Contents lists available at ScienceDirect

Saudi Journal of Biological Sciences

journal homepage: www.sciencedirect.com

Original article

Employment of *Cassia angustifolia* leaf extract for zinc nanoparticles fabrication and their antibacterial and cytotoxicity



لجمعية السعودية لعلوم الحياة AUDI BIOLOGICAL SOCIET

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ARTICLE INFO

Article history: Received 2 January 2021 Revised 21 February 2021 Accepted 22 February 2021 Available online 4 March 2021

Keywords: Cassia angustifolia Nano particles Zinc TEM Antibacterial and cytotoxicity

ABSTRACT

The plant Cassia angustifolia belongs to Saudi Arabia, which is one of the native places and now cultured throughout the global countries. Medical care in the Arab world is an essential outlet for medicinal plants, both because they are crucial elements for prophetic medicine and due to their lengthy background in the Middle East, Cangustifolia is one of the medicinal plants used in the Saudi Arabia. The usage of plant extracts for synthesizing nanoparticles is conducive to other biological material, since it avoids the lengthy phase of cell culture maintenance. Silver nanoparticles attract further attention due to their strong conductivity, stability and antimicrobial activity across different metal nanoparticles. The present study was designed in the Saudi C. angustifolia leaves with the zinc synthesis of nanoparticles and its antibacterial ability. The plant extracts of C. angustifolia was used for synthesis of zinc nanoparticles, antimicrobial activities against bacterial strains have been tested along with transmission electron microscope (TEM), UV spectroscopy and antimicrobial activities have been conducted. This study showed that silver ions may be transferred from the plant extract to silver nanoparticles. AgNPs biogenic capacity to antibacterial with lovo cell with IC₅₀ ranged from 33.5 ± 0.2 µg/mL demonstrated strong antibacterial capacity to antibody. The overall absorption value for the extract was between 420 and 440 nm and the color transition to green was the plasma absorption of the AgNPs. TEM results was showed in 200,000 magnification. The uniqueness of the current study is that Cassia angustifolia leaf extract from Saudi Arabia was used to prepare the metallic nanoparticles. Additionally, ZnCl₂ may also be used as nanoparticles of mineral salt and zinc, which, since their application has been confirmed, are antimicrobial

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1. Introduction

As per the numerous scientific reports, the utilization of medicinal plants in the form of raw extracts, or chemical components are largely associated with a reduced risk of degenerative diseases which are associated oxidative stress, because they contain antioxidants such as phenol, carotenoids, vitamins and flavonoids (Arts and Hollman, 2005). Plants have been extensively used as a therapeutic medicine to treat human wellbeing disturbances in numerous ancient systems. Natural plant extract is commonly

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Peer review under responsibility of King Saud University.



used in the beauty, dairy, color and medicinal industries because of their possible pharmacological and therapeutic use (Al-Ghamdi et al., 2020). Cassia angustifolia Vahl is a traditional medicinal plant actually belongs to Caesalpiniaceae family, frequently classified as cassia senna/makkai (Marpaung, 2020). This traditional plant is a residence of Saudi Arabia and then exist in Yemen, Egypt which is a fast-track growing 5–8 m tall shrub, widely cultivated for its leaves and fruits in limited regions in Pakistan country. Since the ancient times, a number of societies have succumbed to nature, primarily to plants as medicinal and health conscious sources. Presently, a large majority of the global population, especially in developing countries, uses plants to meet the essential needs of medicinal attention (Hafez et al., 2019). This plant is accepted in pharmacopoeia both in the United Kingdom and United States and C angustifolia leaves and pods are used for the treatment in anti-helmentic decoction powder for intestinal worms which is also widely used as an anti-pyretic in typhoid, splenic enlargement, cholera, laxative, anemia, toxicity and genotoxicity induced by E.Coli (Ahmed et al., 2016).

https://doi.org/10.1016/j.sjbs.2021.02.075

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Leguminosae is a family widely known as the family of legumes, peas or beans is one of the largest families of flowering plants, comprising approximately 650 genera and 18,000 species. It is a massive, economically significant species of flowering plants (Hu et al., 2000). Casalpinioideae, Papilionoideae and Mimosoideae are divided between three sub-families with distinct names and are now known to be independent families (Gledhill, 2008). Caesalpinioideae is a genetically engineered plant and are primarily trees distributed in the humid tropical zone. Caesalpinacae, with 152 genera and 2800 species, represents approximately 11% of the known legume flora. Cassia is an important genus of the Caesalpiniaceae, containing some 600 species (Dave and Ledwani, 2012, Abdel Hakim et al., 2019). The anthroquinone glycosides are found in the leaves, pods and sees Cassia Angustifolia Vahl and are used in the pharmaceuticals industry, especially sennoside a and b (Oureshi et al., 2007).

