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## ARTICLE

### In-vitro antioxidant, the antimicrobial and anticarcinogenic activity of environment friendly nano formulation using *Mangifera indica*

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#### ABSTRACT

The green synthesis of nanoparticles (NPs) from plant extracts is an innovative and considered as an effective method, which has many biological and therapeutic benefits. In this regard, silver nanoparticles were synthesized from leaves of *Mangifera indica* (Mango) and their properties were characterized. Morphological characterizations such as UV-Visible Spectroscopy (UV-Vis), Fourier-Transform Infra-red Spectroscopy (FTIR), and X-ray diffraction (XRD) confirm the physical characteristics of silver nano-particles. In addition, *Mangifera indica* phytochemicals possesses DPPH radical scavenging activity, antioxidant, and antibacterial properties. The nano formulation (phytochemicals + NPs) is also tested against the Hep G2 human cell line for anticarcinogenic properties. Nano formulation concentrations at 300µg/ml shown anticarcinogenic effect to Hep G2 human cell lines up to 53.29768%. The Bio pesticidal activity of the paddy pest *Cicadella viridis* (green leaf hopper) was checked through an antifeedant assay and the activity of the pest was decreased with the introduction of nano formulation. These results show better antioxidant, antibacterial and anticarcinogenic properties. Thus, they can be used to replace chemical therapeutic drugs.

## 1. Introduction

The requirement for food is rising as the world population in the world is increasing day by day. Humans and crops are affected by pathogens and pests; hence the usage of chemical pharmaceuticals and pesticides is inevitable. Since 1960, the most common method used for the control of crop pests in India is synthetic pesticides (Kumar & Singh, 2015). Pesticides are agrochemicals used in the agricultural field to protect crops from

various diseases and to increase production. It can cause dermatological, gastrointestinal, neurological, carcinogenic, respiratory, reproductive, and endocrinal effects when consumed by humans and animals. Pesticide residues have been detected in human breast milk (Negatu et al., 2016; Organization, 1990). Chemical fertilizers without regard for their hazardous effect are used extensively in the agricultural sector. The lack of knowledge

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about the optimal use of pesticides among farmers results in soil, water, and air pollution that causes human health risks. To overcome the drawbacks of chemical pesticides, nano formulations from plant extracts can be used as biopesticides (Patil et al., 2022). These are naturally occurring formulations of substances and very small quantities of them can be effective and are biodegradable (Dhuper et al., 2012). Nano formulations from young leaves of mango (*Mangifera indica*) contain highly active chemical compounds of mangiferin and chinonin. These phytochemicals possess antimicrobial, anti-diabetic, antioxidant, anti-inflammatory, anti-allergic and anticarcinogenic properties (Govindarajan et al., 2022; Sastry et al., 2003; Sivaramakrishnan et al., 2019).

Nanoparticles synthesized from mango leaf extract could be used in pesticide formulation and can check the activity against pests of crops like paddy. These studies of the anticarcinogenic activity of silver nanoparticles from various plant sources are progressing. Several studies are going on the activity of AgNPs against colon cancer, breast cancer, etc. The activity of AgNPs against the Hep G2 human cancer cell line is not well studied. The current study, the anticarcinogenic properties of *Mangifera indica* nanoparticle is used against liver cancer (Hep G2) cell lines.

## 2. Materials and Methods

### 2.1 Extraction of *Mangifera indica*

*Mangifera indica* leaves were collected from Coimbatore and shade dried before extraction. 15 g of mango leaves were finely and mixed with 150 ml of distilled water. It is then sterilized and cooled. The extract was then kept in a shaker incubator overnight and filtered using Whatman filter paper.

### 2.2 Phytochemical tests

The preliminary phytochemical analysis was performed by the following tests.

