

KARAKTERISASI KOMPREHENSIF NANO PARTIKEL EKSTRAK DAUN SIRSAK

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Abstrak

Nanoteknologi telah merevolusi berbagai bidang, termasuk kedokteran, dengan memberikan peluang baru untuk penelitian dan aplikasi. Penelitian ini berfokus pada karakterisasi nanopartikel yang berasal dari ekstrak daun sirsak. Nanopartikel disintesis menggunakan polimer kitosan dan ekstrak daun sirsak, dan sifat-sifatnya dianalisis. Hasil penelitian menunjukkan rentang ukuran partikel yang luas, dengan ukuran rata-rata 210,9 nm dan modus 161,4 nm. Nanopartikel menunjukkan potensial zeta negatif sebesar -31,5 mV, yang menunjukkan stabilitasnya dalam larutan. Efisiensi penyerapan ditemukan sebesar 87,90%, dan kapasitas muatan obat mencapai 37,92%. Mikroskop elektron pemindaian mengonfirmasi morfologi nanopartikel yang berbentuk bulat. Temuan ini menyoroti potensi nanopartikel dari ekstrak daun sirsak untuk pengiriman obat dan menekankan pentingnya karakterisasi komprehensif untuk aplikasi di masa depan.

Kata kunci: Nanopartikel, Ekstrak, Sirsak, Karakterisasi

COMPREHENSIVE CHARACTERIZATION OF NANO PARTICLES DERIVED FROM SOURSOP LEAF EXTRACT

Abstract

Nanotechnology has revolutionized various fields, including medicine, by offering new opportunities for research and application. This study focuses on the characterization of nanoparticles derived from soursop leaf extract. Nanoparticles were synthesized using chitosan polymer and soursop leaf extract, and their properties were analyzed. The results showed a wide range of particle sizes, with an average size of 210.9 nm and a mode of 161.4 nm. The nanoparticles exhibited a negative zeta potential of -31.5 mV, indicating their stability in solution. The absorption efficiency was 87.90%, and the drug loading capacity reached 37.92%. Scanning electron microscopy confirmed the spherical morphology of the nanoparticles. These findings highlight the potential of soursop leaf extract nanoparticles for drug delivery and emphasize the importance of their comprehensive characterization for future applications.

Keywords: *Nanoparticles, Extracts, Soursop, Characterization*

INTRODUCTION

Over the past few decades, nanotechnology research has rapidly developed, which has presented novel prospects for its applications in diverse fields, including medicine. The characterization of nanoparticles has garnered the interest of researchers. Nanoparticles are defined as submicron-sized particles, exhibiting distinct physicochemical properties and behavior compared to their larger counterparts. These entities possess distinctive characteristics, such as a significant level of responsiveness, an extensive surface area, and the capability to traverse natural obstructions, such as cellular membranes (Anselmo & Mitragotri, 2016; Biswas & Wu, 2005; De et al., 2008; Mohanraj & Chen, 2006).

Plant extracts have been identified as an essential source for the production of nanoparticles through synthesis. The utilization of plants in traditional medicine has been attributed to their capacity to furnish bioactive compounds. The botanical specimen garnered significant interest in the foliage of the *Annona muricata* plant, commonly known as soursop leaves.

The leaves of the soursop plant are known to contain a range of bioactive compounds, such as alkaloids, flavonoids, phenolics, and triterpenoids (Coria-Télez et al., 2018; Gajalakshmi et al., 2012; Gavamukulya et al., 2014; Hasmila et al., 2019; Vijayameena et al., 2013; Zubaidi et al., 2023). These compounds have been found to exhibit a variety of beneficial properties, including antioxidant (Florence et al., 2014; Muthu & Durairaj, 2015; Nawwar et al., 2012), antibacterial (Haro et al., 2014; Silva et al., 2021; Viera et al., 2010), anticancer (Abdul Wahab et al., 2018; Agu et al., 2018; Prasad et al., 2019; Rady et al., 2018), and antitumor activities (González-Pedroza et al., 2021; Hamizah et al., 2012; Mansour et al., 2018; Merlín-Lucas et al., 2021; Roduan et al., 2019).

In several recent investigations, scholars have achieved the production of nanoparticles utilizing extract derived from soursop leaves. Nanoparticles exhibit significant promise in diverse medical domains, such as cancer management (Gavamukulya et al., 2019,

2021; Jabir et al., 2021) or pharmaceutical transportation (Santos et al., 2023; Vernet-Crua et al., 2023). Before their widespread application, a comprehensive understanding of the characteristics of nanoparticles is imperative. Characterizing nanoparticles derived from soursop leaf extract entails a range of analytical techniques employed to delineate the physical and chemical attributes of these particles (Burleson et al., 2004; Domingos et al., 2009; Hall et al., 2007; A. Kumar & Dixit, 2017; Powers et al., 2007; Titus et al., 2019). This research focused on the characterization of nanoparticles produced from soursop leaf extract.

MATERIALS and METHODS

Materials

In this study, the materials used included chitosan, glacial acetic acid, soursop leaf extract, distilled water, ethanol, DMSO (Dimethyl sulfoxide), uranyl acetate dye, and carbon-coated copper grid.

