GREEN SYNTHESIS OF COPPER OXIDE NANOPARTICLES USING

PANICUM MAXIMUM LEAF EXTRACT

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Abstract

Copper oxide nanoparticles (CuONPs) are considered one of the most important metal nanoparticles, because they are low in cost, easily available, and have good antimicrobial properties, catalytic efficiency, excellent electrical conductivity, etc. "Green Synthesis" is the process of synthesizing nanoparticles by using natural reducing and capping agents, thereby reducing the impact of hazardous chemicals on the environment. The present study reports an eco-friendly way to synthesize CuONPs by using *Panicum maximum* leaf extract and copper sulphate pentahydrate ($CuSO_4.5H_2O$). The effect of three different ratios of plant extract: precursor salt (1:1, 1:2, 2:1) and five different pH values (5.5, 6.5, 7.5, 8.5, and 9.5) on synthesis of CuONPs were studied. To confirm synthesized nanoparticles, UV-Vis, XRD, FTIR and SEM techniques were used. The Surface Plasmon Resonance (SPR) of synthesized NPs showed peak at 270-315 nm. The 2:1 ratio showed the highest particle intensity. It was concluded that the best plant extract for synthesizing CuONPs was precursor salt ratio. The highest particle intensity was observed at pH 8.5. It indicated the formation of CuONPs in the reaction mixture. SEM results revealed the formation of irregularly shaped and aggregated particles in the nano range. The average size of the nanoparticles was calculated by using XRD Data and Scherrer equation, resulting in the 17 nm for *Panicum*-CuONPs. The presence of natural reducing and capping agents on CuONPs were identified by FTIR analysis. According to these results, it is possible to conclude that *Panicum maximum* leaf extract can be used to synthesize CuONPs in an eco-friendly way.

Keywords: Copper nanoparticles, Green Synthesis, *Panicum maximum,* Surface Plasmon Resonance

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INTRODUCTION

"Nanotechnology" is a branch of a novel technology that has promising applications in a range of fields including agriculture, food processing, biomedicines, textile, electronics, and so on. The size of the nanoparticles ranges between 1-100 nm (1 nm = 10^{-9} m). The size of the nanoparticles affects their distribution, targeting ability, biological fate (identity), and toxicity (Mohanraj and Chen, 2006). Copper and Copper oxide nanoparticles (CuONPs) have gained the attention of researchers due to their antimicrobial properties, sustainability, cost-effectiveness, and simplicity. Unlike silver and gold, copper is relatively less expensive. Therefore, these particles can be used over different types of microorganisms including fungi and bacteria. Copper/CuO nanoparticles also have catalytic, optical and superconductive properties (Chakraborty *et al*. 2022). CuONPs have been synthesized by using different types of plant extracts such as *Ocimumbasilicum* (Altikatoglu *et al*, 2017), *Ocimumbasilicum L, Asparagus adscendensRoxb*, *Withaniasomnifera (L.) Dunal* (Chakraborty *et al*, 2022). However, not much research has been done on synthesizing nanoparticles from leaf extract of grassy weeds. Usage of these weedy plants for the synthesis of nanoparticles is simple and cost-effective. Weeds are readily available and easy to grow. Therefore, weeds can be used to produce nanoparticles at the commercial level. *Panicum maxium* is one of the most commonly found perennial weeds in Sri Lanka and it has a high production potential (Jank *et al*., 2013). Even though *P. maximum* is a cheap weedy plant, its leaf extract consists of many essential bio-chemical compounds like terpens, flavonoids, palmitic acids, alkaloids, tannins, saponin and has antimicrobial, anticancer and antileishmanial properties (Kanife *et al*., 2012; Okokon *et al*., 2014). Some literature reports reveal successful synthesis of metal and metal oxide nanoparticles from Panicum species e.g.: Silver Oxide Nanopartcles from *Panicum miliaceum* grains extract (Velsankar *et al*., 2022), CuONPs from *Panicum sumatrense* grains extract (Velsankar *et al*., 2022), Siver nanoparticles from *Panicum virgatum* (switchgrass)) extract (Mason *et al*., 2012). For the very first time, the present study reports biological synthesis of CuONPs using leaf extracts of *Panicum maximum*. The main objective of this study is to synthesize CuONPs using an abundantly available, cheap, and weedy plant like *Panicum maximum*. Further, the present study would help to control the fast-growing weed species like *P. maximum.*

METHODOLOGY

Preparation of plant materials and plant extract
Fresh leaves of *Panicum maximum* were collected and leaves were rinsed with running tap water to wash out dust particles, then washed with distilled water and dried in a dehydrator for 16-18 hrs at 40 °C. Dried leaves were ground into a powder using an electrical grinder. After that, 10 g of powdered leaves were added to 100 ml of distilled water and boiled at 60 °C for 30 minutes. The mixture was then filtered with Whatman no.1 filter paper. A concentrated sulphuric test was performed to test the availability of Flavonoids in the plant extract. The light brown color of the plant extracts had changed into an orange color, indicating the presence of Flavonoids in the plant extract.

