



# STUDY OF ANTIOXIDANT ACTIVITY OF Ag, ZnO & Cu NANO PARTICLES

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## Abstract:

The antioxidant properties of three different types of nanoparticles were examined based on free radical scavenging, reducing power, and bleaching inhibition. The antioxidant capacity of nanoparticle samples was shown to be related to their biological and metal contents.

The nutritional value of nanoparticles is determined by the metal and its source. Nanoparticles have been re-positioned in modern medicine due to their well-known benefits such as "antioxidant" and "antimicrobial" activity. In the current study, nine different types of nanoparticles from three different types, namely Silver, copper and zinc oxide were tested against vitamin C to determine antioxidant activity to provide some possible mechanism by which it can be used in disease management.

The optical density of all the nanoparticles examined ranged from 0.257 to 0.507 with silver nanoparticles having the lowest optical density value of 0.257 followed by copper nanoparticles having an optical density of 0.507 and copper nanoparticles having the highest optical density (Optical density - 0.507).

## Introduction:

They contribute to the respiration of cells and other vital cellular processes, but they are also responsible for aging and the onset of disease. Unstable molecules with unpaired free electrons are known as free radicals. They are very reactive because the free electrons constantly try to form covalent pairs with other electrons. Free radicals deprive other molecules of their electrons during this process. It can harm molecules like carbohydrates, fats, proteins, and nucleic acids in addition to having an impact on how cells are regulated. Free radicals come from a variety of places, including the environment and the cells themselves. The production of free radicals occurs in aerobic organisms.

When metabolic processes are normal, the two main sources are cell respiration in the mitochondrial membrane and electron transfer in the plasma membrane. As a result of the ability of free radicals like superoxide to escape from the electron transport chain, mitochondria are the primary source of oxidative damage<sup>1</sup>. Cells have developed an intracellular antioxidant system to guard against intracellular damage caused by radicals. Free electrons are changed by proteins into a non-reactive form during this process. Antioxidants prevent delay, or hinder the oxidation of the biomolecules to regulate oxidative reactions. Certain components that safeguard and protect proteins are present in the main antioxidant enzymes. Non-enzymatic antioxidants can also neutralize radicals, for example, water-soluble substances such as Vitamin C, and glutathione, or fat-soluble substances such as Vitamin E, and beta-carotene. Synthetic antioxidants such as butylated hydroxyl toluene (BHT) and butylated hydroxyl anisole (BHA) have recently been reported to be harmful for human health. Thus, the search for effective, non-toxic, natural compounds with Antioxidative activity has increased in recent years. Recently, nanomaterials have started playing a fundamental role in human life and health owing to their substantial benefits in biomedical applications like medical imaging, drug delivery, disease diagnosis, cancer treatment, treatment of infectious diseases, treatment of neurodegenerative disorders, including Parkinson's disease, and so on. Moreover, the strong antioxidant property exhibited by some nanomaterials is opening the exciting potential to

develop new regimens with enhanced and targeted actions. For example, Gold, silver, copper, zinc, and selenium nanoparticles have been shown to possess the ability to reduce oxidative stress due to their efficient redox-active radical-scavenging properties.

## MATERIAL AND METHOD

Three types of nanoparticles viz. Silver, Zinc oxide & Copper were synthesized by reported method

Antioxidant activity has been checked by DPPH in different types of Nanoparticles using a spectrophotometer.

The free radical scavenging activity of nanoparticle samples was determined by the 1,1-diphenyl-2-picrylhydrazyl (DPPH) assay as described by Isla et al, with minor modification. The DPPH solution (50 mg/L) was prepared by dissolving 5 mg of DPPH in Ethanol (100 ml)<sup>[9][10]</sup>. A 0.75 ml of aqueous Nanoparticles solution of 20 mg/ml, was added to 2 ml of DPPH solution. The absorbance was measured at 517 nm after 15 min of incubation at 25°C. Methanol was used as a positive control. The ability to scavenge the DPPH was calculated using (I). The concentration of nanoparticle samples required to scavenge 50% of DPPH (IC<sub>50</sub>) was determined based on the ascorbic acid calibration curve (0 – 10mg/L).

Took 0.2 mL of Nanoparticles and mixed them with 1 mL of water. All three types of nanoparticle solutions were made in this manner, with a drop of each nanoparticle mixture deposited on strips of chromatography paper using a capillary tube. For 10 minutes, dry this drop of Nanoparticle combination. All of the strips should be immersed in the DPPH solution at the same time and dried. All of the white dots on the strip appeared at the point of the nanoparticle solution, where the antioxidant quality was present. 517nm of absorbance was measured. The positive control was methanol.