1. Alkaloids test (Wagner's test): Two ml of *Mangifera indica* leaf extract was allowed to react with Wagner's reagent, until the appearance of Red color (or) Brown color precipitate which indicates the presence of alkaloids (Govindarajan et al., 2022).
2. Phenol test (Ferric chloride test): One ml *Mangifera indica* leaf extract and 10% of FeCl<sub>3</sub> solution were added and allowed until the appearance of formation of blue or green colour which indicates the presence of phenols (Vijayakumar & Sumathi, 2016).
3. Fehling's Test (reducing sugar): Two drops of Fehling's reagents added to two ml of *Mangifera indica* leaf extract. The solution was placed in the 40°C water bath until formation of brick red precipitate which indicates the presence of reducing sugars. (Brain & Turner, 1975).
4. Saponins Test (Foam test): Two ml *Mangifera indica* leaf extract with 9 ml of double distilled water and the extract was vigorously shaken. The foam formation indicates the presence of Saponins (Brain & Turner, 1975)
5. Test for Flavonoids: Few drops of NaOH added to 1 ml of *Mangifera indica* extract. The appearance of yellow colour confirms the flavonoids (Sofowora, 1996).
6. Phytosterols Test (Salkowski's test): Few drops of chloroform + 100 µl of acetic anhydride + 100 µl of Conc. H<sub>2</sub>SO<sub>4</sub> were added to 1 ml of *Mangifera indica* extract. The appearance of greenish blue colour signifies phytosterols (Govindarajan et al., 2022).
7. Proteins and Amino acids (Ninhydrin test): One ml of *Mangifera indica* extract and 3 drops of Ninhydrin reagent. The presence of proteins forms purple colour (Govindarajan et al., 2022).
8. Steroids test: One ml of acetic anhydride and few drops of chloroform followed by 2 drops of H<sub>2</sub>SO<sub>4</sub> was added to one ml of *Mangifera indica* leaf extract. The appearance of blue or green colour represents sterols (Govindarajan et al., 2022).
9. Tannin test: One ml of *Mangifera indica* leaf extract and few drops of 1% FeCl<sub>3</sub> reagent. The yellow colour formation indicate to the presence of Tannin. (Saravanan et al., 2020).
10. Glycoside test: 1 ml *Mangifera indica* (*M. indica*) leaf extract and five drops of Conc H<sub>2</sub>SO<sub>4</sub>. The red colour formation indicate to the Cardiac glycosides present. (Saravanan et al., 2020).

### 2.3 Green Synthesis *Mangifera indica* - silver nanoparticle

The reducing effect of *M. indica* leaf extract, from silver nitrate to silver nanoparticles. 10 ml of *M. indica* leaf extract added to 90 ml silver nitrate solution (0.1 M) incubated for 24 hours dark at room temperature. (Dogra et al., 2022).

### 2.4 UV analysis

After 24 hours nano particle was analysed by 260 – 400 nm wavelength by using the labman UV spectrophotometer.

### 2.5 Fourier transforms infrared spectroscopy

For FTIR Analysis, the synthesized *M. indica* silver nanoparticles Sample was centrifuged at 8000 rpm for 10 minutes. The Supernatant was Discard and pellet was washed twice with 10 mL of double Distilled water. The nanoparticle was dried to make powder. The nanoparticles was analysed by FTIR Spectrum. (Govindarajan et al., 2022).

### 2.6 Antioxidant property

Antioxidant activities of *Mangifera indica* leaves and silver nanoparticles synthesized using mango leaves were evaluated by the DPPH method and nitric oxide scavenging method (Ramli et al., 2021).

### 2.6.1 DPPH radical scavenging activity

The free radical scavenging activity of plant extract was measured using 2, 2-diphenyl-1-picrylhydrazyl (DPPH), and the free radicals was quantified based on previous studies (Braca et al., 2001; Wongsu et al., 2022). DPPH solution (0.004%) was prepared using methanol. Then, 3 mL of DPPH solution added to 0.5, 1.0, 1.5, 2.0, and 2.5  $\mu$ l of plant extract and ascorbic acid (control) at similar concentration range. The test samples (leaf extract) were mixed and incubated for 30 min at room temperature. In spectrophotometer (OD<sub>517</sub>) the loss of DPPH pigmentation was analyzed. Similar protocol was used to measure DPPH scavenging activity for control samples and the calculated using the following formula.

$$\text{DPPH Radical Scavenging activity (\%)} = \frac{(\text{control} - \text{test})100}{\text{control}}$$

### 2.6.2 Nitric oxide (NO) scavenging activity

NO scavenging activity of the leaf extract was estimated by adding 400  $\mu$ L of C<sub>5</sub>H<sub>4</sub>FeN<sub>6</sub>Na<sub>2</sub>O<sub>3</sub> (100 mM) + 100  $\mu$ L of PBS (pH: 7.4) were added to 0.5, 1.0, 1.5, 2.0, and 2.5  $\mu$ L of plant extract and allowed to incubate for 2 hr 30 mins at 25 °C. Then 2 mL of Griess reagent was added to 2 the test samples and allowed to incubate for 30 minutes at room temperature. The absorbance of pigmentation changes was observed at OD<sub>540nm</sub>. All the experiments were performed in triplicates, the NO scavenging activity was calculated using the following formula.

