International Journal of Pharmacy and Analytical Research (IJPAR)

IJPAR |Vol.14 | Issue 1 | Jan - Mar -2025

www.ijpar.com

ISSN: 2320-2831

DOI: https://doi.org/10.61096/ijpar.v14.iss1.2025.85-94

Research

Synthesis, Characterisation And Biological Evaluation Of Selenium Conjugated Naringenin Nanoparticles

A. Helen Sonia*, Pandi Sathya, V. Shankarananth

Department of Pharmaceutics, S.A Raja Pharmacy College, Vadakkangulam-627116, Tirunelveli District, Tamil Nadu, India

*Author for Correspondence: A. Helen Sonia
Email: helensoniarajeshanpu@gmail.com

Published on: 17 Feb 2025

Published by:
DrSriram Publications

Abstract

In this study, Selenium Nanoparticles (Se-N-NPs) were synthesized by chemical reduction method using sodium salt of selenium and ascorbic acid, a reducing agent and a bioactive molecule naringenin, a bioflavonoid from citrus fruits. The sodium selenite and ascorbic were mixed together followed by conjugation with naringenin. Se-N-NPs were characterized by UV, FTIR, SEM, TEM, EDAX, X-RD and Zeta Sizer analysis. The UV spectrum indicated the highest peak of absorbance at 293 nm in acetone whereas FTIR analysis of Se-N-NPs indicated absorbance bands at 3336 cm^{-1} for OH group, 2926cm^{-1} for CH_3 and 1631 cm^{-1} C=O group. Drug release studies proved that the NPs release the drug in sustained manned with time. The antioxidant activity of NPs was carried by DPPH method using ascorbic acid as std.. The anticancer activity was studied as MCF-7 cancer cell line by MTT assay method. The obtained result showed that NPs have IC_{50} 260µg which proved anticancer effect of NPs. Apoptosis studies were done with dual dye AO/EB stain. This result proved that the Se-N-NPs inhibited cancer cell growth by inducing apoptosis. Comprehensively it can be concluded that Se-N-NPs can be attempted to develop a nano formulation with anti-cancer potential. However, extensive research is needed to reach a concrete conclusion.

Keywords: naringenin, nanoparticles, anti-cancer, apoptosis

INTRODUCTION

One of the most significant naturally occurring flavonoids is naringenin, which is primarily present in several edible fruits including tomatoes and citrus species[1,2,3] and figs from the Smyrna type Ficuscarica[4]. Naringenin ($C_{15}H_{12}O_5$), also known by its chemical name 2,3-dihydro-5,7-dihydroxy-2-(4-hydroxyphenyl)-4H-1-benzopyran-4-one, is a compound with a molecular weight of 272.26. This ubiquitous molecule dissolves in organic solvents such as alcohol but is insoluble in water. Naringenin's pharmacological effects, such as hepatoprotective, anti-atherogenic, anti-inflammatory, anti-mutagenic, anticancer, and antibacterial, have been supported by mounting evidence from both in *vitro* and in *vivo* animal research. It has even been suggested that naringenin may be used in cardiovascular, infectious, neoplastic, neurological, rheumatological, gastrointestinal,

and other disorders [5,6]. Unfortunately, because of its poor absorption at tumor locations and inferior pharmacokinetics, its widespread applicability in cancer is restricted. The current study's objective was to create nanoparticles laden with naringenin. Selenium nanoparticles as carriers for naringenin offer a versatile and powerful platform for drug delivery, enhancing bioavailability, targeting, and therapeutic efficacy across various delivery systems such as oral, liposomal, solid lipid nanoparticles, and polymeric nanocarriers. Selenium doping the process of incorporating selenium ions into these flavonoid structures further enhances their antioxidant and anticancer properties by leveraging selenium's role in reducing oxidative stress and promoting apoptosis in cancer cells. Selenium-doped quercetin has shown potential for higher bioactivity due to the combination of selenium's cancer-preventive properties and quercetin's already robust antioxidant capabilities. Both selenium-doped compounds exhibit enhanced therapeutic profiles, but the specific applications of selenium-doped quercetin are more aligned with cardiovascular health and cancer therapy, whereas selenium-doped naringenin may be more suitable for metabolic and neuroprotective interventions. Selenium nanoparticles (SeNPs) for naringenin drug delivery represent a promising strategy due to the potential for improved bioavailability, enhanced therapeutic effects, and targeted delivery. By incorporating selenium, an essential trace element with known anticancer and antioxidant properties, into nanoparticle formulations, these limitations can be overcome, opening new avenues for effective drug delivery.

MATERIALS AND METHODS

Naringenin, sodium selenite and ascorbic acid, MTT 3-(4,5-dimethythiazol-2-yl)-2,5-diphenyltetrazolium bromide, DAPI 40-6-diamidino-2-phenylindole, acridine orange/ethidium bromide (AO/EtBr), were purchased from Sigma Aldrich, India. All analytical grade chemicals were obtained from commercial suppliers.

