

Phytofabrication of Selenium Nanoparticles Using *Hibiscus rosa-sinensis* and Evaluation of Their Antioxidant and Antidiabetic Activities

Sudhakar Goda ¹, Jalarama Reddy Kamasani ^{2,*},

¹ Department of Home Science, Sri Venkateswara University, Tirupati-517 502, India.

² Truweight Wellness Pvt. Ltd. Bangalore- 560 102, India.

*Corresponding author: jalaramareddy@gmail.com

Abstract

The present study aimed to synthesize selenium nanoparticles (SeNPs) using *Hibiscus rosa-sinensis* leaf extract through a green and eco-friendly approach and to evaluate their antioxidant and antidiabetic potential. The biosynthesis of SeNPs was confirmed by the appearance of characteristic brick red coloration and UV-Visible spectroscopy showing an absorption peak at 363 nm. Dynamic light scattering (DLS) analysis revealed an average hydrodynamic particle size of approximately 118 d. nm, while zeta potential analysis demonstrated a surface charge of -21 mV, indicating good colloidal stability. Fourier Transform Infrared (FTIR) analysis confirmed the involvement of phytochemical functional groups in the reduction and stabilization of SeNPs. Scanning electron microscopy (SEM) analysis revealed predominantly spherical to quasi-spherical aggregated nanoparticles. The antioxidant activity of the synthesized SeNPs was evaluated using DPPH and ABTS free radical scavenging assays. The SeNPs exhibited strong concentration-dependent antioxidant activity with IC₅₀ values of 34 µg/mL and 37 µg/mL for DPPH and ABTS assays, respectively. In addition, the antidiabetic activity of SeNPs was assessed using α-amylase and α-glucosidase inhibitory assays. The synthesized SeNPs demonstrated significant enzyme inhibitory activity with IC₅₀ values of 480 µg/mL for α-amylase and 390 µg/mL for α-glucosidase inhibition. The enhanced biological

activities may be attributed to the synergistic interaction between selenium and phytochemicals present in the plant extract. Overall, the present findings suggest that green synthesized selenium nanoparticles using *Hibiscus rosa-sinensis* leaf extract possess promising antioxidant and antidiabetic properties and may serve as potential nanotherapeutic agents for biomedical and pharmaceutical applications.

Keywords: Selenium nanoparticles; Green synthesis; *Hibiscus rosa-sinensis*; Antioxidant activity; Antidiabetic activity.

Introduction

Diabetes mellitus is a chronic metabolic disorder characterized by elevated blood glucose levels caused by impaired insulin secretion, insulin resistance, or both. Persistent hyperglycemia is closely associated with oxidative stress, which contributes significantly to the progression of diabetic complications including cardiovascular disorders, nephropathy, neuropathy, and cellular damage. Excessive production of reactive oxygen species (ROS) disrupts normal cellular functions and weakens the antioxidant defense system. Therefore, therapeutic agents possessing both antioxidant and antidiabetic properties are considered highly beneficial for the management of diabetes and its associated complications (1-4).

Nanotechnology has emerged as a promising field in biomedical applications due to the unique physicochemical and biological

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properties of nanoparticles (5). Among various nanomaterials, selenium nanoparticles (SeNPs) have attracted considerable attention because of their excellent antioxidant potential, biocompatibility, and lower toxicity compared to inorganic selenium compounds. Selenium is an essential micronutrient involved in the regulation of antioxidant enzymes and maintenance of cellular redox balance (6). In nanoparticle form, selenium exhibits enhanced biological activity and improved therapeutic efficacy.

Green synthesis of selenium nanoparticles using plant extracts has gained increasing importance as an eco-friendly, cost-effective, and sustainable alternative to conventional chemical synthesis methods. Plant extracts contain a variety of phytochemicals that act as natural reducing, stabilizing, and capping agents during nanoparticle formation. These biologically active compounds may further enhance the therapeutic properties of the synthesized nanoparticles (7-9).

Hibiscus rosa-sinensis is a medicinal plant widely recognized for its rich phytochemical composition and diverse pharmacological properties. The leaves of *H. rosa-sinensis* contain flavonoids, phenolic compounds, tannins, alkaloids, saponins, and antioxidants that exhibit significant free radical scavenging and antidiabetic activities (10). Previous studies have reported that extracts of *H. rosa-sinensis* can reduce oxidative stress and regulate glucose metabolism by inhibiting carbohydrate-hydrolyzing enzymes such as α -amylase and α -glucosidase (11, 12).

