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Green synthesis of silver nanoparticles using *Manihot esculenta* leaves against *Aedes aegypti* and *Culex quinquefasciatus*

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Abstract To investigate the silver nanoparticles synthesis using leaves aqueous extract of *Manihot esculenta* against two important mosquito species, *Aedes aegypti* and *Culex quinquefasciatus*. The synthesized silver nanoparticles were monitored by UV–vis spectrophotometer and further characterized by XRD, FESEM and HRTEM. Aqueous extract of *M. esculenta* appeared to be effective against *A. aegypti* (LC_{50} = 66.14 mg/mL; LC_{90} = 172.41 mg/mL) and *C. quinquefasciatus* (LC_{50} = 61.60 mg/mL; LC_{90} = 184.80 mg/mL). The aqueous silver nitrate 1 Mm solution tested was third instar of *A. aegypti* and *C. quinquefasciatus* (LC_{50} = 76.96 mg/mL; LC_{90} = 230.88 mg/mL and LC_{50} = 84.06 mg/L; LC_{90} = 252.78 mg/mL). The 0.5 mM synthesized Ag PNs against *A. aegypti* (LC_{50} = 4.53 mg/mL; LC_{90} = 13.59 mg/mL); *C. quinquefasciatus* (LC_{50} = 3.46 mg/mL; LC_{90} = 10.38 mg/mL). The 1 mM silver solution synthesized Ag NPs tested were *A. aegypti* (LC_{50} = 3.08 mg/mL; LC_{90} = 9.84 mg/mL) *C. quinquefasciatus* (LC_{50} = 3.21 mg/mL; LC_{90} = 11.24 mg/mL). The control showed nil mortality in the concurrent assay. This is a perfect ecological and inexpensive approach for the control of mosquito larvae.

Introduction

Mosquitoes are one of the best-known vectors of more than a few diseases such as zika, dengue fever and Japanese encephalitis. These diseases are endemic to mainly muggy countries causing millions of deaths. Larviciding is a doing well way of sinking mosquito densities in their propagation places earlier than they appear into adults (Govindarajan et al., 2014).

Insecticide applications, although highly efficacious against the target vector species control, are facing a threat due to the enlargement of resistance to chemical insecticides resulting in rebounding vectorial capability (Mehdi et al., 2012). Green synthesis of nano particle from plant extract may be a suitable alternative vector control approach in this regard (Makarov et al., 2014).

Nanotechnology concerns with the development of tentative processes for the synthesis of nanoparticles of different sizes, shapes and controlled disparity (Ahmed et al., 2016). It

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offers numerous benefits of eco-friendliness and compatibility for pharmaceutical and biomedical applications as they do not use deadly chemicals in the synthesis protocols (Bhosale et al., 2014).

Manihot esculenta (Crantz) is an economic crop of India. Cassava which has been reported to have antimicrobial, antimalarial and leishmanicidal activities (Melo et al., 2009; Punthanara et al., 2009). The aim of this study was to investigate the silver nanoparticles synthesis using leaves aqueous extract of *M. esculenta* against two important mosquito species, *A. aegypti* and *C. quinquefasciatus*.

Materials and methods

Collection of plant sample

M. esculenta leaves were collected from Karadimalai, Vellore district and an identity authenticity plant taxonomists.

Synthesis of Ag NPs

Aqueous solutions of silver nitrate ($AgNO_3$) (Sigma-Aldrich, Mexico) were prepared (0.5 mM and 1 mM) with distilled water. A known volume 20 mL of leaf extract was added, drop by drop, to 80 mL of 0.5 mM and 1 mM aqueous solution of silver, shaking continuously and gauged to 100 mL using distilled water, observing a brownish color, indicating the formation of Ag NPs. The synthesized AgNPs were characterized UV, XRD, FE-SEM and HR-TEM analysis.

Rearing mosquito larvae

Mosquito *A. aegypti* larvae were collected, maintained and reared in the laboratory for larvicidal bioassay for Kamaraj et al. (2008) method, minor modifications.