Formulation of Se-N-NPs

For the preparation of Se-N-NPs, a solution of sodium selenite (Na_2SeO_3) was prepared by dissolving 500 mg of Na_2SeO_3 in 500 mL water. Thereafter, 400 mg Naringenin dissolved in 5 mL of acetone was added to it. The ascorbic acid solution was prepared by dissolving 12 g of ascorbic acid in 100 mL of water and was added drop wise to the above mixture and stirred for 24 h. The brick red colored Se-N-NPs were obtained. The resulting NPs were then centrifuged at 8000 rpm and washed with chilled ethanol to remove unbound Naringenin. Further, unreacted materials like ascorbic acid and Na_2SeO_3 were removed by multiple washings with water. Thereafter, the NPs were lyophilized and stored at -20^{o}C until further use[7-8].

Characterization of Nano-particles

Nanoparticles with their encapsulated particles were characterized using FTIR, TEM, DLS, DSC and X-ray as following:

Fourier transform infrared spectroscopy (FT-IR)

The reactive functional groups of the prepared nanoparticles were explored using Fourier transform infrared spectroscopy FTIR (Jasco, FT/IR 6100, Japan) within the spectral range of 4000–400 cm^{-1}.

Transmission electron microscopy (TEM)

The shape and size of the prepared nanoparticles was examined using TEM measurements [Jeol- TEM-2100 high resolution- transmission electron Microscopy, Japan]. Transmission electron microscopy was running at magnification power: 200 kv, maximum resolution: 1.4 A°.

Zeta-potential and particles size measurements

Zeta potential and particles size of the prepared nanoparticles were investigated using dynamic light scattering (DLS) technique via Zeta-sizer (Nano ZS, Malvern Instruments Ltd., Malvern, UK). The prepared nanoparticles were appropriately diluted before measurements. Then the samples were transferred to a 4 ml quartz cuvette and measured at room temperature (25 °C). Size distributions were investigated in terms of intensity versus particle size

X-ray diffraction (XRD)

XRD is a technique usually used to investigate the crystallinity of the prepared nanoparticles and identify the physical state of the materials; hence confirm the formation and presence of certain nanoparticle. XRD measurements were performed with X-ray difractometer (Bruker D8 Advance, USA).

EDAX

The elemental composition of the SeNPs was determined by energy dispersive X-ray spectroscopy (EDS) using a microprobe for surface microanalysis (Quantax EDS, Bruker, Billerica, Germany).

Cell culture conditions

MCF-7 cancer cell lines were used to evaluate the anticancer activity of Ch/Se nanocomposite. human breast adenocarcinoma (MCF-7) cell line was purchased from the American Type Culture Collection (Rockville, MD, USA) and maintained in Dulbecco's Modified Eagle's Medium (DMEM) supplemented with 10% heat-inactivated fetal bovine serum (FBS), 100 U mL^{-1} penicillin, and 100 U mL^{-1} streptomycin. The cells were grown at 37 °C in a humidified atmosphere of 5% CO_2.

Preparation of phosphate buffer pH 7.4

Phosphate buffer pH 7.4 was prepared as per the method described in I.P 1996 using disodium hydrogen phosphate and sodium hydroxide. The pH was adjusted to 7.4[9].

Preparation of calibration curve for Se-N-NPs

Accurately weighed quantity (100 mg) of naringenin was transferred into a 100 mL volumetric flask and dissolved in small amount of distilled water (D.W) and made up to the volume to make the standard stock solution of 1mg/mL. From the stock, 1mL was taken in 100 mL volumetric flask and made up the volume with the distilled water; from this solution 0.5mL to 3mL solution was transferred to 10 mL volumetric flask and made up to required volume with more distilled water and the resulting concentration ranges from 5 to 50 µg/mL. The absorbance of these solutions was determined at 293nm using UV spectrophotometer. The calibration curve was constructed between the absorbance and concentration.

***In-Vitro* Drug Release Study**

Three different techniques were used to determine the in vitro drug release of Se-N-NPs: dialysis bags, direct addition, and combination. The tests were carried out in sink circumstances with a 0.5% SLS solution in phosphate buffer (pH 7.4) as the drug release medium.

Dialysis bag method

The membrane used in the dialysis bag was regenerated cellulose. 50 mg of drug-loaded nanoparticles were weighed, dissolved in 3 mL of dissolving media, and then added to the donor compartment of the dialysis bag[10]. The dialysis bag ends were clamped and put in a beaker with 200mL of dissolving medium. At ambient temperature, the beaker was agitated with a magnetic bead at 100 rpm. In parallel, control research was conducted to evaluate free drug diffusion through the dialysis bag membrane. NPs solution in dissolving media was put into a dialysis bag instead of nanoparticles dispersion. At various times, a 5-mL aliquot of the dissolving fluid was taken out of the receptor compartment and replaced with an equivalent volume of brand-new media. At 293 nm, the samples were examined using a UV-visible spectrophotometer.

