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Research Article

Fabrication of Silver Nanoparticles Using *Fimbristylis miliacea*: A Cheap and Effective Tool against Invasive Mosquito Vector, *Aedes albopictus*

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Mosquitoes are the most critical group of insects in the context of public health, since they transmit key parasites and pathogens, causing millions of deaths annually. *Aedes albopictus* is an important invasive mosquito vector of dengue fever across urban and semiurban areas of India. In this study, we biofabricated silver nanoparticles (AgNPs) using the *Fimbristylis miliacea* aqueous leaf extract (*Fm*-ALE) as reducing and stabilizing agent. The synthesis of *Fm*-AgNPs was confirmed by the excitation of surface plasmon resonance and orange-brown color using ultraviolet-visible (UV-vis) spectrophotometry. High-resolution scanning electron microscopic (HR-SEM) and transmission electron microscopic (TEM) showed the clustered (size 0.5 μm) and quasi-spherical structures of *Fm*-AgNPs. The formation of AgNPs has been characterized by X-ray diffraction (XRD) spectroscopy. Fourier transform infrared (FTIR) spectroscopy investigated the identity of secondary metabolites, which may act as *Fm*-AgNP capping agents. These results propose that AgNPs synthesized provided from those *Fm*-ALE have the high sources to be improved into the most suitable materials useful for protecting and killing the invasive mosquito vector, *Ae. albopictus* populations. The acute toxicity of *Fm*-ALE synthesized Ag NPs, and a combined treatment testing blends of mosquito vector was evaluated against I, II, III, and IV instar larva's (ILs) of *Ae. albopictus*. The LC_{50} values of *Fm*-ALE (174.39 ppm I-ILs, 214.40 ppm II-ILs, 232.38 ppm III-ILs, and 251.62 ppm IV-ILs) and *Fm*-AgNPs synthesized were 23.78 ppm I-ILs; 27.88 ppm II-ILs; 31.47 ppm III-ILs; 36.68 ppm IV-ILs, respectively. Likewise, *Fm*-AgNP synthesis was more toxic than ALE in the invasive mosquito vector and recorded from UV-vis spectrum, FTIR, TEM, and XRD analysis. These results propose that AgNPs synthesized provided from those *Fm*-ALE have the high sources to be improved into the most suitable materials useful for protecting and killing the invasive mosquito vector, *Ae. albopictus* populations.

1. Introduction

Mosquito and mosquito-borne diseases (MBDs) are successfully spreading throughout the entire world, with an inordinate impact on adolescents and children, which are more important responsible for significant global morbidity and mortality [1]. The insect-borne disease is at risk of developing worldwide following globalization and the enlargement of travel and trade from areas colonized by vectors. *Aedes albopictus* (*Ae. albopictus* and tiger mosquito) is a belligerent, and aggressive, is arising entirely global as a population health danger following its basic process in current Chikungunya virus (CHIKV) and dengue virus (DENV) outbreaks, and is one of the most invasive animal species for one hundred in the world, and in less than 30 years, it has developed across the five continents, colonizing abundant lands [2–4]. Hence, the very quick enlargement of species was caused by the worldwide profession of tires and the ability to release maximum eggs that diapauses and resists almost cold winters of temperate areas [5–8]. Commonly, public alertness and general knowledge of *Aedes*-transmitted diseases may improve the likelihood of patients being discussed with a doctor. Its symptoms are agreeable to an arboviral disease improved soon after coming again from a country, where the disease is endemic [9, 10].

In the current year, bioactivity-way for a fabrication process of metal-NPs has been recommended as sources of natural, ecological-friendly replacement to classic physiochemical methods [11]. Specifically, AgNP synthesis is developing as multiple intention agents, because the reason to their biosynthesis is very easy and inexpensive, protection permanent over time, and most effective on arboviruses and human pathogenic bacteria [12, 13]. Population entering into any region mostly dengue, yellow fever, and disadvantages exist may control utilizing medicinal plant-derived repellents role [14–16].

