

**Research Article****Preparation and antioxidant activity of selenium nanoparticles by using herbal extract****Runjhun Pallavi¹, Prasanth Damarasingu², Asmita Patil³, K Sreedhar Naik⁴, Sowjanya Pulipati⁵, Pundareekaksha Rao⁶, Ikram⁷, Venkata Suresh Jilakara^{8*}**¹Associate Professor, Department of Pharmaceutics, SGT College of Pharmacy, SGT University Gurugram, India²Associate Professor, Department of Pharmacology, School of Pharmacy, Centurion University of Technology and Management, Gopalpur, Balasore, Odisha, 756044, India³Shobhaben Pratapbhai Patel School of Pharmacy & Technology Management, SVKM's NMIMS, Vile Parle (W), Mumbai, Maharashtra, 400049, India⁴Department of Pharmacology, JNTUA-Oil Technological and Pharmaceutical Research Institute, Ananthapuramu, Andhra Pradesh, 515001, India⁵Professor, Vignan Pharmacy College, Vadlamudi, Guntur, Andhra Pradesh, India⁶Professor & HOD, Ayurveda College, Coimbatore, Tamil Nadu, India⁷Principal, Adarsh College of Pharmacy, Jaspur, Uttarakhand, India⁸Professor and Principal, Vathsalya College of Pharmacy, Anantharam, Bhuvanagiri, Telangana, India***Corresponding author: Venkata Suresh Jilakara**, Professor and Principal, Vathsalya College of Pharmacy, Anantharam, Bhuvanagiri, Telangana, India

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ABSTRACT

This research looked into how well Aloe vera juice works as an antioxidant in the production of selenium nanoparticles. A UV-visible spectrophotometer, Transmission electron microscopy and the ABTS and DPPH tests were used to do the testing. The UV-Vis spectrum of aloe vera extract at 350 nm shows that it contains round and hollow particles with sizes between 7 and 48 nm. These particles are more stable over longer periods of time. The results from the ABTS and DPPH tests show that the aloe vera extract that was used to make selenium nanoparticles worked better than the extract that was used in its original form. Selenium nanoparticles were made when proteins in the Aloe vera extract joined together to form a cap and keep the particles stable. Even though it is poisonous, selenite has antioxidant qualities and is now being used as a chemopreventive drug to help find cancer. Selenium that is in the form of elements or nanoparticles is not as dangerous.

KEYWORDS: Synthesis, Antioxidant Activity, Selenium nanoparticles, Aloe vera, DPPH

INTRODUCTION:

Over the past ten years, nanotechnology has been a major area of study. Nanotechnology has become a method that connects different fields in biochemical applications, with the main goal of treating diseases like Alzheimer's and cancer (Attia, *et al.*, 2023). Chemicals and physical methods can both be used to make nanoparticles (Amarachinta, *et al.*, 2021). Microorganisms, enzymes, plants, and plant products could be used instead of expensive physical tools and dangerous chemicals in bio nanotechnology production. Nanotechnology says that a nanoparticle is a very small item with one or fewer dimensions that moves as a single unit (Akhilesh, *et al.*, 2012). Selenium can be in a number of different oxidation states, such as +6 and +4. Biology, chemistry, physics, and health are all areas where selenium shows great promise and has special qualities (Aboud, *et al.*, 2016). Selenium nanoparticles could be used as antioxidants, antimicrobials, and anticancer agents, but they are very toxic, which makes it hard to make steady selenium nanoparticles that can be used safely in living things (Sabareesh, *et al.*, 2020).

A lot of antioxidants, like polyphenolic chemicals, can be found in fruits and veggies. Some nutrients, like vitamin E and C, are thought to help you avoid getting diseases linked to reactive stress the most (Suryawanshi, *et al.*, 2021). Unstable oxygen species, which are very unstable and bad for cells, cause oxidative stress (7). They are made during the redox process in metabolism. Reactive oxygen species (ROS) can be completely removed by using antioxidant enzymes and substances that are not enzymes (Sharma, *et al.*, 2018). A study shows that antioxidants can help protect against diseases that are caused by free radicals. Aloe vera is grown for its usefulness in farming and medicine, but it can also be found naturally in warm areas around the world (Ahire, *et al.*, 2020). This plant can also be grown successfully indoors in a pot as a decoration. Vitamins A, C, and F are the main vitamins that can be found in aloe vera leaves (Sharma, *et al.*, 2016). To do this, the study looked at how well the ABTS and DPPH tests worked. The study also looked at how well Aloe vera leaf could lower the potential for making and keeping selenium nanoparticles (Sahoo, *et al.*, 2014).

