



# Protective Effect of *Jatropha Tanjorensis*-Based Green Selenium Nanoparticles Against Cadmium-Induced Hepatotoxicity in Male Rats

Research Article

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## ABSTRACT

Cadmium (Cd) contamination remains a significant environmental concern due to its adverse effects on the liver and genetic expression. This study explores the protective prospect of green-synthesised selenium nanoparticles using *jatropha tanjorensis* against cadmium-induced hepatotoxicity and TGF $\beta$ 1 mRNA gene expression in rats. Forty male albino rats were grouped into eight (n=5 per group). Group I served as control; rats received 1mL of 0.9% deionised water. Group II, the negative control, received Cadmium (0.5 mg/kg BW), Group III: rats received Gext (800 mg/kg BW). Group IV: rats received SeNPs (0.5 mg//kg BW). Group V: rats received Gext-SeNPs (0.5mg//kg BW). Group VI: rats received Cd (0.5 mg/kg BW) + Gext (800mg/kg BW). Group VII: rats received Cd (0.5 mg/kg BW) + SeNPs (0.5mg/kg BW). Group VIII: rats received Cd (0.5 mg/kg BW) + Gext-SeNPs (0.5mg/kg BW). All treatments were orally administered. The treatment lasted for 28 days. The result indicates that Cd induced hepatotoxicity in rats and elevated TGF $\beta$ 1 mRNA expression. Cd exposure increased liver Cd accumulation (0.164 ppm vs. 0.028 ppm in control), oxidative stress, and TGF $\beta$ 1 mRNA expression, while decreasing catalase and total antioxidant capacity (TAC). Gext-SeNPs significantly reduced Cd accumulation (0.026 ppm), restored antioxidant levels, and downregulated TGF $\beta$ 1 expression, demonstrating protective effects against Cd-induced hepatotoxicity.

**Keywords:** Protective, Nanoparticles, Cadmium, Hepatotoxicity, Synthesised.

## 1 Introduction

In recent years, there has been a noticeable surge in academic exploration regarding the application of nanotechnology in agricultural development, as observed in the work by (Duhan et al, 2017). In particular, inorganic nanoparticles (NPs) have emerged as a promising tool for enhancing animal feed. They hold the potential to augment the characteristics of traditional mineral elements, with a focus on their biological efficiency, bioavailability, and antimicrobial effects (Wang et al, 2017).

Nanoparticles, commonly referred to as NPs, are particles with a diameter of less than 100 nm, produced through either synthetic or biological means. Earlier investigations have demonstrated that NPs can maintain superior bioavailability while reducing toxicity in comparison to inorganic and organic formulations of trace minerals, as discussed by (Wang et al, 2017).



Among the minerals, selenium (Se) has garnered significant attention due to its delicate balance between essentiality and toxicity in living organisms, as elucidated by (Livingstone and Black, 2003). The biological efficacy of selenium hinges on its integration into the active centers of 25 selenoproteins (SeLPs), as detailed by (Papp et al, 2010).

In a different domain, the concept of plant chemotherapy, which applies the principles of pharmacology and medicinal chemistry to the plant kingdom, has gained prominence within the pharmaceutical industry, as recognized by (Ross and Brain, 2000). This has driven increased scientific scrutiny of medicinal herbs, focusing on their therapeutic potential and safety. As a result, physicians now have access to valuable data to guide patients in making informed decisions about their use, as highlighted by (Hara et al, 2001). While it is widely acknowledged that medicinal plants and their derivatives are generally safer than synthetic drugs due to their closer resemblance to the human somatic system, this is often an underexplored aspect, as emphasized by (Gamaniel, 2000).

One such medicinal plant, *Jatropha tanjorensis*, is noted for its low antioxidant properties and minimal haemagglutination titre value, indicating a low level of toxicity to red blood cells. This plant's leaf extract is also reputed for its hypoglycemic properties and is commonly employed as a remedy for diabetes, as reported by (Olayiwola et al, 2004). In Southern Nigeria, it is a popular natural remedy for malaria infection and hypertension, with the juice of the leaves being consumed. Additionally, the leaf extract exhibits antimicrobial properties, inhibiting the growth of *S. aureus* and *E. coli* (Oboh and Masodje, 2009).

Synthesis of nanoparticles from plant extract is termed green synthesis of nanoparticles. The evolution of green and sustainable methods for nanoparticles synthesis has been a recent research focus (Adil et al., 2015). Frequently, plant extracts are effective precursors for the production of metallic nanoparticles (Khan et al., 2018). FeNPs, are frequently synthesized using various plants, such as green tea (Shahwan et al., 2011), Sorghum sp (hybrid sorghum) (Njagi et al., 2011), Eucalyptus globulus (Madhavi et al., 2013), *Tridax procumbens* (Senthil and Ramesh, 2012), Grape seed proanthocyanidin (Narayanan et al., 2012). Green synthesis of selenium nanoparticles (SeNPs) using plant extracts and microorganisms has emerged as an eco-friendly and alternative to conventional methods (Alqaraleh et al., 2024). This approach offers precise control over nanoparticle size and shape, typically producing spherical SeNPs ranging from 40-150 nm (Alagesan & Venugopal, 2018). Plant extracts rich in reducing agents like flavonoids and alkaloids facilitate the reduction of selenium ions to SeNPs (Alqaraleh et al., 2024). Characterization techniques such as UV-Vis spectroscopy, SEM, TEM, and XRD are commonly used to analyze the synthesized SeNPs (Kirupagaran et al., 2016). Green-synthesized SeNPs have demonstrated potential in various applications, including anticancer treatments (Ramamurthy et al., 2013), antibacterial agents (Kirupagaran et al., 2016), and photocatalytic activities (Alagesan & Venugopal, 2018). This method offers advantages such as cost-effectiveness, environmental friendliness, and the production of biocompatible nanoparticles with narrow size distributions (Alqaraleh et al., 2024).

The heavy metal cadmium (Cd) has become a pervasive environmental pollutant, posing significant health risks to both humans and animals. Health hazards associated with heavy metals are primarily linked to Cd, lead (Pb), and nickel (Ni), as pointed out by (Amari et al, 2017). The accumulation of these metals is a multifaceted interplay of edaphic and environmental factors (Hasanuzzaman et al, 2018).

Cadmium is ubiquitously present in the environment, largely attributable to factors such as fossil fuel combustion, metal ore processing, and waste incineration, (Rahimzadeh et al, 2017). Remarkably, unlike other organic pollutants, Cd cannot be biologically degraded by microorganisms. Consequently, Cd accumulates in the food chain, resulting in a significant health risk with a half-life spanning several decades (Gong et al, 2019). This situation underscores the substantial threat Cd poses to global public health and agricultural food security, leading to its classification as a potentially hazardous chemical on a global scale. Consequently, the development of treatment interventions to mitigate Cd poisoning has garnered significant attention.

Selenium (Se) is an essential trace element that is crucial for human health. As a key component of various enzymes and proteins, selenium primarily exerts its biological functions in the form of selenoproteins within the body. Currently, over 30 types of selenoproteins have been identified, with more than 20 of them containing selenocysteine residues. Among these, glutathione peroxidases (GPXs), thioredoxin reductases (TrxRs), and iodothyronine deiodinases (DIOs)



have been widely studied. Selenium boasts numerous biological functions, including antioxidant properties, immune system enhancement, thyroid function regulation, anti-cancer effects, cardiovascular protection, reproductive capability improvement, and anti-inflammatory activity (Bai et al., 2024).