Senna is a spreading shrub, up to 1.8 m tall, abundant in South India and elsewhere. Any taxonomists are considering it as a syndicate of *Senna alexandrina*. Vahl *C. angustifolia*; It's *Senna* linn. The whole plant, particularly for its cathartic properties, is medicinally important and is used for laxation. Crude protein, 10.6%; Pentosan, 5.9%; colored ash, 4.5%; water soluble gum, 34%; extractive alcohol, 9–0%; extractive ether, 5.8% and extractive hexane, 6.1%. Seeds are creamy, dicotyledonous and medium-sized, endosperm-containing seeds of 48–52%. The details are included in Table 1 (Chaubey and Kapoor, 2001).

Nanoparticles have individuality and the ability to improve the metabolism of plants (Giraldo et al., 2014). Nanoparticles have unusual physicochemical properties, i.e. high surfaces, high reactions, tunable poric sizes and particularly morphology of Nanotechnology opens up a broad variety of potential uses within the fields of biotechnology and agriculture (Siddiqui et al., 2015). Based on the characteristics of nanoparticles interact with plants and trigger many morphological and physiological changes. Nanoparticles' effectiveness depends on their chemical structure, their scale, their coverage, their reactivity, and above all their efficacy (Khodakovskaya et al., 2012). Nanotechnology has a function to play in addressing numerous environmental and health concerns that exist in agricultural activities from improper use of chemical fertilizer. Nanoparticles including carbon nanotube, lead, oxide of titanium, gold, sulfur, zinc, iron, silica, apatite, copper and carbon nanoparticles have increased plant growth, and crop production is increased at correct amounts (Sanzari et al., 2019).

Previous investigations have been done on Gold and Silver nanoparticles from plants such as Hibiscus rosa sinensis, Sorbus Auuparia, Cinnamomum Camphora, Citrullus colocynthesis, Catharanthus hirta L, Zingiber officinale, Acalypha indica, Moringa oleifera, Saururus chinenis, Artocarpus sativum, Anacardium Westernale, Nelumbo nucifera or Ocimum sanctuary. In addition, plants have been synthesized in the past for example. In 2012 the connection between nanoparticles was reported by a study from India using Cassia angustifolia leaf extract (Amaladhas et al., 2012). Limited

| Table 1 | | |
|------------------------------|---------------|---------------|
| The seed element of chemical | testing of C. | angustifolia. |

| Chemical list | Seed coat | Endosperm | Germ |
|--------------------------|--------------|---------------|----------|
| Protein | 12.1% | 4.2% | 41.6% |
| Moisture | 5.2% | 11.2% | 7.8% |
| Ether extract | 2.4% | 0.7% | 9.1% |
| Ash | 8.7% | 1.15% | 4.5% |
| Crude fiber | 20.7% | 0.8% | 11.8% |
| Carbohydrates soluble in | Pentose | Galactomannan | Pentose, |
| water | | | Glucose |

Table was adopted from (Chaubey and Kapoor, 2001) studies.

studies found that plant growth and production was positively influenced by nanoparticles. However, there was no study implemented in the *C. angustifolia* leaf in the Saudi Arabia using the Zinc nanoparticles to study the anticancer activity. The present study was designed in the Saudi *C. angustifolia* leaves with the zinc synthesis of nanoparticles and its antibacterial ability.

2. Materials and methods

2.1. Collection of plant material

The aerial italica sections of Gabal Al-Ateeq, Al Madinah Al Munawwwarah (24_24037.500 N 39_32034.000E) were gathered. The plant taxonomy was performed in line with morphological features and library data and checked by a taxonomy (Al-Haidari and Al-Oqail, 2020).