Fimbristylis miliacea (*Fm* and medicinal herbs) sanctified with differential medicinal components are a potential of myriad compounds advantages for plant biologists, zoologist researchers, and human being population around the world, working in pursuit to detect source lead compounds from natural medicinal plant sources. It is all important, cell processes are activated by the potential composed by the metabolism of foods mainly by oxidation reactive, and essential energy with radicals like peroxid (ROS), superoxide and hydroxyl (OH), and imparts oxidative stress on the cells (IOSC). The signification research and recognition of benign phytocompounds from the *Fm*-medicinal plant thus become very significant in current years, and its species of variety plant has been pharmaceutical sources like antioxidant, antinociceptive, anti-inflammatory, antipyretic, antimicrobial, cytotoxic, hepatoprotective, and antidiabetic effects [17]. Since *Fimbristylis miliacea* is an easily available grass type with abundant phytoconstituent in this exploration, we stated a pattern to AgNP synthesis using the *Fm*-ALE, and it is a cheap and eco-friendly fabric process as a highly reducing and stabilizing agent. Here, we demonstrate a convenient eco-friendly green synthesis route for preparing AgNPs using *Fm*-ALE, and they are subjected to subsequent analytical and biological characterization for investigating antibacterial and larvicidal abilities for controlling human disease-transmitting invasive mosquito vector, *Ae. albopictus* as shown in Figure 1.

2. Materials and Methods

2.1. Collection and Preparation. Silver nitrate (Ag^+) was procured from Lakshmi Scientific Chemicals Ltd., Pondicherry in India. *Fm*-fresh leaf was gathered from the Western Ghats Forest (WGF) (10°36′N to 10°14′N latitude and 76°49′E to 76°77′E longitude), Erode District, India. After, the glassware was cleaned thoroughly in acid and washed off with triple-distilled water. The new identification was proved at the Department of Botany, Annamalai University, and specimens were numbered (Authentication Number SVC/BOT/131). Leaves were retained in our department laboratory and ready for use upon request. *Fm*-fresh leaves were dried in the shade, and ALE was produced by blending 30 g of air-dried leaves with 300 mL of water, with stable stirring on a magnetic stirrer. Then, air-dried ALE was left for 3 h and separated by Whatman No. 1 filter paper. It was saved in an amber-colored airtight bottle at 10°C temperature until research.

2.2. Target Medical Pest. Larvae stage, such as I, II, III, and IV-ILs of medical important pest *Ae. albopictus*, was taken from stagnant fresh water at around from side by side from Poompuhar College to Sirkazhi down (11°14′N to 11°23′N latitude and 79°81′E to 79°73′E longitude), Nagapattinam District in India. *Ae. albopictus* is identified by ICMR, Madurai in India, and carried to the laboratory for continuous rearing. Dog biscuits+yeast powder mixed feed was (3 : 1 ratio) utilized to culture the mosquito under (27 ± 2°C, 75 ± 5% relative humidity, with a photoperiod of 12 L:12D) possible environment.

2.3. Larval Bioassay. The protocol was accepted for larval bioassay and concentration range from 10 to 250 μg/mL. The necessary *Fm*-ALE and AgNPs were mixed (249 mL) in triple-distilled water. After, each research was assayed against 3^{rd} instars of twenty-five larvae, repeated 5 times. The lethal concentration (50/90) and other statistical data were calculated by using Probit analysis [19].

2.4. Primary Chemical Analysis. The PCA is followed by the methods [20, 21]. We screened the bioactive chemical constituents (CCs) detecting the presence of secondary metabolites such as alkaloids, flavonoids, saponins, steroids, tannins, terpenoids, tri-terpenoids, anthraquinones, amino acid, phenol, glycosides, carbohydrate, protein, and phytosterols in the *Fm*-ALE.

2.5. Silver Nanoparticle (AgNP) Synthesis. $AgNO_3$ 90 mL Mm added with *Fm*-ALE 10 mL was made in 250 mL conical flasks for decreases into Ag^+ ions. After that, the mixture was kept for 1 h at 27 ± 3°C in the laboratory conditions. Wherein, the first initial stage was detection of *Fm*-AgNPs containing dark brown color change in the mixture (ALE

Figure 1: Schematic illustration of green synthesis of AgNPs using *Fm*-ALE for effective invasive vector mosquito control applications.