In vitro antioxidant activity of Se-N-NPs

The antioxidant activity of the Se-N-NPs was analyzed using the DPPH technique. The stock solution was prepared by dissolving 24mg of DPPH in 100mL of ethanol. Ethanol was used to separate the DPPH stock solution, yielding a beneficial mixture with an absorbance of approximately 0.973 at 517nm [11]. A test tube was used to combine 100µL of Se-N-NPs and 100µl of DPPH working solution. A standard solution is often prepared by dissolving three ml of DPPH solution in 100 ml of ethanol. The tubes were left in absolute darkness for 30 min. Hence, the absorbance was measured at a wavelength of 517 nm, The percentage of antioxidant activity was calculated using the following formula: [(A control-A test) ÷A control] × 100.

In vitro MTT assay on MCF-7 cell line

Cells were obtained from the NCCS Cell Repository in Pune, India. Upon procurement, the cells were maintained in liquid nitrogen and subsequently sub cultured in DMEM supplemented with 10% FBS. Following incubation, the cells containing the Se-N-NPs. were washed with new culture media and then treated with MTT (5 mg/mL in PBS) [12]. The cells treated with the MTT were incubated for an additional 4 h at 37°C to observe their responses. Cell viability was assessed by measuring absorbance at a wavelength of 540 nm using a 96 well plate reader. The ratio of stable cells to that of the control group was evaluated. The ideal dose and IC_{50} values were then calculated. The proliferation inhibition percentage was determined by multiplying the discrepancy between the optical density of the control and the optical density of the test by 100. The Se-N-NPs demonstrated a dose-response curve, showing a 50% reduction in cytotoxicity compared to the control cells [13].

Apoptosis in Se-N-NPs was measured using AO/EB staining.

To investigate the apoptosis induced by Se-N-NPs, MCF-7 cells were subjected to AO/EB staining. A solution of 100µL acridine orange(AO) and 100µL ethidium bromide(EB) was prepared by dissolving them in PBS, resulting in a 200µL dye[14]. The solution was incubated at room temperature for 5 min. The stained cells using a 40x fluorescent microscope.

Measurement of apoptotic induction using acridine orange/ethidium bromide (AO/EB) dual staining method

Principle

AO is permeable and it stains all the MCF-7 cancer cells viable/nonviable cells. It emits green fluorescence if intercalated into double-stranded nucleic acid (DNA) or red fluorescence if bound to single-stranded nucleic acid (RNA). EB is taken up only by nonviable cells that have lost membrane integrity and emits red fluorescence by intercalation into DNA. They distinguished four types of cells according to the fluorescence emission and the morphological aspect of chromatin condensation in the stained nuclei. Viable cells have uniform bright green nuclei with an organized structure. Early apoptotic cells (which still have intact membranes but have started to undergo DNA cleavage) have green nuclei, but perinuclear chromatin condensation is visible as bright green patches or fragments. Late apoptotic cells have orange to red nuclei with condensed or fragmented chromatin. Necrotic cells have uniformly orange to red nuclei with no condensed chromatin.

Reagents

1. Acridine Orange (AO)
2. Ethidium Bromide (EB)
3. PBS (1%)

Procedure

MCF-7 cancer cells were seeded at 5 x 10^4 cells/well in a 96-well plate and incubated for 24 hours. After treatment with NPs (200 and 300 µg/ml) for 24 h, the cells were detached, washed with cold PBS, and then stained with a mixture of AO (100µg ml^{-1})/ EB (100µg ml^{-1}) ratio (1:1) at room temperature for 5 min. The stained cells were observed by a fluorescence microscope at 20x magnifications. At the end of treatment, the cells were collected and washed three times with PBS. The plates were stained with acridine orange/ethidium bromide (AO/EB 1:1 ratio; 100 µg/ml) for 5 minutes and examined immediately under a fluorescent microscope with 20x magnification.

RESULTS AND DISCUSSION

Determination of absorption peak

The UV absorption profile for Se conjugated naringenin was analyzed and an absorption peak was observed at 293 nm.

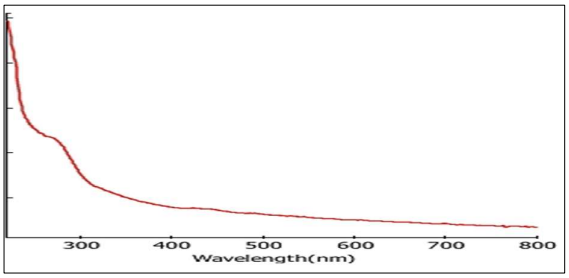

Fig 1: UV curve for Se-N-NPs

Formulation of Se-N-NPs

The brick red colored Se-N-NPs were obtained.

