLES BY ASPERGILLUS NIGER BDU-A4

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Abstract. In this work, the formation of silver nanoparticles was investigated by Aspergillus niger BDU-A4 at different acidity (pH: 4.8; 5.8; 7.4; 8.9 and 9.9) and temperature (20; 25; 30; 37 and 45°C). For this purpose, biomass suspensions of Aspergillus niger BDU-A4 was prepared and incubated in conjunction with Ag-nitrate salt on the given conditions. As a result of UV-Vis absorbance spectrum it was found that maximum amount of silver nanoparticles is produced in pH 7.4 and 37°C.

Keywords: Aspergillus niger, silver nanoparticles, influence, pH, temperature.

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1. Introduction

The synthesis of metal nanoparticles as well as copper, zinc, titanium, magnesium, gold, cadmium and silver has particular importance among the synthesized nanoparticles. So they are widely used in the medical field, optical and electronic equipment manufacturing, chemical industries, also agriculture in the form of elements and compounds. The environmental risks of nanoparticles were investigated also by researchers as well as their synthesis and application [1, 4, 10, 15, 17].

The optimal condition of metal oxides and salts was determined by different researchers for the synthesis of individual metal nanoparticles. Also the influence spectrum of the various physical and chemical parameters were well investigated that affecting the process of the synthesis of nanoparticles [14, 21].

The antibacterial and inhibitor effects of silver along with it’s metal properties make important the research of more effective ways to synthesize the silver. Although the synthesis of silver nanoparticles is available with biological and other methods. Green synthesis is more effective financially and environmentally. The different species of bacteria and fungi that are producent of nanoparticles have been investigated. Fungi synthesize silver nanoparticles from silver ions of their salts through intracellular and extracellular enzymes [2, 5, 11, 12, 20].

To increase the synthesis of silver nanoparticles with minimum investment by microorganisms, is required the selection of the optimum culture conditions and suitable physical parameters for their producents. Because, each of the selected active strains is able to synthesize in different amount and form of silver nanoparticles, depending on following factors: definition of biomass, pH, temperature, light intensity, nutritious environment, concentration, incubation period [3, 18, 22].
It was determined that smaller sized and spherical shaped nanoparticles are synthesized at higher pH, while at lower pH are formed relatively large, rod- or triangle-shaped nanoparticles. It was determined relation between the size of silver nanoparticles synthesized by fungi and temperature. So the increasing of the reaction temperature leads to the reduction of nanoparticle size and their monodispersity [3, 5, 8].

The optimal concentrations of biomass and substrate were studied by researchers to increase the production of silver nanoparticles on an industrial scale by fungi and bacteria. The optimal physico-cultural conditions that affect the synthesis of silver nanoparticles determined by Sonal and his collaborators. They have studied the effect of temperature and pH, biomass and substrate concentrations, as well as the effects of nutrient - rich environment, filtration level of suspension and intensity of light for optimal synthesis of nanoparticles by Fusarium oxysporum [3, 18].

Plant extracts have been investigated as nanoparticle producent and determined their optimal synthesis conditions during the study of rapid synthesis of silver nanoparticles [13, 16].

At previous studies the strains of mold fungus were isolated from soil samples and from the different rotten plant residues, identified and tested their ability to generate silver nanoparticles. It was studied that Aspergillus niger BDU-A4 active strain forming silver nanoparticles intensively. Also, it was studied the relation of silver nanoparticles synthesis by Aspergillus niger BDU-A4 with the incubation period and the amount of biomass [7-9].

The main purpose of the presented study is to test the acidity (pH) and temperature on the formation of silver nanoparticles by Aspergillus niger BDU-A4.

2. Materials and methods

**Fungal culture:** Weekly cultures of Aspergillus niger BDU-A4 that was taken from the cultures collection of Baku State University.

**Reagents:** For the synthesis of silver nanoparticles as the primary source 1mM AgNO₃ solution, also the solution of 1 N HCl and the solution of 1 M NaOH for the preparation of solutions at relevant pH, distilled water for the preparation of solutions. The composition of the nutrient medium (g / l): - Sucrose-20; NaNO₃-3; K₂HPO₄-1; MgSO₄·7H₂O - 0.5; KCl- 0.5; FeSO₄·7H₂O-0.01.

