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Plant Based Biosynthesis of Copper Nanoparticles and its Efficacy on Seed Viability and Seedling Growth in Peanut (*Arachis hypogaea* Linn.)

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Nano-fertilizers can easily adsorb into the plant and can increase the reactive points in the plant and hence are treated as an efficient alternative to the conventional fertilizers. Based on this, the present study was intended to synthesize copper nanoparticles (CuNPs) using aqueous root extract of Schrebera swietenioides Roxb. as green reducing agent. The synthesized nanoparticles were studied for its effectiveness on enhancement of seed germination and plant growth promotion on peanut (Arachis hypogaea Linn.). The formation of nanoparticles was confirmed by observing colour change in the reaction mixture, which shows characteristic absorption maxima at 340 nm. The SEM and TEM analysis confirmed that the nanoparticles were in monodispersed with spherical to irregular shape with an average particle size of 35 nm. The EDX analysis confirmed that the nanoparticles contain 82.5% copper metal. The synthesized nanoparticles were applied for its seed germination enhancement activity on peanut seeds and results confirms that the nanoparticles were significantly enhances the germination of peanut seeds with decrease in mean germination time. The peanut plant growth also enhances when compared with metal solution treatment and untreated plants. The root length of CuNPs treated plants was observed to be 9.27 ± 0.15 cm, which is significantly more than the untreated $(6.40 \pm 0.10 \text{ cm})$ as well as treated copper metal $(7.13 \pm 0.25 \text{ cm})$ plants. The shoot length of 19.13 \pm 0.20 cm was observed for nano-treated plants and is greatly enhanced than the untreated (10.30 \pm 0.20 cm) and treated copper metal $(11.27 \pm 0.25 \text{ cm})$ plants. The protease activity on day 5 of the germination study was found to be 0.904 ± 0.004 , 0.133 ± 0.002 and 0.095± 0.002 units/mL, respectively for the peanut seeds treated with CuNPs, copper metal solution and untreated conditions. The catalase activity at 5th day of seed germination studies the activity was observed to be 45.177 ± 0.192 , 23.691 ± 0.074 and 18.331 ± 0.209 units/ min/g, respectively for CuNPs treated, copper sulphate treated and untreated pea nut seeds. The water update of the nano treated seeds was observed to be very high along with high quantity of photosynthetic pigments when compared with the other treatments in the study. Based on the results achieved, it can be confirmed that the nano-treatment enhances the seed germination and plant growth promotion on peanut seeds.

Keywords: Copper nanoparticles, Nano priming, Seed germination, Growth enhancement, Peanut.

INTRODUCTION

Recently, nanotechnology has played a critical role in different fields, including cosmetic, medicine, agriculture, and food sciences [1]. Currently, the fabrication of metal nanoparticles by using the microbial extracts and bioactive substances of plants is considered the effective green approach to the utilisation of microbes and plants as nano-factories [2]. In the view of challenges faced by communities globally, especially of climate change and population growth, nanotechnology can minimise the adverse impacts of agricultural pesticides on

human health and environments, improve agricultural product quality and increase food production and security. The characteristics of nanomaterials render them excellent candidates to be used for the development and design of novel tools for supporting industries such as agriculture. Nanotechnology can be used to enhance agricultural components, including agricultural product and soil quality, through plant growth simulations or by employing nanoparticles based fertilizers. Additionally, the utilisation of pesticides and fertilizers with nanoparticle based compounds and carriers can be reduced without the lowering of productivity [3].

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Currently, a biological method of nanoparticle synthesis is free from hazardous chemicals and hence was considered as green chemistry synthesis of metal nanoparticles. The green route of nanoparticle synthesis using bioactive compounds from plant extracts has more advantage than microorganism because the microbial process is highly expensive due to the maintenance of microbial culture as well as isolation of targeted microbe [4].

Copper nanoparticles (CuNPs) have received considerable research attention in fields, including biodiesel, solar cells, photocatalysis, supercapacitors, water pollutant removal and electrocatalysis, because of their favourable properties. These properties include nontoxicity, low costs, and easy fabrication. Although some methods offer enhanced selectivity and sensitivity and are highly economical, the electrocatalytic applications of CuNPs are limited [5]. With the advances in technology, the use of CuNPs as the antimicrobial agent has emerged as a common alternative, rendering the production of CuNPs economical [6]. The considerable antimicrobial effect of CuNPs against various microbes is well documented; hence, currently, these nanoparticles are used to control different plant pathogens. Moreover, the controlling plant pathogens mediated through CuNPs are environmentally benign compared with the chemically fabricated fungicides [7].