Table 1: Larvicidal activity of *Fm*-aqueous extract against *Ae. albopictus*.

Stages	LC_{50} (mg/L)	95% confidence limits		LC_{90} (mg/L)	Slope	Regression	χ^2
		LCL	UCL				
Instars 1st	174.39	164.66	184.78	271.29	3.642772	$y = 3.642 \times -2.669$	4.721
Instars 2nd	214.40	201.40	230.41	332.01	2.728162	$y = 2.728 \times -1.352$	3.516
Instars 3rd	232.38	217.07	252.56	357.69	2.448937	$y = 2.448 \times -1.084$	2.508
Instars 4th	251.62	234.66	275.24	371.21	1.648359	$y = 1.648 \times -1.467$	1.911

Values represent the mean of five replications. Mortality of the after 24 h of exposure period LC50 = lethal concentration brings out 50% mortality and LC90 = lethal concentration brings out 90% mortality. LCL = lower confidence limit; UCL = upper confidence limit; χ^2 = chi-square. [a]Significant at *p*⊠0.05.

Table 2: Larvicidal activity of *Fm*-AgNPs synthesized against *Ae. albopictus*.

Stages	LC_{50} (mg/L)	95% confidence limits		LC_{90} (mg/L)	Slope	Regression	χ^2
		LCL	UCL				
Instars 1st	23.78	21.56	25.83	44.40	4.628878	$y = 4.628 \times -0.048$	4.048
Instars 2nd	27.88	25.48	30.20	52.35	3.134263	$y = 3.134 \times +0.989$	0.490
Instars 3rd	31.47	29.10	33.91	56.61	2.447822	$y = 2.447 \times +0.935$	0.422
Instars 4th	36.68	34.32	39.32	60.95	1.462074	$y = 1.462 \times +0.659$	0.326

Values represent the mean of five replications. Mortality of the after 24 h of exposure period LC50 = lethal concentration brings out 50% mortality and LC90 = lethal concentration brings out 90% mortality. LCL = lower confidence limit; UCL = upper confidence limit; χ^2 = chi-square. [a]Significant at *p*⊠0.05.

+$AgNO_3$). The entire reaction is granted in a dark place to avoid photoactivation. For the purification activity, acquired *Fm*-AgNPs allowed to ultracentrifugation above 6,000 rpm for 20 min. After the centrifugation, the supernatant was rejected, and the pellet was cautiously diluted with triple-distilled H_2O [22]. Further, mixed material was placed in the laboratory, labeled, and stocked for further analysis.

2.6. Characterization of AgNPs. After the Ag^+ ion solution was intently watched carefully by using UV-vis spectroscopy, purified *Fm*-AgNPs (biomolecules) in the FTIR spectroscopy, and obtained to dry at 60°C, the air-dried powder was inflicted to XRD spectroscopy to identify their exact structure and material [23]. *Fm*-ALE mediated synthesis of AgNP process such as UV, FT-IR, TEM, and XRD analyses.

2.7. Data Analysis. The larvicidal activity (%) data of invasion mosquito, *Ae. albopictus* larvae, were subjected to different statistical baggage, LC_{50}/LC_{90}, LCL, UCL, regression, chi-square, slope, etc. All the values were calculated by (IBM) SPSS statistical new version 20.0 version.

3. Results and Discussion

3.1. Larvae Bioassay of Fm-ALE Extract and AgNP Synthesis. The larval bioassay of *Fm*-ALE treatment was tested against I, II, III, and IV-ILs of important invasive mosquitoes and is shown in Table 1. The LC_{50}/LC_{90} values of *Fm*-ALE appeared to be effective against I-ILs (174.39/271.29 μg/mL), II-ILs (214.40/332.01 μg/mL), III-ILs (232.38/357.69 μg/mL), and IV-ILs (251.62/371.21 μg/mL) invasive

Figure 2: Phytochemical test present in the *Fm*-ALE.

mosquito larvae, *Ae. albopictus*. Table 2 shows the treatment of *Fm*-AgNPs synthesized and had the following LC_{50} and LC_{90} values; ILs had LC_{50}/LC_{90} values of 23.78/44.40 μg/mL; II-ILs had values of 27.88/52.35 μg/mL; III-ILs had values of 31.47/56.61 μg/mL; IV-ILs had values of 36.68/60.95 μg/mL. A control contained nil mortality in the same time assay, and the χ^2 value was significant at $p < 0.05$ level.