FT IR spectroscopical characterization

The macromolecular structure of naringenin was analyzed via FT-IR and its characteristic peaks were identified at 3336.31 cm^{-1} for the free OH that is part of the glycerol diglycidyl ether moiety. The aryl CH_3 had a stretching vibration at 2926 cm^{-1}. The keto group gave a distinct vibration at 1043.05 cm.

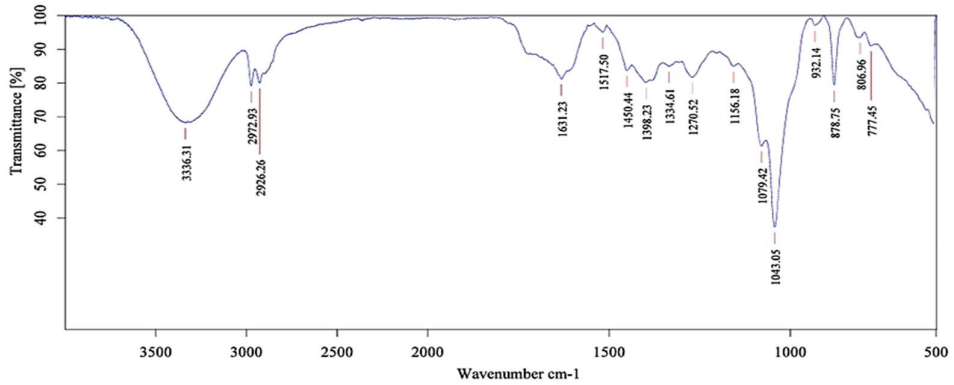

Fig 2: FT-IR of Se-N-NPs

SEM Analysis

Microstructural analysis of selenium doped naringenin was analyzed and the SEM images are depicted below [15]. From the images, it can be seen that the selenium conjugated particles are highly agglomerated and the average width of the agglomerated particles was measured to be 31.87 nm.

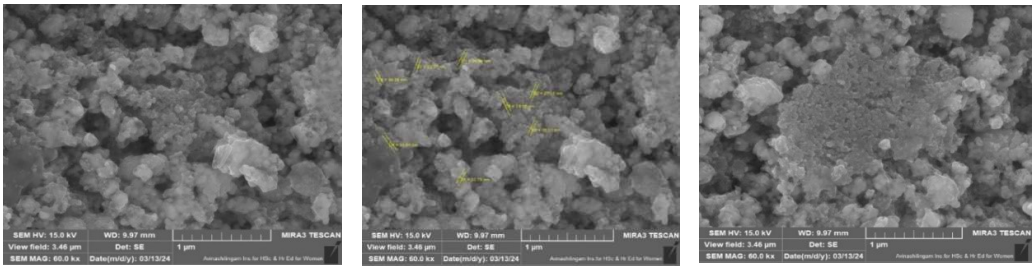

Fig 3: SEM images for Se-N-NPs

TEM Analysis

The transmission electron microscopy analysis was carried out to determine the morphology of the synthesized samples. From the TEM images we can witness the spherical structure of the selenium conjugated naringenin [16]. The measured size of the spherical particles varies roughly between 20 nm to 50 nm. The selected area electron diffraction (SAED) analysis of the particles observed through TEM instrument indicated the polycrystalline nature of the sample.

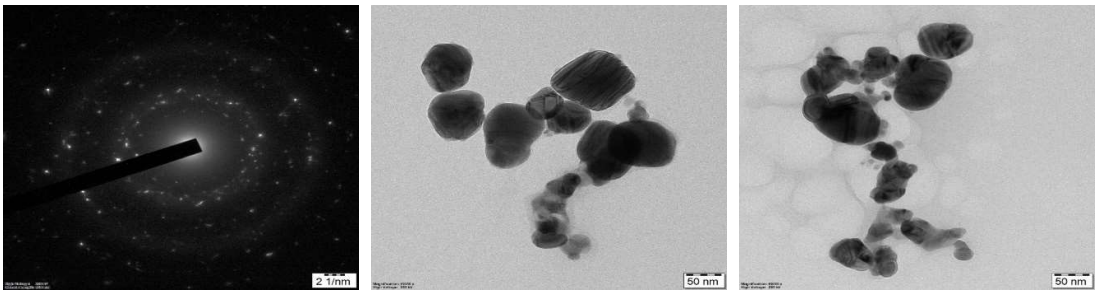

Fig 4: Transmission Electron Microscopy images for Se-N-NPs.

XRD Analysis

The selenium doped naringenin was subjected to XRD analysis and the diffraction profile indicated the crystalline peaks for naringenin at 23.66°, 26.43°, and 28.43°, respectively [17]. Small shifts in 2θ angles or peak intensities can suggest strong interactions between selenium and naringenin at molecular level. The XRD peaks in selenium-conjugated naringenin nanocomposites are expected to show a mix of selenium's crystalline peaks and possibly broader peaks for naringenin, reflecting the nanoparticle size, interaction, and degree of crystallinity in the composite.