In this view, this study prepared CuNPs through a green approach by using a biological reducing agent, *i.e.* the aqueous extract of *Schrebera swietenioides* Roxb. roots. The incorporation of a bioactive compound with CuNPs in *S. swietenioides* is highly beneficial in the treatment of various ailments. This green approach was developed to avoid toxic chemicals, which act as capping and reducing agents during the chemical reaction. The biologically prepared CuNPs were analyzed by employing different methods, including FT-IR, UV-visible, FESEM, EDX, TEM and X-ray diffraction. Furthermore, the synthesized CuNPs were used to enhance peanut seed germination and promote its growth.

EXPERIMENTAL

All the chemicals used for the synthesis of CuNPs and its application study were of analytical reagent grade and obtained from Merck Chemicals Private Limited, India.

Collection of *S. swietenioides* plant roots: The fresh roots of *S. swietenioides* were collected from the live plant growing in the location of Tirumala hills, Tirupati and authenticated by the bonatist from P.B. Sidddharth College of Arts & Science, Vijaywada, India. The sand and dirt from the collected roots were cleaned by washing with distilled water several times and then water was removed using sterile cotton and tissue paper. Then the roots were cut into small pieces dried until fixed weight obtained in two successive measurements. Then the dried roots were powdered and the powdered root material was used for the synthesis of CuNPs.

Preparation of aqueous root extract: In a 250 mL beaker containing 100 mL of distilled water, 10 g of accurately weighed root powder was added. The content was stirred using a magnetic stirrer and incubated at 40 °C for 80 min with constant stirring.

Then the solution was filtered through Whatman filter paper and filtered extract was used for the synthesis of CuNPs [8].

Synthesis of copper nanoparticles: The green synthesis of copper nanoparticles using aqueous root extract of *S. swietenioides* was carried as per the procedure reported by Suresh *et al.* [9] with slight modifications briefly, in a 500 mL volumetric flask containing 400 mL copper sulfate (5 mM) solution, 50 mL of root extract was added and the content was mixed well for 2 min. The solution pH was adjusted to 7 using 1 N NaOH solution. The formation of CuNPs was monitored by observing the colour change in the reaction mixture. The pure nanoparticles was collected by centrifugation followed by washing with distilled water several times and dried in an air oven at 60 °C for 2 h. The collected nanoparticles were stored in an Amber coloured bottle.

Characterization of copper nanoparticles: The synthesized nanoparticles was characterized using various techniques such as UV-visible spectrophotometer, FE-SEM, TEM, EDS and XRD studies. The UV-visible spectrophotometer (JASCO, Japan) was used for the determination of optical absorption characteristics of the nanoparticles and the optical absorption was measured in the range of 800-400 nm. FE-SEM (NOVA NANOSEM 450, FEI, USA) and TEM (Jeol/JEM 2100, Japan) analysis was carried for the determination of size, shape, topography and surface morphology of the CuNPs. The EDS (Rontec Xflash 3001, Japan) analysis was carried for the evaluation of the elemental composition of the synthesized nanoparticles. The lattice parameters and lattice nature of the nanoparticles was evaluated using XRD (Rigaku Corporation, Japan) studies which were performed at a scan speed of 2°/min in 20° to 80° diffraction angles (2θ) [10,11].

Effect of CuNPs on the germination of peanut seeds: The impact of synthesized CuNPs on the germination of peanut seeds was evaluated based on the procedure reported by Liu *et al.* [12]. Uniform size healthy peanut seeds were selected by visual observation and the seed dormancy was simulated by keeping the seeds in a refrigerator at 4 °C for 24 h. The surface sterilization of the selected seeds was carried using 0.5% by weight mercury(II) chloride for 10 min. Then the seeds were washed using distilled water for several times until complete removal of mercury(II) chloride from the seed surface. The seed germination study was carried on an incubator which was conducted in darkness at 30 °C using three treatments as given in Table-1.