$$\text{NO scavenging activity (\%)} = \frac{(\text{control} - \text{test})100}{\text{control}}$$

## 2.7 Antimicrobial activity

The antibacterial effect to AgNPs was tested with certain infection causing pathogens such as *Escherichia coli*, *Pseudomonas species*, *Klebsiella species*, and *Proteus species*. These bacterial species were prone to gut inflammation (Govindarajan et al., 2020; Bhuyar et al., 2021). Urinary Tract Infections (Govindarajan et al., 2022), and abundantly available in the environment which transmits the infection to humans (Shanmugasundarasamy et al., 2022).

### 2.7.1 Media and culture condition

Culture media, Luria Britani (LB) media were used for all bacterial strains in this study at neutral pH under aseptic conditions.

### 2.7.2 Preparation of the bacterial inoculum

The bacterial strains were inoculated from single colonies in streak plates of stock culture. A loop full bacterial colony were sub-cultured to test tubes of 50 ml LB broth and incubated 12 h at

37°C on a shaking incubator which is used as overnight cultures for all bacterial assays (Govindarajan et al., 2022).

### 2.7.3 Well diffusion method

LB agar plates were prepared adding 1.2% agar to LB media and allowed for solidification. After solidification, 20  $\mu$ l of overnight bacterial cultured were swabbed on agar surfaces in different LB plates. Then the agar surfaces were punctured uniformly (5 mm) using well puncture. About 25, 50, 75, and 100  $\mu$ L of plant extract was pipetted into four different wells for all bacterial strains along with 10  $\mu$ L of commercial antibiotics and allowed to incubate overnight at 37 °C. The zone of inhibition was measured (in diameter) from the clear zone around the well in mm.

## 2.8 In-vitro anticarcinogenic activity

### 2.8.1 Cell line:

The human liver cell line (HEP G2) was procured from National Centre for Cell Science (NCCS), Pune. The cell line was grown in Eagles Minimum Essential Medium enriched with 10 % fetal bovine serum (FBS). The cell line was incubated at 37 °C at CO<sub>2</sub> incubator. The culture medium was replaced with fresh medium to maintain cell line at live state.

### 2.8.2 Cell treatment procedure

The monolayer cells were separated using trypsin-ethylene diamine tetra acetic acid (EDTA) to develop single-cell suspensions. The viable cells were counted using a haemocytometer and diluted to 1x10<sup>5</sup> cells/ml using 5 % FBS. 100  $\mu$ l of cells were pipetted to wells in 96-well plates allowed to incubate for 24 h at 37 °C CO<sub>2</sub> incubator for the cells to adhere on well surfaces. After incubation, the cells were treated with 100 L different concentrations of nano formulated extract followed by adding fresh dimethyl sulfoxide (DMSO) and allowed incubate for 24 h at 37 °C CO<sub>2</sub> incubator. Then the samples were imaged in fluorescent microscope to estimate the anticarcinogenic effect of nano formulated extract. A control sample was prepared with similar protocol without adding the nano formulated extract. All the experiments were done in triplicates.

## 3. Results and discussion

### 3.1 Phytochemical analysis of *M. indica* extract

The phytochemical analysis for the *M. indica* extract shows presence of various compounds, tabulated below (Table.1).

**Table.1:** Phytochemical availability in *M. indica* extract

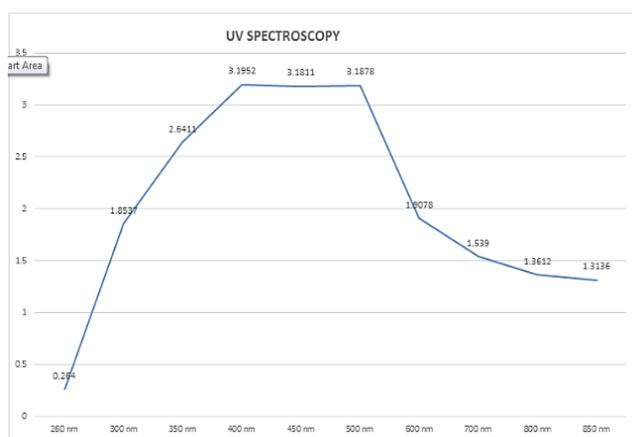
S. No	Compounds	Present / Absent
1.	Phenol	+++
2.	Tannin	+++
3.	Alkaloid	++