Nanoparticle Preparation

Soursop leaf extract nanoparticles were prepared using chitosan polymer (Desmiaty et al., 2016) One gram of chitosan was dissolved in 100 mL of 1% glacial acetic acid solution using a magnetic stirrer to produce a chitosan solution with a concentration of 1%. A total of 500 mg of soursop leaf extract was dissolved in 135 mL of distilled water, 10 mL of ethanol, and 15 mL of 1% DMSO, then the solution was filtered. Furthermore, 80 mL of 1% chitosan solution was added to the soursop leaf extract solution so that the concentration in the solution became 0.3%. The stirring process was carried out using a magnetic stirrer at 400 rpm to form nanoparticles. The formation of nanoparticles is characterized by homogeneous turbidity. The solution was stirred using a magnetic stirrer for 30 minutes to form a stable solution of soursop leaf extract nanoparticles. Furthermore, the stability of the soursop leaf extract nanoparticle solution was observed for 5 days, including observations of changes in color, turbidity, and precipitate.

Characterization of Nanoparticles

a. Examination of particle size, particle distribution and polydispersity index

The Particle Size Analyzer was employed in this study to determine the particle size and distribution, along with the polydispersity index. The experimental data were collected at a temperature of 25 degrees Celsius. The samples underwent dilution with distilled water prior to analysis. Subsequently, the specimen is inserted into the cuvette, which is then positioned within the apparatus's holder. Subsequently, the NIBS (Non-Invasive Back-Scatter) technique was employed to determine the diameter (Behera et al., 2012; Chiriac et al., 2009; Kaszuba & Connah, 2006; Kathad & Gajera, 2014; Zimmerman et al., 2014).

b. potential zeta analysis

The investigation used a zeta potential analyzer to measure the zeta potential value at a temperature of 25°C. Prior to analysis, the samples underwent dilution with distilled water. Subsequently, the specimen is introduced into the cuvette, which is inserted into the apparatus's receptacle. Subsequently, the user selects the option from the menu of the zeta potential analyzer to measure the zeta potential value in millivolt units (Dissanayake et al., 2021; Lunardi et al., 2021; Marín et al., 2017; Wu et al., 2005; Zhang et al., 2008).

c. Absorption efficiency and DLC analysis

To begin the experiment, 3 mL of nanoparticles extracted from soursop leaves (NEDS) should be transferred into a centrifugation tube. Next, the NEDS should be dissolved in 30 mL of distilled water and subjected to centrifugation at a rate of 10,000 revolutions per minute for a duration of 30 minutes. Following the completion of the centrifugation procedure, the supernatant was extracted and subjected to analysis of its absorbance utilizing a UV-vis spectrophotometer with a wavelength of 422.5 nanometers (Amoli-Diva et al., 2016; Chiang et al., 2011; Marciano et al., 2008; Nagaonkar et al., 2015; Peckus et al., 2017).

d. Examination of particle morphology

A scanning electron microscope was utilized to analyze particle morphology. Several 1-3 drops of the sample solution are placed on a carbon-coated copper grid. Then, uranyl acetate dye was added and left for 30 minutes

at room temperature. This process is carried out using a voltage of 120 KVA. The sample to be analyzed is prepared with the minimum possible thickness so that electrons can penetrate it, and the results of this electron penetration are then processed into an image (Evans et al., 2011; Hailstone et al., 2009; J. Liu, 2005; Mühlfeld et al., 2007; Schaffer et al., 2009).

RESULT

Particle size, particle size distribution, and polydispersity index

The NanoPartica SZ-100V2 Series was utilized to conduct particle size analysis on nanoparticles derived from soursop leaf extract. The results indicate that the average particle size was 210.9 nm, with a standard deviation of 134.4 nm. Additionally, the mode of the particle was found to be 161.4 nm, as illustrated in Figure 1. The findings suggest notable fluctuations in the particle size distribution of nanoparticles derived from soursop leaf extract, with a considerable span of particle sizes.

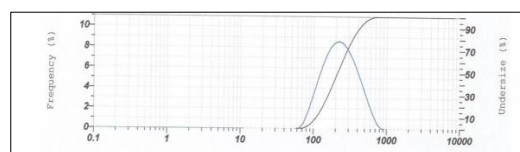


Figure 1. particle size analysis

The particle size distribution analysis reveals that the average particle value indicates larger particles at 210.9 nm. In contrast, the mode at 161.4 nm suggests particle clusters exhibiting a higher frequency at that particular size.

The polydispersity index is a quantitative measure employed to describe the distribution of particle sizes within a given sample. The Polydispersity Index (PDI) is a unitless quantity that ranges from 0 to 1. A Polydispersity Index (PDI) value near zero denotes a homogeneous distribution of particles, whereas a PDI value in proximity to one denotes a heterogeneous distribution of particles. The soursop leaf extract nanoparticles exhibited a polydispersity index value of 0.381, which suggests a non-uniform particle size distribution.

Although the value is close to homogeneity, it suggests the existence of particle size variation within the sample. The polydispersity index value of 0.381 suggests that the particle size distribution of the sample is heterogeneous to a certain extent but not wholly non-uniform when viewed through a scientific lens. While there could be inevitable fluctuations in the size of particles, the general composition remains relatively uniform. This interpretation demonstrates the likelihood of multiple particles with diverse dimensions within a specified sample.

Zeta potential

The NanoPartica SZ-100V2 Series was utilized to obtain measurements, which revealed that the mean zeta potential value of nanoparticles derived from soursop leaf extract was -31.5 mV. The findings presented in Figure 2 were obtained through the utilization of the equipment.