MATERIALS AND METHODS

Preparation of extract of Aloe vera:

30 grams of aloe vera leaves that have been cleansed with great care. In twenty milliliters of sterile distilled water, mince the ingredients very finely. Following that, the extract was filtered using Whatman filter paper and then put away at a temperature of 4 degrees Celsius for use at a later time (Ramkanth, *et al.*, 2018 and Behera, *et al.*, 2010).

Synthesis of selenium nanoparticles:

Five millimoles of sodium selenite were mixed together in 25 ml of water and put in a flask that was set on a magnetic stirrer. After that, small amounts of aloe vera extract were added to the flask holding the sodium selenite solution until the color of the solution changed in a way that could be seen (Surana and Mahajan, 2022). Someone got 5 milliliters (ml) of this solution and used that as the test. The last 20 milliliters of fluid were kept in a dark place and stirred for 72 hours. It was seen that the color of the solution had changed after a few days (Reddy and Gandla, 2022).

UV Visible spectroscopic analysis of selenium nanoparticles:

The UV-Vis spectrum was examined subsequent to a color transition that spanned a duration of ten to fifteen minutes in order to discern the reduction in metallic selenium ions. A minute specimen was isolated from the solution through the utilization of a UV-Vis spectrophotometer,

with a precise wavelength range of 250 to 700 nanometers (Surana, *et al.*, 2022 and Keservani, *et al.*, 2010).

Transmission Electron Microscopic analysis of selenium nanoparticles:

This Techni 20 was used to do Transmission Electron Microscopic (TEM) research. To create an extremely thin layer of the sample on a carbon-coated copper grid, an extremely small quantity of the sample was applied. A 10 minute centrifugation at 13000 rpm was used on the Aloe vera juice that had Se nanoparticles in it. To get rid of any organic impurities that might be in the nanoparticles, the solid that had been gathered was centrifuged at 13,000 revolutions per minute for 10 minutes after being re-suspended in deionized water. The last step was to freeze dry the pellet using a lyophilizer (Yeola, *et al.*, 2023 and Pawar, *et al.*, 2023).

Evaluation of Antioxidant Activity of selenium nanoparticles:

ABTS (2, 2-azino-bis-3-ethylbenzothiazoline-6-sulphonic acid) assay:

The way the ABTS test was done was changed in a few small ways. We used the plant extracts' ability to get rid of the cationic free radical ABTS to figure out what percentage of ABTS radical suppression the extracts caused (Deepika, *et al.*, 2020 and Ahmad, *et al.*, 2017). There were different amounts of aloe vera juice (from 100 to 600 µg/ml) and Se nanoparticles made by living things. Each specimen was combined with 3 ml of 0.1 mmol ABTS and allowed to rest in the dark for 15 minutes. A wavelength of 745 nm was used to measure the amount of decolorization. In this test, Rutin was used as a reference, and the control solution was just ABTS reagent with no sample (Gandra, 2020 and Kakar, *et al.*, 2010). The following formula was used to figure out what amount of ABTS+ scavenging inhibition the extract had:

$$\% \text{ Inhibition} = (\text{Abs control} - \text{Abs sample}) / \text{Abs control} * 100$$

DPPH (Diphenyl picryl hydrazine) assay:

In order to assess the antioxidant properties of plant products and their ability to eliminate free radicals, we examined their capacity to convert the stable free radical DPPH to diphenyl picryl hydrazine. Using spectrophotometry, the quantity of color change from purple to yellow over a range of 517 nanometers was determined. Se nanoparticles produced by living organisms were combined with 3 ml of 0.1 mmol DPPH and allowed to remain in the dark for 15 minutes. A variety of concentrations of aloe vera extract were combined, spanning from 0 to 100 µg/ml. In the experiment, rutin was employed as the standard, while a control consisting of DPPH methanol solution devoid of any sample was utilized (Sonawane, *et al.*, 2023). After complete mixing, the reaction mixture was left at ambient temperature for 30 minutes without any visible light. Spectrophotometry was employed to quantify the absorption at a 517 nm wavelength. Using the aforementioned technique, the efficacy of the plant extract in removing free radicals was determined (Govindarajan, *et al.*, 2022 and Aher, *et al.*, 2023)

$$\% \text{ DPPH Scavenging inhibition} = (\text{Abs control} - \text{Abs sample}) / \text{Abs control} * 100$$