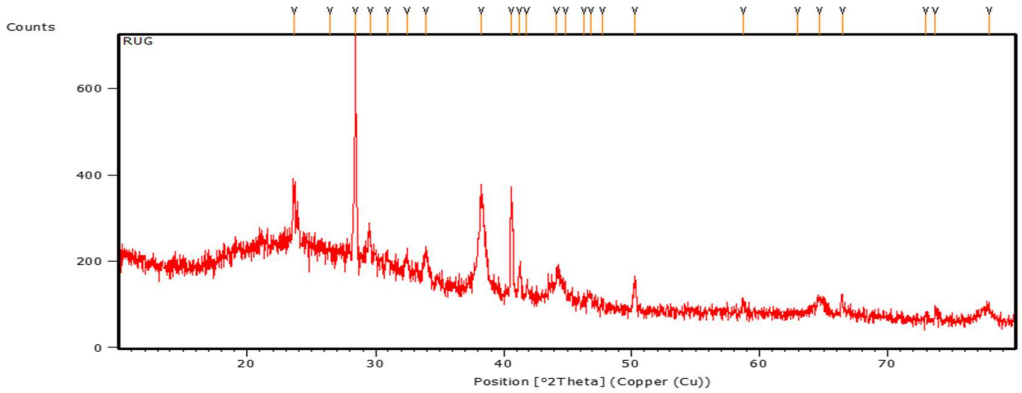

Fig 5: XRD of Se-N-NPs

EDAX Analysis

The elemental composition of the prepared samples was analyzed through EDAX analysis[18]. The presence of C, O, P, K, Ca, Cu, and Se was observed, wherein, the carbon and oxygen had a weight percentage of 60.10% and 11.22% indicating the dominance of naringenin. Additionally, the successful incorporation of Se was observed, with a high weight percentage of 12.32 %. The presence of P, K, Ca, and Cu may be due to minor contaminants from the precursors used.

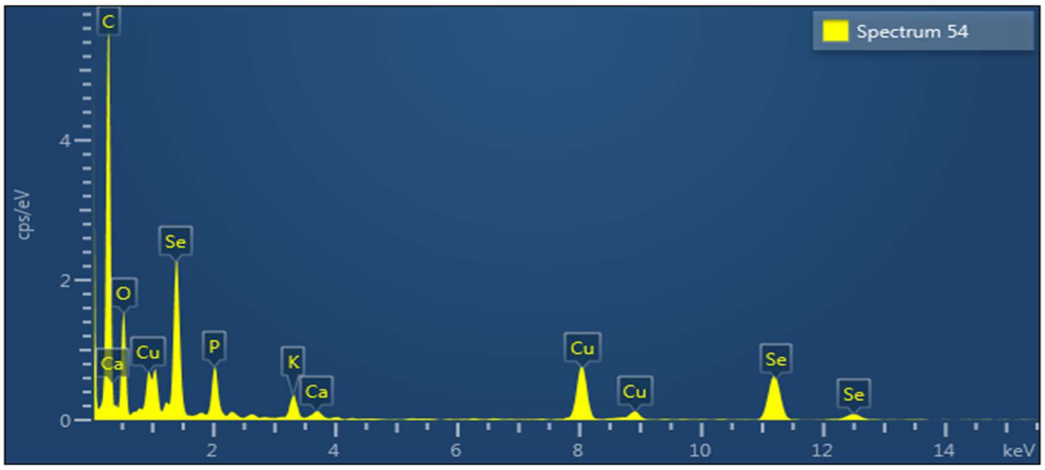

Fig 6: Elemental composition of Se-N-NPs.

Table 1: Elemental composition of Se-N-NPs

Element	Line type	k Factor	k Factor type	Absorption correction	Wt.%	Wt.% Sigma	Atomic %
C	K series	2.769		1.00	60.10	1.37	80.50
O	K series	2.020		1.00	11.22	0.76	11.28
P	K series	1.052		1.00	4.25	0.39	2.21
K	K series	1.009		1.00	2.05	0.25	0.84
Ca	K series	0.986		1.00	0.68	0.18	0.27
Cu	K series	1.247		1.00	9.38	0.55	2.38
Se	K series	1.683		1.00	12.32	0.72	2.51
Total:					100.00		100.00

ZETA potential Analysis

Zeta potential analysis was taken for the selenium conjugated naringenin nanoparticles and the size distribution analysis and zeta potential distribution was determined. From the figure, the formulation presented submicrometric particle diameters of 282.1 nm and a polydispersity index of 0.134. PDI > 0.7 is regarded as being too polydisperse to assess the size distribution, while PDI < 0.05 indicates a totally monodisperse and monomodal distribution. Between these two boundaries, the selenium conjugated naringenin nanoparticles has shown the results. Thus, the particles are homogeneous, have a narrow size distribution, and are quasi-monodisperse. The results showed adequate particle size, with a unimodal distribution and low polydispersity index. The method of interfacial deposition of pre-formed polymer was shown to be relatively straightforward and rapid, and offer reproducible particle size with a narrow distribution.