Larvicidal bioassay

One gram of aqueous extract was first dissolved in 100 mL of distilled water (stock solution). The larvicidal activity was assessed following WHO (1996) as per the method of Rahuman et al. (2000), minor modification.

Data analysis

The larvae mortality data were subjected to probit analysis by Reddy et al. (1992).

Results

Characterization of the synthesized nanoparticles

UV–vis spectrum analysis

Initially the aqueous extract was light green and upon providing the silver salt, it turned light brown color to dark brown color. The presence of nanoparticles was confirmed by obtaining a spectrum in UV-vis range of 200–800 nm. The 0.5 mM peak at 422 nm was obtained due to the SPR of silver nanoparticles. The highest values were reached with 1 mM $AgNO_3$ the system became stable after 24 min and band of 426 nm.

XRD analysis

The XRD patterns of vacuum dried synthesized using aqueous leaf extract of *M. esculenta*. The 0.5 mM synthesized a number of Bragg reflections with 2θ values of 17.50°, 28.84° and 35.44° sets of lattice planes were observed which may be indexed to the (002), (121) and (220) facts of silver. The 1 mM synthesized peaks at 38.02°, 34.69°, 44.39°, 64.62 and 77.40° correspond to the (1 0 1), (1 0 0), (2 0 4), (1 4 4) and (2 0 2), face centered cubic structures.

FESEM analysis

FESEM micrograph shows the synthesized nanoparticles were (0.5 and 1 mM) spherical and aggregated shape. The EDX attachment with the FESEM was known to provide information on the chemical analysis of the fields that are being investigated or the composition at specific locations. In representative profile of the spot EDX analysis was obtained by focusing on Ag NPs.

HR-TEM analysis

The biosynthesized Ag NPs were analyzed by HR-TEM to find the information of morphology size of nanoparticles. These micrographs show individual silver particles as well as a number of aggregated and spherical shapes. Under careful inspection of such images, these assemblies were found to be aggregates of silver nanoparticles.

Larvicidal activity

In the present study, the larvicidal aqueous crude leaf extracts, $AgNO_3$ and synthesized Ag NPs of *M. esculenta* were prominent; however, the highest mortality was found in synthesized Ag NPs against third instar larvae of *A. aegypti* and *C. quinquefasciatus* at the concentration of 10 mg/L. The larvae mortality was observed aqueous leaf extract of *M. esculenta* (Fig. 1). The LC_{50} values of *M. esculenta* aqueous leaf extract appeared to be effective against *A. aegypti* (LC_{50} = 66.14 mg/mL; LC_{90} = 172.41 mg/mL) and *C. quinquefasciatus* (LC_{50} = 61.60 mg/mL; LC_{90} = 184.80 mg/mL) (Table 1). Most considerable mortality was evident after the treatment of silver nanoparticles.

The aqueous $AgNO_3$ 1 Mm solution tested was third instar of *A. aegypti* and *C. quinquefasciatus* showed in (Fig. 2). The

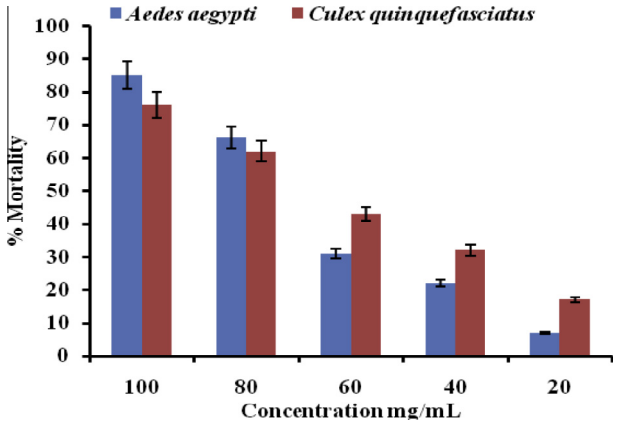

Figure 1 Larvicidal activity of *M. esculenta* aqueous leaf extract.