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ANTIOXIDANT ACTIVITY OF COPPER NANOPARTICLES SYNTHESISED FROM THE LEAF EXTRACT OF *OCIMUM SANCTUM*

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ABSTRACT

The green synthesised copper nanoparticles from the leaf extract of Ocimum sanctum was evaluated for the antioxidant activity. The antioxidant activity of CuNps was studied by DPPH (1,1-diphenyl-2-picrylhydrazyl) free radical scavenging assay. It showed 81.3 % of scavenging activity which indicates that the synthesised Copper nanoparticles is a potential antioxidant agent.

KEY WORDS

Antioxidant, copper nanoparticles, DPPH assay, Ocimum Sanctum.

I. INTRODUCTION:

The metal nanoparticles are focused mainly for the research work due to its unique properties and pharmaceutical applications. Many researchers have used green synthesis methods for different metal nanoparticles due to their growing need of eco-friendly properties [1]. The plant extract has been used as capping and reducing agent for the synthesis of copper nanoparticles due to their reducing properties present in the leaf extract [2,3]. The natural plants have antioxidant free radical scavenges which help in preventing pathologies like heart disease, cancer, arthiritis and liver disease [4]. Natural antioxidants are useful in protecting cells from oxidative damage [5]. It also plays a protective role in a number of diseases such as Cardiovascular and neurodegenerative diseases in which oxidative stress and free radicals are the major contributors [6].

The present work highlights the antioxidant activity of the green synthesised copper nanoparicles from the leaf extract of *Ocimum sanctum.* DPPH free radical scavenging assay is used to study the antioxidant activity of the copper nanoparticles.

II. MATERIALS AND METHODS:

2.1 Materials

All the Chemicals used were of Analytical grade. Double distilled water was used throughout the work. Filteration was done by using Whatmann No.1 filter paper

2.2 Preparation of *ocimum sanctum* leaf extract

The *Ocimum Sanctum* leaves were collected and rinsed with double distilled water to remove all the dust and impurities. The washed leaves were cut into small pieces. About 100 gm of leaves were weighed separately and transferred into 100 mL distilled water and boiled for 15 minutes at 60°C and cooled down. The prepared solution was filtered by Whatmann No.1 filter paper to get a clear solution. This filterate was known as *Ocimum sanctum* leaf extract and stored at 25°C for future work [7].

2.3 Synthesis of copper nanoparticles from the leaf extract of *ocimum sanctum*

For the synthesis of Copper nanoparticles, 50 mL of *Ocimum Sanctum* leaf extract was added to 50 mL of 0.05M Copper sulphate solution and kept for incubation for 24 hours. The formation of Copper nanoparticles was confirmed by the colour change from green to black colour. The CuNps solution was purified by repeated

centrifugation at 6000 rpm for 10 mins. The synthesized CuNps were lyophilized and stored in an air tight container [7].

2.4 Characterisation of copper nanoparticles

The synthesised copper nanoparticles were characterised by UV-VIS, FT-IR, SEM-EDX and XRD. The UV-VIS spectrum confirmed the formation of CuNps. The FT-IR analysis of CuNps showed that the flavonoids, alkaloids and polyphenols present in the *Ocimum Sanctum* leaf extract act as a capping and reducing agent which was surrounded by the CuNps. SEM Analysis reveals that the morphology of CuNps was spherical and uniform Floret shape. EDX of the synthesized CuNps showed strong Copper signals along with O, N, C and S peaks which may originate from the biomolecules that was bound to the surface of the CuNps. XRD Analysis confirmed the FCC (face centered cubic) lattice of CuNps with high crystallinity and its average size was found to be 29 nm using Debye-Scherrer equation [7].

2.5 Antimicrobial activity of copper nanoparticles

The antimicrobial activity of copper nanoparticles was established by Resazurin microtitre assay method. The result showed that it had excellent antibacterial activity towards *E. coli* and *B.subtilis*. It may act as a good substituent for the standard *Streptomycin.* It also showed good antifungal activity against *C.albicans*[7].

2.6 Antioxidant activity of copper nanoaprticles

The percentage of antioxidant activity (AA %) of each substance was assessed by DPPH (1,1-diphenyl-2-picrylhydrazyl) free radical scavenging assay. Different concentrations of CuNps were added to all the tubes except blank. Then 3 mL of ethanol and 0.3 mL of 0.5 mM DPPH radical solution in ethanol was added. The control solution was prepared by mixing ethanol (3.5 mL) and DPPH radical solution (0.3 mL). Absorbance was read at 517 nm after 30 min of reaction. Same procedure was done for the standard BHT and its absorbance was also measured [8]. The scavenging activity percentage (AA %) was calculated using the below formula

$$\% \text{ Antioxidant activity} = \frac{(\text{absorbance at blank}) - (\text{absorbance at test})}{(\text{absorbance at blank})} \times 100$$

III. RESULTS AND DISCUSSION:

3.1 Antioxidant activity by DPPH assay

DPPH is a stable free radical at room temperature often used to evaluate the antioxidant activity of several natural compounds. The reduction capacity of DPPH radicals was determined by decrease in its absorbance at 517 nm, which is induced by antioxidants [9].