A zeta potential > ±30 mV dictates a stable dispersion while < ±30 mV indicates an unstable dispersion. The measured zeta potential found was -34.8 mV. Irrespective of the charge over the particle, the larger magnitude indicate the strong repulsive forces that prevent its aggregation [124] . This is indicative that the synthesized nanoparticles can be attained as a stable dispersion. This value of close to -35 mV can be considered high enough to maintain system stability and prevent agglomeration when dispersed in water

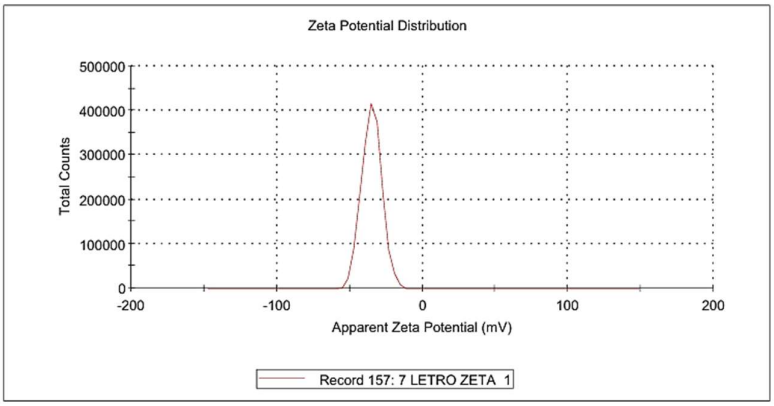

Fig 7: Zeta Potential Distribution for Se-N-NPs.

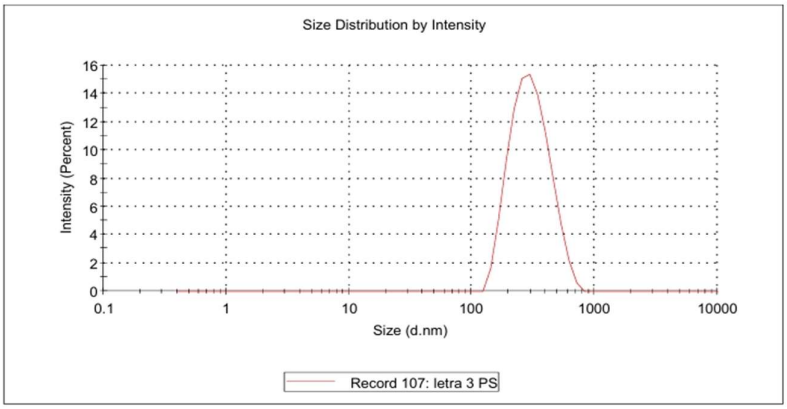

Fig 8: Size Distribution by intensity for Se-N-NPs

Table 2: Standard calibration curve for Se-N-NPs

Concentration (µg/ml)	% of inhibition (Ascorbic acid)	% of inhibition (A)
100	92.1	72.21
50	88.2	78.4
25	84.6	67.9
12.5	83.3	62.3
6.25	80.1	56.4
3.12	77	50.1

Table 3: Invitro drug release studies

Time (min)	%Cumulative drug release Dialysis bag method
0	0
15	13.5
30	24.23
60	66.56
120	78.34
240	94.23
480	97.13

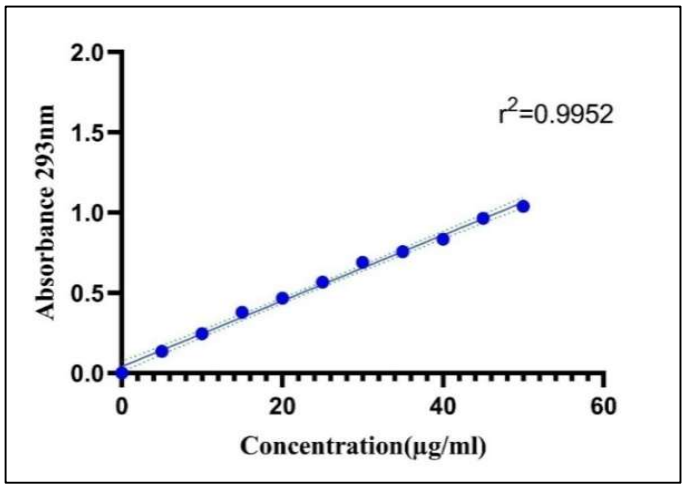

Fig 9: Standard calibration curve for Se-N-NPs.