The emergence of Nano-biotechnology has primarily been driven by the utilization of selenium (Se), a vital trace mineral essential for various bodily functions. Se nanoparticles (SeNPs) have attracted considerable interest from researchers in the past decade due to their exceptional properties. The synthesis of *Jatropha tanjorensis*-mediated selenium nanoparticles for the removal of cadmium-induced hepatotoxicity is a novel research work hoped to offer solution to the removal of cadmium contamination.

### 1.1 Aim

To evaluate the protective effects of green-synthesised selenium nanoparticles using *Jatropha tanjorensis* leaf extract against cadmium-induced hepatotoxicity in male Wistar rats.

### 1.2 Objective

- To synthesize and characterize green selenium nanoparticles using *Jatropha tanjorensis* leave extract
- To determine the toxic effects of cadmium on Cadmium -induced albino rats.
- To estimate the protective potentials of *Jatropha tanjorensis* -based selenium nanoparticles on hepatotoxicity of Cadmium-induced albino rats
- To determine the expression of TGFB1 apoptosis related gene on Cadmium -induced albino rats
- To investigate the effect of *Jatropha tanjorensis* -based selenium nanoparticles on TGFB1 gene expression.

## 2 Literature Review

Nanotechnology, the study and manipulation of materials at the nanoscale (0.1-100 nm), has diverse applications across multiple fields (Rajak, 2018). In medicine, nanoparticles are used for drug delivery, cancer therapy, and early disease detection. Nanotechnology contributes to environmental protection, energy reduction, and pollution control (Afzal et al., 2022). It has significant potential in information technology, homeland security, and food safety (Afzal et al., 2022). The technology is advancing rapidly, with applications in dentistry, ophthalmology, surgery, and tissue engineering (Nikalje, 2015). Nanosensors are being developed for health monitoring and disease identification. Nanotechnology also shows promise in personalized medicine, tailoring treatments to individual genetic profiles (Alghamdi et al., 2022). As the field continues to grow, it is expected to revolutionize various industries and create new, improved products (Rajak, 2018).

In recent years, there has been a noticeable surge in academic exploration regarding the application of nanotechnology in agricultural development (Duhan et al, 2017). In particular, inorganic nanoparticles (NPs) have emerged as a promising tool for enhancing animal feed. They hold the potential to augment the characteristics of traditional mineral elements, with a focus on their biological efficiency, bioavailability, and antimicrobial effects, as highlighted by (Wang et al, 2017).

Synthesis of nanoparticles from plant extract is termed green synthesis of nanoparticles. The evolvement of green and sustainable methods for nanoparticles synthesis has been a recent research focus (Adil et al., 2015). Frequently, plant extracts are effective precursors for the production of metallic nanoparticles (Khan et al., 2018). FeNPs, are frequently synthesized using various plants, such as green tea (Shahwan et al., 2011), Sorghun sp (hybrid sorghum) (Njagi et al., 2011), Eucalyptus globulus (Madhavi et al., 2013), *Tridax procumbens* (Senthil and Ramesh, 2012), Grape seed proanthocyanidin (Narayanan et al., 2012).

*Jatropha tanjorensis*, a member of the Euphorbiaceae family, is well known perennial plant has been documented for its medicinal attributes. Traditionally, the consumption of *Jatropha tanjorensis* plant leaves in Nigeria has been widespread, predominantly utilized in soups and tonics, with claims of enhancing blood volume. Furthermore, these



leaves have a long-standing reputation in traditional medicine for treating conditions such as anaemia, diabetes, and cardiovascular diseases, as documented by (Orhue et al., 2008), (Omoriegie and Osagie, 2011), and (Oyewole et al., 2012). *Jatropha tanjorensis* ethanol leaf extract (JELE) and *Jatropha tanjorensis* acetone leaf extract (JALE) significantly inhibited the corrosion of aluminium in HCl solution (Okon and Faith 2017).

Selenium (Se) is an essential trace element that is crucial for human health. As a key component of various enzymes and proteins, selenium primarily exerts its biological functions in the form of selenoproteins within the body. Currently, over 30 types of selenoproteins have been identified, with more than 20 of them containing selenocysteine residues. Among these, glutathione peroxidases (GPXs), thioredoxin reductases (TrxRs), and iodothyronine deiodinases (DIOs) have been widely studied. Selenium boasts numerous biological functions, including antioxidant properties, immune system enhancement, thyroid function regulation, anti-cancer effects, cardiovascular protection, reproductive capability improvement, and anti-inflammatory activity (Bai et al., 2024).

Nowadays, selenium nanoparticles are widely being investigated for their anti-cancer activity against breast, lung, kidney, and osteosarcoma cancers based on *in vivo* and *in vitro* experiments (Ramamurthy et al, 2013). However, due to selenium toxicity and bioavailability elemental selenium and naturally sourced selenium (selenocysteine and selenomethionine) cannot be used in cancer therapies.

Nano-selenium has advantages over other natural sources of selenium because of its size, porosity, and bio-dispersion. Thus, *Polyporus umbellatus* polysaccharide (PUP) capped SeNPs have been utilized to detect the *in vitro* anti-proliferation effect with the MTT assay. Particularly, PUP-SeNPs inhibit the cell growth of four types of human cancer:

- a. MDA-MB-23 cells causing breast cancer in humans
- b. HepG2 cells responsible for human liver cancer
- c. HeLa cells known for human cervical cancer
- d. HT 29 for colon cancer

It is shown by researchers that, by using nano-selenium of the above nature, no toxicity to normal human cells such as liver cells (LO2), embryonic kidney cells (293T), and mouse embryonic fibroblast cells (NIH3T3) was observed (Shi et al., 2020).

Cadmium is a toxic element to which man can be exposed at work or in the environment. Cd's most salient toxicological property is its exceptionally long half-life in the human body. Once absorbed, Cd accumulates in the human body, particularly in the liver. The cellular actions of Cd are extensively documented, but the molecular mechanisms underlying these actions are still not resolved. The liver manages the cadmium to eliminate it by a diverse mechanism of action. Still, many cellular and physiological responses are executed in the task, leading to worse liver damage, ranging from steatosis, steatohepatitis, and eventually hepatocellular carcinoma. The progression of cadmium-induced liver damage is complex, and it is well-known the cellular response that depends on the time in which the metal is present, ranging from oxidative stress, apoptosis, adipogenesis, and failures in autophagy (Souza-Arroyo et al., 2022).