3.2. Preliminary Analysis. Fm-ALE was screened for the presence of major phytochemicals (MPCs) such as alkaloids (Figure 2(a)), flavonoids (Figure 2(b)), saponins (Figure 2(c)), steroids (Figure 2(d)), tannins (Figure 2(e)), terpenoids (Figure 2(f)), tri-terpenoids (Figure 2(g)), phenol (Figure 2(h)), carbohydrate (Figure 2(i)), protein (Figure 2(j)), phytosterols (Figure 2(k)), and all for test tube (Figure 2(l)) except anthraquinones, amino acid, and glycosides responsible of mosquitocidal activity (Table 3).

3.3. UV, XRD, SEM, and TEM Analysis of Fm-AgNPs. The *Fm*-ALE of AgNP ($AgNO_3$+ALE) composite was indicated and confirmed via the orange-brown color change (Figure 3). Figure 4 shows the $AgNO_3$ solution turned brown within 2 min with the addition of *Fm*-ALE and the control $AgNO_3$ solution (without ALE) showed no change of any color. There was no absorption peak in the UV-vis spectrum, which can reduce Ag^+ ions and produce AgNPs of incubation at the highest p^H (Figures 5 and 6). The preparation of the AgNPs synthesized from *Fm*-ALE was

Table 3: Phytochemical screening of *Fm*-ALE.

S. no.	Phytoconstituents	Aqueous
1	Alkaloids	+++
2	Flavonoids	+
3	Saponins	++
4	Steroids	—
5	Tannins	+
6	Terpenoids	+++
7	Tri-terpenoids	++
8	Anthraquinones	—
9	Amino acid	—
10	Phenol	+
11	Glycosides	—
12	Carbohydrate	++
13	Protein	+
14	Phytosteroids	++

+++: strongly positive phytochemical group; ++: positive phytochemical group; +: trace phytochemical group; —: absence of phytochemical group.

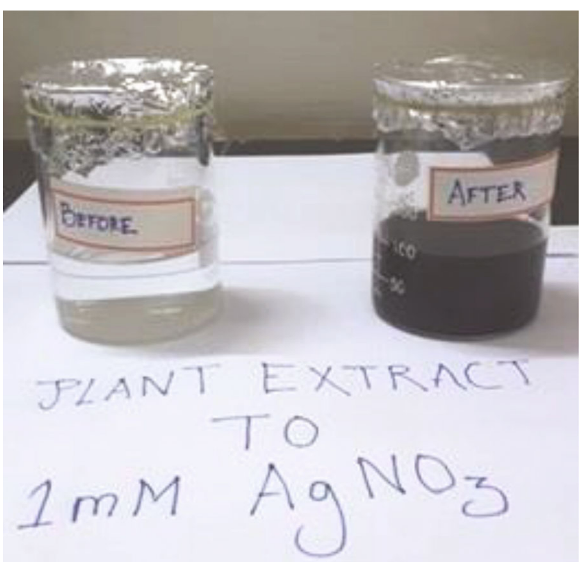

Figure 3: *Fm*-ALE-mediated synthesis of AgNPs process.

evaluated through a spectrophotometer in a range of wavelengths from 200 to 1200 nm.