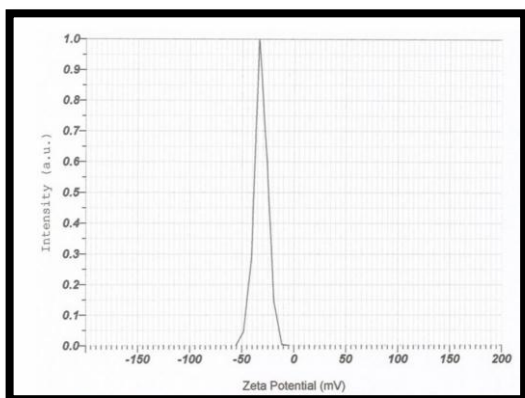


Figure 2. Potential Zeta Measurement

Upon conducting calculations, it was determined that the electrophoretic mobility of nanoparticles generated from soursop leaf extract was $-0.000248 \text{ cm}^2/\text{Vs}$.

Absorption Efficiency

The efficacy of nanoparticle entrapment can be evaluated by determining the ratio of particles adsorbed by the adsorption system or medium. According to the analysis, the nanoparticles exhibit an entrapment efficiency of 87.90%. The data suggests that a significant % of the total number of nanoparticles, precisely 87.90%, are efficiently adsorbed by the adsorption device or material employed. The efficacy of

nanoparticle adsorption in this study may be influenced by various factors, such as the physical and chemical characteristics of the nanoparticles, their size, the nature of the adsorption medium, and other experimental parameters (Dhakar et al., 2010, 2012; Lv et al., 2018; Song et al., 2008; Were et al., 2003).

Drug Loading Capacity

The drug loading capacity pertains to the quantity of drug that can be accommodated within the delivery system, specifically in the nanoparticles of soursop leaf extract. The drug loading capacity was determined to be 37.92%. The nanoparticle system derived from soursop leaf extract exhibits a remarkable capacity for drug delivery. The findings indicate that nanoparticles synthesized from soursop leaf extract possess the potential to serve as a dependable vehicle for drug delivery.

Particle morphology

Figure 3 depicts the spherical shape and morphological characteristics of the nanoparticles derived from soursop leaf extract, as determined through scanning electron microscopy (SEM). The spherical morphology suggests that the particles possess a spherical architecture with a relatively consistent diameter.

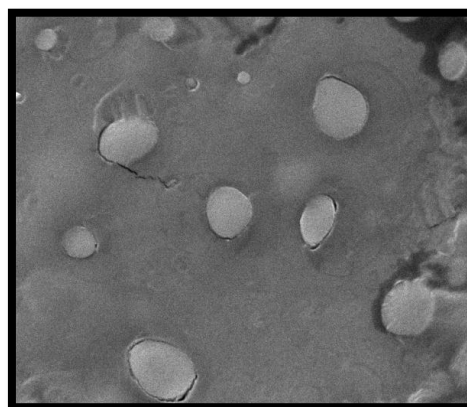


Figure 3. SEM nano particles of soursop leaf extract

Whilst spherical particles are frequently observed through SEM analysis, the findings of this study exhibit the wide range of particle shapes that can be observed. The observed particle diversity can be attributed

to the methodology employed, which yields nanoparticles exhibiting a range of spherical morphologies.

DISCUSSION

Since the inception of nanoparticles, their multifaceted impacts, particularly in healthcare, have been widely recognized (Dilnawaz & Acharya, 2023; Haleem et al., 2023; R. Liu et al., 2023; Verma et al., 2023). Consequently, numerous advancements have been made to enhance the stability and efficacy of nanoparticles. The impact of basic sciences, particularly physics, and chemistry, on the advancement of nanoparticles has been significant (Apsokardu & Johnston, 2018; He & Alexandridis, 2015; G. V. P. Kumar et al., 2007; Qi & Wang, 2004; Sizochenko et al., 2014; Sylvestre et al., 2004; Yli-Juuti et al., 2013). Several pharmaceutical companies are competing to develop this technology, with numerous pharmaceutical scientists dedicated to producing nanoparticles possessing a range of desirable attributes (Ahmadi Shadmehri et al., 2019; Rodríguez-Luis et al., 2016; Singh et al., 2021; Sridhar & Ramakrishna, 2013).

Diverse techniques employed for the synthesis of nanoparticles yield varying sizes of the resultant nanoparticles. The size of nanoparticles is influenced by the type of extract utilized during their production. Several research studies have demonstrated that a uniform technique, such as the production of silver nanoparticles, can yield particles of varying sizes. Diverse plant parts of a single plant species have been observed to yield varying sizes (Elavazhagan & Arunachalam, 2011; Jain et al., 2009; P. P. N. V. Kumar et al., 2014; Kumarasamyraja & Jeganathan, 2013; Nakkala et al., 2014; Sun et al., 2014; Vijayaraghavan et al., 2012).

Based on the results of making nanoparticles using soursop leaf extract, it is known that the nanoparticle size is 161.4 nm. The same method might produce different sizes if using different types of extracts. However, further research is needed to prove this.

The average zeta potential value of the soursop leaf extract nanoparticles obtained (-31.5 mV) indicates that the surface of the

nanoparticles has a negative charge. This could be due to the functional groups and chemical bonds in the soursop leaf extract, which give the particles a negative charge. A negative zeta potential indicates electrostatic repulsion between the particles in the solution, preventing agglomeration or agglomeration of the particles (Ferrari et al., 2010; Huo et al., 2019; Jiang et al., 2009; Schultz et al., 2008; Sis & Birinci, 2009).

Nanoparticles derived from soursop leaf extract that exhibit a negative zeta potential have a greater propensity for stability in solution. Additionally, the electrophoretic mobility of nanoparticles derived from soursop leaf extract (-0.000248 cm²/Vs) represents the velocity of particle movement under the influence of an electric field. Particles exhibiting negative electrophoretic mobility are observed to move in a direction that is opposite to the direction of the electric field that has been applied. The observation above is by the negative charge on the surface of nanoparticles derived from soursop leaf extract. A greater electrophoretic mobility indicates an increased velocity of particle movement when subjected to an externally applied electric field (Ito et al., 2004; Streich et al., 2015).