**UV-Vis spectral analysis of silver nanoparticles:** For UV(UV-Vis) spectrophotometer was used "Analytic Jena" spectrophotometer (model: Specord - 250 pulse, Jena/Germany). UV spectrophotometer (UV-vis) is a valuable tool for determination, characterization of nanomatherials. UV-vis is equipment which calculates the dispersed or absorbed light of a given sample at the solution. The suspension of silver nanoparticles has dark golden-yellow color through the surface plasmonic resonance. These are the motions of electrical conductance as a response to electromagnetic waves. Silver nanoparticles have a characteristic absorption spectrum at the "vision" circle of UV-vis. Spectral characterization of silver nanoparticles depends strongly from their size, shape, interparticular gaps and surroundings [17].

**Synthesis of silver nanoparticles:** The fungal culture was incubated in 250 ml Erlenmeyer flasks that contain 100 ml nutrient medium, at 28°C, at 120 rpm, within 120 hours. Received biomass washed by distilled water, it was incubated again within 24
hours with 100 ml of distilled water at 28 °C, to remove of nutrient from cell completely. Then the suspension is drained through Watman filter paper (№1) and the gained wet biomass is weighted. For the synthesis of silver nanoparticles were prepared 100 ml of suspension that consists of 10 grams of biomass, 1 ml of distilled water and 1 mM AgNO₃ in 250 ml Erlenmeyer flasks. Suspensions were incubated at different pH and temperature in dark conditions.

**Selection of the optimal acidity and temperature for the synthesis of Ag nanoparticles:** In order to ensure fast and stable synthesis of silver nanoparticles by *Aspergillus niger* BDU-A4 has been studied the effects of following factors: medium acidity (pH), temperature. The experiments in the study of each of the physical and cultural indicators were carried out fourfold. The biomass was incubated in solutions with pH 4.8, 5.8, 7.4, 8.9 and 9.9 to study the effect of pH on the formation of nanoparticles. The suspension was incubated in a thermostat at stable temperatures of 20, 25, 30, 37 and 45°C to study the effect of temperature on the formation of nanoparticles.

3. **Results and discussion**

The formation of silver nanoparticles by *Aspergillus niger* BDU-A4 were investigated at different pH values (4.8, 5.8, 7.4, 8.9, 9.9). Obtained results of spectrofotometer analysis have been shown in Fig.1. According to results of UV spectra, the synthesis of silver nanoparticles has been observed at all tested pH values. But it has been shown that the maximum synthesis of silver nanoparticles occurs in an alkaline medium (8.9 and 9.9 pH values). According to reduction of pH value from pH 7.4 toward acidic environment the amount of synthesized nanoparticles are decreased, and when the pH level reaches 4.8, it results in aggregation of particles. After incubation the flocculation of particles was not observed in none of the solutions with different pH values. However, after one week the aggregation of particles has been occurred in a solution of 4.8pH. But, in solutions of alkaline was observed stable diffusion of the nanoparticles.

![Fig.1. UV spectra of silver nanoparticles formation by *Aspergillus niger* BDU-A4 depending on acidity (pH)](image_url)
The formation of silver nanoparticles by *Aspergillus niger* BDU-A4 were studied at different temperatures of 20, 25, 30, 37, 45°C. Spectrophotometrical analysis of results are shown in Fig. 2. It has been shown that absorption of silver nanoparticles increase gradually as the temperature increases.

**Fig. 2.** The effect of temperature on the formation of silver nanoparticles by *Aspergillus niger* BDU-A4

The highest density of nanoparticles is observed at 37°C, but reduced at 45°C respectively.

4. **Conclusion**

The effect of different pH values were analyzed in UV-Vis spectroscopy and it was revealed that, the optimum acidity value for the highest synthesis of silver nanoparticles is pH 7.4. According to UV spectrophotometrical analysis of temperature effect, the optimum temperature was 37°C for getting the highest density of silver nanoparticles.

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**References**