Many studies demonstrate the effectiveness of natural products in counteracting the toxic effects of cadmium however, to our knowledge, no previous studies of *Jatropha tanjorensis* –mediated selenium nanoparticle GSeNPs or *Jatropha tanjorensis* leaf extract (GEXT) on cadmium-induced hepatotoxicity is available. The present study aimed initially to synthesize and characterize green SeNPs with an ethanolic *J. tanjorensis* –mediated selenium nanoparticle (GSeNPs). Secondly, the study compared, for the first time, the *in vivo* protective outcomes of green- synthesized *J. tanjorensis* –mediated selenium nanoparticle (GSeNPs) MOLE-SeNPs conjugate or GEXT as treatments for hepatotoxicity caused by cadmium in male rats



### 3 Materials and Methods

#### 3.1 Plant extract preparation, *Jathropa tanjorinesis* leaves extract (Gext)

*Jathropa tanjorinesis* leaves (Gext) were obtained from Ado-Ekiti and authenticated by a Botanist. Dried *J. tanjorinesis* leaves were crushed into a powder using a high-speed milling device. Three hundred grams of powder was extracted in absolute ethanol for 48 hours then filtered twice through filter paper with 2  $\mu\text{m}$  pore size. The resultant extract was concentrated and evaporated to dryness using a rotary evaporator at 40–45 °C. The residual yield of Gext extract was 17.8 g/300 g of dried powder. The obtained extract was kept at 4°C until use.

#### 3.2 Synthesis of Selenium Nanoparticles (SeNPs)

A bottom-up approach was adopted in the synthesis of selenium nano-particles and green selenium nanoparticles. The following experimental procedure was adopted for the synthesis of selenium nano-particles using sodium selenite, sodium hydroxide, and ascorbic acid. 0.5 M sodium hydroxide was prepared by dissolving 20 g of sodium hydroxide pellets in 1 L of deionized water.

5 g of sodium selenite was added to 100 mL of deionized water in a 250 mL flask and stirred until the sodium selenite was completely dissolved. 10 mL of 0.5 M sodium hydroxide solution was added to the flask containing the sodium selenite solution, and stirred continuously with a magnetic stir bar. The pH of the solution was monitored with a pH meter, and adjusted as necessary to maintain a pH of around 10-11.

0.02 M ascorbic acid solution was added to the flask containing the sodium selenite solution, and stirred continuously with a magnetic stir bar. The reaction mixture turned yellow to orange as the reduction of selenite ions to selenium nanoparticles occurs. The solution was stirred cautiously for 2 hours at 90°C temperature. 50 mL of ethanol was added to the flask to stop the reaction, and then stirred for an additional 30 minutes to ensure complete mixture. The reaction mixture was centrifuged at 5000 rpm for 15 minutes to separate the selenium nanoparticles from the solution. The supernatant was discarded and the selenium nanoparticles were washed several times with deionized water to remove any remaining impurities. Few drops of hydrochloric acid were added to the selenium nanoparticle suspension to stabilize the nanoparticles and the pH adjusted to 3. The selenium nanoparticle suspension was kept in a dark, cool place until ready for use.

#### 3.3 Synthesis of *Jatropha Tanjorensis*-Mediated Leaf Extract Selenium Nanoparticles (Gext -SeNPs)

The synthesis of green selenium nanoparticles was done using sodium selenite, extract *Jatropha tanjorensis*, and ascorbic acid. 3.0 g of sodium selenite was dissolved in 100 mL of deionized water in a beaker. 0.02M of ascorbic acid was further dissolved in 50 ml of deionized water in a separate beaker. *Jatropha tanjorensis* was extracted by adding 10 g of dried and powdered leaves to 100 mL of distilled water in a flask. The mixture was stirred for 30 minutes, at the temperature of 70°C and then the extract was filtered using Whatman filter paper. 10 mL of the *Jatropha tanjorensis* extract was added to 100mL of deionized water and the solution was added to sodium selenite solution and stirred for 10 minutes. 2 mL of the ascorbic acid solution was also added to the mixture and stirred for an additional 10 minutes. 1 M NaOH solution was added to the solution until the pH reaches 10. The colour of the solution changed from clear to yellow-green and to brick red. The mixtures were stirred for 2 hours. UV-Vis's spectrophotometer was used to monitor the absorption spectrum of the mixture at different time intervals until the maximum absorption peak was reached, which was around 320 nm. The mixture was centrifuged at 10,000 rpm for 20 minutes to separate the green selenium nanoparticles. The supernatant was discarded and the nanoparticles were washed several times with deionized water to remove any impurities. The nanoparticles were dried in an oven at 90°C for 30 minutes. The resulting green selenium nanoparticles were characterized using various techniques such as transmission electron microscopy (TEM), X-ray diffraction (XRD), and Fourier-transform infrared spectroscopy (FTIR).



### 3.4 Animals and study protocol

Forty (40) male rats of comparable weight and age, twenty (20 +4 days) weeks old male albino rats, with average BWs of 120–185 g were housed in Animal house, School of Science and Computer studies, Department of Science Technology, Federal polytechnic Ado-Ekiti, Ekiti State Nigeria. All animals were cared for humanely according to the conditions stated in the Guide for the Care and Use of Laboratory Animals of the National Academy of Science (NAS), published by the National Institute of Health. The study also followed National Institutes of Health's recommendations for the care and use of laboratory animals, and adequate procedures were made to ensure that the animals were not subjected to excessive pain and discomfort during the study. Rats were randomly allocated into eight (8) equal groups of (n =5).

### 3.5 Experimental procedure

Animals were randomly allocated into eight groups of five rats each and administered orally in the following ways: Group I: control rats received 1mL of 0.9% deionized water. Group II: negative control received Cadmium (0.5 mg/kg BW), Group III: rats received Gext (800 mg/kg BW). Group IV: rats received SeNPs (0.5 mg/kg BW). Group V: rats received GSeNPs (0.5mg/kg BW). Group VI: rats received Cd (0.5 mg/kg BW) + Gext (800mg/kg BW). Group VII: rats received Cd (0.5 mg/kg BW) + SeNPs (0.5mg/kg BW). Group VIII: rats received Cd (0.5 mg/kg BW) + GSeNPs (0.5mg/kg BW). Gavage was done day after day for 28 days, using a feeding needle. All treatments were orally administered. All animals were carefully observed throughout the experiment for any signs of intoxication or mortality.

#### 3.5.1 Blood and tissue sample collection

Twenty-four hours after the last treatment, the overnight-fasted rats were weighed and anaesthetised with ketamin at the end of the dosing period. Two mL blood samples were collected from the rats via cardiac puncture. Blood was collected using BD Vacutainer PST II Tubes, left for coagulation, then centrifuged at 322 x g for 20 min for serum separation. Sera were preserved at -20 °C until biochemical analysis. Kidney, liver and testis tissues were collected and divided into four portions. Thirty mg was washed in cold saline and mixed with RNA Shield for storage before RNA extraction.

#### 3.5.2 RT-PCR analysis of apoptosis and stress related gene expression TGFβ1

Table 1: Primer sequence

S/N	Gene	Forward (5–3')	Reverse (5–3')
2	TGFβ1	CCTGGAAAGGGCTCAACAC	CAGTTCTTCTCTGTGGAGCTGA

The second portion was homogenized by Ultra Turrax homogenizer in a cold solution of 0.015 M KHPO<sub>4</sub> buffer and 0.15 M KH<sub>2</sub>O<sub>4</sub> (1:6 w/v; pH 7.8) for homogenate preparation for biochemical measurement.

#### 3.5.3 Analyses of antioxidant biomarkers

Superoxide dismutase (SOD), glutathione peroxidase (GPx), malondialdehyde (MDA), catalase (CAT), and glutathione concentrations (GSH) were assessed using serum samples and the analysis kits as instructed by the manufacturer (Nanjing Jiancheng Bioengineering Institute, Nanjing, China). Mobi Microplate spectrophotometer (Sunnyvale, CA) was used for all analyses.

