

Anticancer Activity of Silver Nan Particles against Cell Line MCF-7–Human - Breast Cancer through MTT Assay

C. K. Senthilkumaran^{1*} and S. Sugapriya²
sks.apsc@psgtech.ac.in, sugapriya0314@gmail.com

¹Department of Applied Science, PSG College of Technology, Coimbatore, India.,

²Department of Chemistry, Coimbatore Institute of Technology, Coimbatore, India.

Abstract. Silver nanoparticles were synthesized by simple chemical precipitation method demonstrated a considerable activity of anticancer against cell line MCF 7 human - breast cancer at various doses. Above 60 % of cell death has been observed in maximum concentration level of 1000 μ L. Results showed that at higher concentration levels have significant cell mortality. After the 48 hours of incubation the inhibitory effect was observed. Consequently, 50% on killing the cancer cell decides the inhibitory. Concentration I, C50 of the prepared silver nanoparticles in case of cell line MCF-7 human breast cancer, holds at round 846 μ L. In addition, the light microscopic observation of control and different concentration of silver nanoparticles sample treated cell lines MCF-7 was studied.

Keywords: Silver nanoparticles, Cytotoxicity, MCF-7 and MTT assay.

1 Introduction

Cancer cells exhibit an uncontrolled division, which would lead to the increasing the number of dividing cells [1,2]. It is an extraordinary kind of tissue growth but due to its steady growth progress it can be a preventable disease [3]. The utilization of cultured tissues and cells has risen due to *In vitro* cytotoxicity testing [4]. This testing procedure reduces the use of laboratory animals [5]. *In vitro* cytotoxicity testing cultures can be grown under an optimized environment like conditional temperature with proper humidity and supply of controlled oxygen, pH etc resulting to minimizing the experimental errors. Recent decade Breast cancer is the utmost frequently pinpointing cancer for women worldwide. Cytotoxicity or Anticancer has been expounded as the tissue cell dispatch property of a chemical compound. Nowadays there is an increasing demand for anticancer therapy [6]. To measure the cytotoxic properties MTT assay has been defined as rapid and reproducible manner used to screen the cytotoxicity drugs.

The nanoparticles like Ag, Au, ZnO, TiO₂, NiO etc., have significant properties due to their cytotoxic activity [7, 8]. Nanoparticles or nanotechnology provides a more prominent and significant part in the treatment of cancer cells [9]. In this research work, we focused on the cytotoxicity assessment by the synthesized nanoparticles on human - breast cancer cells. Ag nanoparticles synthesized by chemical precipitation method are taken to anticancer activity.

2 Process With The Reagents

2.1 Materials

3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl-tetrazolium bromide, a yellow tetrazole, phosphate buffered saline (PBS), Dimethyl sulphoxide (DMSO), The cell chemical culture such as FBS – Fetal Bovine Serum and EMEM – Eagles Minimum Essential Medium from sigma Aldrich chemical.

2.2 Cell culture

The Michigan Cancer Foundation-MCF-7 human - breast cancer cell line cultured has been acquired out of NCCS - National Centre, for Cell. Science, Pune. The obtained cell was cultivated in EMEM containing FBS 10% and kept at 37°C, CO₂ 5%, air 95% and with 100% relative humidity.

2.3 Stock Solution

Stock solution was prepared by using the prepared nano powders like Ag nanoparticles. By taking the accurate weight, one mg of prepared samples was dissolved with 2 mL of double distilled or de-ionized water.

Cytotoxicity assessment of prepared samples using MTT assay based on the perception of mitochondrial dehydrogenase venture in the living cells. The cytotoxicity of prepared silver nanoparticles on the cell line MCF-7 human - breast cancer has been discussed by MTT Assay.

3 Principle

3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl-tetrazolium bromide, a yellow tetrazole in phosphate buffered saline has been cleared by mitochondrial dehydrogenase of feasible cells, releasing a measurable purple colour formazan crystals product. The formed purple colour formazan crystals production is inversely consistent to the degree of cytotoxicity corresponding to the feasible cell number.