Figure 7 shows the high-resolution scanning electron microscopic (HR-SEM) and transmission electron microscopic (TEM) analysis supplies the information about the sizes, and morphology of AgNPs was obtained ranging from 41 to 60 nm. The morphology of the AgNPs is quasispherical as seen in the SEM image. There was cluster formation of an average size of 0.5 μm due to evaporation of suspended liquid causing the particles to cluster around the outer edge to form quasispherical structures. Figure 5 displays the formation of *Fm*-AgNPs characterized using XRD analysis. The six well defined characteristic diffraction peaks at 27.8°, 32.2°, 46.1°, 54.8°, 57.5°, and 76.7° correspond to the face-centered cubic crystal-shaped structure of metallic silver. The interplanar spacing (dl) values were obtained as 3.196, 2.769, 1.963, 1.671, 1.600, and 1.240°A using Bragg's formula from the XRD pattern and were further corroborated crystalline nature of *Fm*-AgNPs. The lattice constant was calculated was 3.196°A which was well-matching with standard data (JCPDS PDF04-0783). The peak broadening was observed due to the formation of nanoparticles. The peak intensity of the (corresponding 111 to 311) plane indicated the purity of AgNPs. It is significant to record that the intensity ratio between 36° and 53° peaks is lesser than the value of the standard (0.48 versus 0.5).

3.4. FT-IR Analysis. Figure 6 shows the that FTIR spectrum showed major peaks at 3571.77, 3423.39, 1986.17, 1055.96, 1038.12, 981.81, 726.78, 600.26, 567.83, 495.03, 454.77, and 428.76 cm^{-1}. Above the peak value, they correspond to functional groups like the alcohol group in lower (C=O band stretch, alkoxy, 3571.77 cm^{-1}), alcohol and phenol group in strong and broad (O-H stretch, H-bonded, 3423.39 cm^{-1}), aromatic group in strong (C-H bend stretch, 1986.17 cm^{-1}), aliphatic amines group in medium (C-N stretch, 1055.96, 1038.12 cm^{-1}), alkenes group in strong (C-H rock, 981.81, 726.78 cm^{-1}), and alkyl halides group in medium (C-Br stretch, 600.26, 567.83 cm^{-1}).

4. Discussion

According to the latest World malaria report, there were 241 million cases of malaria in 2020 compared to 227 million cases in 2019. The estimated number of malaria deaths stood at 627 000 in 2020—an increase of 69 000 deaths over the previous year [22]. Mosquitocide resistance requires the growth of approaches for prolonging the use of more effective vector control compounds. The use of mixed differential insecticides and phytochemicals is one such approach that may be acceptable for mosquito control [23, 24]. Phytochemical compounds (PCMs) may act as the most suitable alternative to synthetic insect-killing activity in the future and are readily obtained in many research areas in the global because are comparably safe and inexpensive. Medicinal plants not only contain some AgNPs but are proven to be nonlethal for aquatic life stages of the mosquito vector, *Ae. albopictus*, and are easily biodegradable in the environment. In the current research, *Fm*-AgNPs synthesized were more toxic than ALE in the invasive mosquito vector and resulted from UV-vis spectrum, FTIR, TEM, and XRD analyses. Similarly, UV-vis spectrum, FTIR, TEM, and EDX analyses from *Helitropiumindicum*-AgNPs and were highly larval killing activity (LC_{50}/LC_{90} values of 72.72/126.86 μg/mL) [25]. Almost, spherical and cubic NPs are the very most general materials of AgNP synthesis green potentials [26]. Also, previously researched, the *Sargassum muticum*-derived synthesized AgNPs signify that they were very strong scattered, with AgNPs a size range of 43-79 nm [27]. This research is in agreement with a previous result; SEM focused that the *Hygrophila auriculata*-AgNPs synthesized were an almost spherical or cubic shape, with a mean size ranging from 9.0 to 30 nm and XRD acute peaks at 2Θ values of 38.13 (111),