Figure 1: Color change in the *Panicum maximum* leaf extract from brown to orange after adding Con. H_2SO_4

Green synthesis of Copper Oxide nanoparticles (CuONPs)

Samples were prepared by mixing various amounts of plant extracts and 1 mM $CuSO_4.5H_2O$ solution in the ratios of 1:1, 1:2, and 2:1. All samples were stirred on a magnetic stirrer at ambient room temperature for 2 hrs. All samples were then centrifuged at 14 000 rpm for 15 minutes. The pellets containing CuONPs were washed with distilled water and kept for characterization.

Effect of pH on the synthesis of CuONPs

The effect of pH on the synthesis of CuONPs nanoparticles was observed under 5 different pH values *i.e.* pH 5.5, 6.5, 7.5, 8.5 and 9.5. The pH value of the samples was adjusted by using a 10% NaOH solution.

Scanning Electronic Microscope (SEM) XRD (X-ray Diffractometer) and FTIR (Fourier-transform Infrared Spectroscopy) analysis

The reaction mixture was centrifuged at 15000 rpm for 15 minutes and pellets were washed 3 times with de-ionized water. Pellets were then dried in the laboratory oven at 50 °C for 17 - 19 hrs and ground into a fine powder. SEM analysis was done by using SEM microscope (Zeiss™ - Germany) and particles were observed at 5.00 kV under 4 different magnifications 1.00 KX, 4.00 KX, 14.00 KX and 25.00 KX. FTIR analysis of the synthesized nanoparticles was done by using ATR mode. XRD analysis was done by using the Powder X-ray diffraction and the raw data of XRD analysis were further analyzed by using **OriginLab software** and **Match!3** software.

RESULTS AND DISCUSSION

After adding 1 mM $CuSO_4.5H_2O$ solution into plant extract, colour of the plant extract changed from light brown to light green and then changed to a dark greenish-brown within 2 hrs (Figure 2). This colour change indicates the formation of nanoparticles (CuO) (Iliger *et al*, 2021).

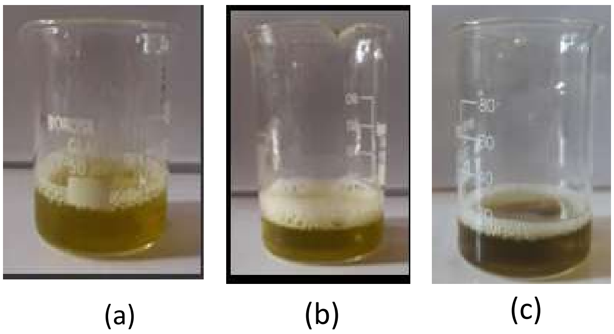

Figure 2: *Panicum maximum* after 2hrs of stirring: (a) $CuSO_4$: Plant extract -2:1 (b) $CuSO_4$:Plant extract -1:1 (c) $CuSO_4$:Plant extract -1:2