The production of soursop leaf extract nanoparticles results in the formation of spherical-shaped particles, which offer numerous benefits. It is widely acknowledged that spherical nanoparticles exhibit the highest surface area-to-volume ratio compared to other particle geometries. Using a spherical morphology confers favourable mechanical robustness to nanoparticles when suspended in a medium. Particles with a spherical shape exhibit improved inter-particle rubbing, mitigating the likelihood of undesired agglomeration or settling. This process enhances the dispersion and stability of particles, thereby enabling their homogeneous distribution in the liquid medium (Calderón-Jiménez et al., 2022; Chitra & Annadurai, 2013; Jindal, 2017; Radoń et al., 2018; Sundrarajan et al., 2017).

This suggests that the production of nanoparticles from soursop leaf extract holds promise as a viable alternative for

nanoparticle design, with potential implications for the advancement of medical science.

CONCLUSIONS and SUGGESTION

Conclusion

Soursop leaf extract nanoparticles in this study have a broad size distribution, with an average size of 210.9 nm and a negative zeta potential of -31.5 mV, indicating stability in the solution. The absorption efficiency of the nanoparticles reached 87.90%, and the drug loading capacity reached 37.92%. Morphological analysis showed that the nanoparticles were spherical. These findings highlight the potential use of soursop leaf extract nanoparticles as reliable vehicles for drug delivery.

Suggestion

Future research can carry out more in-depth analyses such as stability analysis, biocompatibility testing, and potential toxicity of nanoparticles from soursop leaf extract.

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REFERENCE

Abdul Wahab, S. M., Jantan, I., Haque, M. A., & Arshad, L. (2018). Exploring the leaves of *Annona muricata* L. as a source of potential anti-inflammatory and anticancer agents. *Frontiers in Pharmacology*, *9*, 661.

Agu, K. C., Okolie, N. P., Falodun, A., & Engel-Lutz, N. (2018). In vitro anticancer assessments of *Annona muricata* fractions and in vitro antioxidant profile of fractions and isolated acetogenin (15-acetyl guanacone). *Journal of Cancer Research and Practice*, *5*(2), 53–66.

Ahmadi Shadmehri, A., Namvar, F., Miri, H., Yaghmaei, P., & Nakhaei Moghaddam, M. (2019). Assessment of antioxidant and antibacterial activities of Zinc Oxide nanoparticles, Graphene and Graphene decorated by Zinc Oxide nanoparticles.

International Journal of Nano Dimension, *10*(4), 350–358.

Amoli-Diva, M., Pourghazi, K., & Hajjaran, S. (2016). Dispersive micro-solid phase extraction using magnetic nanoparticle modified multi-walled carbon nanotubes coupled with surfactant-enhanced spectrofluorimetry for sensitive determination of lomefloxacin and ofloxacin from biological samples. *Materials Science and Engineering: C*, *60*, 30–36.

Anselmo, A. C., & Mitragotri, S. (2016). Nanoparticles in the clinic. *Bioengineering & Translational Medicine*, *1*(1), 10–29.

Apsokardu, M. J., & Johnston, M. V. (2018). Nanoparticle growth by particle-phase chemistry. *Atmospheric Chemistry and Physics*, *18*(3), 1895–1907.

Behera, S. S., Patra, J. K., Pramanik, K., Panda, N., & Thatoi, H. (2012). *Characterization and evaluation of antibacterial activities of chemically synthesized iron oxide nanoparticles*.

Biswas, P., & Wu, C.-Y. (2005). Nanoparticles and the environment. *Journal of the Air & Waste Management Association*, *55*(6), 708–746.

Burleson, D. J., Driessen, M. D., & Penn, R. L. (2004). On the characterization of environmental nanoparticles. *Journal of Environmental Science and Health, Part A*, *39*(10), 2707–2753.

Calderón-Jiménez, B., Montoro Bustos, A. R., Pereira Reyes, R., Paniagua, S. A., & Vega-Baudrit, J. R. (2022). Novel pathway for the sonochemical synthesis of silver nanoparticles with near-spherical shape and high stability in aqueous media. *Scientific Reports*, *12*(1), 882.

Chiang, C.-K., Chen, W.-T., & Chang, H.-T. (2011). Nanoparticle-based mass spectrometry for the analysis of biomolecules. *Chemical Society Reviews*, *40*(3), 1269–1281.

Chiriac, A. P., Niță, L. E., Neamțu, I., & Bercea, M. (2009). Contribution to polymer nanoparticles analysis by laser light scattering. *Polymer Testing*, *28*(8), 886–890.

