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Study of Silver nano Particales for Anticancer Activity K

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ABSTRACT
Cancer is a leading causeof death world wide,Accounting for nearly 10 million deaths in 2020,According to world health organization.(WHO).About 50 million people worldwide living with cancer. Silvernanoparticles have gained significant attention in recent years for their potent role in cancer prevention and treatment. They contribute to cancer prevention by apoptosis induction,antiproliferative effectsand angiogenic effects.
By using green synthesis methods we can synthesize silver nano particales from bacteria ,fungi, plants and animals. In our present study we are synthesizing silver nanoparticles from bacteria through green synthesis and studying for their anti canceractivity. Samples are collected from oil contaminated soils near oil industry in Kamareddy district.
Sample are used to isolate bacteria. Isolated bacteria were grown in high amount and centrifuged at 10,000 rpm to get the bacteria.100 ml of supernatant containing bacteria is mixed with 1 ml of silver nitrate and incubated at 30c for 24 hours. After 24 hours yellow colored solution turned into brown indicating formation of silver nano particales
Anticancer activity of silver nano particales was evalued by using the MTT assay.formazan crystals formed were dissolved in DMSO, and absorbance was measured at 570 nm using spec photometer the biologically synthesized silver particales were observed at 430 nm.Anticancer activity of AgNp-SD against AS49 , MCF-7, HT29 and CHO-KI Cell lineswere measured.The study evaluated the biocompatibility and cytotoxic effects of biosynthesized silver nanoparticles (AgNPs) compared to cisplatin across four cell lines.

Keywords: Anti-cancer activity, Silver nano particles, MTT assay, AS49, MCF-7, HT29, CHO-KI

INTRODUCTION

The growing resistance of bacteria to conventional antibiotics has underscored the urgent need for alternative methods to combat microorganisms responsible for severe, life-threatening infections (Jindal et al., 2015; Yuan et al., 2017). At the same time, cancer remains a leading cause of global mortality, emphasizing the necessity for innovative therapeutic strategies (Jeyaraj et al., 2013). Among the diverse range of nanomaterials, silver nanoparticles (AgNPs) have attracted significant attention due to their exceptional physicochemical properties, including large surface area, stability, and versatility (Abdeen et al., 2014). These characteristics make AgNPs highly suitable for a variety of applications, such as antimicrobial treatments, cancer therapies, biomolecular detection, biolabeling, catalysis, and even microelectronics (Rai et al., 2009; Zarina and Nanda, 2014a; Golinska et al., 2014).

Green synthesis methods involving biological agents such as bacteria, fungi, plants, and algae offer a sustainable and eco-friendly alternative to traditional chemical and physical synthesis methods. These approaches are not only cost-effective but also harness the natural reducing and capping properties of biological agents. (Rafique M., 2017) Plants and plant extracts, in particular, are increasingly preferred due to their faster synthesis rates compared to microorganisms like bacteria and fungi. However, soil bacteria remain a promising option because of their adaptability and ability to produce nanoparticles with well-defined sizes and shapes, critical for biomedical applications.

AgNPs have demonstrated remarkable potential in both antimicrobial and anticancer applications, largely due to their ability to interact with and disrupt cellular structures. The environmentally friendly synthesis of AgNPs using soil bacteria offers a sustainable alternative to conventional chemical and physical methods. Soil bacteria, known for their resilience and adaptability, can reduce silver ions to form nanoparticles with defined sizes and shapes, which are crucial for effective biomedical applications.

This paper investigates the biosynthesis of silver nanoparticles using soil bacteria and evaluates their anticancer properties, aiming to contribute to the development of novel strategies in the fight against microbial infections and cancer.

MATERIALS AND METHODS

Soil sampling and bacterial isolation

Soil samples were gathered from highly oil-contaminated regions in kamareddy district, Telangana, at a depth of 5 cm beneath the soil surface. These samples were carefully placed in sterile containers and transported to the laboratory for further analysis.