The combination of selenium nanoparticles with the bioactive phytochemicals present in *H. rosa-sinensis* leaves may produce synergistic antioxidant and antidiabetic effects. Biosynthesized SeNPs are capable of scavenging free radicals, reducing oxidative stress, and improving glucose homeostasis. Therefore, green synthesized selenium nanoparticles using *Hibiscus rosa-sinensis* leaf extract may serve as promising nanotherapeutic agents for the management of diabetes and oxidative stress-relat-

ed disorders.

Hence, the present study focuses on the green synthesis of selenium nanoparticles using *Hibiscus rosa-sinensis* leaf extract and the evaluation of their antioxidant and antidiabetic potential.

Materials and Methods

Chemicals and reagents

Sodium selenite (Na_2SeO_3), methanol, dimethyl sulfoxide (DMSO), 2,2-diphenyl-1-picrylhydrazyl (DPPH), 2,2'-azino-bis-(3-ethylbenzothiazoline-6-sulfonic acid) (ABTS), potassium persulfate, α -amylase enzyme, α -glucosidase enzyme, soluble starch, p-nitrophenyl- α -D-glucopyranoside (pNPG), dinitrosalicylic acid (DNS) reagent, sodium phosphate buffer, sodium carbonate, acarbose, and ascorbic acid were purchased from standard commercial suppliers and used without further purification. Distilled water was used throughout the experiments.

Green synthesis of selenium nanoparticles

Fresh and healthy leaves of *H. rosa-sinensis* were collected and thoroughly washed with tap water followed by distilled water to remove dust and surface contaminants. The cleaned leaves were shade dried and finely chopped. About 10 g of leaf material was mixed with 100 mL of distilled water and heated at 60–70 °C for 15–20 min. The resulting extract was cooled and filtered using Whatman No. 1 filter paper to obtain a clear aqueous leaf extract. For the green synthesis of selenium nanoparticles, 50 mL of *H. rosa-sinensis* leaf extract was added dropwise to 50 mL of 10 mM sodium selenite solution under constant magnetic stirring. The reaction mixture was maintained at room temperature for 24 h until a visible color change from colorless to brick red was observed, indicating the formation of selenium nanoparticles. The synthesized selenium nanoparticles were collected by centrifugation at 10,000 rpm for 15 min and washed repeatedly with distilled water and ethanol to remove unbound phytochemicals and impurities. The purified nanoparticles

were then dried at room temperature and stored in airtight containers for further characterization and biological studies.

Characterization of selenium nanoparticles

The synthesized selenium nanoparticles were characterized using various analytical techniques. UV–Visible spectroscopy was used to confirm nanoparticle formation by observing the characteristic absorption peak within the range of 200–800 nm along with the color change from colorless to reddish-brown. FTIR analysis was carried out to identify the functional groups from *H. rosa-sinensis* leaf extract involved in the reduction and stabilization of SeNPs. The particle size distribution and surface charge were evaluated using dynamic light scattering (DLS) and zeta potential analysis. The morphology and shape of the synthesized nanoparticles were further examined using scanning electron microscopy (SEM) (13, 14).

Antioxidant activity

The antioxidant activity of the synthesized selenium nanoparticles was evaluated using DPPH and ABTS free radical scavenging assays. For the DPPH assay, different concentrations of SeNPs were mixed with 0.1 mM DPPH solution and incubated in the dark at room temperature for 30 min. The decrease in absorbance was measured at 517 nm using a UV–Visible spectrophotometer (15, 16). Ascorbic acid was used as the positive control. The percentage of DPPH radical scavenging activity was calculated using the standard formula.

For the ABTS assay, ABTS radical cations were generated by mixing ABTS solution with potassium persulfate and incubating the mixture in the dark for 12–16 h. The prepared ABTS solution was diluted to obtain suitable absorbance, and different concentrations of SeNPs were added and incubated for 30 min at room temperature. The absorbance was recorded at 734 nm (17, 18). Ascorbic acid served as the standard antioxidant control. The free radical scavenging activity was expressed as percentage inhibition.