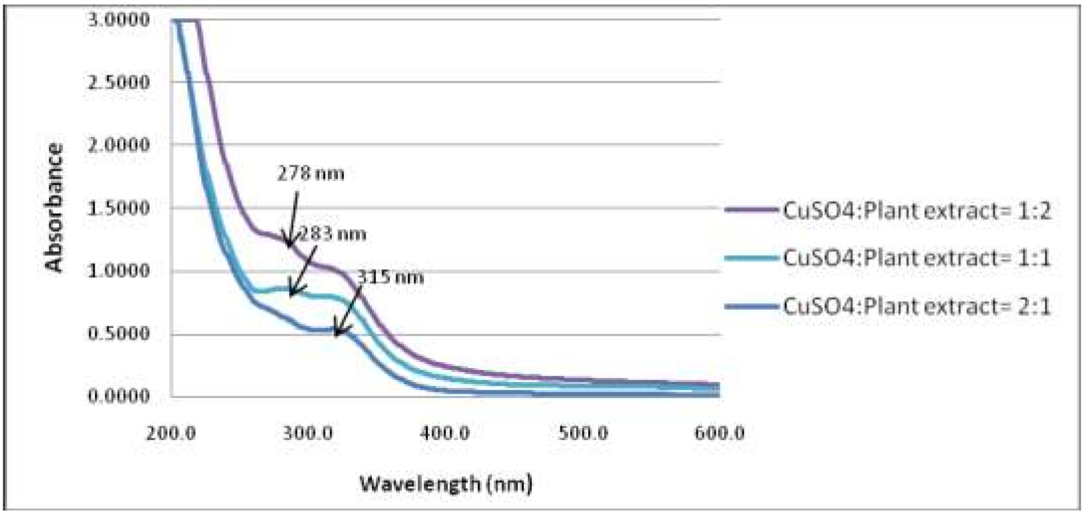

Figure 3: Absorbance spectra of *Panicum maximum*-CuONPs synthesized by using different ratios of CuSO4: plant extract

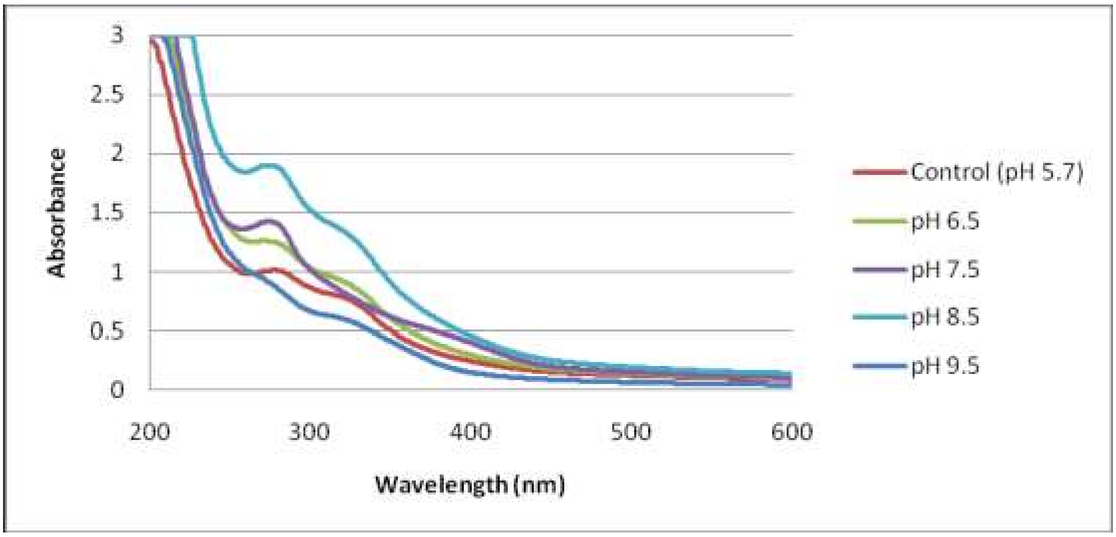

Figure 4: UV-visible absorption spectra of synthesized CuONPs using *Panicum maximum* leafextract atdifferent pH conditions.

The SPR (Surface Plasmon Resonance) for the samples was observed in the wavelength range of 200-350 nm. This indicates the presence of CuONPs in the samples. Altikatoglu *et al* (2017), Amaliyah *et al* (2020) reported similar results on absorption peak for CuNPs. As shown in Figure 3, the intensity of SPR bands were increased with increasing volume of plant extract in the solution. As Akintelu *et al* (2021) reported, the number of particles synthesis in the reaction mixture increases with the increasing volume of plant extract. According to Figure 4, *P. maximum*-CuONPs at pH 5.6 (control), pH 6.5, pH 7.5, pH 8.5 and pH 9.5 showed SPR absorbance at 278 nm, 276 nm, 275 nm, 273 nm and 316 nm respectively. The highest absorbance (1.89 Abs.) was observed for nanoparticles that synthesized at pH 8.5. The lowest absorption (0.6 Abs.) was observed at pH 9.5. As pH value increased up to pH 9.5, the peak absorbance of the synthesized CuONPs was declined. According to Traiwatcharanon *et al.* (2017), this may be because of the high amount of hydroxide ions, and in basic media, presence of a high concentration of hydroxide ions, nanoparticles tend to bind to the surface of adjacent nanoparticles through Brownian diffusion. This leads to aggregation of nanoparticles and as a result the particle intensity of reaction mixture declines. In addition, SEM analysis also confirmed the formation of CuONPs. However, these particles are seemed aggregated and compacted. (Figure 5)