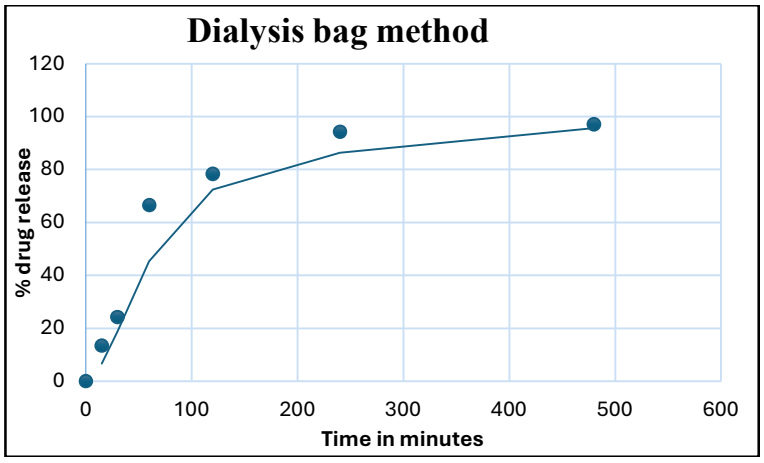

Fig 10: Invitro drug release studies
Drug is released in a sustained fashion with time

Table 4: Cell Viability of Se-N-NPs on MCF-7 cell line

Control	50 µg	100 µg	200 µg	300 µg	400 µg
100	99.21	82.62	64.58	35.3	22.25
100	95.36	80.21	62.29	35.99	22.11
100	86.31	83.43	56.53	36.96	25.15

Average	93.653	82.06	61.133	36.08	23.17
SD	6.32	3.23	3.13	2.43	2.00

From the above table, IC_{50} value was calculated as 254µgm

Measurement of apoptotic induction using acridine orange/ethidium bromide (AO/EB) dual staining method

MCF-7 cancer cells were treated with NPs (200 and 300 µg/ml) for 24 h, stained with dual dye AO/EB, and then analyzed by fluorescence microscopy (Zoe Fluorescent Cell Imager, Biorad). Live cells show green fluorescence with normal nuclear appearance. Early apoptotic cells with fragmented nuclear show yellow fluorescence with condensed chromatin. Late apoptotic cells show orange fluorescence with chromatin condensation or fragmentation (uniformly red/orange-stained cell nuclei).

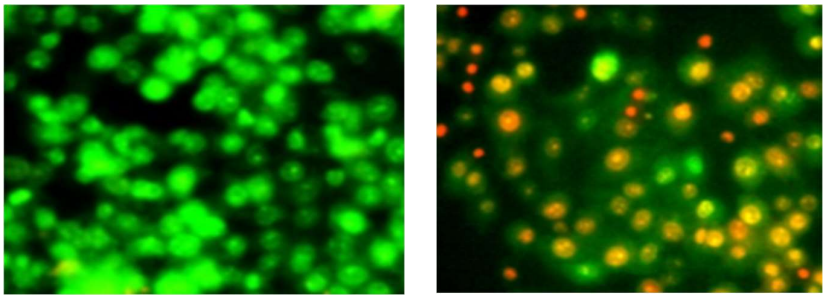

Fig 11: Apoptotic induction using acridine orange/ethidium bromide (AO/EB) dual staining method of Se-N-NPs.

CONCLUSION

This work focuses on the synthesis, characterization, and biological evaluation of selenium conjugated naringenin nanoparticles to explore their potential as anticancer and antioxidant agents. Selenium, a trace element with known anticancer and antioxidant properties, is conjugated with naringenin, a natural flavonoid renowned for its powerful antioxidant activity and anticancer potential, to create a nanostructured therapeutic system. The synthesis process involves optimizing various nanoparticle fabrication methods to ensure efficient conjugation of selenium and naringenin, aiming for nanoparticles with high stability, uniform size, and surface characteristics favorable for cellular interaction and absorption. The synthesized nanoparticles are thoroughly characterized using advanced techniques such as Zeta potential for size distribution, scanning electron microscopy (SEM) for morphology, X- ray diffraction (XRD) for structural integrity, and Fourier-transform infrared spectroscopy (FTIR) for confirming successful conjugation of selenium and naringenin. For anticancer activity assessment, the synthesized nanoparticles are tested on cancer cell line such as MCF- 7 (breast cancer), through standard in vitro cytotoxicity assays like MTT. In addition, induction of apoptosis, and potential modulation of molecular pathways involved in cell proliferation and survival were also studied and correlated. The project also explored the antioxidant capacity of selenium-conjugated naringenin nanoparticles via radical scavenging assays DPPH measuring their capacity to neutralize free radicals and protect against oxidative stress compared to free naringenin and selenium. By examining both anticancer and antioxidant properties, this research aims to understand how selenium and naringenin can jointly arrests cancer cell growth at a molecular level. The outcomes of this study could provide significant insights into the synthesis of nanoparticle-based drug delivery systems that exploit the natural biological activities of selenium and naringenin, suggesting a promising path for advancing nanotechnology applications in medicine.