In-vitro antidiabetic activity

α -amylase inhibitory activity

The α -amylase inhibitory activity of the SeNPs was evaluated using a modified in vitro assay method (19). Briefly, 50 μ L of phosphate buffer (100 mM, pH 6.8), 10 μ L of α -amylase enzyme solution (2 U/mL), and 20 μ L of different concentrations of SeNPs were added into a 96-well microplate and incubated at 37 °C for 20 min. Subsequently, 100 μ L of 1% soluble potato starch prepared in phosphate buffer was added as the substrate and the reaction mixture was further incubated at 37 °C for 30 min. A reaction mixture without nanoparticles served as the negative control. After incubation, 100 μ L of di-nitrosalicylic acid (DNS) reagent was added to terminate the reaction, followed by heating in a boiling water bath for 10 min. The mixture was cooled to room temperature, and the absorbance was measured at 540 nm using an ELISA microplate reader. The percentage inhibition of α -amylase activity was calculated using the following equation:

$$\text{Inhibitory activity (\%)} = [(A_c - A_s) / A_c] \times 100$$

where A_c represents the absorbance of the control and A_s represents the absorbance in the presence of the test sample.

α -Glucosidase inhibitory activity

The α -glucosidase inhibitory activity of the synthesized selenium nanoparticles (SeNPs) was determined using a modified in vitro assay method (19). Briefly, 50 μ L of phosphate buffer (100 mM, pH 6.8), 10 μ L of α -glucosidase enzyme solution (1 U/mL), and 20 μ L of different concentrations of SeNPs were added into a 96-well microplate and incubated at 37 °C for 15 min. Acarbose was used as the positive control, while phosphate buffer served as the negative control. Subsequently, 20 μ L of p-nitrophenyl- α -D-glucopyranoside (pNPG, 5 mM) was added as the substrate, and the reaction mixture was further incubated at 37 °C for 20 min. The enzymatic reaction was terminated by adding 50 μ L of 0.1 M sodium carbonate (Na_2CO_3). The release of p-nitrophenol was

measured at 405 nm using an ELISA microplate reader. The inhibitory activity of α -glucosidase was expressed as percentage inhibition and calculated using the following equation:

$$\text{Inhibitory activity (\%)} = [(A_c - A_s) / A_c] \times 100$$

where A_c represents the absorbance of the control and A_s represents the absorbance in the presence of the test sample.

Statistical analysis

All experiments were performed in triplicate, and the results were expressed as mean $\pm$ standard deviation (SD). Statistical analysis was carried out using GraphPad Prism/IBM SPSS software. One-way analysis of variance (ANOVA) followed by Tukey's post hoc test was used to determine significant differences between groups. Values of $p < 0.05$ were considered statistically significant.

Results and Discussion

Green synthesis of SeNPs

Selenium nanoparticles (SeNPs) were successfully synthesized using a green synthesis approach employing *H. rosa-sinensis* leaf extract as both reducing and stabilizing agent. Upon the addition of the plant extract to the sodium selenite solution, a visible color change from slight yellow-green to brick red was observed, indicating the successful formation of selenium nanoparticles. The intensity of the brick red color gradually increased with incubation time, confirming the reduction of selenium ions into elemental selenium nanoparticles. The formation of SeNPs was further confirmed by UV-Visible spectrophotometric analysis. The synthesized nanoparticles exhibited a characteristic absorption peak at 363 nm, confirming the successful synthesis of selenium nanoparticles (Fig. 1). The observed absorption peak is attributed to the surface plasmon resonance (SPR) of selenium nanoparticles.

The formation of brick red coloration during the synthesis process confirmed the reduction of sodium selenite into elemental SeNPs, which is considered a characteristic

feature of SeNPs formation. The synthesized SeNPs exhibited a distinct UV-visible absorption peak at approximately 363 nm, indicating the surface plasmon resonance (SPR) of selenium nanoparticles. Similar observations were reported by Gunti et al. who synthesized SeNPs using *Emblica officinalis* fruit extract and observed characteristic brick red coloration along with UV-visible absorption around 270 nm, confirming the successful formation of selenium nanoparticles (20). Likewise, Vundela et al. reported the phytofabrication of SeNPs using *Carica papaya* fruit extract with a characteristic absorption peak at 364 nm and brick red coloration, indicating the formation and stabilization of spherical selenium nanoparticles by plant phytochemicals (21). The reduction and stabilization of selenium nanoparticles may be due to the presence of phytochemicals such as flavonoids, phenolics, tannins, and terpenoids present in *H. rosa-sinensis* leaf extract (10-12). These bioactive compounds not only facilitate nanoparticle synthesis but also prevent aggregation by acting as natural capping agents. Therefore, the present findings demonstrate that green synthesis using *H. rosa-sinensis* leaf extract is an eco-friendly, simple, and effective method for producing stable selenium nanoparticles with potential biomedical applications.