2.2. Plant storage

Samples were identified and deposited for further processing in polythene bags at 4 °C. The plant components were fermented and dried with water. With the aid of a milling machine, dried samples were well ground in a fine powder. The powder was stored for extraction and further analysis in air-sealed plastic containers at room temperature.

2.3. Synthesis of Zinc-Nanoparticles

By inserting 10 g of powder in 100 mL of solvent, aquatic and ethanolic extracts were produced from collected plant materials. Heat therapy was administered on the aqueous extract for 10 min at 80 °C to avoid enzymes' operation. The solution was then sorted by the nominee No. 1 (pore scale 125 mm). In addition, the ethanol extract has been maintained overnight and filtered over the same nominee for Whatman alluded to above. For the extract concentration, Filtrate was heated and stored for future use. 10 mL of each prepared extract as a reduction and reduction for the zinc nanoparticle synthesis. Capping agents were mixed in a flask of Erlenmeyer with 90 mL of a 1 mMAgNO3 solution which enabled for 48 h of room temp. Each sample was prepared for repeatability three times. AgNPs have been processed at 4 °C temperature for further analysis (Mohammed et al., 2018).

2.4. Bacterial strains

In this study, we have used 4 bacterial species such as P. *aerug-inosa, E. Coli, S. aureus* and *Bacillus subtilis* (*B. subtilis*) were received from the Biology Department Faculty of Sciences, University of Princess Nora Bint Abdulrahman.

2.5. Phytochemical screening

In order to classify active constituents, standard methods were used to create phytochemical screens of the extract (Selim et al., 2020).

2.6. Transmission electron microscopy

The cell-level distribution of PFOS and PFOA in root cross sections was defined by TEM-EDS in order to understand the findings of DESI-MS. New from the Foshan nursery plants were obtained Ci, Td, Ca, Pa and Ad with heights of around 20 cm, cultivated at 1/2 force Hoagland for 2 weeks. Then these plants had been incubated at a concentration of 50 μ g / L for 7 days in a 1/2 intensity Hoagland-nutrient solution spiked in PFOA (or PFOS). Fresh root

parts in root hair zone Ci, Td, Ca, Pa and Ao were subsequently dehydrated by graduated ethanol (30 percent - 100 per cent - v /v), embedded in the Epon – 812 resin with a leica UCT ultramicrotome in 2 mm bits, set to 2.5 percent (v | v) glutaraldehyde and 1 percent (v | v) osmium tetroxide successively. The copper grids (50-70 nm) also had uranyl acetate and plum citrate tainted by 4% (w / v) then under electron microscopy (JEM-1400 Plus, Hitachi, Tokyo, Japan). In each sample we scaned the root slice surface at the first stage to see if the elementF was usable. The energy spectra in this spring were conducted with the energy dispensing X-ray spectrometer (IXRF Systems, Houston, Tex, USA). In addition, the epidermic-to-xyle vessels were studied in the areas that contained the wall of the cells and the gap between the cells and separate cell organules (e.g. nucleus, chondriosome, Golgi and cytoplasm). Fluorine's (F) energy range was not contained in either of the blank community root samples and did not find PFASs or any of the other fluorinated chemicals in the blank samples. Therefore, the occurrence of a fluorine energy range (F) in the PFOS (or PFOA) root cross sections of the treatment population has been detected (Wang et al., 2020)

2.7. UV spectroscopy

The following is used for biogenic AgNPs to detect UV–visible spectrophotometer (Shimadzu, Tokyo, Japans), as well as for dynamic light screening, zeta potential, electron electric field microscopy (Peabody, MA, USA), and transmission electron microscopy (Peabody, MA, USA). Double-beam UV-2450 (200–800 nm) was used to search for the decline of pure Ag + ions. The absorption range of green ZnO. NPs displayed a normal peak of 374 nm. (Bellmann et al., 2020).