TABLE-1 TREATMENT CONDITIONS STUDIED IN GERMINATION STUDY OF PEANUT SEEDS Treatment Treatment solution Untreated Sterile distilled water Metal treated 10 mg/L copper sulfate solution Nanoparticle treated 10 mg/L CuNPs solution

In all three treatment conditions, healthy peanut seeds were cultured in sterile Petri dishes ($100 \text{ mm} \times 15 \text{ mm}$) with 10 seeds per dish on Whatman filter paper in the culture container. The filter paper was moistened with selected treatment solution and each treatment was carried in three replicates

and the same volume of the treatment solution was added every day to prevent drying. In all the treatment studies, the emergence of radical to 2 mm or more was considered as germination and the number of seeds germinated was recorded in every 24 h of incubation at room temperature.

The impact of nanoparticles treatment on the germination of peanut seeds was confirmed by ascertaining the germination rate, mean germination time and final germination percentage as per the procedure described by Arturo *et al.* [13]. The formulae used in the seed germination study are given as eqns. 1-3.

$$FGP = \frac{A_e \times M}{100} \tag{1}$$

where FGP (final germination percentage) is the germination capacity of peanut seeds that germinated completely at a given time; $A_{\rm e}$ is the germination accumulated until the last evaluation; and M is the total of sowed peanut seeds during the study.

$$MGT = \frac{\Sigma(nt)}{(\Sigma n)}$$
 (2)

where MGT (mean germination time) is the average time require to germinate the peanut seeds; n is the number of peanuts newly germinated at time t; and t is the number of days from sowing.

Germination rate =
$$\sum \frac{n}{t}$$
 (3)

Water uptake of peanut seeds: During the imbibition stage of seed germination, the effect of nanoparticles treatment on the water uptake of peanut seeds was evaluated based on the procedure described by Guan *et al.* [14]. The study was carried in triplicates and each batch consists of 25 number of uniform sized seeds. The seeds were weighed and placed in a plastic box containing saturated sterile cotton and incubated at 25 °C. At intervals of 40, 80 and 120 min, all the seeds were removed, blotted dry and weighed. Changes in weight due to imbibition were expressed as the amount of water absorbed per seed dry weight, which was calculated using eqn. 4:

Water uptake =
$$\frac{\text{Fresh weight of seed} - \text{Dry weight of seed}}{\text{Dry weight of seed}} \times 100 \text{ (4)}$$

Green house study for assessment of peanut plant growth: The effect of nanoparticles treatment on the growth promotion activity of peanut plant was evaluated by conducting a greenhouse study using 25 cells plastic seedling trays and the experiment was carried in triplicates in each treatment based on the procedure reported by Acharya et al. [15]. The seeds used in the seed germination study conducted previously were individually planted in the seedling trays containing soil. All the experimental trays were thoroughly moistened using distilled water for control, copper sulphate and CuNPs solutions. The seedlings were grown for up to 7 days. The impact of nano-treatment on the growth of peanut plant was evaluated by determining the numbers of plants grown in each treatment studied, shoot length, root length of reach plant in each treatment. The copper metal uptake and accumulation on the roots, shoots and leaves of peanut plants in each treatment was assessed using atomic absorption spectroscopy (Shimadzu, Japan) as per the procedure described by Tariq & Ashraf [16].

Hydrolytic enzymes assay

Preparation of crude enzyme extract from germinating seeds: In each day at fixed time of seed germination study, the seeds were separated, the seed coat was removed and weighed. The weighed sample was homogenized using a mortar and pestle by addition 10 mL of 0.1 M ice-cold pH 7.6 phosphate buffer. Then the homogenate was centrifuged at 10,000 rpm and kept the sample at 4 °C for 15 min. Then, the pellet was discarded and supernatant was preserved and utilized as crude enzyme extract. The crude enzyme extract was used for evaluation of the enzymatic assays as well as the total protein analysis.

Amylase activity: The α - and β -amylase activity of the enzyme extract was measured as per the procedure described by Bernfeld [17]. The crude enzyme was directly used for the determination of β -amylase. The crude extract was exposed to 70 °C for 15 min to denature β -amylases and the β -amylase free crude extract was subjected to α -amylase activity. The enzyme activity was expressed as mg maltose produced mg⁻¹ protein.