RESULTS AND DISCUSSION

Selenium Nanoparticles observation:

A consistent alteration in the color of the reaction medium occurs when biomolecules aid in the reduction of metal compounds to metal nanoparticles. The continuous investigation reveals that the colorless sodium selenite solution (A) undergoes a transformation into a yellowish (B) upon the progressive addition of Aloe vera leaf extract at the onset of the experiment. As the reduction

process continues for a period of 24 hours, the reaction medium progressively transforms into a hue of deep pink, as depicted in figure 1.

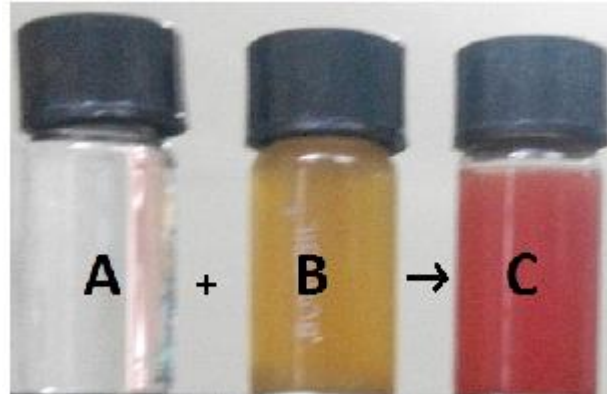


Figure 1: Change in color of Selenium Nanoparticles

UV Visible spectroscopic analysis of selenium nanoparticles:

Spectral scanning was utilized to determine whether or not the aloe vera juice contained selenium nanoparticles. The scanning process utilized wavelengths between 250 and 700 nanometers. It can be seen in Figure 2 that a colloidal solution absorbed the most light at a range of 350 nm. In the beginning, the colloidal solution was white, but after being left alone for 24 hours, it changed color and became dark brown. There is a clear sign that particles are being made gradually during the incubation time because the absorbance shows a peak at 268 nm.

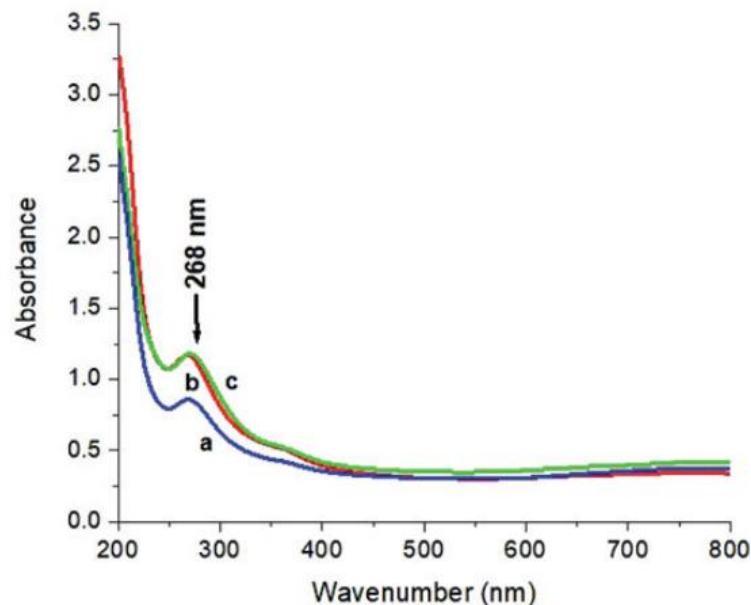


Figure 2: UV Spectrum of selenium nanoparticles

Transmission Electron Microscopic analysis of selenium nanoparticles:

A TEM study showed that selenium nanoparticles were formed in the colloidal solution. The range of particle sizes made by Aloe vera juice is shown in Figure 3. They are between 7 and 48 nm. The fact that different-sized particles are present shows that the aloe vera juice can make polydisperse nanoparticles.