REFERENCES

1. Trantas EA, Koffas MAG, Xu P, Ververidis F. When plants produce not enough or at all: metabolic engineering of flavonoids in microbial hosts. *Front Plant Sci.* 2015;6.
2. Nabavi SM, Nabavi SF, Habtemariam S, Moghaddam AH, Latifi AM. Flavonoid biosynthetic pathways in plants: Versatile targets for metabolic engineering. *Biotechnol Adv.* 2020; 38:107316.
3. Koopman F, Beekwilder J, Crimi B, Van Houwelingen A, Hall RD, Bosch D, et al. De novo production of the flavonoid naringenin in engineered *Saccharomyces cerevisiae*. *Microb Cell Fact.* 2012; 11:155.
4. Wu J, Zhou T, Du G, Zhou J, Chen J. Modular optimization of heterologous pathways for de novo synthesis of (2S)-naringenin in *Escherichia coli*. *PloS One.* 2014;9: e101492.

5. Lyons MM, Yu C, Toma RB, Cho SY, Reiboldt W, Lee J, et al. Resveratrol in raw and baked blueberries and bilberries. *J Agric Food Chem.* 2003;51(20):5867–70.
6. Yauk Y-K, Liang L, Hung M-S, Cheung W-L, Zhang T, Ngai SM, et al. Naringenin chalcone carbon double-bond reductases mediate dihydrochalcone biosynthesis in apple leaves. *Plant Physiol.* 2024; doi:10.1093/plphys/kiae515.
7. Kumari M, Purohit MP, Patnaik S, Shukla Y, Kumar P, Gupta KC. Curcumin loaded selenium nanoparticles synergize the anticancer potential of doxorubicin contained in self-assembled, cell receptor targeted nanoparticles. European Journal of Pharmaceutics and Biopharmaceutics. 2018 Sep 1;130:185-99.
8. A.Helen Sonia, Menaka Sundaram, V.Shankarananth, R. Kamalraj. Evaluation of antioxidant potential of silver nanoparticles derived from erythrina indica leaf extract., International Journal of Pharmacy and Industrial Research, Vol. 14 No. 2 (2024): 2024 Volume -14 - Issue 2, 191-97.
9. Pasaribu, S. P., Kaban, J., Ginting, M. & Sinaga, K. R. Synthesis and evaluation antibacterial activity of phosphate buffer solution (pH 7.4) - soluble acylated chitosan derivative. in 020025. 2018, doi:10.1063/1.5082430.
10. Yu M, Yuan W, Li D, Schwendeman A, Schwendeman SP. Predicting drug release kinetics from nanocarriers inside dialysis bags. J Controlled Release. 2019;315:23–30.
11. Dumore NS, Mukhopadhyay M. Antioxidant properties of aqueous selenium nanoparticles (ASeNPs) and its catalysts activity for 1, 1-diphenyl-2-picrylhydrazyl (DPPH) reduction. J Mol Struct. 2020;1205:127637
12. Maatouk, M. et al. Effect of heated naringenin on immunomodulatory properties and cellular antioxidant activity. Cell Stress Chaperones21, 1101–1109 (2016).
13. Jiang Y, Ma X, Feng J, Xiong S. Selenium in nitrogen-doped microporous carbon spheres for high-performance lithium–selenium batteries. Journal of Materials Chemistry A. 2015;3(8):4539-46.
14. Krishnan V, Loganathan C, Thayumanavan P. Green synthesized selenium nanoparticle as carrier and potent delivering agent of s-allyl glutathione: anticancer effect against hepatocarcinoma cell line (HepG2) through induction of cell cycle arrest and apoptosis. J Drug Deliv Sci Technol. 2019;53:101207.
15. Cai, J. Naringenin: A flavanone with anti-inflammatory and anti-infective properties. (2023).
16. Wu, C. et al. Naringenin-loaded solid lipid nanoparticles: preparation, controlled delivery, cellular uptake, and pulmonary pharmacokinetics. Drug Des. Devel. Ther. 911 (2016). doi:10.2147/DDDT.S97738.
17. Krishnakumar, N., Sulfikkarali, N., RajendraPrasad, N. & Karthikeyan, S. Enhanced anticancer activity of naringenin-loaded nanoparticles in human cervical (HeLa) cancer cells. Biomed. Prev. Nutr.1, 223–231 (2011).
18. Serhan Uru, S. U., Ahmet Akir, A. A. & Brahim Ethem Ulu Ay and Ali Aslan, B. E. U. A.A. A. A. The Metal Complexes of Natural Compounds, Usnic Acid and Naringenin Immobilized on Mesoporous Silica: Solid Phase Extraction and Oxidative Catalytic Properties. J. Chem. Soc. Pak.43, 682–682 (2021).