Production of biomass

The bacterial isolates were cultured aerobically and incubated at38°C in Orbital Shaker with continuous agitation in 250 rpm. After 24 hours of growth, the microbial biomass was harvested and centrifuged at 10,000 rpm for 10 minutes.

Screening and Synthesis of silver nanoparticles

Synthesis of Ag nanoparticles was done by the reference to the Kalishwaralal et al., 2008 and Vaidyanathan et al. (2010). For the preparation of AgNPs, 100 ml of supernatant was mixed with one ml of silver nitrate solution (1 mM) and the reaction mixture was prepared without AgNO3 that used as a control test. The supernatant were then incubated at 30°C for 24 hours, protected from light to prevent any photochemical reactions. Over the incubation period, the solutions transitioned in colour from yellow to brown which indicates the formation of silver nanoparticles.

Anticancer activity

The anticancer activity of the test agent was evaluated using the MTT assay. Cells were seeded in a 96-well plate (50,000 cells/well) and grown overnight. After adding varying concentrations of the test agent, the plate was incubated for 24 hours at 37°C with 5% CO_2. MTT reagent (0.5 mg/mL) was then added, and the plate was incubated for 3 hours. Formazan crystals formed were dissolved in DMSO, and absorbance was measured at 570 nm using a spectrophotometer. The IC50 value was determined using the logarithmic equation Y=Mln(x)+CY = M\ln(x) + CY=Mln(x)+C, derived from the viability graph.

RESULTS AND DISCUSSIONS

In recent years, therapeutic and diagnostic strategies leveraging nanotechnology have shown remarkable promise in improving cancer treatment. (Ratan 2020;) The field of cancer nanotechnology has evolved into a multidisciplinary domain, bridging chemistry, biology, medicine, and engineering, and has been pivotal in advancing cancer detection, prevention, and therapy. (Zivyar2021) Nanoparticles (NPs) have become a focal point of scientific research, celebrated for their exceptional effectiveness and safety. (Wang 2010)The growing applications of nanotechnology in medicine have led to the recent approval of several nanotechnology-based anticancer drugs by the US FDA. These include Myocet™ (Perrigo, Dublin, Ireland), DaunoXome® (Gilead Sciences, Foster City, CA, USA), Doxil® (Johnson & Johnson, New Brunswick, NJ, USA), and Abraxane® (Celgene, Summit, NJ, USA). (Nguyen2011).

The study aimed to investigate the extracellular biological synthesis of silver nanoparticles utilizing bacteria isolated from the oil contaminated region of soil. Bacterial isolates were chosen for their performance under various stress conditions and biochemical characteristics, qualifying them for the biological extracellular synthesis of silver nanoparticles. The isolated bacterial were screened by green synthesis method. The aqueous silver ions (Ag+) were reduced to AgNPs when added to the cell-free supernatant of bacterial cultures within 1h after incubation in dark condition. During the incubation period, the yellow color was changed to brown colour and the control showed no change in colour shown in the figure (1)Silver nanoparticles (AgNPs) synthesized by bacterial isolate kmr009 werecharacterized by UV-Vis spectrum. The Surface Plasmon Resonance was found to bein the range of 200-600 nm silver nanoparticles. The biologically synthesized silver nanoparticles of kmr009 were observed at 430nm. The SEM picture was shown in the figure (1)