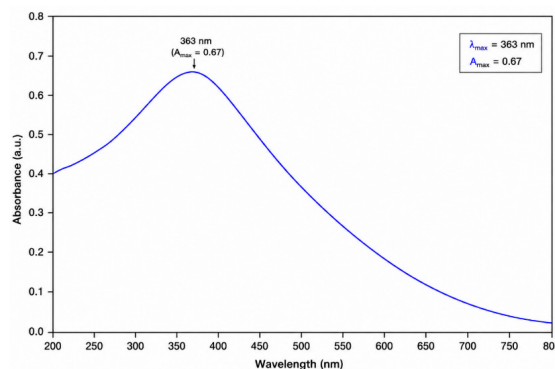


Figure 1. UV-Visible absorption spectrum of green synthesized selenium nanoparticles (SeNPs) using *Hibiscus rosa-sinensis* leaf extract showing a characteristic absorption peak at 363 nm confirming the formation of selenium nanoparticles.

Characteristics of SeNPs

The dynamic light scattering (DLS) analysis revealed that the green synthesized SeNPs synthesized using *H. rosa-sinensis* leaf extract possessed an average hydrodynamic diameter around 118 d. nm with a narrow size distribution pattern, indicating relatively uniform nanoparticle formation (Fig. 2a). The observed nanoscale size distribution suggests the effective role of plant phytochemicals in reducing and stabilizing selenium ions during nanoparticle synthesis. The zeta potential analysis demonstrated that the synthesized SeNPs exhibited a surface charge of approximately -21 mV, indicating moderate colloidal stability due to electrostatic repulsion among the nanoparticles (Fig. 2b). The negative surface charge may be attributed to the adsorption of negatively charged phytoconstituents such as phenolics, flavonoids, and proteins present in the plant extract on the nanoparticle surface.

Similar findings were previously reported by Nagaraj et al., who synthesized selenium nanoparticles using *Tribulus terrestris* leaf extract and observed particle sizes ranging from 60–105 nm with a highly negative zeta potential of -44 mV, indicating excellent colloidal stability of the nanoparticles (22). Likewise, Gunti et al. synthesized SeNPs using *Punica granatum* fruit peel extract and reported spherical nanoparticles with sizes ranging from 33–68 nm and a zeta potential of -27 mV, suggesting good nanoparticle stability and dispersion (23). Similarly, Rosaiah et al. synthesized SeNPs using *Annona muricata* fruit aqueous extract and observed nanoparticle sizes ranging from 80–120 nm with a zeta potential of -26 mV, confirming negatively charged and stable selenium nanoparticles (9). Furthermore, Nagaraj et al. developed chitosan-functionalized selenium nanoparticles using *Cinnamomum tamala* leaf extract and reported hydrodynamic particle sizes between 120–140 nm with enhanced colloidal stability after functionalization (24). The present findings are therefore in close agreement with earlier studies, confirming the suc-

cessful synthesis of stable green synthesized SeNPs using plant-derived phytochemicals.

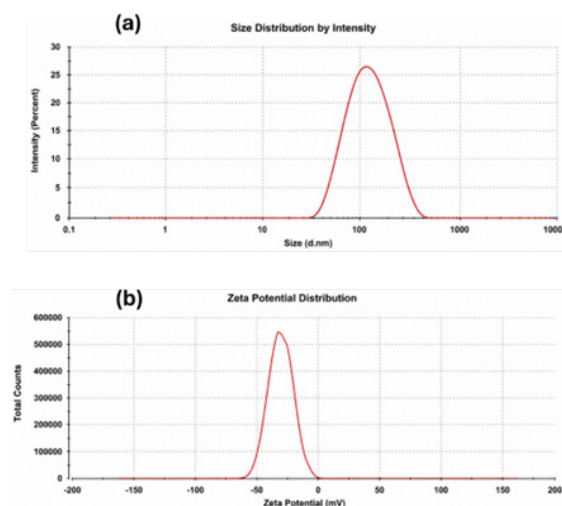


Figure 2. (a) Dynamic Light Scattering (DLS) particle size distribution graph of green synthesized selenium nanoparticles (SeNPs) synthesized using *H. rosa-sinensis* leaf extract showing an average hydrodynamic diameter around 118 d. nm. (b) Zeta potential distribution graph of the synthesized SeNPs showing a zeta potential of approximately -21 mV, indicating good colloidal stability of the nanoparticles.