4.	Reducing sugar	++
5.	Steroids	+
6.	Phytosterols	+
7.	Amino Acid	-
8.	Protein	-
9.	Flavonoids	-
10.	Glycosides	-

+++ (high), ++ (moderate), + (low), and - (absent)

### 3.2 UV-Vis Spectroscopy confirms AgNPs:

The AgNPs synthesized using *M. indica* leaves extract was detected by UV-VIS Spectroscopy. Absorbance peak of the nanoformulation of *M. indica* in Figure.1 shows a maximum absorbance peak at 400 nm, which depicts the spectral property of the silver nanoparticle in the solution. The UV spectrum values were tabulated in Table.2.



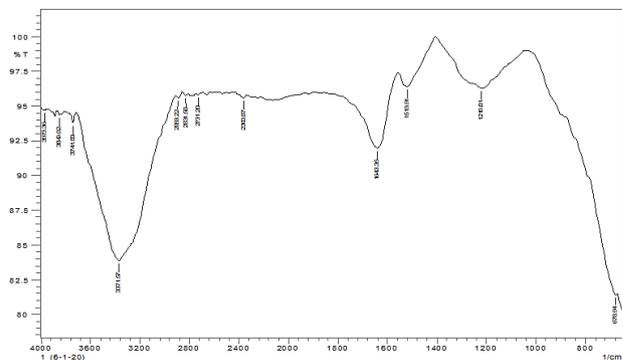
**Figure 1** UV spectroscopy graph depicts the maximum absorbance peak of *M. indica* nano formulation

**Table 2** UV-Vis spectrum values for AgNPs.

Sample Name	<i>Mangifera indica</i>
Abs (260nm)	0.2640
Abs (300nm)	1.8537
Abs (350nm)	2.6411
Abs (400nm)	3.1592
Abs (450nm)	3.1811
Abs (500nm)	3.1878
Abs (600nm)	1.9078
Abs (700nm)	1.5390
Abs (800nm)	1.3612
Abs (850nm)	1.3136

### 3.3 FTIR Analysis

FTIR spectrum of *M. indica* AgNPs was shown in figure.2 depicts the presence functional groups of different compounds. The spectrum was recorded in the region between 600 and 4000/cm. From table.3, the FTIR peaks at 3371.57cm<sup>-1</sup> show the presence of OH groups, and 1643.35 cm<sup>-1</sup> shows C=O groups.



**Figure 2** FTIR spectrum of silver nanoparticle synthesized from *Mangifera indica*.

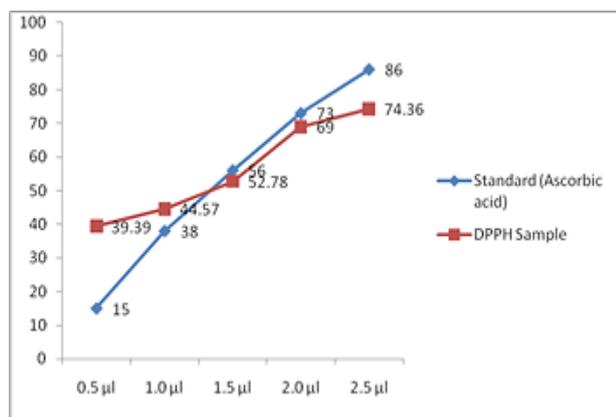
**Table 3** FTIR analysis of *Mangifera indica* nanoparticle's functional compounds

Absorbance (cm <sup>-1</sup> )	Group	Compound class
3973.36	O-H stretching	Alcohol
3849.92	O-H stretching	Alcohol
3741.90	O-H stretching	Alcohol
3371.57	O-H stretching	Alcohol
2893.22	C-H stretching	Aldehyde
2831.50	C-H stretching	Aldehyde
2731.20	C-H stretching	Aldehyde
2360.87	O=C=O stretching	Carbon dioxide
1643.35	C=O stretching	Imine/Oxime
1519.91	N-O stretching	Nitro compound
1219.01	C-O stretching	Vinyl ether