- Chitra, K., & Annadurai, G. (2013). Antimicrobial activity of wet chemically engineered spherical shaped ZnO nanoparticles on food borne pathogen. *International Food Research Journal*, 20(1).
- Coria-Téllez, A. V, Montalvo-González, E., Yahia, E. M., & Obledo-Vázquez, E. N. (2018). *Annona muricata*: A comprehensive review on its traditional medicinal uses, phytochemicals, pharmacological activities, mechanisms of action and toxicity. *Arabian Journal of Chemistry*, 11(5), 662–691.
- De, M., Ghosh, P. S., & Rotello, V. M. (2008). Applications of nanoparticles in biology. *Advanced Materials*, 20(22), 4225–4241.
- Desmiaty, Y., Rahmat, D., & Maulidina, N. S. (2016). Pembuatan Nanopartikel Berbasis Kitosan dari Infus Daun Sirsak (*Annona muricata* LINN) Sebagai Antioksidan. *Journal of Tropical Pharmacy and Chemistry*, 3(4), 307–312.
- Dhakar, R. C., Maurya, S. D., Sagar, B. P. S., Bhagat, S., Prajapati, S. K., & Jain, C. P. (2010). Variables influencing the drug entrapment efficiency of microspheres: A pharmaceutical review. *Der Pharmacia Lettre*, 2(5), 102–116.
- Dhakar, R. C., Maurya, S. D., & Saluja, V. (2012). From formulation variables to drug entrapment efficiency of microspheres: a technical review. *Journal of Drug Delivery and Therapeutics*, 2(6).
- Dilnawaz, F., & Acharya, S. (2023). Nanoparticle-based CRISPR/Cas delivery: an emerging tactic for cancer therapy. *Current Medicinal Chemistry*.
- Dissanayake, K., Midekessa, G., Lättেকivi, F., & Fazeli, A. (2021). Measurement of the size and concentration and zeta potential of extracellular vesicles using nanoparticle tracking analyzer. *Next Generation Culture Platforms for Reliable In Vitro Models: Methods and Protocols*, 207–218.
- Domingos, R. F., Baalousha, M. A., Ju-Nam, Y., Reid, M. M., Tufenkji, N., Lead, J. R., Leppard, G. G., & Wilkinson, K. J. (2009). Characterizing manufactured nanoparticles in the environment: multimethod determination of particle sizes. *Environmental Science & Technology*, 43(19), 7277–7284.
- Elavazhagan, T., & Arunachalam, K. D. (2011). Memecylon edule leaf extract mediated green synthesis of silver and gold nanoparticles. *International Journal of Nanomedicine*, 1265–1278.
- Evans, J. E., Jungjohann, K. L., Browning, N. D., & Arslan, I. (2011). Controlled growth of nanoparticles from solution with in situ liquid transmission electron microscopy. *Nano Letters*, 11(7), 2809–2813.
- Ferrari, L., Kaufmann, J., Winnefeld, F., & Plank, J. (2010). Interaction of cement model systems with superplasticizers investigated by atomic force microscopy, zeta potential, and adsorption measurements. *Journal of Colloid and Interface Science*, 347(1), 15–24.
- Florence, N. T., Benoit, M. Z., Jonas, K., Alexandra, T., Désiré, D. D. P., Pierre, K., & Théophile, D. (2014). Antidiabetic and antioxidant effects of *Annona muricata* (Annonaceae), aqueous extract on streptozotocin-induced diabetic rats. *Journal of Ethnopharmacology*, 151(2), 784–790.
- Gajalakshmi, S., Vijayalakshmi, S., & Devi, R. V. (2012). Phytochemical and pharmacological properties of *Annona muricata*: a review. *International Journal of Pharmacy and Pharmaceutical Sciences*, 4(2), 3–6.
- Gavamukulya, Y., Abou-Ellella, F., Wamunyokoli, F., & AEl-Shemy, H. (2014). Phytochemical screening, anti-oxidant activity and in vitro anticancer potential of ethanolic and water leaves extracts of *Annona muricata* (Graviola). *Asian Pacific Journal of Tropical Medicine*, 7, S355–S363.
- Gavamukulya, Y., Maina, E. N., El-Shemy, H. A., Meroka, A. M., Kangogo, G. K., Magoma, G., & Wamunyokoli, F. (2021). *Annona muricata* silver nanoparticles exhibit strong anticancer activities against cervical and prostate adenocarcinomas through regulation of CASP9 and the

CXCL1/CXCR2 genes axis. *Tumor Biology*, 43(1), 37–55.

Gavamukulya, Y., Maina, E. N., Meroka, A. M., El-Shemy, H. A., Magoma, G., & Wamunyokoli, F. (2019). In search of new anticancer drugs: Data for cytotoxic activities of green synthesized silver nanoparticles from ethanolic extracts of fruits and leaves of *Annona muricata* and 5-Fluorouracil against HeLa, PC3 and PNT1A cell lines. *Data in Brief*, 26, 104442.

González-Pedroza, M. G., Argueta-Figueroa, L., García-Contreras, R., Jiménez-Martínez, Y., Martínez-Martínez, E., Navarro-Marchal, S. A., Marchal, J. A., Morales-Luckie, R. A., & Boulaiz, H. (2021). Silver nanoparticles from *Annona muricata* peel and leaf extracts as a potential potent, biocompatible and low cost antitumor tool. *Nanomaterials*, 11(5), 1273.

Hailstone, R. K., DiFrancesco, A. G., Leong, J. G., Allston, T. D., & Reed, K. J. (2009). A study of lattice expansion in CeO₂ nanoparticles by transmission electron microscopy. *The Journal of Physical Chemistry C*, 113(34), 15155–15159.

Haleem, A., Javaid, M., Singh, R. P., Rab, S., & Suman, R. (2023). Applications of Nanotechnology in Medical field. *Global Health Journal*.

Hall, J. B., Dobrovolskaia, M. A., Patri, A. K., & McNeil, S. E. (2007). *Characterization of nanoparticles for therapeutics*.

Hamizah, S., Roslida, A. H., Fezah, O., Tan, K. L., Tor, Y. S., & Tan, C. I. (2012). Chemopreventive potential of *Annona muricata* L leaves on chemically-induced skin papillomagenesis in mice. *Asian Pacific Journal of Cancer Prevention*, 13(6), 2533–2539.

Haro, G., Utami, N. P., & Sitompul, E. (2014). Study of the antibacterial activities of soursop (*Annona muricata* L.) leaves. *International Journal of PharmTech Research*, 6(2), 575–581.