Protease activity: The protease assay of the crude enzyme was carried as per the procedure reported by Reimerdes & Meyer [18] using casein standard. The measurement was carried out by estimating the release of tyrosine calculated from the standard curve prepared with tyrosine. One unit of protease activity was defined as the amount of enzyme required for liberating 1 mg of tyrosine in 30 min at 45 °C.

Catalase activity: The catalase activity of the crude enzyme was determined based on the procedure reported by Hugo [19] and the activity was expressed as mol/min/mg protein.

Determination of protein content: The quantity of the protein in the germinating seed was determined based on the standard protocol reported by Lowry *et al.* [20].

Determination of chlorophyll content in plant leaves: The quantity of photosynthetic pigments *i.e.* chlorophyll A and B in the fresh leaves of peanut plants that are grown in the green house study was evaluated based on the procedure reported by Shinde *et al.* [21] and the results were subjected to the formula described by Arnon [22] for summarizing the quantity of the pigments.

RESULTS AND DISCUSSION

The present work focused on the green synthesis of copper nanoparticles using *S. swietenioides* aqueous root extract as green reducing agent and further the synthesized nanoparticles were applied for its seed germination and plant growth promotion activity on peanut seeds. The observation of colour change by reacting the plant extract and copper sulphate confirms the reduction of copper into nano sized particles by reaction with bioactive compounds in the root extract. The colour change was observed within a short period of 1 h confirms that the nanoparticles formation was completed within a short period of 1 h. The root extract of *S. swietenioides* contains various chemical constituents that are having the ability to convert reduction of Cu⁺ to Cu⁰ and hence nanoparticles were formed

[23]. Further the formation of nanoparticles in the reaction mixture was confirmed by measuring the maximum absorption wavelength using UV-visible absorption spectrophotometer and the spectra shows the characteristic absorption maxima at 340 nm (Fig. 1). This confirms that the formation of nanosized copper particles and the results are in correlation with the previous findings [6,8].

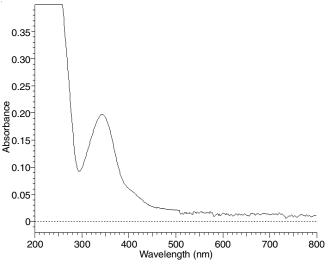


Fig. 1. UV-visible absorption spectra of copper nanoparticles

FE-SEM analysis: The FE-SEM analysis was carried to determine the morphology of the synthesized CuNPs and analysis confirmed that the CuNPs were with an average particle size of around 35 nm (Fig. 2a) particles size was distributed nearly monodispersed. Further it was confirmed that the FE-SEM analysis were in aggregates with spherical in shape and some of the particles were irregular shape. The EDX analysis was carried for the determination of elemental composition of the synthesized CuNPs. The EDX spectra (Fig. 2b) shows major elemental peak at 0.93 and 8.02 keV that is specific to the Cu metal. The EDX spectra also shows the peaks corresponds to carbon and oxygen were identified which may be due to the plant biomolecules that were used for the capping of the CuNPs. The % copper content in the synthesized CuNPs was 82.5 %. Similar type size distribution and shape of the CuNPs was reported along with the elemental composition findings supports the results achieved in present study [8].

TEM studies: TEM analysis was carried to evaluate the morphological characteristic, orientation and size distribution of the chemical and biological samples and nanoparticles. The TEM analysis as shown in Fig. 3a confirms that the nanoparticles were spherical to irregular in shape with broad size distribution [24]. The nanoparticles were widely dispersed in the range of 15-45 nm size with an average particle size of 35 nm. The XRD analysis was carried to evaluate the crystalline