#### 3.5.4 Serum and tissue cadmium analysis

One mL/mg of serum/ liver tissues were digested with 10 mL analytical grade H<sub>2</sub>O<sub>2</sub> (hydrogen peroxide) and HNO<sub>3</sub> (nitric acid) (1:1) in a Teflon beaker. The solution was then made up to 25 mL with distilled water. Cd concentration

in samples were analysed with thermal decomposition coupled atomic absorption spectrophotometer (AAS) Santos et., al.2017; Santos et., al.2020).

### 3.6 Statistical analysis of data

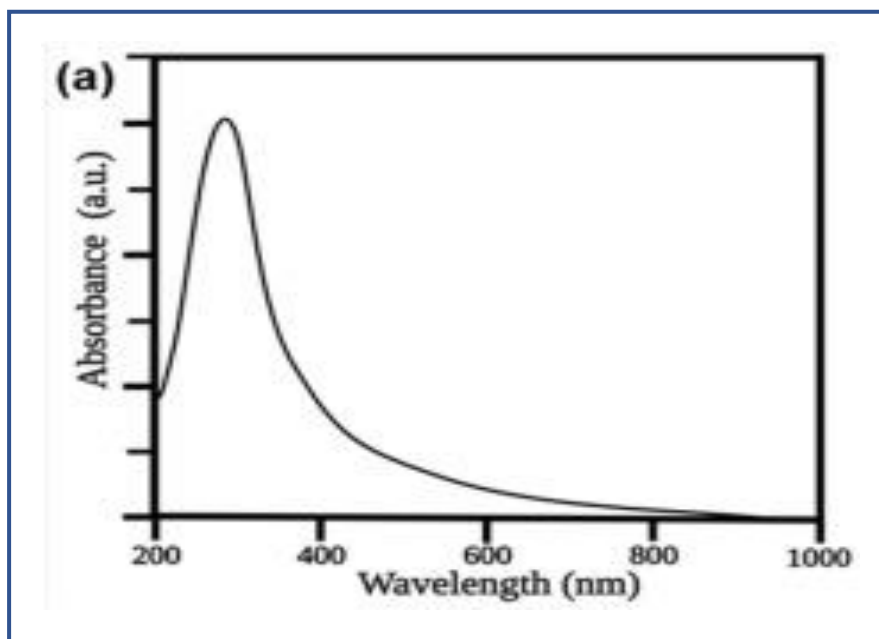
All obtained raw data were analysed using a one-way analysis of variance (ANOVA) by using the SPSS 25.0 computer program followed by post hoc Tukey HSD multiple comparisons to determine the statistically significant variations between the various parameters in the experimental replicates. A p-value of  $< 0.05$  was considered statistically significant. Data were shown as means  $\pm$  SD. All graphs were generated using GraphPad Prism 8 (GraphPad Software Inc., San Diego, 189 CA, USA).

## 4 Materials and Methods

All experimental groups showed no altered clinical observations or behavioural alterations except for Cd-exposed rats displayed a decreased activity, food consumption, and body weights with frequently increased urination when compared with the other experimental groups. Meanwhile, rats from the Cd- Gext, Cd-SeNP and Cd- Gext -SeNP groups were clinically normal and did not exhibit any signs of toxicity.

### 4.1.1 UV-visible spectroscopic analysis of Se-NPs

SeNPs were synthesized by reduction of Se into Se-NPs after the addition of Gext filtrate. The stimulation of the surface plasmon resonance of Se-NPs gave the reaction solution its distinctive brick-red colour, which served as a useful spectroscopic hallmark of their production (Fig. 1). UV-Visible spectrophotometer, revealed that selenium was absorbed at 320 nm absorbance peak which was unique to Se-NPs.



**Figure 1:** The UV-visible absorption spectrum of green synthesized Se-NPs



**Figure 2:** FTIR analysis of Green Se-NPs

#### 4.1.2 Characterization Of Selenium Green Nano-Particles

The FTIR spectra of the green synthesized Se-NPs highlighted the functional groups present in Se-NP (Figure 3). The peaks found in between 3500 and 3000  $\text{cm}^{-1}$ , 1609.71  $\text{cm}^{-1}$ , 1312.71  $\text{cm}^{-1}$  and 1029.53  $\text{cm}^{-1}$  were assigned to stretching vibration of O–H bond of alcoholic group, stretching vibration C O bond of aldehydes, ketones, carboxylic acids, and esters, bending vibration of C–C and stretching vibration of C–N bond, respectively (Karade et al., 2019). The occurrence of these bonds can be attributed to the presence of the phenolic hydroxyl group, phenolic acids, terpenoids-phenols, and aliphatic amines. These functional groups are also responsible for the reduction of  $\text{Se}^{2+}$  ions and behave as the capping agent of the nanoparticles (Karade et al., 2019).

#### 4.1.3 XRD Results of SeNps

The XRD measurement is frequently found to be a valuable analytical tool for determining the crystalline nature of freshly generated compounds and their phases. The XRD pattern of the Se-NPs, shown in Figure 4, During this measurement, a sequence of diffraction peaks was detected that were consistent with the theory. The prominent peaks of the XRD pattern suggest that the green synthesized Se-NPs were well-crystallized.