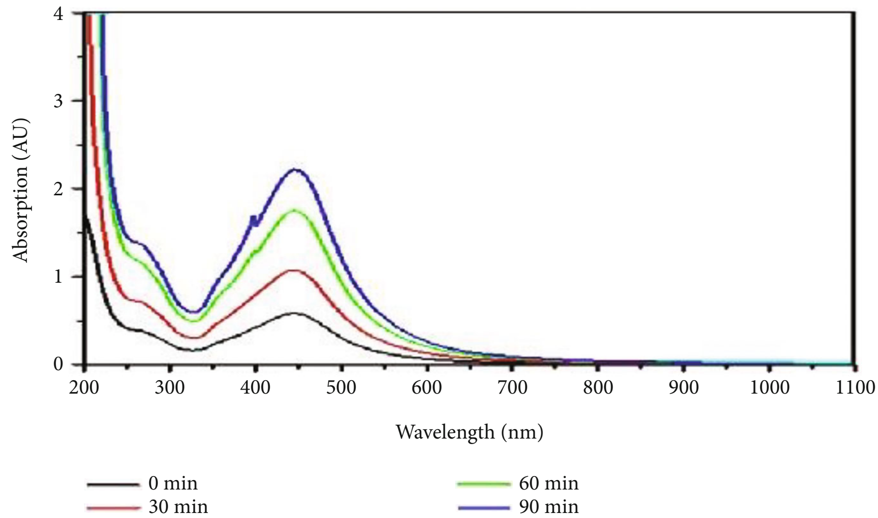

Figure 4: UV-vis spectral of AgNO3 with *Fm*-ALE.

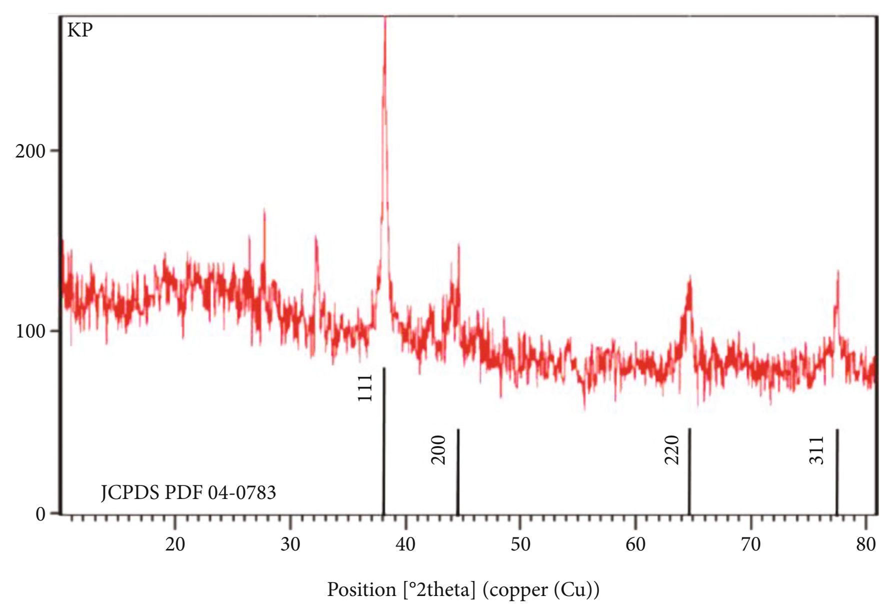

Figure 5: X-ray diffraction (XRD) analysis of AgNPs synthesized using *Fm*-ALE.

44.31 (200), 64.44 (220), 77.37 (311) planes of the cubic face-centered Ag [28]. FTIR analysis of the purified *Chomeliaasiatica*-AgNPs, and the bands due to hydrogen-bonded (O-H stretch, 3222.44 cm^{-1}), alkenes (C-H stretch, 2922.23 cm^{-1} and 2853.35 cm^{-1}), nitriles (-C≡N (triple bond) stretch, 2209.12 cm^{-1}), aromatics (C-C stretch, 1593.43 cm^{-1}), aliphatic amines (C-N stretch, 1240.74 cm^{-1}), alkyl halides (C-H stretch, 1167.81 cm^{-1}), alcohol (C-O stretch, 1102.98 cm^{-1}), and alkyl halides (C-C1 stretch, 833.39 cm^{-1}) [24].