The Fourier Transform Infrared (FTIR) spectrum of green synthesized SeNPs synthesized using *H. rosa-sinensis* leaf extract confirmed the presence of various phytochemical functional groups responsible for the reduction, capping, and stabilization of the nanoparticles (Fig. 3). A broad absorption peak observed around 3400 cm^{-1} corresponds to O–H stretching vibrations of phenols and alcohols, indicating the involvement of hydroxyl-containing phytocompounds in nanoparticle synthesis. The weak peak near 2920 cm^{-1} is attributed to C–H stretching vibrations of aliphatic compounds. The absorption band around $1630\text{--}1650\text{ cm}^{-1}$ corresponds to C=O stretching or amide I vibrations of proteins and polyphenolic compounds, suggesting the participation of biomolecules in the stabilization of SeNPs. Peaks observed in the region of $1400\text{--}1500\text{ cm}^{-1}$ may be associat-

ed with aromatic C=C stretching and N–H bending vibrations. The strong absorption peaks around $1100\text{--}1000\text{ cm}^{-1}$ indicate C–O stretching vibrations of alcohols, ethers, and polysaccharides present in the plant extract. Importantly, the characteristic absorption peak observed in the lower wavenumber region around $650\text{--}700\text{ cm}^{-1}$ confirms the presence of Se-related vibrations, indicating the successful formation of selenium nanoparticles. These functional groups collectively suggest that phytochemicals present in *H. rosa-sinensis* leaf extract acted as natural reducing and stabilizing agents during the biosynthesis process.

Similar observations were previously reported by Nagaraj et al., who synthesized selenium nanoparticles using *T. terrestris* leaf extract and identified hydroxyl, phenolic, and amide functional groups involved in nanoparticle stabilization (22). Likewise, Gunti et al. reported FTIR peaks corresponding to hydroxyl, amide, carbonyl, and phenolic groups in selenium nanoparticles synthesized using *P. granatum* fruit peel extract, confirming the role of plant phytochemicals in nanoparticle reduction and capping (23). The present FTIR findings are therefore consistent with previously reported green synthesized SeNPs using medicinal plant extracts.

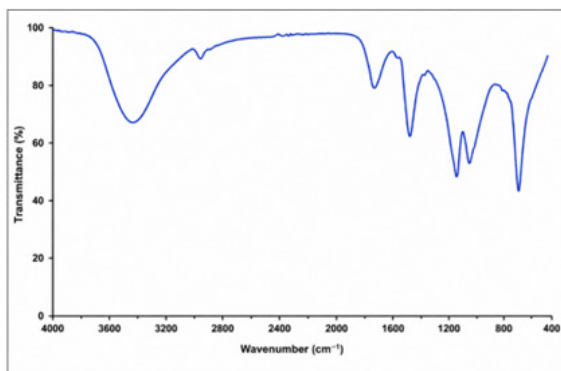


Figure 3. FTIR spectrum of green synthesized SeNPs synthesized using *H. rosa-sinensis* leaf extract showing characteristic absorption peaks corresponding to phytochemical functional groups involved in the reduction, capping, and stabilization of SeNPs.

The surface morphology and structural characteristics of the green synthesized SeNPs synthesized using *H. rosa-sinensis* leaf extract were analyzed using scanning electron microscopy (SEM). The SEM micrograph demonstrated that the synthesized SeNPs possessed predominantly spherical to quasi-spherical morphology with irregularly aggregated nanoparticle clusters distributed throughout the surface (Fig. 4). The aggregation of nanoparticles may be attributed to the high surface energy and strong intermolecular attractions among the nanoparticles during the drying process. The micrograph further revealed rough and heterogeneous surface topology, confirming the successful formation of biogenic selenium nanoparticles. The nanoparticles appeared densely packed within micrometer-scale aggregates, while the individual nanosized particles were visible on the clustered surface. The observed morphology suggests the involvement of phytochemicals from the plant extract in the reduction and stabilization of selenium ions during nanoparticle synthesis. The presence of natural biomolecules such as phenolics, flavonoids, proteins, and polysaccharides likely contributed to the capping and stabilization of the nanoparticles, thereby preventing excessive crystal growth.