Hasmila, I., Natsir, H., & Soekamto, N. H. (2019). Phytochemical analysis and antioxidant activity of soursop leaf extract

(*Annona muricata* Linn.). *Journal of Physics: Conference Series*, 1341(3), 032027.

He, Z., & Alexandridis, P. (2015). Nanoparticles in ionic liquids: interactions and organization. *Physical Chemistry Chemical Physics*, 17(28), 18238–18261.

Huo, W., Zhang, X., Gan, K., Chen, Y., Xu, J., & Yang, J. (2019). Effect of zeta potential on properties of foamed colloidal suspension. *Journal of the European Ceramic Society*, 39(2–3), 574–583.

Ito, T., Sun, L., Bevan, M. A., & Crooks, R. M. (2004). Comparison of nanoparticle size and electrophoretic mobility measurements using a carbon-nanotube-based coulter counter, dynamic light scattering, transmission electron microscopy, and phase analysis light scattering. *Langmuir*, 20(16), 6940–6945.

Jabir, M. S., Saleh, Y. M., Sulaiman, G. M., Yaseen, N. Y., Sahib, U. I., Dewir, Y. H., Alwahibi, M. S., & Soliman, D. A. (2021). Green synthesis of silver nanoparticles using *Annona muricata* extract as an inducer of apoptosis in cancer cells and inhibitor for NLRP3 inflammasome via enhanced autophagy. *Nanomaterials*, 11(2), 384.

Jain, D., Daima, H. K., Kachhwaha, S., & Kothari, S. L. (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.

Jiang, J., Oberdörster, G., & Biswas, P. (2009). Characterization of size, surface charge, and agglomeration state of nanoparticle dispersions for toxicological studies. *Journal of Nanoparticle Research*, 11, 77–89.

Jindal, A. B. (2017). The effect of particle shape on cellular interaction and drug delivery applications of micro-and nanoparticles. *International Journal of Pharmaceutics*, 532(1), 450–465.

Kaszuba, M., & Connah, M. T. (2006). Protein and nanoparticle characterisation using light scattering techniques. *Particle &*

Particle Systems Characterization, 23(2), 193–196.

Kathad, U., & Gajera, H. P. (2014). Synthesis of copper nanoparticles by two different methods and size comparison. *Int J Pharm Bio Sci*, 5(3), 533–540.

Kumar, A., & Dixit, C. K. (2017). Methods for characterization of nanoparticles. In *Advances in nanomedicine for the delivery of therapeutic nucleic acids* (pp. 43–58). Elsevier.

Kumar, G. V. P., Shruthi, S., Vibha, B., Reddy, B. A. A., Kundu, T. K., & Narayana, C. (2007). Hot spots in Ag core– Au shell nanoparticles potent for surface-enhanced Raman scattering studies of biomolecules. *The Journal of Physical Chemistry C*, 111(11), 4388–4392.

Kumar, P. P. N. V., Pammi, S. V. N., Kollu, P., Satyanarayana, K. V. V., & Shameem, U. (2014). Green synthesis and characterization of silver nanoparticles using Boerhaavia diffusa plant extract and their anti bacterial activity. *Industrial Crops and Products*, 52, 562–566.

Kumarasamyraja, D., & Jeganathan, N. S. (2013). Green synthesis of silver nanoparticles using aqueous extract of acalypha indica and its antimicrobial activity. *Int J Pharm Bio Sci*, 4(3), 469–476.

Liu, J. (2005). Scanning transmission electron microscopy and its application to the study of nanoparticles and nanoparticle systems. *Microscopy*, 54(3), 251–278.

Liu, R., Xu, Y., Zhang, N., Qu, S., Zeng, W., Li, R., & Dai, Z. (2023). Nanotechnology for Enhancing Medical Imaging. *Nanomedicine*, 99.

Lunardi, C. N., Gomes, A. J., Rocha, F. S., De Tommaso, J., & Patience, G. S. (2021). Experimental methods in chemical engineering: Zeta potential. *The Canadian Journal of Chemical Engineering*, 99(3), 627–639.

Lv, Y., He, H., Qi, J., Lu, Y., Zhao, W., Dong, X., & Wu, W. (2018). Visual validation of the measurement of entrapment efficiency of drug nanocarriers.

International Journal of Pharmaceutics, 547(1–2), 395–403.

Mansour, H. H., Elkady, A. A., Elrefaei, A. H., & Hafez, H. F. (2018). *Radioprotective, antioxidant and antitumor efficacy of Annona muricata L. leaf extract*.

Marciano, F. R., Bonetti, L. F., Pessoa, R. S., Marcuzzo, J. S., Massi, M., Santos, L. V., & Trava-Airoldi, V. J. (2008). The improvement of DLC film lifetime using silver nanoparticles for use on space devices. *Diamond and Related Materials*, 17(7–10), 1674–1679.

Marín, R. R. R., Babick, F., & Hillemann, L. (2017). Zeta potential measurements for non-spherical colloidal particles—practical issues of characterisation of interfacial properties of nanoparticles. *Colloids and Surfaces A: Physicochemical and Engineering Aspects*, 532, 516–521.

Merlín-Lucas, V., Ordoñez-Razo, R. M., Calzada, F., Solís, A., García-Hernández, N., Barbosa, E., & Valdés, M. (2021). Antitumor potential of annona muricata linn. An edible and medicinal plant in mexico: In vitro, in vivo, and toxicological studies. *Molecules*, 26(24), 7675.