#### 4.1.4 DLS Result of SeNPs

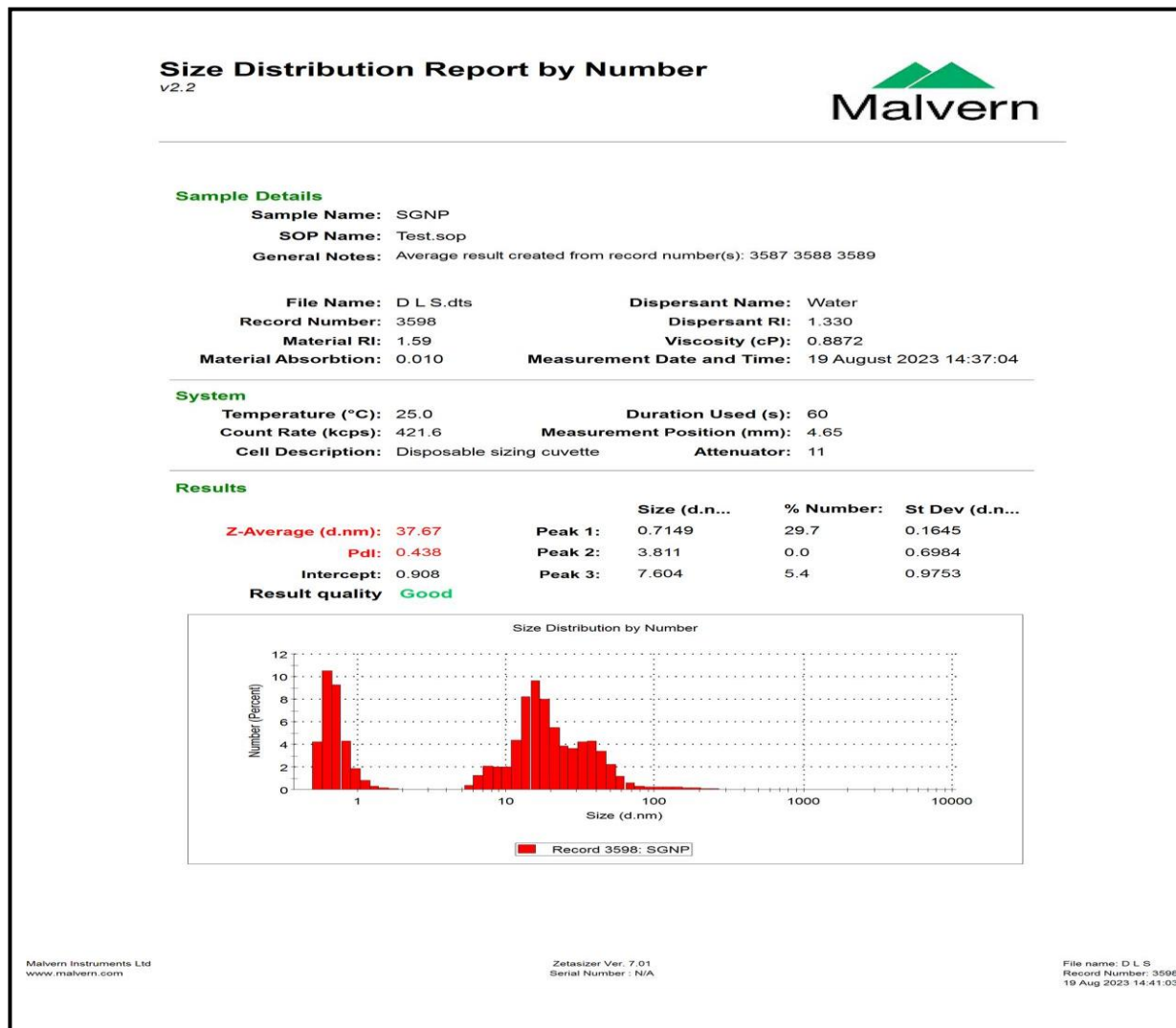


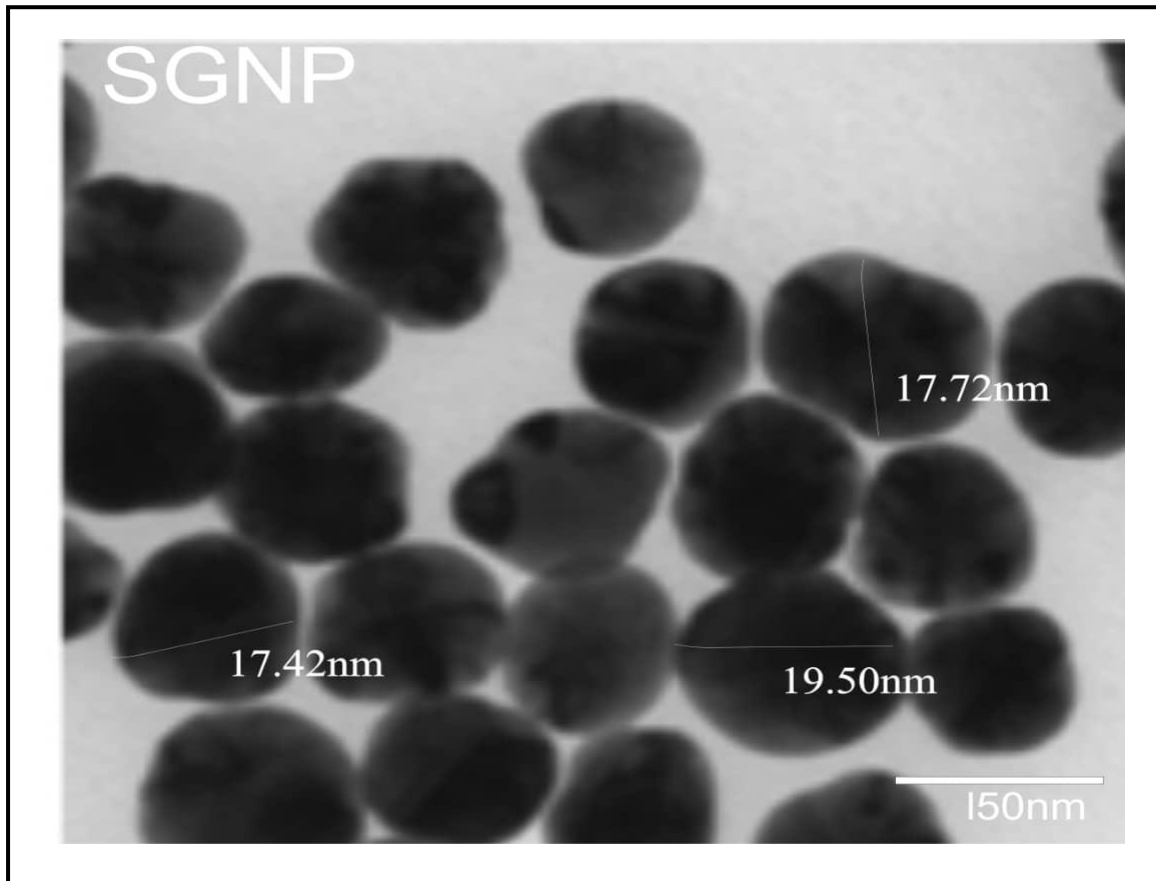
Figure 3: Particle size distribution

Dynamic Light Scattering (DLS) involves applying a laser beam to the sample and monitoring fluctuations in the scattering intensity which results from the Brownian motion of the particles. The magnitude of the scattered intensity is a function of several parameters including particle size; therefore, by applying a scattering autocorrelation function and several assumptions, the average hydrodynamic diameter of particles in the sample can be calculated.

From the information above, we observed that the average particle distribution is 37.67nm with a short period of 0.438per second while the particle size ranges from 0.7- 7.6nm, with particles of 0.7 making a greater percentage. Gext-SeNPs dispersant rate was 1.59 with absorption speed of 0.010.

#### 4.1.5 Transmission Electron Microscopy (TEM) Analysis

TEM the transmission electron microscopy is a very powerful tool for material science. TEM can be used to analyze the quality, shape, size and density of a quantum well. As shown below, the particle sizes and shapes are relatively excellent TEM analysis.



**Figure 4:** Particle sizes, shapes and distributions of the green nanoparticles

#### 4.4 PCR Analysis

##### 4.4.1 Relative mRNA levels of apoptosis-related genes (FGFB-1) in renal tissues

The probability of Cd-inducing liver cell apoptosis was investigated via measuring the transcriptional levels of apoptosis-related genes TGF $\beta$ 1 data are shown in (Figure 6). Cd- induced increase expression of apoptosis related gene. However, administration of Gext, SeNPs and Gext-SeNP in rats ameliorates the toxic effect of cadmium, by decreasing the gene expression as shown below. The amelioration was most effective in those that received Gext -SeNP.