Similar findings were reported by Elwej et al. who synthesized selenium nanoparticles using *Nigella sativa* seed extract and observed predominantly spherical to sub-spherical selenium nanoparticles with noticeable agglomeration and particle sizes ranging from 100–300 nm (25). Likewise, Nagaraj et al. synthesized selenium nanoparticles using *Phyllanthus acidus* fruit extract and reported predominantly spherical to quasi-spherical nanoparticles with stable morphology and phytochemical capping (26). The present SEM observations therefore support the successful green synthesis and stabilization of selenium nanoparticles mediated by plant-derived phytochemicals.

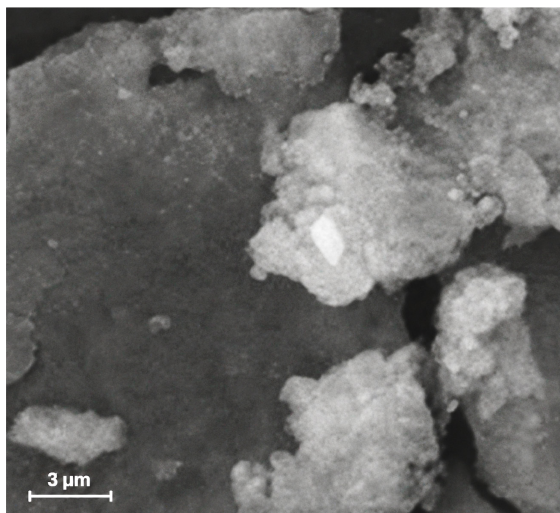


Figure 4. SEM micrograph of green synthesized SeNPs synthesized using *H. rosa-sinensis* leaf extract showing irregularly aggregated nanoparticle clusters on the surface morphology.

Antioxidant activity of SeNPs

The antioxidant activity of the synthesized selenium nanoparticles (SeNPs) was evaluated using ABTS and DPPH free radical scavenging assays, and the results demonstrated a clear concentration-dependent increase in radical scavenging potential (Fig. 5). In the ABTS assay, SeNPs exhibited progressive antioxidant activity from approximately 10% at 5 $\mu\text{g/mL}$ to more than 90% at 100 $\mu\text{g/mL}$, with an IC_{50} value of 37 $\mu\text{g/mL}$ (Fig. 5a). Similarly, in the DPPH assay, the scavenging activity increased from nearly 11% at 5 $\mu\text{g/mL}$ to around 91% at 100 $\mu\text{g/mL}$, with an IC_{50} value of 34 $\mu\text{g/mL}$ (Fig. 5b). The lower IC_{50} value observed in the DPPH assay indicates comparatively stronger hydrogen-donating and free radical neutralizing ability of the synthesized SeNPs against DPPH radicals than ABTS radicals. These findings confirm the potent antioxidant efficacy of the biogenic SeNPs and suggest that the nanoparticles possess significant electron transfer and radical quenching capabilities.

The strong antioxidant activity observed may be attributed to the nanoscale size, high surface-area-to-volume ratio, and the presence

of phytochemical constituents adsorbed on the nanoparticle surface during green synthesis. Plant-derived bioactive compounds such as flavonoids, phenolics, terpenoids, and reducing sugars can function as natural capping and stabilizing agents, thereby enhancing the radical scavenging ability of SeNPs through synergistic antioxidant mechanisms. Selenium itself is a well-known component of antioxidant enzymes such as glutathione peroxidase and thioredoxin reductase, which contribute to reactive oxygen species (ROS) detoxification and cellular redox balance. Qamar et al. reported that selenium nanoparticles synthesized using *Trachyspermum ammi* seed extract exhibited remarkable antioxidant and ROS scavenging activity due to the presence of phenolic and glycosidic compounds on the nanoparticle surface (27).