Mohanraj, V. J., & Chen, Y. (2006). Nanoparticles—a review. *Tropical Journal of Pharmaceutical Research*, 5(1), 561–573.

Mühlfeld, C., Rothen-Rutishauser, B., Vanhecke, D., Blank, F., Gehr, P., & Ochs, M. (2007). Visualization and quantitative analysis of nanoparticles in the respiratory tract by transmission electron microscopy. *Particle and Fibre Toxicology*, 4(1), 1–17.

Muthu, S., & Durairaj, B. (2015). Evaluation of antioxidant and free radical scavenging activity of Annona muricata. *European Journal of Experimental Biology*, 5(3), 39–45.

Nagaonkar, D., Gaikwad, S., & Rai, M. (2015). Catharanthus roseus leaf extract-synthesized chitosan nanoparticles for controlled in vitro release of chloramphenicol and ketoconazole. *Colloid and Polymer Science*, 293, 1465–1473.

- Nakkala, J. R., Mata, R., Gupta, A. K., & Sadras, S. R. (2014). Biological activities of green silver nanoparticles synthesized with *Acorous calamus* rhizome extract. *European Journal of Medicinal Chemistry*, *85*, 784–794.
- Nawwar, M., Ayoub, N., Hussein, S., Hashim, A., El-Sharawy, R., Wende, K., Harms, M., & Lindequist, U. (2012). Flavonol triglycoside and investigation of the antioxidant and cell stimulating activities of *Annona muricata* Linn. *Archives of Pharmacal Research*, *35*, 761–767.
- Peckus, D., Tamulevičius, T., Meškinis, Š., Tamulevičienė, A., Vasiliauskas, A., Ulčinas, O., Gulbinas, V., & Tamulevičius, S. (2017). Linear and nonlinear absorption properties of diamond-like carbon doped with Cu nanoparticles. *Plasmonics*, *12*, 47–58.
- Powers, K. W., Palazuelos, M., Moudgil, B. M., & Roberts, S. M. (2007). Characterization of the size, shape, and state of dispersion of nanoparticles for toxicological studies. *Nanotoxicology*, *1*(1), 42–51.
- Prasad, S. K., Varsha, V., & Devananda, D. (2019). Anti-cancer properties of *Annona muricata* (L.): A Review. *Medicinal Plants-International Journal of Phytomedicines and Related Industries*, *11*(2), 123–134.
- Qi, W. H., & Wang, M. P. (2004). Size and shape dependent melting temperature of metallic nanoparticles. *Materials Chemistry and Physics*, *88*(2–3), 280–284.
- Radoń, A., Łukowiec, D., Kremzer, M., Mikula, J., & Włodarczyk, P. (2018). Electrical conduction mechanism and dielectric properties of spherical shaped Fe₃O₄ nanoparticles synthesized by co-precipitation method. *Materials*, *11*(5), 735.
- Rady, I., Bloch, M. B., Chamcheu, R.-C. N., Banang Mbeumi, S., Anwar, M. R., Mohamed, H., Babatunde, A. S., Kuate, J.-R., Noubissi, F. K., & El Sayed, K. A. (2018). Anticancer properties of graviola (*Annona muricata*): a comprehensive mechanistic review. *Oxidative Medicine and Cellular Longevity*, 2018.
- Rodríguez-Luis, O. E., Hernandez-Delgado, R., Sánchez-Nájera, R. I., Martínez-Castañón, G. A., Niño-Martínez, N., Sánchez Navarro, M. del C., Ruiz, F., & Cabral-Romero, C. (2016). Green synthesis of silver nanoparticles and their bactericidal and antimycotic activities against oral microbes. *Journal of Nanomaterials*, 2016.
- Roduan, M. R. M., Abd Hamid, R., Cheah, Y. K., & Mohtarrudin, N. (2019). Cytotoxicity, antitumor-promoting and antioxidant activities of *Annona muricata* in vitro. *Journal of Herbal Medicine*, *15*, 100219.
- Santos, I. L., Rodrigues, A. M. da C., Amante, E. R., & Silva, L. H. M. da. (2023). Soursop (*Annona muricata*) Properties and Perspectives for Integral Valorization. *Foods*, *12*(7), 1448.
- Schaffer, B., Hohenester, U., Trügler, A., & Hofer, F. (2009). High-resolution surface plasmon imaging of gold nanoparticles by energy-filtered transmission electron microscopy. *Physical Review B*, *79*(4), 041401.
- Schultz, N., Metreveli, G., Franzreb, M., Frimmel, F. H., & Syldatk, C. (2008). Zeta potential measurement as a diagnostic tool in enzyme immobilisation. *Colloids and Surfaces B: Biointerfaces*, *66*(1), 39–44.
- Silva, R. M. da, Silva, I. de M. M. da, Estevinho, M. M., & Estevinho, L. M. (2021). Anti-bacterial activity of *Annona muricata* Linnaeus extracts: a systematic review. *Food Science and Technology*, *42*.
- Singh, A., Mittal, A., & Benjakul, S. (2021). Chitosan nanoparticles: Preparation, food applications and health benefits. *Sci. Asia*, *47*(2021), 1–10.
- Sis, H., & Birinci, M. (2009). Effect of nonionic and ionic surfactants on zeta potential and dispersion properties of carbon black powders. *Colloids and Surfaces A: Physicochemical and Engineering Aspects*, *341*(1–3), 60–67.
- Sizochenko, N., Rasulev, B., Gajewicz, A., Kuz'min, V., Puzyn, T., & Leszczynski, J. (2014). From basic physics to mechanisms of toxicity: The “liquid drop” approach

applied to develop predictive classification models for toxicity of metal oxide nanoparticles. *Nanoscale*, 6(22), 13986–13993.