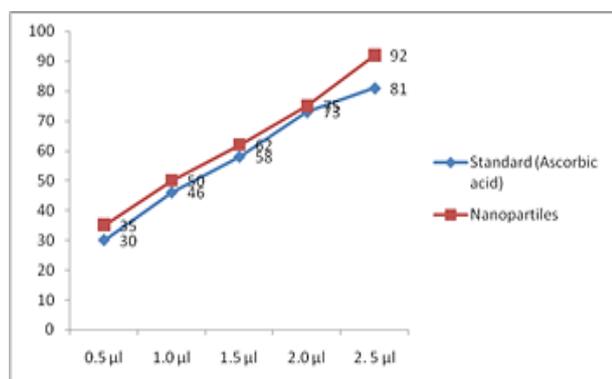
### 3.4 Antioxidant property

Antioxidants properties of cells provides protection against free radicals, which poses a major role in cardiovascular disease, cancer, and other infectious diseases (Bhuyar et al., 2021). Free radical molecules were produced at the breakdown of food molecules during digestion process, irradiation effect, and smoking tobacco. In this experiment, the synthesized silver nanoparticles showed good antioxidant properties scavenging properties. Figure 3 visualizes Nitric oxide scavenging activity and Figure 4 supports the DHHP scavenging activity and these

both assays were done to establish the antioxidant property of the AgNPs which shows satisfactory good results.



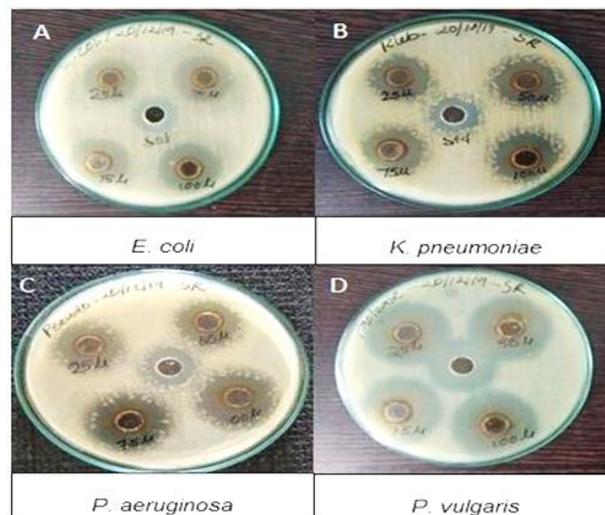
**Figure 3** DPPH activity of *M. indica* extract with AgNPs.



**Figure 4** Nitric oxide scavenging activity of *Mangifera indica* nanoparticles.

### 3.5 Antibacterial activity

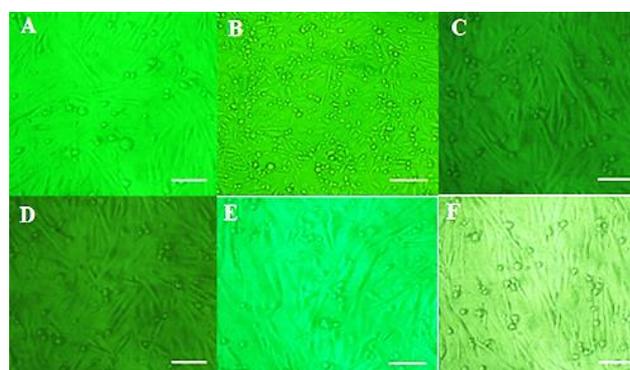
Figure 5 shows the antibacterial activity of *M. indica* extract with AgNPs against bacterial strains of *Escherichia coli*, *Pseudomonas aeruginosa*, *Klebsiella pneumonia*, and *Proteus vulgaris*. All the strains used in the experiment were gram-negative. Among these organisms, the data from Table 7 shows the nanoformulation synthesized from *Mangifera indica* showed high inhibitory action, especially against *Proteus vulgaris*.



**Figure 5** Antibacterial activity of *M. indica* extract with AgNPs.

### 3.6 Anticarcinogenic activity

*In vitro* cytotoxicity of the silver nanoparticles synthesized from *Mangifera indica* was evaluated against Hep G2 which is a human liver cancer cell line. Fig-6 shows that with increasing concentration of the nanoparticles, the cytotoxicity against HEP G2 cancer cell lines increase. From figure 6, we can visualize the increase in the cytotoxic activity of the nanoparticles with an increase in concentration. Table 8 displays the experimental data.



**Figure 6** Anti-carcinogenic activity of nano formulation against HEP G2 cell line.

**Table 4** Anti-bacterial activity (zone of inhibition) of *M. indica* extract with AgNPs.

Concentration* of Nano formulation (µl)	Zone of inhibition (diameter (mm)) on bacterial strains.			
	<i>Escherichia coli</i>	<i>Klebsiella pneumoniae</i>	<i>Pseudomonas aeruginosa</i>	<i>Proteus vulgaris</i>
25	10	12	13	16
50	15	18	15	18
75	1.7	20	18	19
100	2	22	20	22
Standard (µl) 10	10	10	10	12

\* Standard Concentration of antibiotics is 0.1 mg/ml whereas; the plant-extract concentration is (1 mg/ml).