2.8. Antibacterial activity

A well-gar diffusion approach has been used to calculate the anti-bacterial active action of silver nanoparticles. Three bacteria forms, the gram-positive Staphylococcus aureus and the aeruginosa Pseudomonas and gram-negative community Escherichia coli, have been examined. Microorganism pure communities. On Mueller-Hinton Agar, they were sub-cultured. Swabbed 0.2 mL (CFU 2.5*10⁵ mL) of bacteria strains. Sterile swabs are put randomly on each individual agar plate. Three of them spaced correctly Wells (holes) with a diameter of 4 mm per plate were produced with the sterile cultivation agar surface. Borer with metal cork. The usage of 0.2 mL extract in each hole was aseptic and held in room. Temperature for 1 h to enable the extracts to be diffused and incubated into the agar medium. The comparison negative regulation was sterile purified water. Plates at 37 °C have been incubated for 18 - 24 h. Inhibition areas were analyzed that existed as a clear region around the pools (Mohammed et al., 2018).

2.9. Statistical analysis

Each evaluation was carried out for repetitions at least three times in this analysis and standard deviations and means have been determined by Microsoft Excel 2013 (Khan et al., 2019).

3. Results

Results of the current study indicate that the accumulation of the *Staphylococcus aureus* and the *aeruginosa Pseudomonas* and gram-negative community *Escherichia coli* to the silver in the form of a nitrate (0.1 mM) induced a color shift in the mixture from yellowish to brown, which suggests that AgNPs have a good plasmon resonance. Our study is in agreement with the previous study (Veerasamy et al., 2011). After a couple of days, the color shift of the mixture was steady and dark brown. It was quite long. UV–vis spectroscopy was used to observe the bio-reduction mechanism between Ag ions and silver nanoparticles. Silver surface plasmon resonance of 420, 425 nm, 430, 440 nm and 430 nm for silver nanoparticle mixtures formed by the *Staphylococcus aureus* and the *aeruginosa Pseudomonas* and gram-negative community *Escherichia coli* are observed in the mixture of silver nanoparticles morphology, surface structure and scale were studied using TEM. The phytochemical screening and TEM photos are seen in Figs. 1 and 2, which are prepared in the present study *Cassia angustifolia* plant.

Silver nanoparticles have been biosynthesized in this analysis as a promising medicinal candidate. Documentation has been made on the biosynthesis of nanoparticles with biological agents such as microbes, fungi, actinomycetes, yeast, algae and herbs (Mohammed, 2015). The silver nanoparticles synthesis with leaves of *C. angustifolia* and AgNO₃ solution. The color change identified in C.angustifolia leaf. The cytotoxic potency of ZnNPs was tested using MTT against MDA and MCF-7 cell line that were cultured in 96 wells plate. Different concentrations of the ZnNPs used to the cell lines and tested after 72 h of incubation at 37°C in 5% CO₂. IC₅₀ were observed at different ZnNPs (Table 2 and Fig. 3). By determining a cell viability percentage, cytotoxic activity of silver nanoparticles was calculated using an MTT trial. The concentration-based cytotoxic behavior in cells of breast cancer revealed synthesized AgNPs. Phytosynthesized silver nanoparticles have caused 50% (IC_{50}) , at a concentration of 17.9 ± 1.4 µg / mL, of anti-cancer activity to MCF 7-cells. Extract of aqueous flora C. Angustifolia displayed mild cell line activity. Fig. 3 indicates the concentration of the silver nanoparticles against the percentage of MCF 7 in this sample.

Viability
$$\% = \frac{OD \text{ sample}}{OD \text{ control}} \times 100$$

The second leading cause of death in the world has been cancer, the malignant case of unregulated cell proliferation. Colorectal and lung tumors are troubling and correlated with multiple forms of cancer Mortality of the highest. New cancer medications with reduced adverse effects and increased selectivity and potency are among the most difficult areas of contemporary clinical science (Ali et al., 2012, Fatima et al., 2019).



Fig. 1. Color change of zinc nanoparticles obtained using *Cassia angustifolia* leaf extracts A. and seed extract B.