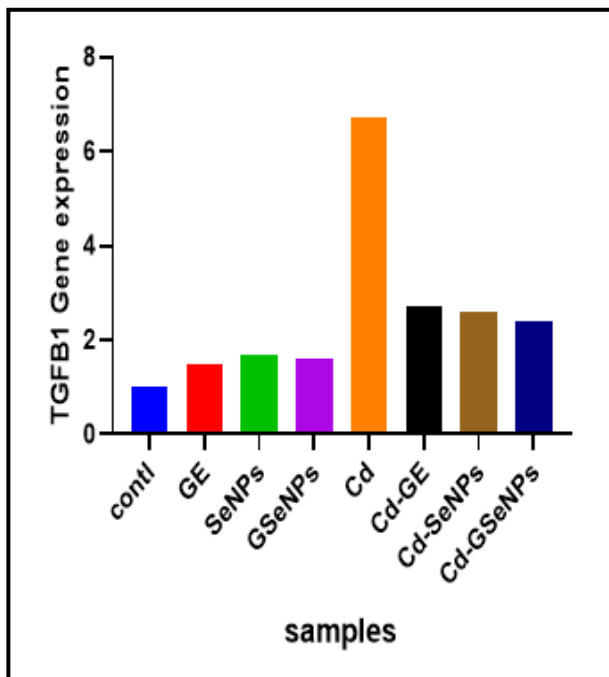


Figure 5: TGFB1 Expression

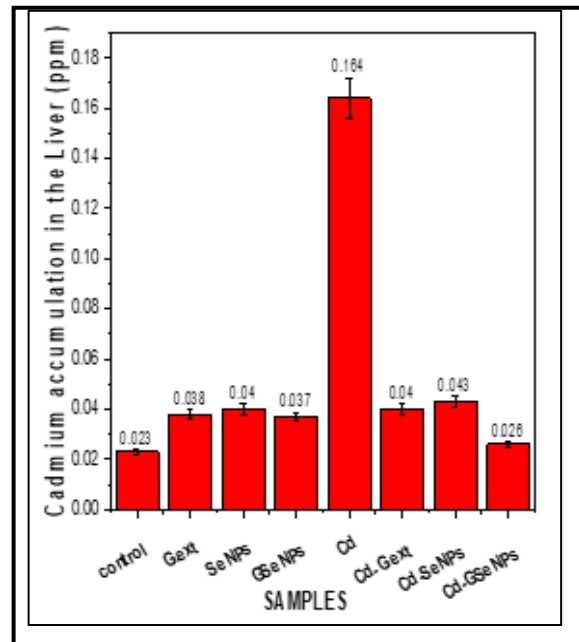


Figure 6: Cadmium accumulation in the liver

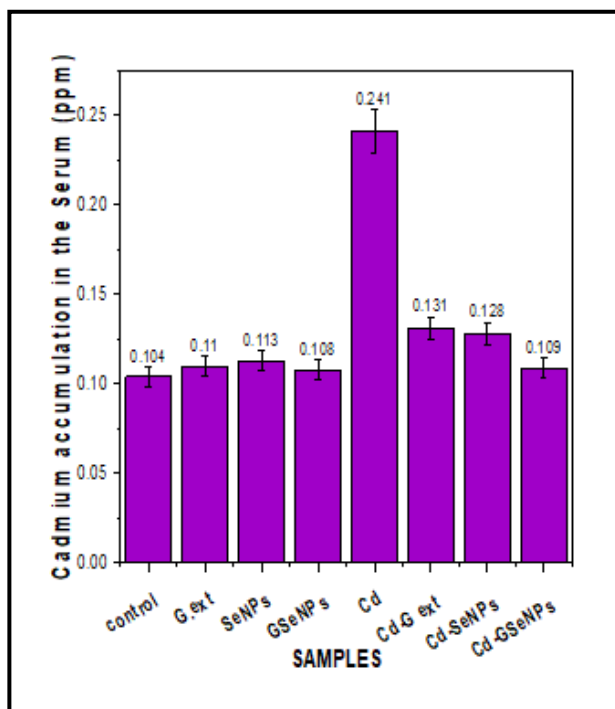


Figure 7: Cadmium accumulation in the serum



Figure 5 shows that Cd exposure significantly increased TGFβ1 mRNA expression in liver tissues (6.4-fold vs. control). Treatment with Gext, SeNPs, and Gext-SeNPs reduced TGFβ1 expression, with Gext-SeNPs showing the most significant reduction (2.1-fold, Figure 5).

Figure 6 shows that Cadmium accumulation in the liver. The accumulation of Cadmium in the liver was shown in figure 6 above. Cadmium (Cd) is a heavy metal that can accumulate in the body. One of the primary targets of Cd toxicity is the liver, where it can induce a range of biochemical damages including the alterations of various enzymes and proteins activities., Impaired bile production, Hepatic enzyme leakage, Reduced protein synthesis, Oxidative stress and so on. However, Gext, Cd-SeNPs, Cd-Gext-SeNPs have shown that they have the potentials of ameliorating the toxic effect of cadmium as demonstrated in Figure 6 above.

Cadmium accumulation is also noticed in the serum as expressed in Figure 7 above. However, just as in the case of liver the biosynthesized selenium nanoparticles (Cd-Gext-SeNPs) was able to reduce it accumulation in the serum. (Figure 7).

#### 4.5 PCR Analysis

Gext, Cd-SeNPs and Cd-Gext-SeNPs conjugate impact on Cd-induced changes in oxidative stress biomarkers in male rat liver was shown in Table 2 below. There was significant increase in MDA in rats that were exposed to Cd but this was reversed with Gext, Cd-SeNPs and Cd-Gext-SeNPs. This reverse was prominent in those that received Cd-Gext-SeNPs. as they are similar to the control group. Similar trend was observed in LDH. There was significant decrease in SOD activities in rats that were exposed to Cd. Nevertheless, its activity was ameliorated with Cd-Gext-SeNPs. Similar trend was observed in other oxidative stress parameters such as Catalase, GSH, GPx and TAC.

**Table 2:** Oxidative stress in liver

	Control	Gext	SeNP	Gext-SeNP	Cd	Cd-Gext	Cd-SeNP	Cd-Gext-SeNP
MDA (uM)	1.761±	3.993±	3.675±	3.831±	7.267±	3.104±	2.906±	2.514±
	0.219	0.320	0.320	0.253	0.411	0.213	0.234	0.241
SOD (U/mL)	2.822±	1.876±	1.617±	1.711±	0.563±	1.461±	1.517±	2.351±
	0.210	0.125	0.130	0.083	0.046	0.231	0.161	0.213
CATALASE (umol/ml/mins)	8.214±	4.505±	4.822±	4.493±	2.905±	4.987±	5.433±	6.418±
	0.360	0.351	0.316	0.300	0.239	0.397	0.341	0.395
GSH (mM)	0.344±	0.175±	0.195±	0.185±	0.032±	0.181±	0.202±	0.271±
	0.028	0.018	0.015	0.017	0.021	0.052	0.016	0.012
GPX (U/L)	89.570±	67.643±	69.624±	68.465±	43.103	68.231±	67.154±	79.817±
	9.064	4.147	5.074	4.416	2.862	4.284	5.721	4.605



TAC (mM/g tissue)	1.165±	0.705±	0.710±	0.739±	0.326±	0.743±	0.694±	0.971±
	0.022	0.042	0.020	0.040	0.024	0.021	0.043	0.046
LDH (U/L)	46.446±	73.643±	70.091±	71.579±	187.324±	73.624±	76.151±	57.488±
	6.745	4.128	0.586	2.749	9.864	0.632	2.766	6.462

Where GExt=green extract, SeNPs= selenium nanoparticles, GextSeNPs = green selenium nanoparticles, Cd = cadmium

#### 4.6 Discussion

This study demonstrates that Cd induces hepatotoxicity, oxidative stress and increase expression of TGFβ1 gene in albino rats administered with Cd. The work further showed that green synthesized selenium nanoparticles can ameliorate the TGFβ1 gene expression

The findings of this study are consistent with other researchers' findings especially in antioxidant and hepatoprotective properties of jatropha tanjorinesis and SeNP against various toxicants. Report of Oyagbemi et al., (2010) showed that Gext prevented carbon tetrachloride-induced oxidative stress and liver damage in rats by enhancing antioxidant enzyme activities and reducing lipid peroxidation. Similarly, El-Batal et al., (2016) demonstrated that SeNP ameliorated acetaminophen-induced oxidative stress and liver injury in mice by increasing glutathione level and decreasing MDA level.