#### 4. Discussion

This study shown the efficiency of AgNPs from *M. indica* as an anti-carcinogenic agent. The excitation of surface plasmon vibration induces the change of colour from golden yellow to dark brown and denotes the generation of silver nanoparticles in the aqueous solution (Govindarajan *et al.*, 2022). The phytochemical studies of the synthesized nanoparticle revealed the presence of phenol, reducing sugar, flavonoids, tannin, and alkaloid in the nano formulation which have pharmacological properties such as antioxidant and antimicrobial properties (Ayeleso *et al.*, 2014). By using UV-Vis spectroscopy maximum absorption peak of nano formulation was found between 400nm and 500nm. Other studies reported that the characteristics of Ag nanoparticles normally appear at a wavelength interval of 400nm–600nm and the maximum absorbance of AgNPs occurs at 421 nm (Ayeleso *et al.*, 2014; Vasireddy *et al.*, 2012).

FTIR spectroscopy is a reliable and sensitive methods for the detection of bio-molecular fractions of various plant extracts (Joshi, 2012; Palanisamy *et al.*, 2021). In this study, the functional groups such as alcohol, aldehyde, nitro compound, vinyl ether, and amine were identified by using FTIR. XRD analysis revealed eight important peaks present in the (20-80) 2 $\theta$  range. In another study, four important XRD peaks for silver nanoparticles synthesized from *Mangifera indica* in the (20-80) 2 $\theta$  range were reported (Aljabali *et al.*, 2018; Jain *et al.*, 2009). In this study, the synthesized nano formulation showed a strong action against *Escherichia coli*, *Proteus vulgaris*, *Klebsiella pneumoniae*, and *Pseudomonas aeruginosa* where all are gram-negative stains. Similar studies observed that silver nanoparticles undergoing an interaction with bacterial cells displayed strong action against wide range of bacterial strains (Birla *et al.*, 2009; Kumar *et al.*, 2012; Reddy., 2014).

The high antioxidant activity of the silver nanoparticles that are synthesized using the leaf extracts of *Mangifera indica* is due to the capping agent and its free radical scavenging potential. The results of DPPH free radical scavenging activity showed that the nanoparticles exhibited antioxidant activity and a maximum percentage of inhibition was observed at 74.36. A similar study done on the silver nanoparticles generated from the extracts of *Piper longum* fruit has been reported where a maximum percentage of inhibition was observed at 55 (Gul *et al.*, 2016).

Natural products with high antioxidant activities were investigated for their anticarcinogenic activities, i.e., oxidative stress. The phytochemical present in the *Mangifera indica*, mangiferin is a potential compound with active C-glycosylated xanthose. Polyphenol mangiferin reduced DNA damage by scavenging free radicals through its antioxidant properties. In this study, the synthesized nano formulation revealed high potency against Hep G2 which is a human liver cancer cell line. In similar studies *in vitro* cytotoxic potential of AgNPs synthesized from *Mangifera indica* in MCF-7 and HCT-116 cells were observed.

#### 5. Conclusion

In this study, AgNPs were synthesized from mango leaf extract through a simple, single-step, low cost, and green process. Subsequently, the synthesized silver nanoparticles were characterized by UV-Vis, XRD, and FTIR to verify the morphology, production, and crystallinity. The nano formulation contains more antimicrobial activity against *Escherichia coli* and *Proteus vulgaris*. The antioxidant property was confirmed by DPPH and Nitric oxide scavenging activity. Silver nanoparticles from *Mangifera indica* can be potent natural antioxidants due to the presence of phytochemicals like phenol, phytosterols, tannin, steroids, and reducing sugar. *Mangiferin* is a therapeutically active C-glycosylated xanthone of *Mangifera indica*. Polyphenols of mangiferin exhibit antioxidant properties and tend to decrease oxygen-free radicals, thereby reducing DNA damage. The AgNPs synthesized showed high efficiency against Hep G2 which is a human liver cell line. The nano formulation using *M. indica* leaves has anticarcinogenic, antioxidant, antimicrobial, and insecticidal activity. The silver nanoparticle synthesized from *Mangifera indica* has shown high efficiency against the Hep G2 cell line. So, it can be used in Hep G2 cancer cell line treatment.