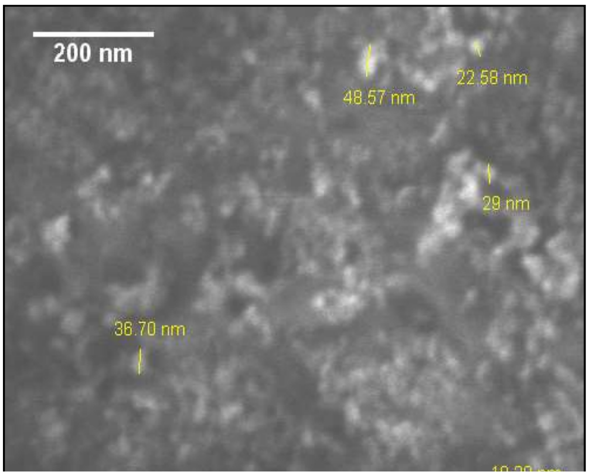

Figure 5: SEM images of biosynthesized CuONPs by using *Panicum maximum* leaf extract observed under 14.00 KX after 1 week of synthesis

The results of FTIR analysis (Figure 6) reveal the details of the functional groups of the chemical compounds which are responsible for synthesis of CuONPs. The probable functional groups and chemical compounds were identified by using FTIR Tables and Charts and previous studies. The peaks at 3777.71 cm^{-1}, 3773.71 cm^{-1} and 3661.32 cm^{-1} are due to the presence of O-H and/or N-H stretching vibrations (Movasaghi *et al*, 2008).

The O-H groups and N-H groups might be from alcohols and amines respectively (Sigma, 2023). The peak at 3265.47 cm^{-1}, is probably from O-H stretching of alcohols present in the extract (Samanta *et al*, 2011; Sigma Aldrich, 2023). The peaks observed at 2921.71 cm^{-1} and 2358.45 cm^{-1} represent the presence of C-H bending of alkanes (Kannan *et al*, 2020). The strong peak at the 1632.01 cm^{-1} represents C=O stretching/ C-C from phenyl ring (Movasaghi *et al*, 2008; Karthik *et al*, 2014). The weak peaks observed at 1547.80 cm^{-1}, 1530.59 cm^{-1}, 1462.48 cm^{-1}, 1451.23 cm^{-1}, 1407.87 cm^{-1}, 1233.05 cm^{-1}, 1070.90 cm^{-1} represent nitro compounds from amides (Movasaghi *et al*, 2008; Sigma Aldrich, 2023), C=N and/or C=C stretching of adenine (Movasaghi *et al*, 2008; Sigma Aldrich, 2023), C-H bending probably from methylene groups (Nagaonkar and Rai, 2015; Sigma Aldrich, 2023) asymmetric CH_3 bending modes of the methyl groups of proteins (Movasaghi *et al*, 2008), C-O stretching probably from flavonoids (Jagdish *et al*, 2021), asymmetric vibration of sulphate groups (Mahdavi *et al*., 2013) and C-O stretching probably from nucleic acids or a sugar monomer (Movasaghi*et al*, 2008) respectively. Likewise, Kumari *et al* (2017) observed that the small peaks below 700 cm^{-1} on the FTIR spectrum represents Cu-O vibrations.