The present investigation shows that the LC_{50}/LC_{90} values of *Fm*-ALE appeared to be effective against 1st instars (174.39/271.29 μg/mL), 2nd instars (214.40/332.01 μg/mL), 3rd instars (232.38/357.69 μg/mL), and 4th instars (251.62/371.21 μg/mL) invasive mosquito larvae, *Ae. albopictus*. The detection of the current investigation corresponds with some of the next other previous research, the LC_{50}/LC_{90} values of *Polygonum hydropiper*-oil were 194.63/199.65 ppm, and confertifolin was 2.02/3.16 ppm against the 2nd and 4th instars larvae of tiger mosquito [29]. *Fm*-AgNPs synthesized had the following: 1st instars larvae had LC_{50}/LC_{90} values of 23.78/44.40 μg/mL; 2nd instars larvae had values of 27.88/52.35 μg/mL; 3rd instars larvae had values of 31.47/56.61 μg/mL; and 4th instars larvae had values of 36.68/60.95 μg/mL. Similarly, several previous reports supported the mosquito-killing activity (larval, eggs, and adults) potential of different indigenous medicinal plant-aqueous extract and AgNPs against important vector mosquitoes (IVMs) [26, 30–34]. Particularly, a biochemical method has been used to constant AgNPs synthesized that was researched opposite to *Ae. aegypti* larval killing activity

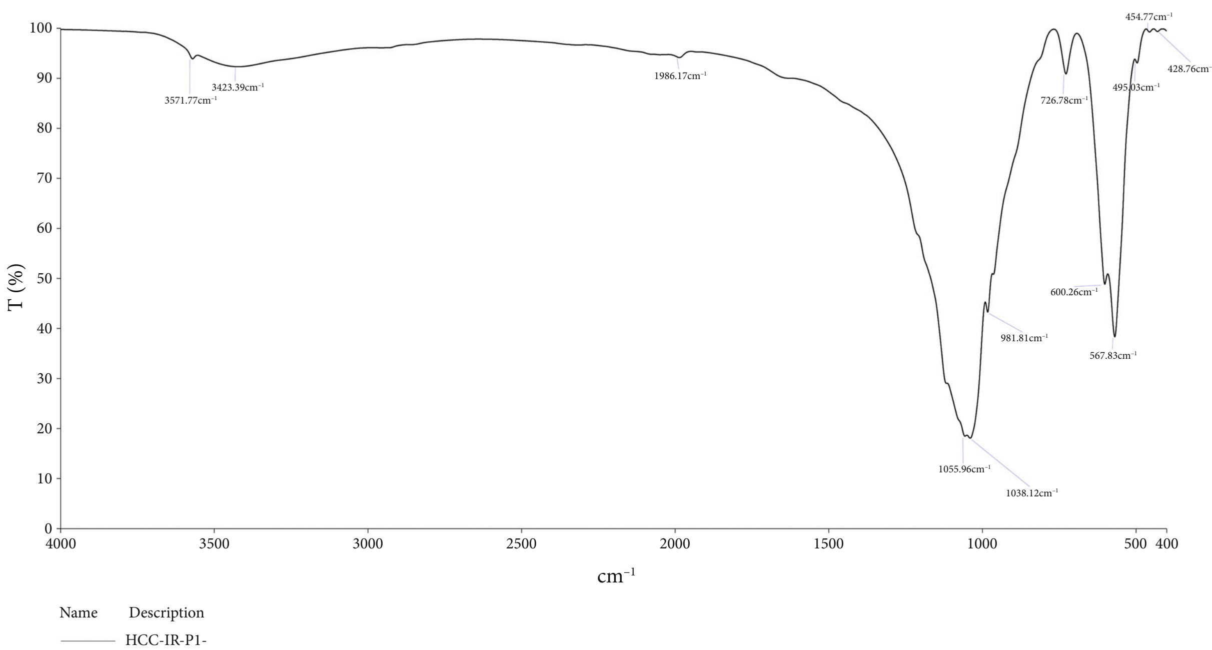

Figure 6: FTIR spectrum of *Fm*-AgNP synthesis.