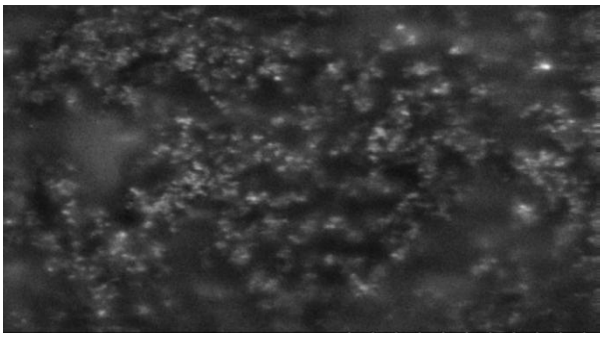

Figure 1

The study evaluated the biocompatibility and cytotoxic effects of biosynthesized silver nanoparticles (AgNPs) compared to cisplatin across four cell lines. Against the CHO-K1 cell line, serving as a control, AgNPs displayed minimal toxicity, with cell viability exceeding 99% at lower concentrations and 33.80% at the highest concentration, making the determination of an IC50 value unfeasible. In the HT29 colorectal adenocarcinoma cell line, AgNPs showed significant cytotoxicity, with a gradual reduction in viability to 33.80% at 100% concentration and an IC50 of 49.4%. Similarly, for the A549 lung carcinoma cell line, AgNPs induced high mortality with an IC50 of 33.78%, as cell viability dropped to 26.80% at the highest concentration. The MCF7 breast adenocarcinoma cell line exhibited lower sensitivity, with an IC50 of 90%, as cell viability reached 48.32% at the highest concentration. Finally, the AgNP-2 sample demonstrated toxicity after a 24-hour treatment period, with IC50 values of 90%, 49.4%, and 33.78% for the MCF7, HT29, and A549 cell lines, respectively. These findings indicate that AgNPs exhibit concentration-dependent cytotoxicity, with greater effectiveness against cancerous cell lines (HT29 and A549) compared to the relatively lower toxicity observed in normal (CHO-K1) and MCF7 cell lines.

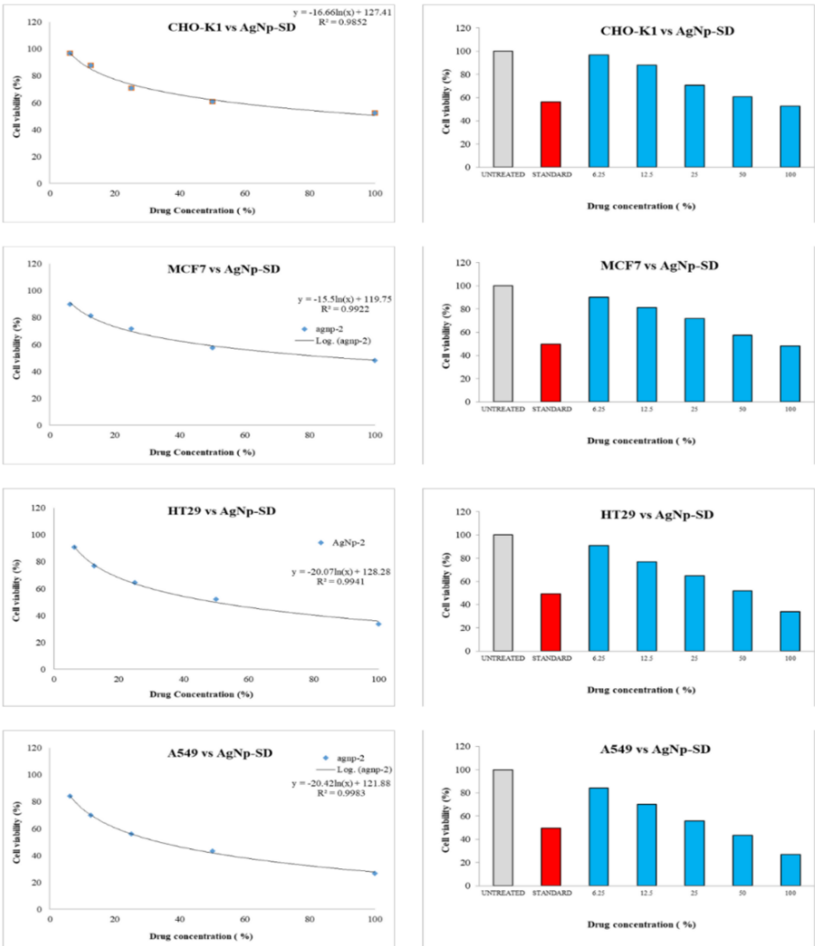

Figure 2: Anticancer activity of AgNp-SD against AS49 , MCF-7, HT29 AND CHO-KI Cell lines

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