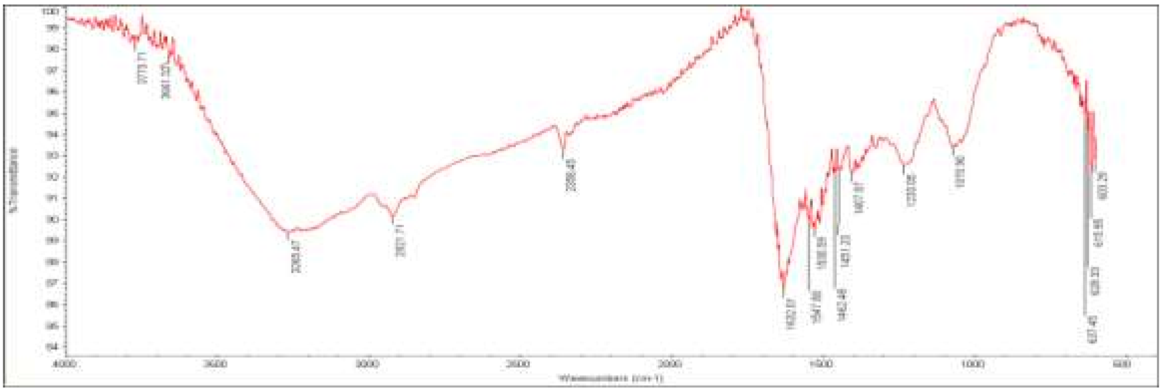

Figure 6: FTIR spectrum of CuONPs synthesized using *Panicum maximum* leaf extract

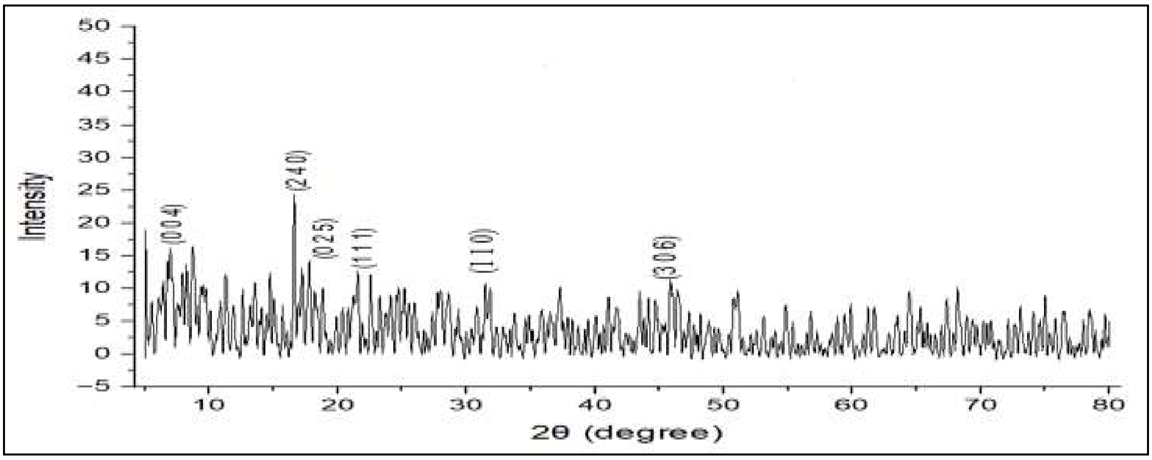

Figure 7: XRD pattern of synthesized CuONPs by using *P. maximum* leaf extract

Fig.7 presents the XRD patterns of *Panicum maximum* leaf extract-mediated CuONPs. The diffraction peaks of synthesized CuONPs by using *Panicum maximum* leaf extract were observed at 2θ values 7.8, 16.6, 17.7, 21.4, 31.6, 46.1 that can be ascertained to plains of (0 0 4), (2 4 0), (0 2 5), (1 1 1), (1 1 0) and (3 0 6) respectively. *Panicum maximum* leaf extract-mediated CuONPs showed a significant amount of low-intensity peaks on diffraction spectra. This may be due to the impurities in the sample (Holder and Schaak, 2019). The average particle size was calculated by using Deby-Scherrer equation **(**$D = K\lambda / \beta \cos\theta$). The average crystallite size of *Panicum*-CuONPs was found to be around 17 nm.

CONCLUSION

In the present study, copper nanoparticles were produced using *Panicum maximum* leaf extracts. It is economical to synthesize nanoparticles using weedy plants than using commercially valuable plants like turmeric (*Curcuma longa*), Basil (*Ocimumbasilicum*) *etc.* The number of nanoparticles increases with the increasing concentration of plant extract in the reaction mixture. SPR of synthesized NPs showed peak at 270-315 nm. The 2:1 ratio showed the highest particle intensity, concluding as the best plant extract: precursor salt ratio for synthesizing CuONPs. The highest particle intensity was observed at pH 8.5, indicating formation of CuONPs in the reaction mixture. SEM results revealed the formation of irregularly shaped and aggregated particles in the nano range. The functional groups of chemical components including amides, alkanes, alcohol etc., included in the plant extract were identified with FTIR analysis. In conclusion, *Panicum maximum* leaf extract can be used to synthesize CuONPs in an eco-friendly way.

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