### ZnO

Antioxidant activity was carried out by DPPH assay using a modified method of Brand–Williams. DPPH (in its oxidized form) is a stable free radical with a purple color. In the presence of an antioxidant, which can donate an electron to the DPPH radical, the latter decays, and the change in absorbance at 520 nm is followed, which can be measured spectrophotometrically. Exactly 39.4 mg of DPPH was dissolved in 100 ml of methanol to get a 0.14 mM concentration of DPPH in the assay. Ascorbic acid standard stock I (conc. 200 mg/ml) was prepared by dissolving 2 mg of ascorbic acid and making up a volume of 10 ml with de-ionized water. For making the standard graph of ascorbic acid, a concentration range of 2, 4, 6, 8, and 10 mg/ml was used. In brief, to an 860 l of 50% methanol/ascorbic acid/test sample with various concentrations, 140 l of 1 mM DPPH was added, mixed, and incubated at 37 °C for 30 min. The absorbance was measured at 520 nm against a 50% methanol blank using a spectrophotometer. A control sample was maintained without the addition of the test sample. The antioxidant activity was measured concerning the standard ascorbic acid absorbance values. The actual absorbance was taken as the absorbance difference between the control and the test sample, and the IC<sub>50</sub> value was determined.

### Silver

The DPPH free radical scavenging assay was carried out in the same manner as before. Separate concentrations (12.5-200 g/ml) of synthesized AgNPs and standard vitamin C were mixed with 3 ml of a 0.1 methanolic DPPH solution and incubated in the dark for 20 minutes. After incubation, the absorbance of the solutions was measured at 517 nm against methanol using a UV-visible spectrophotometer (Agile microplate reader, ACTGENE, Inc., NJ, USA). As a control, the methanolic solution DPPH without a sample was used,

### Cu

CuNPs (10-100 mg/ml) dispersed in methanol were added to different test tubes, and the volume was filled with methanol to. 3 ml of methanol containing 0.004% DPPH was added to this and incubated for about 30 minutes at room temperature. The DPPH radical scavenging activity was calculated using the absorbance at 517 nm. The radical scavenging activity was calculated by reported method. For comparison, the DPPH radical scavenging activity of ascorbic acid was measured. The tests were carried out in triplicate.

## RESULTS :

### Zn

DPPH, a stable free radical with a characteristic absorption at 517–520 nm, was used to study the radical scavenging effects. The decrease in absorption is taken as a measure of the extent of radical scavenging. The radical-scavenging activity (RSA) values were expressed as the ratio of the percentage of sample absorbance

decrease and the absorbance of DPPH solution in the absence of extract at 520 nm. The ZnO nanoparticles were proven to inhibit the DPPH free radical scavenging activity with an IC<sub>50</sub> value of 10.8 mg/ml.

### Silver

DPPH is a stable compound that can accept hydrogen or electrons from silver nanoparticles. The results of DPPH radical scavenging activity tend to increase as the concentration of AgNPs increases. The maximum percentage inhibition observed in AgNPs was 55%, which was lower than the standard vitamin C concentration of 200 g/ml (93%). Similar findings with enhanced DPPH scavenging activity by AgNPs in *Piper longum* fruit.

### Cu

DPPH is a stable nitrogen-centered free radical with a distinctive absorption at 517 nm that changes colour from violet to yellow upon reduction. The antioxidants react with DPPH and decolorize it to 1,1-diphenyl-2-picrylhydrazine. CuNPs have antioxidant properties comparable to standard ascorbic acid. This is because CuNPs are oxidized efficiently and quantitatively. CuNPs inhibited DPPH activity by donating an electron.

Sr. No.	Types of Nanoparticles	Optical Density			Average	DPPH scavenging activity %.
1	Silver	0.257	0.255	0.260	0.257	53%
2	Zinc oxide	0.502	0.506	0.510	0.506	50%
3	Copper	0.507	0.502	0.514	0.507	44%
4	Vitamin C standard	0.500	0.504	0.506	0.503	93%

DPPH Scavenging activity was calculated by reported method.

### CONCLUSION:

It is possible to conclude that silver nanoparticles have the maximum scavenging activity, implying that they have the highest antioxidant properties when compared to other particles.

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