Song, X., Zhao, Y., Hou, S., Xu, F., Zhao, R., He, J., Cai, Z., Li, Y., & Chen, Q. (2008). Dual agents loaded PLGA nanoparticles: systematic study of particle size and drug entrapment efficiency. *European Journal of Pharmaceutics and Biopharmaceutics*, 69(2), 445–453.

Sridhar, R., & Ramakrishna, S. (2013). Electrospayed nanoparticles for drug delivery and pharmaceutical applications. *Biomatter*, 3(3), e24281.

Streich, C., Koenen, S., Lelle, M., Peneva, K., & Barcikowski, S. (2015). Influence of ligands in metal nanoparticle electrophoresis for the fabrication of biofunctional coatings. *Applied Surface Science*, 348, 92–99.

Sun, Q., Cai, X., Li, J., Zheng, M., Chen, Z., & Yu, C.-P. (2014). Green synthesis of silver nanoparticles using tea leaf extract and evaluation of their stability and antibacterial activity. *Colloids and Surfaces A: Physicochemical and Engineering Aspects*, 444, 226–231.

Sundrarajan, M., Bama, K., Bhavani, M., Jegatheeswaran, S., Ambika, S., Sangili, A., Nithya, P., & Sumathi, R. (2017). Obtaining titanium dioxide nanoparticles with spherical shape and antimicrobial properties using *M. citrifolia* leaves extract by hydrothermal method. *Journal of Photochemistry and Photobiology B: Biology*, 171, 117–124.

Sylvestre, J.-P., Poulin, S., Kabashin, A. V., Sacher, E., Meunier, M., & Luong, J. H. T. (2004). Surface chemistry of gold nanoparticles produced by laser ablation in aqueous media. *The Journal of Physical Chemistry B*, 108(43), 16864–16869.

Titus, D., Samuel, E. J. J., & Roopan, S. M. (2019). Nanoparticle characterization techniques. In *Green synthesis, characterization and applications of nanoparticles* (pp. 303–319). Elsevier.

Verma, M., Ozer, I., Xie, W., Gallagher, R., Teixeira, A., & Choy, M. (2023). The

landscape for lipid-nanoparticle-based genomic medicines. *Nat Rev Drug Discov*.

Vernet-Crua, A., Cruz, D. M., Mostafavi, E., Truong, L. B., Barabadi, H., Cholula-Díaz, J. L., Guisbiers, G., & Webster, T. J. (2023). Green-synthesized metallic nanoparticles for antimicrobial applications. In *Nanomedicine* (pp. 297–338). Elsevier.

Viera, G. H. F., Mourão, J. A., Ângelo, Â. M., Costa, R. A., & Vieira, R. H. S. dos F. (2010). Antibacterial effect (in vitro) of *Moringa oleifera* and *Annona muricata* against Gram positive and Gram negative bacteria. *Revista Do Instituto de Medicina Tropical de São Paulo*, 52, 129–132.

Vijayameena, C., Subhashini, G., Loganayagi, M., & Ramesh, B. (2013). Original Research Article Phytochemical screening and assessment of antibacterial activity for the bioactive compounds in *Annona muricata*. *Int. J. Curr. Microbiol. Appl. Sci*, 2, 1–8.

Vijayaraghavan, K., Nalini, S. P. K., Prakash, N. U., & Madhankumar, D. (2012). One step green synthesis of silver nano/microparticles using extracts of *Trachyspermum ammi* and *Papaver somniferum*. *Colloids and Surfaces B: Biointerfaces*, 94, 114–117.

Were, L. M., Bruce, B. D., Davidson, P. M., & Weiss, J. (2003). Size, stability, and entrapment efficiency of phospholipid nanocapsules containing polypeptide antimicrobials. *Journal of Agricultural and Food Chemistry*, 51(27), 8073–8079.

Wu, Y., Yang, W., Wang, C., Hu, J., & Fu, S. (2005). Chitosan nanoparticles as a novel delivery system for ammonium glycyrrhizinate. *International Journal of Pharmaceutics*, 295(1–2), 235–245.

Yli-Juuti, T., Barsanti, K., Hildebrandt Ruiz, L., Kieloaho, A.-J., Makkonen, U., Petäjä, T., Ruuskanen, T., Kulmala, M., & Riipinen, I. (2013). Model for acid-base chemistry in nanoparticle growth (MABNAG). *Atmospheric Chemistry and Physics*, 13(24), 12507–12524.

Zhang, Y., Yang, M., Portney, N. G., Cui, D., Budak, G., Ozbay, E., Ozkan, M., &

Ozkan, C. S. (2008). Zeta potential: a surface electrical characteristic to probe the interaction of nanoparticles with normal and cancer human breast epithelial cells. *Biomedical Microdevices*, *10*, 321–328.

Zimmerman, N., Pollitt, K. J. G., Jeong, C.-H., Wang, J. M., Jung, T., Cooper, J. M., Wallace, J. S., & Evans, G. J. (2014). Comparison of three nanoparticle sizing instruments: The influence of particle morphology. *Atmospheric Environment*, *86*, 140–147.

Zubaidi, S. N., Mohd Nani, H., Ahmad Kamal, M. S., Abdul Qayyum, T., Maarof,

S., Afzan, A., Mohmad Misnan, N., Hamezah, H. S., Baharum, S. N., & Mediani, A. (2023). *Annona muricata*: Comprehensive Review on the Ethnomedicinal, Phytochemistry, and Pharmacological Aspects Focusing on Antidiabetic Properties. *Life*, *13*(2), 353.