4 Procedure

Concerning to make single cell suspension the monolayer cell has been detached along with trypsin-ethylenediaminetetraacetic acid (EDTA) and feasible cells have been calculated using a hemocytometer. Then it was diluted with phosphate buffered saline 5% to provide a density of 1×10^5 cells/mL. Concerning 100 μ L each disc about suspension culture has been sown in 96-disc sheet close to layer density of 10,000 disc. It has been hatched to grant in-favour-of unit loyalty at 37°C, CO₂ 5%, air 95% and with 100% relative humidity. Later 24 hours 100 μ L of medium was treated with different dosages 50, 100, 250, 500 and 1000 μ L prepared Ag nanoparticles. Each nanoparticles treated cells were then hatched or incubated at 37°C, CO₂ 5%, air 95% and with 100% relative humidity for 48 hours. In order to get results, there were control (medium without sample) and triplicate was maintained for all the prepared concentrations.

Subsequently, the cells have been subjected for MTT assay after the cell treatment process. 100 μ L of MTT in phosphate buffered saline has been prepared and 15 μ L of MTT was added in each prepared nanoparticle treated disc also hatched close to 37°C in-favour-of 4 hours.

Standard along MTT has been released as well as developed purple colour formazan crystals have been dispersible along 100 μ L, Dimethyl Sulphoxide. (DMSO) also devours around 570nm over ELISA disc reader. In-vitro cytotoxicity has been calculated at the percentage of viable cells at different concentration of sample relative to untreated (Control) cell. Optical density (OD) value has been exposed to segregate the proportion of cell inhibition by make use of below ritual,

$$\text{Percentage of, viability (\%)} = \frac{\text{Mean OD. value to experimental, sample.}}{\text{Mean OD. value to experimental control. (Untreated)}} \times 100$$

$$\text{Percentage of cytotoxicity (\%)} = 100, - \text{Percentage of viability (\%)}$$

Consequently, 50% on killing the cancer cell decides the Inhibitory. Concentration $I_{C_{50}}$ with the use about data generated plot of dose-response curve.

5 Microscopic Observation

MCF-7 cells have been grown and hatched or incubated with prepared nanoparticles sample belonging to $I_{C_{50}}$ concentration, the prepared Ag nanoparticles treated and untreated discs have been monitored under a light microscope to identify the morphological changes and recorded as photographed.

6 Results And Discussion

Silver nanoparticles have synthesized by simple chemical precipitation method demonstrated a considerable cytotoxicity upon cell lines MCF-7-breast cancer at various doses. Figure 1 depicts that 62.42 % of cell death was observed in maximum concentration level (1000 μ L) of the prepared Ag nanoparticles. More than 60 % of cell death was observed in maximum concentration level of 1000 μ L. Results showed that at higher concentration levels have significant cell mortality. The inhibitory effect has been observed later 48 hours of propagation. Table shows difference in the percentage of feasibility, cytotoxicity also inhibition with concentration of the Ag nanoparticles upon cell line MCF-7-breast cancer. Consequently, 50% on killing the cancer cell decides the Inhibitory. Concentration $I_{C_{50}}$ of the prepared nanoparticles upon cell line MCF-7-human - breast cancer, holds at round 846 μ L (Figure 1). In addition, the light microscopic observation of control and different concentration of Ag nanoparticles i.e., 50 μ L, 100 μ L, 250 μ L, 500 μ L and 1000 μ L treated cell lines MCF-7 is showed in Figure 2.

Table 1: MTT assay on Ag nanoparticles against cell line MCF-7

Concentration (Dosages) (μ L)	% inhibition	R ² value	$I_{C_{50}}$
50	9.2	0.982	846 μ L
100	17.34		
250	31.32		
500	43.71		
1000	62.42		