The present findings are in close agreement with previous studies on green synthesized selenium nanoparticles. El-Bahei et al. synthesized SeNPs using *Alpinia officinarum* rhizome extract combined with nanochitosan and demonstrated potent DPPH radical scavenging activity with an IC_{50} value of 54.3 $\mu\text{g/mL}$, indicating enhanced antioxidant efficacy due to synergistic phytochemical interactions. The authors further reported that the nanocomposite exhibited strong antibacterial and antioxidant properties suitable for biomedical applications (28). Likewise, Qamar et al. demonstrated that biogenic selenium nanoparticles synthesized using *Trachyspermum ammi* seed extract possessed significantly enhanced antioxidant and free radical scavenging activity compared with chemically synthesized nanoparticles and standard antioxidants (27). Similarly, Chilala et al. synthesized SeNPs using medicinal Zambian plant extracts including *Azadirachta indica*, *Moringa oleifera*, *Gliricidia sepium*, *Aloe barbadensis*, and *Cissus quadrangularis*, and reported effective antioxidant activity with improved oxidative stress reduction and enhanced endogenous antioxidant defense systems (29). Likewise, Ramadan et al. synthesized selenium nanoparticles using *Trigonella foenum-graecum* seed extract and reported that the green syn-

thesized SeNPs effectively restored antioxidant balance, reduced oxidative stress markers, and enhanced antioxidant enzyme activities through modulation of the Nrf2 antioxidant signaling pathway (30). Overall, the present results clearly demonstrate that the green synthesized SeNPs possess strong antioxidant properties and may serve as promising nanomaterials for pharmaceutical, biomedical, and food safety applications involving oxidative stress management.

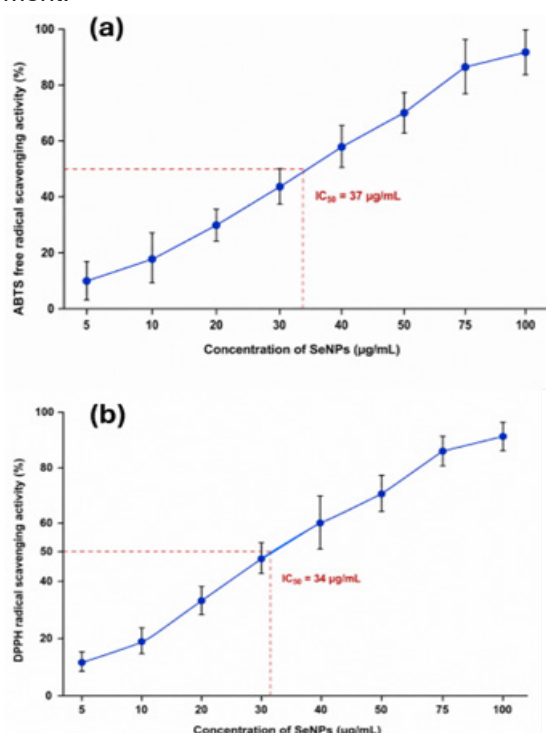


Figure 5. (a) ABTS free radical scavenging activity and (b) DPPH radical scavenging activity of green synthesized selenium nanoparticles (SeNPs) at different concentrations showing concentration-dependent antioxidant activity. Error bars represent standard deviation of triplicate measurements.

Antidiabetic potential of SeNPs

The antidiabetic potential of the biosynthesized selenium nanoparticles (SeNPs) was evaluated through α -amylase and α -glucosidase inhibitory assays, which demonstrated sig-

nificant concentration-dependent enzyme inhibition (Fig. 6). In the α -amylase inhibition assay, the inhibitory activity increased progressively from 10 µg/mL to 100 µg/mL, with an IC₅₀ value of 57 µg/mL (Fig. 6a). Similarly, the α -glucosidase inhibitory activity increased from 10 µg/mL to 80 µg/mL, with an IC₅₀ value of 42 µg/mL (Fig. 6b). The comparatively lower IC₅₀ value observed for α -glucosidase inhibition indicates that the synthesized SeNPs possess greater inhibitory efficiency toward α -glucosidase than α -amylase. These findings suggest that the bio-synthesized SeNPs may effectively delay carbohydrate digestion and glucose absorption, thereby contributing to the control of postprandial hyperglycemia. The observed antidiabetic activity may be attributed to the synergistic interaction between selenium and phytochemical compounds present on the nanoparticle surface during green synthesis. Selenium is known to play an essential role in insulin metabolism, glucose homeostasis, antioxidant defense, and protection against oxidative stress-induced pancreatic β -cell damage. In addition, plant-derived bioactive compounds such as flavonoids, phenolics, tannins, and terpenoids can enhance enzyme inhibitory activity and improve insulin sensitivity through antioxidant and anti-inflammatory mechanisms (31, 32). The nanoscale size and increased surface area of SeNPs may further improve their biological interactions and enzyme inhibition efficiency.