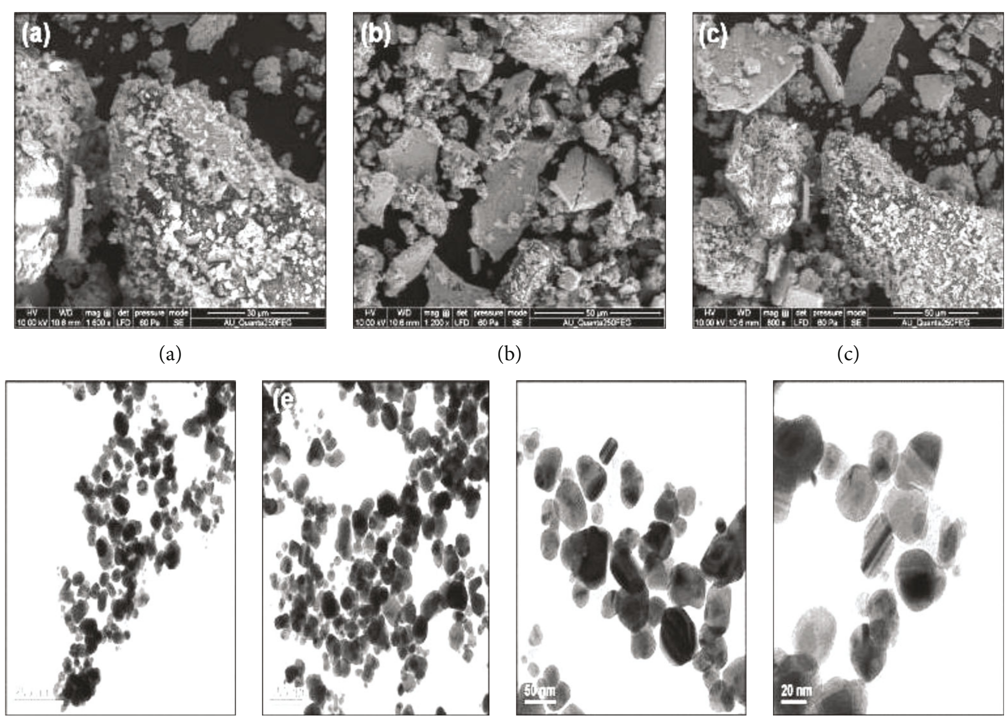

Figure 7: SEM (a–c) and TEM (d–g) images of spherical AgNPs synthesized using *Fm*-ALE.

[35]. Ginger was more effective with the lowest LC_{50} values of 1st instar 7 ppm, 2nd instar 23 ppm, 3rd instar 33 ppm, and 4th instar 35 ppm, after 8 and 16 h against dengue vector, *Ae. albopictus* [36].

The *Bryopsis pennata*-chloroform extract exhibited strong larval killing activity (LC_{50} value was 250.5 μg/mL) against the tiger mosquito vector, *Ae. albopictus* [37]. *Cochliobolus lunatus*-AgNP synthesis of larval killing activity was tested and LC_{50}/LC_{90} values of *Ae. aegypti* 2nd instars 1.29/3.08 ppm, 3rd 1.48/3.33 ppm, and 4th instars 1.58/3.41 ppm [38]. Similarly, larval death rates were recorded from *Jussiaea repens*-LEE, and MPC has larvicidal activity with an LC_{50}/LC_{90} value of *Ae. albopictus* that was 118.3/229.9 μg/mL [39]. *Eucalyptus camaldulensis*- and *Eucalyptus* *urophylla*-EO noticed 60/100% larval death rates at100 μg/mL tested against invasive mosquito, *Ae. Albopictus* [7, 40].

5. Conclusion

In conclusion, the green plants showed that the environmentally benign and revived source of *Fm*-ALE is utilized as an effective lowering agent AgNP synthesis. *Fimbristylis miliacea* is an easily available grass plant with immense medical value utilized for its insect repellant activity. Following this, our reports recommend that *Fm*-plant can be used for the development of insect repellent pesticides in the future prospect. This research was aimed at defining key aspects, such as the nature of the specific molecules responsible for the insecticidal effect observed. Further, the promising effects showed these chemicals in the environment and population health and the field logistics required for utilize of such a local insecticide. The instability in the bioefficacy of medicinal plants tested against mosquito vectors is generally known by variations in the quality and quantity of active compounds and AgNPs. We convey advice that these *Fm*-ALE and AgNPs should be studied in the field for the confirmed potential control of invasive mosquito, *Ae. albopictus*.

Data Availability

The data used to support the findings of this study are included in the article.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

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