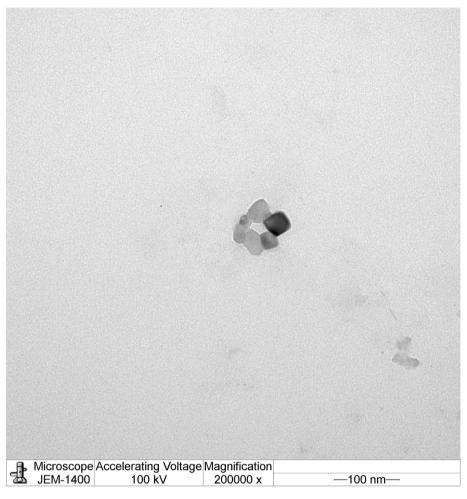


Fig. 2. TEM image of zinc nanoparticles obtained using Cassia angustifolia leaf extracts A. and seed extract B. Magnification is 100,000 (A) and 200,000 (B) and scale bar represents 200 nm (A) and 100 nm (B).

4. Discussion

The present invention relates to green synthesis of metal nanoparticles, and particularly, synthesis of Zinc nano-particles using *Cassia angustifolia* leaf extracts as a reducing and capping agent. Nanotechnology is currently assumed as a robust medical tool, which involves formation of different nanomaterials. For such formation, some metals have been employed such as copper, magnesium, gold and silver. For their unique physicochemical characteristics, nanoparticles (NPs) are gaining the interest of scientist since nanoparticles display totally innovative properties that vary from their original bulk, such as a high surface area, capability in drug delivery, and decreased toxicity. Chemical and physical methods are the well-known method in the synthesis of nanomaterials. However, environmental pollution might be expected therefore, biological synthesis of nanomaterials is environmentally friendly technology since it could be easily available, cost effective with no side effects could be expected. Biosynthesis of nanoparticles using honey from two different flora sources have been investigated. Due to their antimicrobial activity, NPs can be used in a different application, including foodstuff, medication, sunscreens and cosmetics. Thus, a method of producing metal nanoparticles utilizing *Cassia angustifolia* thus solving the aforementioned problems is desired.

A method of preparing metal nanoparticles using *Cassia angus*tifolia leaf extract from Saudi Arabia. Comprises offering solution

Table 2

Effect of cytotoxic potency of ZnNPs.

| | | Silver Nanoparticles sample | Average particle size (nm)* | Zeta potential (mV)* | Inhibition zone + gram (mm) | Inhibition zone - gram (mm) | Cytotoxicity IC ₅₀ |
|--------------------------|-----|--------------------------------|---------------------------------|-------------------------|-----------------------------------|--------------------------------|----------------------------------|
| NPs from plant leaves | 185 | -6.7 | Staphylococcus sp. 26.53 | E. coli 27.6 | Bacillus subtilis 27.93 | <i>MDA</i> 33.5 ± 0.2 | MC7 17.9 ± 1.4 |
| Plant leaves extract | - | - | 17.6 | 7.6 | 9.3 | - | - |

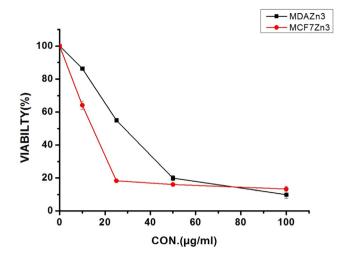


Fig. 3. TEM image of zinc nanoparticles obtained using *Cassia angustifolia* leaf extracts A. and seed extract B. Magnification is 100,000 (A) and 200,000 (B) and scale bar represents 200 nm (A) and 100 nm (B).

include metal salt; combine *Cassia angustifolia* leaf extracts with the metal salt solution (1 mM) to produce metal nanoparticles. The metal salt can be a mixture of Zinc chloride (Zncl). The metal nanoparticles can be zinc nanoparticles that have a mean diameter in the range of about 88 nm to 200 nm. The nanoparticles can be used as an antimicrobial and anticancer. The zinc nanoparticles can be used as an antimicrobial and anticancer agent. Features of the current design will become readily clear upon additional information of the following description and illustrations. A method of preparing metal nanoparticles using *Cassia angustifolia* leaf extract from Saudi Arabia. Comprises offering solution include metal salt; combine *Cassia angustifolia* leaf extracts with the metal salt solution (1 mM) to produce metal nanoparticles. The metal salt can be a mixture of Zinc chloride (Zncl). The metal nanoparticles can be zinc nanoparticles that have a mean diameter in the range of about 88 nm to 200 nm. The nanoparticles can be used as an antimicrobial and anticancer. The zinc nanoparticles can be used as an antimicrobial and anticancer agent. Features of the current design will become readily clear upon additional information of the following description and illustrations. In the below Fig. 4, the spectroscopy results have been documented.