Dauplais et al., (2013), reports that the underlying mechanism of the protective Se effect on Cadmium tissue distribution is attributed to the formation of a colloidal Cadmium–Selenium complex. The Cd–Se compound was first revealed in rat plasma in early in vitro and in vivo studies conducted by Gasiewicz and Smith. The researchers reported that erythrocytes reduced selenite (SeO<sub>3</sub><sup>2-</sup>) to selenide (H<sub>2</sub>Se), which was released to plasma forming a biologically inert, stable Cd–Se complex with Cd<sup>2+</sup>. The synergistic effect of Gext-SeNP combination may be due to their complementary mechanisms of action, such as chelating Cd, scavenging ROS, modulating antioxidant enzymes, restoring glutathione level, inhibiting inflammation and apoptosis, and enhancing liver regeneration. This study demonstrates that Cd induces hepatotoxicity, oxidative stress, and TGFβ1 overexpression in albino rats. Gext-SeNPs significantly ameliorated these effects, likely due to the formation of a Cd-Se complex (Dauplais et al., 2013) and enhanced antioxidant activity from *Jatropha tanjorensis* phytochemicals.

#### 5 Conclusion and Recommendations

This work reveals that green synthesized selenium nanoparticles (Gext-SeNP) have protective effects on oxidative stress parameters in liver tissue of rats exposed to Cd. Gext-SeNPs are promising candidates for mitigating Cd-induced hepatotoxicity and oxidative stress. Further studies should explore their long-term efficacy, mechanisms, and potential against other heavy metal toxicities.

#### 6 Conflict Of Interest Declaration

I declare that I or any of the co- authors did not have conflict of interest, it is our collective decision to publish our work in Techsphere journal. Nwobegu James Sylvester, for and on behalf of the Authors.



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## References

- Alagesan, V. and Sujatha Venugopal (2018). Green Synthesis of Selenium Nanoparticle Using Leaves Extract of *Withania somnifera* and Its Biological Applications and Photocatalytic Activities *BioNanoScience*. <https://doi.org/10.1007/s12668-018-0566-8>.
- Adil, S. F., Assal, M. E., Khan, M., Al-Warthan, A., Siddiqui, M. R., and Liz-Marzán, L. M. (2015). Biogenic synthesis of metallic nanoparticles and prospects toward green chemistry. *Dalton transactions (Cambridge, England : 2003)*, 44(21), 9709–9717. <https://doi.org/10.1039/c4dt03222e>
- Afzal, O., Altamimi, A. S. A., Nadeem, M. S., Alzarea, S. I., Almalki, W. H., Tariq, A., Mubeen, B., Murtaza, B. N., Iftikhar, S., Riaz, N., & Kazmi, I. (2022). Nanoparticles in Drug Delivery: From History to Therapeutic Applications. *Nanomaterials (Basel, Switzerland)*, 12(24), 4494. <https://doi.org/10.3390/nano12244494>
- Alghamdi, M. A., Fallica, A. N., Virzi, N., Kesharwani, P., Pittalà, V., & Greish, K. (2022). The Promise of Nanotechnology in Personalized Medicine. *Journal of personalized medicine*, 12(5), 673. <https://doi.org/10.3390/jpm12050673>
- Alqaraleh, Y Samer Wael A. Al-Zereini, Nesrin R. Mwafi, Sahar M. Jaffal, Aiman I.(2024).The Green Synthesis of Selenium Nanoparticles: A Comprehensive Review on Methodology, Characterization and Biomedical Applications. *Al-Qtatait Research Journal of Pharmacy and Technology*. <https://doi.org/10.52711/0974-360x.2024.00629>
- Amari T, Ghnaya T, and Abdelly C. (2017). Nickel, cadmium and lead phytotoxicity and potential of halophytic plants in heavy metal extraction. *South African Journal of botany volume 111*, pg 99 – 110
- Bai, S., Zhang, M., Tang, S., Li, M., Wu, R., Wan, S., Chen, L., Wei, X., & Feng, S. (2024). Effects and Impact of Selenium on Human Health, A Review. *Molecules (Basel, Switzerland)*, 30(1), 50. <https://doi.org/10.3390/molecules30010050>
- Chen M, Li X, Fan R, Yang J, Jin X, Hamid S, Xu S (2018): Cadmium induces BNIP3-dependent autophagy in chicken spleen by modulating miR-33-AMPK axis. *Chemosphere* 194:396–402.
- Chen J, Pan T, Wan N, Sun Z, Zhang Z, Li S (2017): Cadmium-induced endoplasmic reticulum stress in chicken neutrophils is alleviated by selenium. *J Inorg Biochem* 170:169–177.
- Dauplais M, Lazard M, Blanquet S, Plateau P (2013): Neutralization by metal ions of the toxicity of sodium selenide. *PLoS One* 8(1):e54353.
- Duhan JS, Kumar R, Kumar N, Kaur P, Nehra K, Duhan S. (2017): Nanotechnology: the new perspective in precision agriculture. *Biotechnol Rep.*;15:11–23
- El Batal H, Hasib A, Dehbi F, Zaki N, Quatnane A and Abdelali B.(2016). Assessment of nutritional composition of carob pulp (*Ceratonia Siliqua L.*) collected from various locations in morocco. *Journal of material sci.*7(9) :3278-3285
- Gamaniel K S. (2000): Toxicity from Medicinal Plants and their Products. *J .Nat Proc and Med.*, 4:49.
- Hara O, Kiefer G, Farrell and Kemper K (2001): A Review of Twelve Commonly Used Medicinal Herbs. *Arch. Fam. Med.*, 7: 523-526.
- Hasanuzzaman M, Nahar K, Anee I. T. and Masayuk F.(2017).Glutathione in plants: biosynthesis and physiological role in environmental stress tolerance. *Physiol mol boil plants* 23(2) 249- 268.
- Karade S., Sen S., Shergill S., Jani K, Shouche Y and Gupta R. M.(2019). Whole genome sequence of colistin-resistant *E.coli* from western India. *Medical journal armi*. DOI:101016/j.mjafi.202010/008 .
- Keshan Disease Research Group (2020): Observations on effect of sodium selenite in prevention of Keshan disease. *Chin Med J* 92:471–476.
- Khan, M., Shaik, M.R., Adil, S.F., Khan, S.T., Al-Warthan, A., Siddiqui, M., Tahir, M.N., and Tremel, W., (2018). Plant extracts as green reductants for the synthesis of silver nanoparticles: lessons from chemical synthesis. *Dalton Trans.* 47, 11988–12010.
- Kirupagaran, R. Saritha, A and Bhuvanewari S. (2016), ‘Green Synthesis of Selenium Nanoparticles from Leaf and Stem Extract of *Leucas lavandulifolia*. Sm.and Their Application
- Livingstone MBE, and Black AE, (2003): Biomarkers of nutritional exposure and nutritional status. *Journal of Nutr.* 895–920
- Madhavi V, Prasad T.N.V.K.V, Reddy A.V.B, Ravindra Reddy B, and Madhavi G,(2013) Application of phyto-genic zero valent iron nanoparticles in the adsorption of hexavalent chromium,” *Spectrochimica Acta A: Molecular and Biomolecular Spectroscopy*, vol.116, pp.17–25,2013.
- Munisamy R, Norkhadajah S, Ismail S and Praveena M. S, (2013). Cadmium exposure via food crops: a case study of intensive farming area. *American journal of applied science* 10(10): 1252 – 1262
- Narayanan S, Sathy B.N, Mony U, Koyakutty M, Nair S.V, and Menon D,(2012). “Biocompatible magnetite /gold nanohybrid