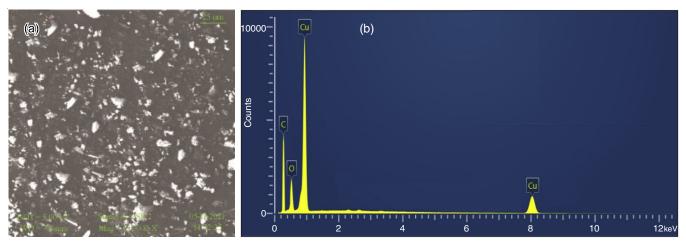


Fig. 2. SEM (a) and EDX (b) analysis results of copper nanoparticles

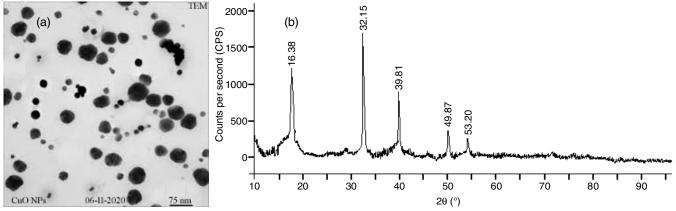


Fig. 3. TEM (a) and XRD (b) analysis results of copper nanoparticles

phase of the synthesized CuNPs. The XRD spectra shows five characteristic peaks identified at 2θ angles of 16.38° , 32.15° , 39.81° , 49.87° and 53.20° . These diffraction peaks correspond to the h,k,l value of the reflections from (110), (111), (220), (800) and (713) and diffractions were corroborating with standard values of the JCPDS card 89-5899 [25]. The Scherrer equation was adopted for calculating the average crystalline size of the CuNPs and results obtained as 32 ± 4 nm on the (111) plane.

Studies have reported that nanoparticles have positive impact on the germination rate and seedling growth [26] and in view of this a laboratory test was conducted to verify the impact of CuNPs on the seed germination rate on peanut seeds. The results observed in the seed germination study of CuNPs on peanut seeds are summarized in Table-2. The results of the seed germination study on peanut seeds confirms that there will be significantly enhanced seed germination activity of peanut seeds when treatment with CuNPs. The nanoparticles treatment significantly decreases the MGT of seeds than metal treated as well as intreated peanut seeds. As all the seeds are healthy and viable there is no considerable variation was observed in FGP of peanut seeds in all the treatment studies. Hence the results confirms that there is a significant decrease in MGT of peanut seeds was observed when they are treated with CuNPs. The photographs observed while seed germination study are shown in Fig. 4. The possible reason behind the enhanced seedling growth rate could be the efficient water and nutrient uptake by the treated seeds as CuNPs can penetrate through the seed coat and may activate the embryo. During penetration, CuNPs cause more new pores that remain helpful in carrying nutrients, efficiently leading to fast germination and growth rate [27]. The obtained outcome could be helpful

to enhance the seed germination rate and seedling growth especially in dormant seeds.

Green house studies: A greenhouse experiment was conducted to determine the effect of CuNPs treatment on the growth enhancement on peanut seeds. The results obtained in this study confirms that the CuNPs treated peanut seeds shows enhanced growth with a greater number of leaves with high shoot length. The root length of CuNPs treated plants was observed to be 9.27 ± 0.15 cm, which is significantly more than the untreated $(6.40 \pm 0.10 \text{ cm})$ as well as metallic copper treated (7.13 \pm 0.25 cm) plants. The shoot length of 19.13 \pm 0.20 cm was observed for nano treated plants and is greatly enhanced than the untreated (10.30 \pm 0.20 cm) and copper metal treated (11.27 \pm 0.25 cm) plants. Chlorophyll A and B contents of the plant were estimated and the results proved that both chlorophyll A and B in the nano-treated plants was observed to be very high than the untreated and metal treated plants (Table-2). The photosynthetic potential and primary production of the plants greatly effects its chlorophyll content. It is also related to the stress in plants. An enhanced chlorophyll content in the CuNPs treated plants shows high photosynthesis capacity, which reflect the morphological as well as physiological characteristics of peanut plants. Hence, the nano-treatment enhances the plant chlorophyll content as well as growth of the plant. The growth promotion activity study results are shown in Fig. 5.