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#### Reference

- Aljabali, A. A., Akkam, Y., Al Zoubi, M. S., Al-Batayneh, K. M., Al-Trad, B., Abo Alrob, O., Evans, D. J. 2018. Synthesis of gold nanoparticles using leaf extract of *Ziziphus zizyphus* and their antimicrobial activity. *Nanomaterials*, 8(3), 174.
- Ayeleso, A. O., Oguntibeju, O. O., & Brooks, N. L. 2014. In vitro study on the antioxidant potentials of the leaves and fruits of *Nauclea latifolia*. *The Scientific World Journal*, 2014.
- Birla, S., Tiwari, V., Gade, A., Ingle, A., Yadav, A., & Rai, M. 2009. Fabrication of silver nanoparticles by *Phoma glomerata* and its combined effect against *Escherichia coli*, *Pseudomonas aeruginosa* and *Staphylococcus aureus*. *Letters in Applied Microbiology*, 48(2), 173-179.
- Braca, A., De Tommasi, N., Di Bari, L., Pizza, C., Politi, M., & Morelli, I. (2001). Antioxidant principles from *baubhinia tarapotensis*. *Journal of natural products*, 64(7), 892-895.
- Brain, K., & Turner, T. 1975. The practical evaluation of phytochemicals. *Wright Scientechina, Bristol*, 57-58.
- Bhuyar, P., Maniam, G. P., & Govindan, N. (2021). Isolation and characterization of bioactive compounds in medicinal plant *Centella asiatica* and study the effects on fungal activities. *Journal of microbiology, biotechnology and food sciences*, 10(4), 631-635.
- Dhuper, S., Panda, D., & Nayak, P. 2012. Green synthesis and characterization of zero valent iron nanoparticles from the leaf extract of *Mangifera indica*. *Nano Trends: J Nanotech App*, 13(2), 16-22.