contrast agents via green chemistry for MRI and CT bioimaging,” *ACS Applied Materials and Interfaces*, vol.4, no.1, pp.251–260, 2012

- Nikalje Anna Pratima (2015). Nanotechnology and its Applications in Medicine. , *Med chem*, 5:2 DOI: 10.4172/2161-0444.1000247
- Oboh F.O.J and Masodje H.I. (2009): Nutritional and Antimicrobial Properties of *Jatropha tanjorensis* leaves. *American – Eurasian Journal of Scientific Research*, 4(1): 7-10
- Okon U Abakedi, and Faith V Eshiet (2017). Adsorption characteristics and inhibition effect of *Jatropha tanjorensis* leaf extracts on aluminium corrosion in hydrochloric acid solution. *International Journal of Chemical Science Online* ISSN: 2523-2843, Print ISSN: 2523-6075 Impact Factor: RJIF 5.22 [www.chemicaljournals.com](http://www.chemicaljournals.com) Volume 1; Issue 2;; Page No. 54-59
- Olayiwola G, Iwalewa E.O, Omobuwajo O.R, Adeniyi A.A, and Verspohi E.J., (2004): The antidiabetic potential of *Jatropha tanjorensis* leaves. *Nig J Nat Prod Med.*;8:55–8.
- Omorieg ES. and Osagie AV. (2011): Phytochemical Screening of *Jatropha tanjorensis* leaf in protein malnourished Rat Plant *Archives* vol. 7 No. 2, pp 509-516
- Orhue ES, Idu M, Ataman J.E, and Ebite L.E., (2008): Haematological and Histopathological studies of *Jatropha tanjorensis* (J.L. Ellis and Soraja) leaves in rabbits. *Asian J Biol Sci.*;1(2):84–89
- Oyewole O.I, Oladipupo O.T, Bukola V.A., (2012): Assessment of renal and hepatic functions in rats administered methanolic leaf extract of *Jatropha tanjorensis*. *Ann Biol Res.*;3(2):837–41.
- Oyegbami A, Oboh G, and Omueti O.(2010). ‘Cassava processors’ awareness of occupational hazards associated with cassava processing in south western Nigeria. *African journal of food agriculture, nutrition and development* 10( 2):2176 - 2186
- Papp L.V, Holmgren A, Khanna K.K. (2010): Selenium and selenoproteins in health and disease. *Antioxid Redox Signal*, vol. 12. New York: Mary Ann Liebert, Inc.; p. 793–5.
- Rahimzadeh M. R, Rahimzadeh M. R, Karzemi S, and Moghadamnia A, (2017). Cadmium toxicity and its treatment: An update. *Caspian J. Intern Med* 8(3): 135 - 145
- Rajak Anupam (2018). Nanotechnology and Its Application. *J Nanomed Nanotechnol*, 9:3 DOI: 10.4172/2157-7439.1000502
- Ramamurthy, C. Sampath, K. P. Arunkumar, M. S. Kumar, V. Sujatha, K. Premkumar, and C. Thirunavukkarasu (2013). Green synthesis and characterization of selenium nanoparticles and its augmented cytotoxicity with doxorubicin on cancer cells *Bioprocess and biosystems engineering* (Print) · <https://doi.org/10.1007/s00449-012-0867-1>
- Rani A, Kumar A, Lia A, and Pant M.(2014). Cellular mechanism of cadmium-induced toxicity: a review. *Int J. Environ Health Res* 24(4) 378 -99
- Ross M.S.T and Brain K.R (2000): *An Introduction to Phytopharmacy*. Pitman Medical publishing company Ltd. pp 4-5.
- Santos, C.S., Sotillo, A., Gupta, T., Delgado, S., Müller, W., Stienen, E.W., de Neve, L., Lens, L, Soares, A.M. and Monteiro, M.S. (2020). Mercury Uptake Affects the Development of *Larus fuscus* Chicks. *Environmental Toxicology and Chemistry* 39 (10):2008-2017.
- Santos, C.S.A., Blondel, L., Sotillo, A., Müller, W., Stienen, E.W.M., Boeckx, P., Soares, A.M.V.M., Monteiro, M.S., Loureiro, S., de Neve, L. and Lens, L. (2017). Offspring Hg exposure relates to parental feeding strategies in a generalist bird with strong individual foraging specialization. *Science of The Total Environment* 601-602:1315-1323.
- Senthil M. and Ramesh, C.( 2012) “Biogenic synthesis of Fe<sub>3</sub>O<sub>4</sub> nanoparticles using *Tridax procumbens* leaf extract and its antibacterial activity on *Pseudomonas aeruginosa*,” *Digest Journal of Nanomaterials&Biostructures*, vol.7, no.4, pp.1655–1661
- Shahwan T, Abu S, Sirriah M. Nairat L,(2011). “Green synthesis of iron nanoparticles and their application as a Fenton-like catalyst for the degradation of aqueous cationic and anionic dyes,”*Chemical Engineering Journal*, vol.172, no.1, pp.258–266.
- Shi X. D, Tian Y. Q., Wu J. L., and S. Y. (2020): Wang, nano-selenium shows no toxicity to normal human cells such as liver cells (LO<sub>2</sub>), embryonic kidney cells *Crit. Rev. Food Sci. Nutr.*, 0, 1–12
- Souza-Arroyo, V., Fabián, J. J., Bucio-Ortiz, L., Miranda-Labra, R. U., Gomez-Quiroz, L. E., & Gutiérrez-Ruiz, M. C. (2022). The mechanism of the cadmium-induced toxicity and cellular response in the liver. *Toxicology*, 480, 153339. <https://doi.org/10.1016/j.tox.2022.153339>
- Wang L, Hu C, Shao L. (2017): The antimicrobial activity of nanoparticles: present situation and prospects for the future. *Int J Nanomedicine.*;12: 49 -1227