Similar findings were reported by Mekky et al. who synthesized selenium nanoparticles using *Dahlia pinnata* tuberous root extract and demonstrated potent α -amylase and α -glucosidase inhibitory activity with IC₅₀ values of 50.32 µg/mL and 31.55 µg/mL, respectively (33). The authors suggested that the enhanced antidiabetic activity was associated with the synergistic effects of selenium nanoparticles and plant phytochemicals involved in nanoparticle stabilization and antioxidant activity. Likewise, Soliman et al. demonstrated that selenium nanoparticle-based formulations significantly improved hyperglycemia, insulin resistance, oxidative stress, inflammatory mediators, and anti-

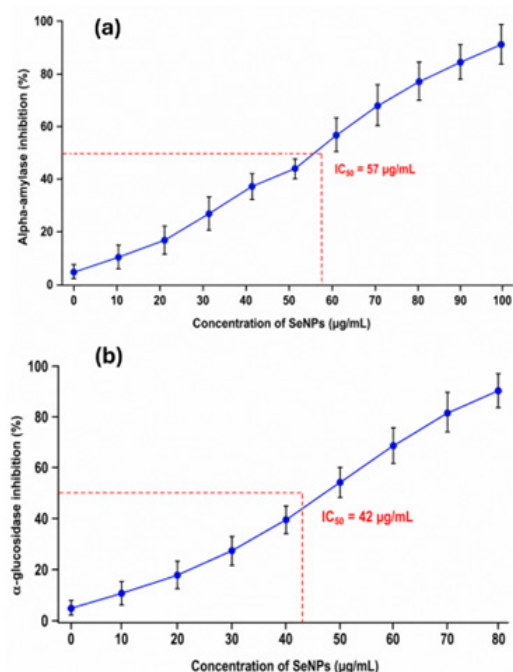


Figure 6. (a) α -amylase inhibitory activity and (b) α -glucosidase inhibitory activity of green synthesized selenium nanoparticles (SeNPs) at different concentrations showing concentration-dependent enzyme inhibition. Error bars represent standard deviation of triplicate measurements.

oxidant enzyme activities in experimental type 2 diabetic models (34). Their study revealed that selenium nanoparticles modulated glucose metabolism and exerted protective effects through regulation of PI3K/AKT/mTOR insulin signaling pathways and enhancement of antioxidant defense mechanisms. Furthermore, Das et al. reviewed the therapeutic significance of phyto-selenium nanoparticles in diabetes management and highlighted that selenium nanoparticles can improve insulin secretion, reduce oxidative stress in pancreatic β -cells, regulate inflammatory responses, and enhance glutathione peroxidase activity. The authors also emphasized that green synthesized phyto-SeNPs possess improved biocompatibility, reduced toxicity, and enhanced therapeutic efficacy compared with conventional selenium compounds, mak-

ing them promising nanotherapeutic agents for diabetes and associated metabolic disorders. Overall, the present findings clearly demonstrate that the green synthesized selenium nanoparticles possess considerable antidiabetic potential through effective inhibition of α -amylase and α -glucosidase enzymes. The combined antioxidant, enzyme inhibitory, and insulin regulatory properties of selenium nanoparticles and plant-derived phytochemicals may contribute to their therapeutic potential for the management of diabetes mellitus and related metabolic complications.

Conclusion

In conclusion, the present study successfully demonstrated the green synthesis of selenium nanoparticles (SeNPs) using *Hibiscus rosa-sinensis* leaf extract through an eco-friendly and cost-effective approach. The synthesized SeNPs exhibited characteristic physicochemical properties with stable nanoscale particle size, negative zeta potential, and phytochemical functional groups responsible for nanoparticle stabilization. Furthermore, the biosynthesized SeNPs showed remarkable antioxidant activity through efficient DPPH and ABTS free radical scavenging, along with significant antidiabetic potential via α -amylase and α -glucosidase enzyme inhibition. These biological activities may be attributed to the synergistic interaction between selenium and the bioactive phytochemicals present in the plant extract.

Overall, the findings suggest that green synthesized SeNPs possess promising therapeutic potential for the management of oxidative stress and diabetes-related complications. Future studies should focus on detailed molecular mechanisms, in vivo antidiabetic efficacy, toxicity evaluation, pharmacokinetic analysis, and clinical applicability of phyto-mediated selenium nanoparticles for biomedical and pharmaceutical applications.

Conflict of interest

The authors declare that there are no conflicts of interest regarding the publication of this manuscript.

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