The % of DPPH radical scavenging activity of CuNps is presented in Fig 3. The free radical in DPPH can be neutralised by the antioxidants present in CuNps by transferring either their electron or hydrogen atoms to DPPH, thereby changing the colour from purple to yellow coloured diphenylpicrylhydrazine[9]. The Fig 1 and 2 shows the colour change occurred in DPPH assay with the standard BHT and the sample CuNps. The antioxidant activity of the standard BHT and synthesised CuNps are represented in Table 1 & Table 2.

TABLE 1: ANTIOXIDANT ACTIVITY OF THE STANDARD BHT

Concentration	Std BHT Blank -0.59	
	O. D	% inhibition
100 µg	0.36	38.9
200 µg	0.27	54.2
300 µg	0.17	71.1
400 µg	0.15	74.5
500 µg	0.11	99.8

Standard –BHT (Butylated hydroxytoluene) – 1 mg/mL

TABLE 2: ANTIOXIDANT ACTIVITY OF THE SYNTHESISED CuNps

Concentration	CuNps Blank – 0.75	
	O. D	% inhibition
100 µg	0.52	30.6
200 µg	0.45	40.0
300 µg	0.37	50.6
400 µg	0.27	64.0
500 µg	0.14	81.3

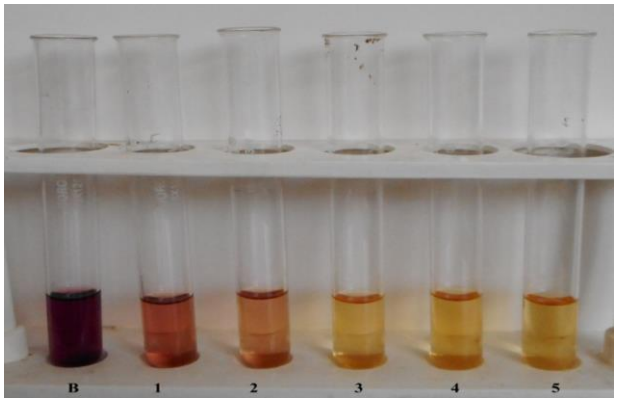

Fig 1: DPPH Assay of the standard BHT

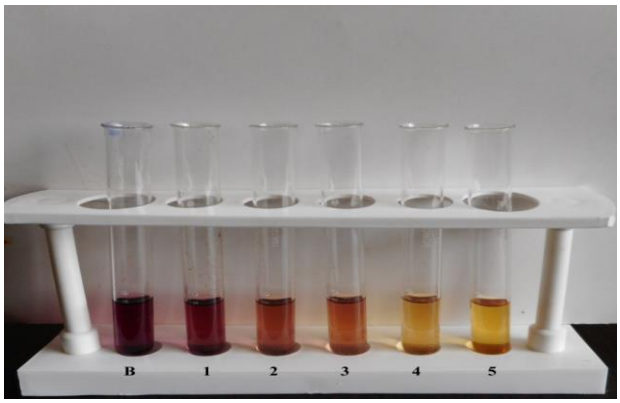

Fig 2: DPPH Assay of the synthesised CuNps

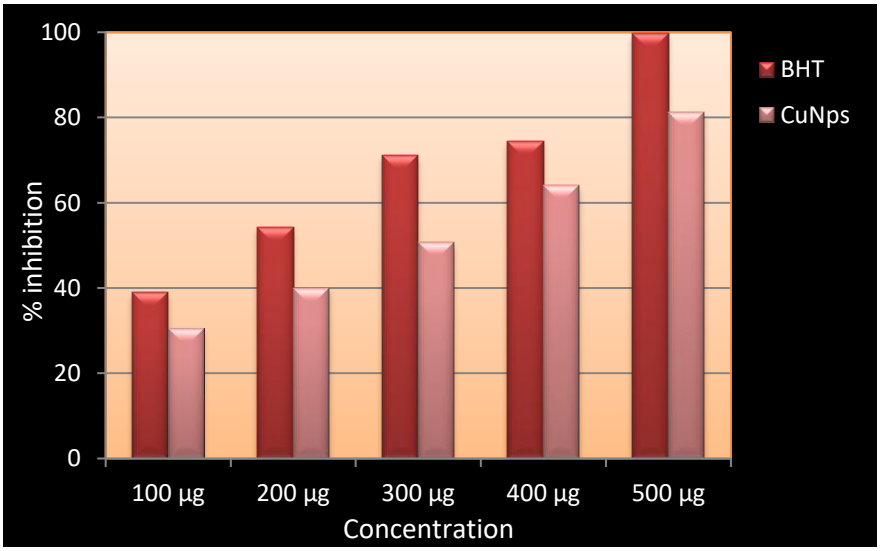

Fig 3: Scavenging activity of the CuNps by DPPH assay

In the DPPH assay, the antioxidant activity of the CuNps synthesised from the leaf extract of *Ocimum Sanctum* shows 81.3% of scavenging activity whereas the standard BHT shows 99.8% of scavenging activity. Hence, the synthesised Copper nanoparticles shows good antioxidant activity.

IV.CONCLUSION:

The Copper nanoparticles synthesised from the leaf extract of *Ocimum Sanctum* is evaluated for the antioxidant activity by DPPH free radical scavenging assay. DPPH assay is used to study the antioxidant activity of the copper nanoparticles. It reveals that it is the potent antioxidant agent. Due to the better activity in free radical scavenging, it can be used in a clinical therapeutic application and also in the pharmaceutical applications in future.

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