Amylase activity: The amylases (both $\alpha \& \beta$) were situated in the cotyledons of the seeds and hence the activity was identified in the cotyledons of the seeds and the activity was changed during the seed germination process. Hence, the $\alpha \& \beta$ amylase activity of peanut seeds during the seed germination was carried and the results obtained in nano treated,

TABLE-2 SEED GERMINATION AND PLANT GROWTH PROMOTION ACTIVITY RESULTS ON PEANUT						
Treatment	FGP	MGT (h)	Root length (cm)	Shoot length (cm)	Chlorophyll (mg/g fresh weight)	
					A	В
Control	95.00 ± 1.00	153.43 ± 1.42	6.40 ± 0.10	10.30 ± 0.20	7.40 ± 0.26	4.43 ± 0.40
CuNPs	99.33 ± 0.58	89.83 ± 1.31	9.27 ± 0.15	19.13 ± 0.20	14.30 ± 0.20	8.50 ± 0.10
Metal	96.67 ± 0.58	145.67 ± 1.36	7.13 ± 0.25	11.27 ± 0.25	8.22 ± 0.02	5.10 ± 0.20



Fig. 4. Seed germination study results [(A) control (no treatment); (B) metal treatment (treated with zinc acetate solution); (C) nano treatement that treated with *S. swietenioides* root extract mediated CuNPs]



Fig. 5. Pictorial view of peanut plants in growth promotion activity study [(A) control (no treatment); (B) metal treatment (treated with zinc acetate solution); (C) nano treatment that treated with *S. swietenioides* root extract mediated CuNPs; (D) comparsion of peanut plant growth identified in three treatemts]

metal treated as well as untreated were compared in Fig. 6A and 6B for α & β amylase activity, respectively. The results confirms that both the amylase activities were changed during the germination and the activity was very less in the early stage of germination. The activity of the seeds treated with nanoparticles shows maximum activity at 4th day of germination study. Whereas the seeds treated with metal solution and untreated shows maximum activity at 5th and 6th days of germination study. This confirmed that the seed germination time was decreased when the seeds treated with nanoparticles. Hence, it can be confirmed that the CuNPs treated seeds was observed to be enhanced seed germination activity with decreased seed germination time than the copper sulphate treated as well as untreated peanut seeds.

Protease activity: In the germination process of seeds, the storage proteins present in the seeds are hydrolyzed by proteolytic enzymes and thus the nutrients required for the development and growth of seedlings was obtained. Hence the protease enzyme activity of peanut seeds in the germination stage was studied for all treatments. The results confirmed that the activity was observed to be very high in the 5th day of germination study for the seeds treated with nanoparticles whereas the seeds treated with copper metal and untreated shows high activity at 6th and 7th days respectively (Fig. 6C).

The protease activity on day 5 of the germination study was found to be 0.904 ± 0.004 , 0.133 ± 0.002 and 0.095 ± 0.002 units/mL, respectively for the peanut seeds treated with CuNPs, copper metal solution and untreated conditions.

Total protein content: It is also observed that the quantity proteins in the germinating seeds was decreased with increase in the germination time. The protein content in first day of germination was observed to be 256.60 ± 1.229 , 257.67 ± 1.716 and 261.87 ± 0.833 , whereas the protein content in fifth day of germination was observed to be 112.13 ± 1.026 , 174.73 ± 0.473 and 199.27 ± 0.586 for CuNPs treated, copper metal solution treated and untreated peanut seeds, respectively. At mean germination time, the quantity of protein was significantly decreased due to maximum hydrolysis of protein in the seeds that facilitates the seed germination. The results observed in protease analysis and protein content in the germinating seeds was in good argument with each other and proved that the nano-treatment enhances the seed germination.

Catalase activity: The tolerance of germinating seeds to oxidative stress is the effective component during the seed germination process and catalase is the antioxidant enzyme that responsible the tolerance of germinating seeds to stress and is also responsible for germination of seeds and growth of seedlings. In the first day of seed germination study, the catalase

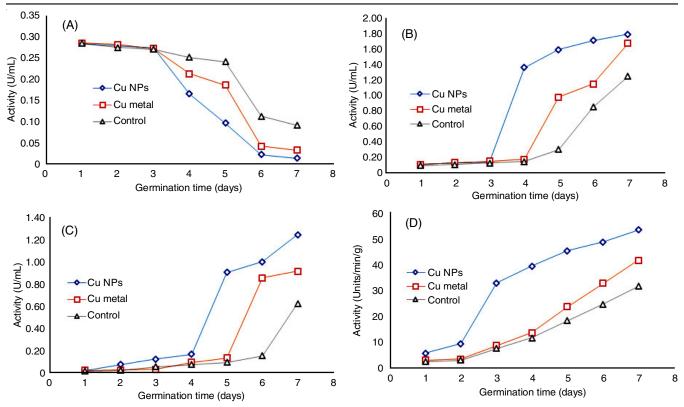


Fig. 6. The enzymatic study results observed during the seed germination activity study on peanut seeds [(A) α -amylase enzyme activity; (B) β -amylase enzyme assay; (C) protease activity assay; (D) Catalase activity]