Our current study has created important results on the basis of previous studies which can be presented in practical applications. These benefits involve the success of the plant extracts of AgNO3, the prepared nanoparticles show antimicrobial activity against small silver for the fast formation of tiny silver nanoparticles (67.8–155.7 nm). *S. aureus, E. coli and P. aeruginosa* similar to and in certain instances much bigger than the commercial antibiotics. In addition, the plant extracts prepared by nanoparticles possessed LoVo cytotoxicity; hence plant extracts used for silver nanoparticle synthesis can be seen as additional values for the production of nano-medicine and pharmaceutical products for cancer cell and pathogenic microorganisms (Mohammed et al., 2018).

TEM photos verified the synthetic ZnO and nanoparticles hexagonal shape. This arrangement suggests that the three 2D ZnO have more ionicity and consequently improved catalytic activity. Structures nanoparticles. Due to the greater surface to volume proportion, decreasing particle size has been well kno wn to increase their role as antimicrobial and anti-carcinogenic agent (Azizi et al., 2017)

| 1.59 0.010 | | Magazza | Viscosity (cP): | | |
|---|--|--|--|--|--|
| 0.010 | | Manur | | | |
| | | measurem | ent Date and Time: | Sunday, November 11, | 2018 10: |
| 25.0 | | | Duration Used (s): | 60 | |
| 327.4 | | Measurem | nent Position (mm): 3.00 | | |
| Disposable micro cuvette (40µI) Attenuato | | | Attenuator: | 10 | |
| | | Size (d.nm): | % Intensity: | St Dev (d.nm): | |
| 185.8 | Peak 1: | 184.2 | 100.0 | 49.50 | |
| 0.240 | Peak 2: | 0.000 | 0.0 | 0.000 | |
| 0.927 | Peak 3: | 0.000 | 0.0 | 0.000 | |
| Good | | | | | |
| | | | | | |
| 1 | 10 | 100 | 100 |) 10000 | |
| | | Size (d.nm) | | | |
| | 327.4 Disposable r 185.8 0.240 0.927 Good | 327.4 Disposable micro cuvette (40 185.8 Peak 1: 0.240 Peak 2: 0.927 Peak 3: Good Size Distr | 327.4 Measurem Disposable micro cuvette (40μl) Size (d.nm): 185.8 Peak 1: 184.2 0.240 Peak 2: 0.000 0.927 Peak 3: 0.000 Good Size Distribution by Intensity | 327.4 Measurement Position (mm): Disposable micro cuvette (40µl) Attenuator: Size (d.nm): % Intensity: 185.8 Peak 1: 184.2 100.0 0.240 Peak 2: 0.000 0.0 0.927 Peak 3: 0.000 0.0 Good Size Distribution by Intensity 1 10 100 1000 | 327.4 Measurement Position (mm): 3.00 Disposable micro cuvette (40µl) Attenuator: 10 Size (d.nm): % Intensity: St Dev (d.nm): 185.8 Peak 1: 184.2 100.0 49.50 0.240 Peak 2: 0.000 0.0 0.000 0.927 Peak 3: 0.000 0.0 0.000 Good Size Distribution by Intensity |

Fig. 4. Size distribution and zinc nanoparticles obtained using Cassia angustifolia leaf extracts A and seed extract B.

5. Conclusion

Cassia angustifolia leaf extract from Saudi Arabia was used to prepare the metallic nanoparticles. The research consists of the preparation of mineral salt and plant flower extract in a blend that mixes leaf extract and mineral salt (1 mL) to create nanoparticles of the metal by means of a ratio of 9:1. ZnCl₂ can be used as the mineral salt and zinc nanoparticles with an average diameter of approximately 185 nm can be used. The nanoparticles are antimicrobial and anticancer after their usage have been proven.

Declaration of Competing Interest

The authors declared that there is no conflict of interest.

Acknowledgments

This research was funded by the Deanship of Scientific Research at Princess Nourah bint Abdulrahman University through the Fasttrack Research Funding Program.

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