- Dogra, S., Sharma, M. D., Tabassum, S., Mishra, P., Bhatt, A. K. & Bhuyar, P. (2022). Green biosynthesis of silver nanoparticles (AgNPs) from *Vitex negundo* plant extract and its phytochemical screening and antimicrobial assessment next to pathogenic microbes. *Journal of Microbiology, Biotechnology and Food Sciences*, e5993.
- Feng, J., Wang, T., Zhang, S., Shi, W., & Zhang, Y. 2014. An optimized SYBR Green I/PI assay for rapid viability assessment and antibiotic susceptibility testing for *Borrelia burgdorferi*. *PloS one*, 9(11), e111809.
- Govindarajan, D. K., & Kandaswamy, K. 2022. Virulence factors of uropathogens and their role in host pathogen interactions. *The Cell Surface*, 8, 100075.
- Govindarajan, D. K., Meghanathan, Y., Sivaramakrishnan, M., Kothandan, R., Muthusamy, A., Seviour, T. W., & Kandaswamy, K. 2022. *Enterococcus faecalis* thrives in dual-species biofilm models under iron-rich conditions. *Archives of Microbiology*, 204(12), 1-11.
- Govindarajan, D. K., Selvaraj, V., Selvaraj, A. S. J. M., Hameed, S. S., Pandiarajan, J., & Veluswamy, A. 2022. Green synthesis of silver micro-and nanoparticles using phytochemical extracts of *Cymbopogon citratus* exhibits antibacterial properties. *Materials Today: Proceedings*.
- Govindarajan, D. K., Viswalingam, N., Meganathan, Y., & Kandaswamy, K. 2020. Adherence patterns of *Escherichia coli* in the intestine and its role in pathogenesis. *Medicine in Microecology*, 5, 100025.
- Govindarajan, D. K., Viswalingam, N., Meganathan, Y., Devaraj, B. S., Sivamani, S., & Sivarajasekar, N. 2022. Response surface methodology optimization for extraction of pectin from waste rinds of *Citrus medica*. Paper presented at the AIP Conference Proceedings.
- Gul, S., Ismail, M., Khan, M. I., Khan, S. B., Asiri, A. M., Rahman, I. U., Kambh, M. A. 2016. Novel synthesis of silver nanoparticles using melon aqueous extract and evaluation of their feeding deterrent activity against housefly *Musca domestica*. *Asian Pacific Journal of Tropical Disease*, 6(4), 311-316.
- Jain, D., Daima, H. K., Kachhwaha, S., & Kothari, S. 2009. Synthesis of plant-mediated silver nanoparticles using papaya fruit extract and evaluation of their anti-microbial activities. *Digest journal of nanomaterials and biostructures*, 4(3), 557-563.
- Joshi, D. D. 2012. *Herbal drugs and fingerprints: evidence based herbal drugs*: Springer Science & Business Media.
- Kumar, P., Senthamil, S. S., Lakshmi P. A., Prem, K. K., Ganeshkumar, R., & Govindaraju, M. 2012. Synthesis of silver nanoparticles from *Sargassum tenerrimum* and screening phytochemicals for its antibacterial activity. *Nano Biomed Eng*, 4(1), 12-16.
- Kumar, S., & Singh, A. 2015. Biopesticides: present status and the future prospects. *J Fertil Pestic*, 6(2), 100-129.
- Negatu, B., Kromhout, H., Mekonnen, Y., & Vermeulen, R. (2016). Use of chemical pesticides in Ethiopia: a cross-sectional comparative study on knowledge, attitude and practice of farmers and farm workers in three farming systems. *The Annals of occupational hygiene*, 60(5), 551-566.
- Organization, W. H. 1990. Public health impact of pesticides used in agriculture: World Health Organization.
- Palanisamy, K. M., Paramasivam, P., Maniam, G. P., Rahim, M. H. A., Govindan, N., & Chisti, Y. 2021. Production of lipids by *Chaetoceros affinis* in media based on palm oil mill effluent. *Journal of biotechnology*, 327, 86-96.
- Ramli, A. N. M., Hamid, H. A., Zulkifli, F. H., Zamri, N., Bhuyar, P., & Manas, N. H. A. (2021). Physicochemical properties and tenderness analysis of bovine meat using proteolytic enzymes extracted from pineapple (*Ananas comosus*) and jackfruit (*Artocarpus heterophyllus*) by-products. *Journal of Food Processing and Preservation*, 45(11), e15939.
- Reddy, N. J., Vali, D. N., Rani, M., & Rani, S. S. 2014. Evaluation of antioxidant, antibacterial and cytotoxic effects of green synthesized silver nanoparticles by *Piper longum* fruit. *Materials Science and Engineering: C*, 34, 115-122.
- Saravanan, Y., Devaraj, B. S., Velusamy, N. K., Soundirarajan, P. S., & Kandaswamy, K. 2020. Phytochemical Extracts of *Leucas aspera* and *Dahlia pinnata* Exhibit Antimicrobial Properties in *Escherichia coli* and *Enterococcus faecalis*. *Current Biotechnology*, 9(4), 297-303.
- Sastry, M., Ahmad, A., Khan, M. I., & Kumar, R. 2003. Biosynthesis of metal nanoparticles using fungi and actinomycete. *Current science*, 162-170.
- Shanmugasundarasamy, T., Govindarajan, D. K., & Kandaswamy, K. 2022. A review on pilus assembly mechanisms in Gram-positive and Gram-negative bacteria. *The Cell Surface*, 8, 100077.
- Sivaramakrishnan, M., Jagadeesan Saravanan, V., Karaiyagowder Govindarajan, D., Meganathan, Y., Devaraj, B. S., Natesan, S., Kandaswamy, K. 2019. Green synthesized silver nanoparticles using aqueous leaf extracts of *Leucas aspera* exhibits antimicrobial and catalytic dye degradation properties. *SN Applied Sciences*, 1(3), 1-8.
- Sofowora, A. 1996. Research on medicinal plants and traditional medicine in Africa. *The Journal of Alternative and Complementary Medicine*, 2(3), 365-372.
- Vasireddy, R., Paul, R., & Mitra, A. K. 2012. Green synthesis of silver nanoparticles and the study of optical properties. *Nanomaterials and Nanotechnology*, 2, 8.
- Vijayakumar, S., & Sumathi, A. 2016. Preliminary phytochemical and GC-MS analysis of bioactive compounds from *Moringa concanensis* Nimmo leaves family: Moringaceae. *Int. J. Recent Adv. Multidiscipl. Res*, 3, 1257-1259.
- Wongsa, P., Bhuyar, P., Tongkoom, K., Spreer, W., & Müller, J. (2022). Influence of hot-air drying methods on the phenolic compounds/allicin content, antioxidant activity and  $\alpha$ -amylase/ $\alpha$ -glucosidase inhibition of garlic (*Allium sativum* L.). *European Food Research and Technology*, 1-13.