activity was observed to be 5.809 ± 0.089 , 3.077 ± 0.035 and 2.511 ± 0.056 whereas the fifth day of seed germination study the activity was observed to be 45.177 ± 0.192 , 23.691 ± 0.074 and 18.331 ± 0.209 units/min/g respectively for CuNPs treated, copper sulphate treated and untreated peanut seeds (Fig. 6D). The results confirms that the nanoparticles treatment shows the enhanced activity.

The nanoprimed seeds imbibed water faster than other primed groups at the very beginning (24 h) of imbibition. At 48 h, seed water uptake of nano priming treatment was still higher than hydropriming and copper sulphate priming treatments. This confirms that the nano treatment enhances the water adsorption of the seeds that facilitates the rapid germination of peanut seeds.

Chlorophyl contents: The impact of nanopriming on the photosynthetic pigments was confirmed by evaluating the chlorophyll A and B content in the grown peanut leaves of three treatment studies. The chlorophyll A was observed to be 14.30 $\pm\,0.20,\,8.22\pm0.02$ and 7.40 ± 0.26 mg/g, respectively whereas chlorophyll B content was found to be $8.50\pm0.10,\,5.10\pm0.20$ and 4.43 ± 0.40 mg/g, respectively for CuNPs treated, copper sulphate treated and untreated peanut plants. The results confirmed that the peanut plants treated with CuNPs is significantly enhances the biosynthesis of main photosynthetic pigments.

Conclusion

The present work reports a green methodology for the synthesis of CuNPs by utilizing *Schrebera swietenioides*

aqueous root extract as green reducing agent. The formation of nanoparticles completed within the shortest time of less than 1 h confirms that the reduction process is rapid. The synthesized nanoparticles were spherical to irregular shape with an average particle size of 35 nm with an elemental copper composition of 82.5 %. The synthesized nanoparticles treatment on peanut seeds shows enhances the water intake and germination of peanut seeds. The nanopriming also enhances the formation of leaves and plant growth of the peanut plants. The root length of CuNPs treated plants was observed to be 9.27 ± 0.15 cm which is significantly more than the untreated (6.40 ± 0.10) cm) as well as metallic copper treated (7.13 \pm 0.25 cm) plants. The shoot length of 19.13 ± 0.20 cm was observed for nano treated plants and is greatly enhanced than the untreated (10.30) \pm 0.20 cm) and copper metal treated (11.27 \pm 0.25 cm) plants. The photosynthetic pigments such as chlorophyll A and B content, enzymatic activities such as amylase, protease, catalase were found to be high active in the seeds treated with CuNPs than the copper metal solution treatment as well as untreated peanut. Hence, based on the finding achieved in the present study, it can be concluded that the CuNPs synthesized having remarkable enhancement on the peanut seed germination and its growth.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interests regarding the publication of this article.