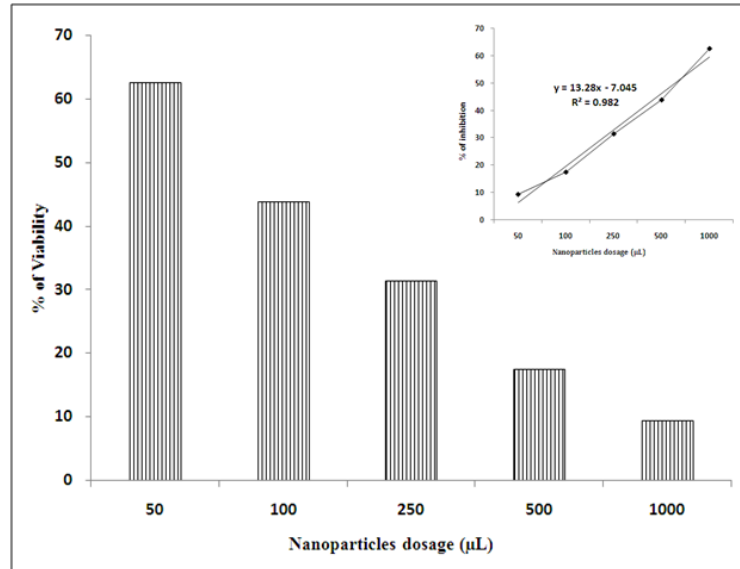


Figure 1 Cytotoxicity of Ag nanoparticles on cell line MCF-7

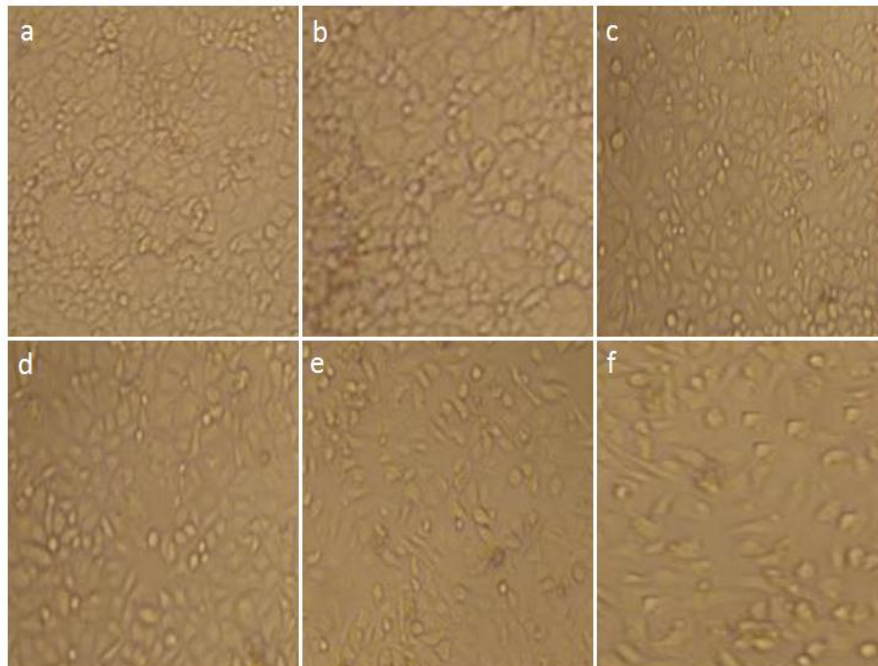


Figure 2: Morphological observation of different concentration of Ag nanoparticles treated cell line MCF-7 under light microscope. a) Control, b) 50µL, c) 100µL, d) 250µL, e) 500µL and f) 1000µL.

The silver nanoparticles have been synthesized by simple chemical precipitation method demonstrated a considerable anticancer activity upon cell lines MCF-7-human - breast cancer at various doses. Above 60 % of cell death was observed in maximum concentration level of 1000 μ L. Results showed that at higher concentration levels have significant cell mortality. After the 48 hours of incubation the inhibitory effect was observed. Consequently, 50% onkilling the cancer cell decides the Inhibitory. Concentration $I_{C_{50}}$ of the prepared Agnanoparticles upon cell line MCF-7-human - breast cancer, holds at round 846 μ L.

Silver nanoparticles was synthesized by chemical precipitation method shows anticancer activity or cytotoxicity on cell line MCF-7-human - breast cancer. In this report, concentration on dosage level increases with increasing the percentage of inhibition. It is due to the increase in the number of prepared samples of Silver nanoparticles. From here the percentage concerning the inhibition on cell line MCF-7- human -breast cancer depicts that at higher concentration levels has significant cell mortality. It decides the Inhibitory. Concentration $I_{C_{50}}$ the prepared Agnanoparticles upon cell line MCF-7-human - breast cancer, holds at round 846 μ L.

7 Conclusion

Silver nanoparticles was synthesized by chemical precipitation method shows anticancer activity or cytotoxicity upon cell line MCF-7-human - breast cancer. This report, concentration on dosage level increases with increasing the percentage of inhibition. It is due to the increase in the number of prepared samples of Silver nanoparticles. From this study the percentage concerning the inhibition on cell line MCF-7- human - breast cancer depict that at higher concentration levels has significant cell mortality. It decides the Inhibitory. Concentration $I_{C_{50}}$ the prepared Ag nanoparticles upon cell line MCF-7-human - breast cancer, holds at round 846 μ L.

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