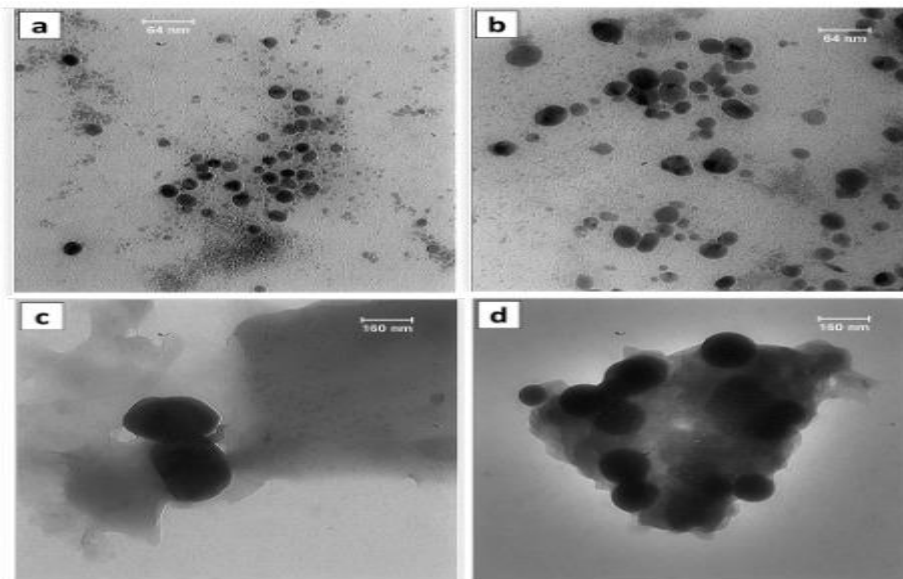


Figure 3: Transmission Electron Microscopic analysis of selenium nanoparticles

Evaluation of Antioxidant Activity of selenium nanoparticles:

ABTS (2, 2-azino-bis-3-ethylbenzothiazoline-6-sulphonic acid) assay:

Rutin was employed as a positive control in the ABTS scavenging experiment to assess the antioxidant activity of selenium nanoparticles generated biogenetically and determine the potency of the compounds in mitigating damage. To make Selenium nanoparticles for this study, different amounts were used. The nanoparticles were able to stop 75% of the molecules on average. But as the quantity of Aloe vera extract went up, the inhibitory effect got stronger, and the average level of inhibition was 55% (Table 1). The picture in Figure 4 shows that selenium nanoparticles made in a lab using aloe vera juice are much better at getting rid of ABTS radicals.

Table 1: Percentage inhibition of synthesized nanoparticles in ABTS assay

Concentration ($\mu\text{g/ml}$)	Percentage Inhibition		
	Selenium nanoparticle	Se-Aloe vera	Rutin
100	35	60	62
200	40	65	67
300	45	70	72
400	50	75	78
500	55	80	83
600	60	85	89

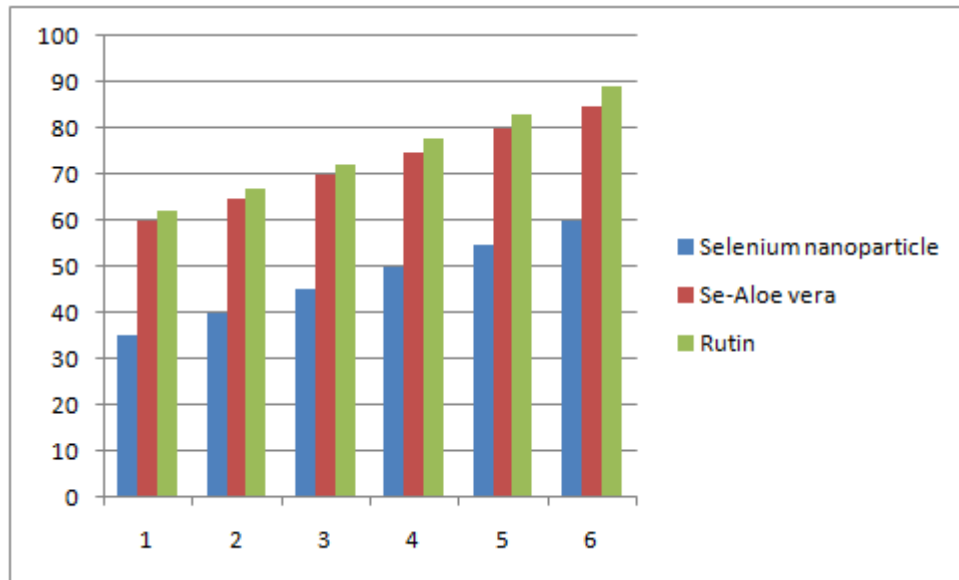


Figure 4: synthesize selenium nanoaprticles' increased antioxidant activity as demonstrated by the ABTS assay