REFERENCES

- J. Danie Kingsley, S. Ranjan, N. Dasgupta and P. Saha, J. Pharm. Res., 7, 200 (2013);
 - https://doi.org/10.1016/j.jopr.2013.02.021
- P. Singh, Y.-J. Kim, D. Zhang and D.-C. Yang, *Trends Biotechnol.*, 34, 588 (2016); https://doi.org/10.1016/j.tibtech.2016.02.006
- J.S. Duhan, R. Kumar, N. Kumar, P. Kaur, K. Nehra and S. Duhan, Biotechnol. Rep., 15, 11 (2017); https://doi.org/10.1016/j.btre.2017.03.002
- S.K. Chandraker, M. Lal and R. Shukla, RSC Adv., 9, 23408 (2019); https://doi.org/10.1039/C9RA03590G
- S. Sukumar, A. Rudrasenan and D.P. Nambiar, ACS Omega, 5, 1040 (2020);
 - https://doi.org/10.1021/acsomega.9b02857
- S. Wu, S. Rajeshkumar, M. Madasamy and V. Mahendran, *Artif. Cells Nanomed. Biotechnol.*, 48, 1153 (2020); https://doi.org/10.1080/21691401.2020.1817053
- S. Mahmoodi, A. Elmi and S. Hallaj-Nezhadi, *J. Mol. Pharm. Org. Process Res.*, 6, 140 (2018); https://doi.org/10.4172/2329-9053.1000140
- 8. L. Dong, L. Han, T. Duan, S. Lin, J. Li and X. Liu, *RSC Adv.*, **10**, 2027 (2020);
 - https://doi.org/10.1039/C9RA07799E
- S.C. Mali, A. Dhaka, C.K. Githala and R. Trivedi, *Biotechnol. Rep.*, 27, e00518 (2020); https://doi.org/10.1016/j.btre.2020.e00518
- M. Siddique, N. Fayaz and M. Saeed, *Physica B Condens. Matter*, 602, 412504 (2021); https://doi.org/10.1016/j.physb.2020.412504
- P.T.S.R.K. Pavani Peddi, P. Rao, N.U. Rani and S.L. Tulasi, *J. Genet. Eng. Biotechnol.*, 19, 131 (2021); https://doi.org/10.1186/s43141-021-00229-9
- S. Liu, C. Yang, W. Xie, C. Xia and P. Fan, *Procedia Environ. Sci.*, 16, 293 (2012); https://doi.org/10.1016/j.proenv.2012.10.041

- N. Pariona, M.I. Arturo, H. Hernandez-Flores and R. Clark-Tapia, Sci. Total Environ., 575 869 (2016); https://doi.org/10.1016/j.scitotenv.2016.09.128
- Y. Tian, B. Guan, D. Zhou, J. Yu, G. Li and Y. Lour, *The Scient. World*, 2014, 834630 (2014);
 - https://doi.org/10.1155/2014/834630
- P. Acharya, G.K. Jayaprakasha, K.M. Crosby, J.L. Jifon and B.S. Patil, Sci. Rep., 10, 5037 (2020); https://doi.org/10.1038/s41598-020-61696-7
- S.R. Tariq and A. Ashraf, Arab. J. Chem., 9, 806 (2016); https://doi.org/10.1016/j.arabjc.2013.09.024
- P. Bernfeld, Methods Enzymol., 1, 149 (1955); https://doi.org/10.1016/0076-6879(55)01021-5
- H.K. Schachman, Methods Enzymol., 4, 32 (1957); https://doi.org/10.1016/0076-6879(57)04050-1
- H. Alebi, Methods Enzymol., 105, 121 (1984); https://doi.org/10.1016/s0076-6879(84)05016-3
- O.H. Lowry, N.J. Rosebrough, A.L. Farr, and Randall, *J. Biol. Chem.*, 1, 193 (1951).
- S. Shinde, P. Paralikar, A.P. Ingle and M. Rai, *Arab. J. Chem.*, 13, 3172 (2020); https://doi.org/10.1016/j.arabjc.2018.10.001
- 22. D.I. Arnon, *Plant Physiol.*, **24**, 1 (1949); https://doi.org/10.1104/pp.24.1.1
- I. Khan, K. Saeed and I. Khan, Arab. J. Chem., 12, 908 (2019); https://doi.org/10.1016/j.arabjc.2017.05.011
- J. Suárez-Cerda, H. Espinoza-Gómez, G. Alonso-Núñez, I.A. Rivero, Y. Gochi-Ponce and L.Z. Flores-López, J. Saudi Chem. Soc., 21, 341 (2017); https://doi.org/10.1016/j.jscs.2016.10.005
- J. Ramyadevi, K. Jeyasubramanian, A. Marikani, G. Rajakumar, A.A. Rahuman, T. Santhoshkumar, A.V. Kirthi, C. Jayaseelan and S. Marimuthu, *Parasitol. Res.*, 109, 1403 (2011); https://doi.org/10.1007/s00436-011-2387-3
- A. Parveen and S. Rao, J. Cluster Sci., 26, 693 (2015); https://doi.org/10.1007/s10876-014-0728-y
- 27. C. Srinivasan and R. Saraswathi, Curr. Sci., 99, 274 (2010).