DPPH (Diphenyl picryl hydrazine) assay:

Antioxidants are chemicals that stop the oxidative chain reaction from moving forward, which stops the oxidation of important biological proteins. The DPPH scavenging assay employed rutin as a standard to quantify the reducing capacity of compounds and the antioxidant activity of biogenic selenium nanoparticles. The picture in Figure 5 shows that selenium nanoparticles made in a lab using aloe vera juice are much better at getting rid of DPPH radicals. The goal of this study was to find out how well selenium nanoparticles made from aloe vera juice work as antioxidants. Researchers have been working on making a new group of organic antioxidants that are very good at fighting oxidative stress and stopping diseases from getting worse, while also taking into account the bad effects of man-made antioxidants. The DPPH scavenging activity test for the study showed that the nanoparticles made were very active (Table 2).

Table 2: Percentage inhibition of synthesized nanoparticles in DPPH assay

Concentration ($\mu\text{g/ml}$)	Percentage Inhibition		
	Selenium nanoparticle	Se-Aloe vera	Rutin
0	35	60	62
20	40	65	67
40	45	70	72
60	50	75	78
80	55	80	83
100	60	85	89

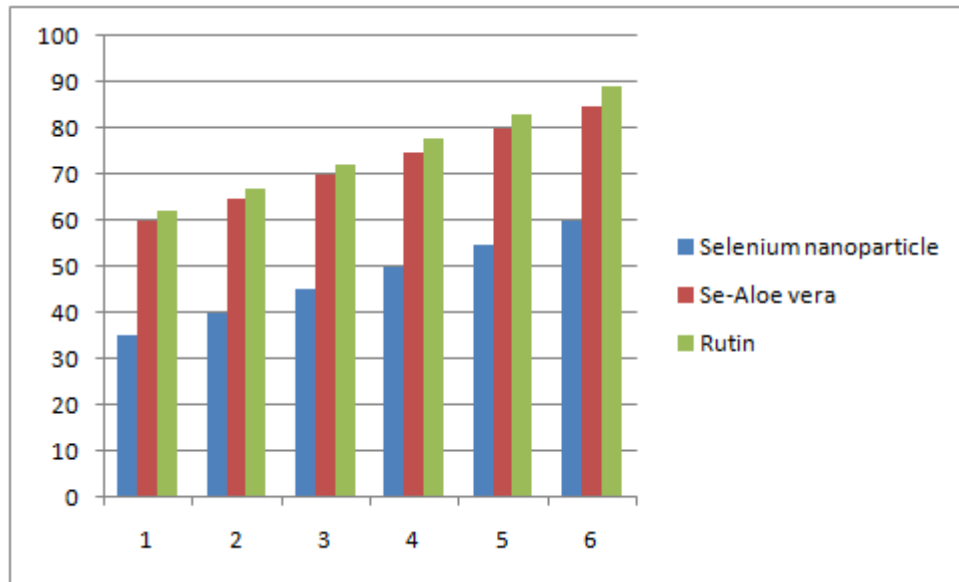


Figure 5: synthesize selenium nanoaprticles increased antioxidant activity as demonstrated by the DPPH assay

CONCLUSION

Using an aloe vera flower product to produce synthetic selenium nanoparticles was the objective of this research. The Aloe vera extract proteins bound and stabilized the selenium nanoparticles, thereby facilitating their formation. The UV-Vis spectrum of aloe vera extract at 350 nm shows that it contains round and hollow particles with sizes between 7 and 48 nm. These particles are more stable over longer periods of time. The TEM showed more proof that nanoparticles were formed. Even though it is poisonous, selenite has antioxidant qualities and is now being used as a chemopreventive drug to help find cancer. Selenium that is in the form of elements or nanoparticles is not as dangerous. The results from the ABTS and DPPH tests show that the aloe vera extract that was used to make selenium nanoparticles worked better than just the extract itself.

DECLARATIONS:

Ethics approval and consent to participate:

Not applicable.

Consent for publication:

All the authors approved the manuscript for publication.

Availability of data and material:

All required data is available.

Competing interests:

All authors declare no competing interests.

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