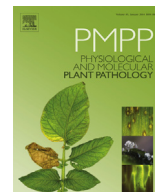




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## *Suaeda maritima*-based herbal coils and green nanoparticles as potential biopesticides against the dengue vector *Aedes aegypti* and the tobacco cutworm *Spodoptera litura*

Udaiyan Suresh <sup>a</sup>, Kadarkarai Murugan <sup>a, b</sup>, Chellasamy Panneerselvam <sup>c</sup>,  
Rajapandian Rajaganesh <sup>a</sup>, Mathath Roni <sup>a</sup>, Al Thabiani Aziz <sup>c</sup>,  
Hatem Ahmed Naji Al-Aoh <sup>d</sup>, Subrata Trivedi <sup>c</sup>, Hasibur Rehman <sup>c</sup>, Suresh Kumar <sup>e</sup>,  
Akon Higuchi <sup>f</sup>, Angelo Canale <sup>g</sup>, Giovanni Benelli <sup>g, \*</sup>

<sup>a</sup> Division of Entomology, Department of Zoology, School of Life Sciences, Bharathiar University, Coimbatore, 641046, Tamil Nadu, India

<sup>b</sup> Thiruvalluvar University, Serkkadu, Vellore 632 115, Tamil Nadu, India

<sup>c</sup> Biology Department, Faculty of Science, University of Tabuk, Tabuk, Saudi Arabia

<sup>d</sup> Chemistry Department, Faculty of Science, University of Tabuk, Tabuk, Saudi Arabia

<sup>e</sup> Department of Medical Microbiology and Parasitology, Universiti Putra Malaysia, 43400 Serdang, Slangor, Malaysia

<sup>f</sup> Department of Chemical and Materials Engineering, National Central University, No. 300, Jhongda RD., Jhongli, Taoyuan, 32001 Taiwan

<sup>g</sup> Department of Agriculture, Food and Environment, University of Pisa, Via Del Borghetto 80, 56124 Pisa, Italy

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## ABSTRACT

The overuse of synthetic pesticides to control insect pests leads to physiological resistance and adverse environmental effects, in addition to high operational cost. Insecticides of botanical origin have been reported as useful for control of agricultural and public health insect pests. This research proposed a novel method of mangrove-mediated synthesis of insecticidal silver nanoparticles (AgNP) using *Suaeda maritima*, acting as a reducing and stabilizing agent. AgNP were characterized by UV–vis spectroscopy, Fourier transform infrared (FTIR) spectroscopy, scanning electron microscopy (SEM), energy-dispersive X-ray spectroscopy (EDX), and X-ray diffraction (XRD) analysis. *S. maritima* aqueous extract and mangrove-synthesized AgNP showed larvicidal and pupicidal toxicity against the dengue vector *Aedes aegypti* and the tobacco cutworm *Spodoptera litura*. In particular, LC<sub>50</sub> of AgNP ranged from 8.668 (larva I) to 17.975 ppm (pupa) for *A. aegypti*, and from 20.937 (larva I) to 46.896 ppm (pupa) for *S. litura*. In the field, the application of *S. maritima* extract and AgNP (10 × LC<sub>50</sub>) led to 100% mosquito larval reduction after 72 h. Smoke toxicity experiments conducted on *A. aegypti* adults showed that *S. maritima* leaf-, stem- and root-based coils evoked mortality rates comparable or higher if compared to permethrin-based positive control (62%, 52%, 42%, and 50.2 respectively). In ovicidal experiments, egg hatchability was reduced by 100% after treatment with 20 ppm of AgNP and 250 ppm of *S. maritima* extract. Furthermore, low doses of the AgNP inhibited the growth of *Bacillus subtilis*, *Klebsiella pneumoniae* and *Salmonella typhi*. Overall, our results highlighted the potential of *S. maritima*-based herbal coils and green nanoparticles as biopesticides in the fight against the dengue vector *A. aegypti* and the tobacco cutworm *S. litura*.

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## 1. Introduction

According to the report of FAO, US \$120 billion losses worldwide

were caused by 20–40% decrease in crop yield, due to the attack from pathogenic organisms and insect pests [109]. Agriculture is the backbone of the Indian economy and nearly 75% of the rural areas of Indian villagers are depending on agriculture. The amount of food production is greatly deteriorated due to the crop pests and diseases, which lead to agricultural damage either directly by causing economic losses to the crops in the field or indirectly by

\* Corresponding author.

E-mail addresses: [cpselva@gmail.com](mailto:cpselva@gmail.com) (C. Panneerselvam), [benelli.giovanni@gmail.com](mailto:benelli.giovanni@gmail.com) (G. Benelli).

causing organoleptic alterations and/or the production of toxic substances [6].

The tobacco cutworm *Spodoptera litura* (Fab.) (Lepidoptera: Noctuidae), is one of the major pests of many important crop plants. Since cutworm larvae can defoliate many economically important crops possessing a high dispersal capability, this pest often leads to high levels of agricultural losses [18,27]. This pest attacks more than 112 species of cultivated crops. Currently, large quantities of insecticides have been used to fight cutworm infestations on different crops [72]. Chemical pesticides play a significant role in increasing agricultural production by controlling the insect pests. Also, molecular research has revealed the interaction of autophagy-related protein 1 with autophagy-related protein 5 in *S. litura* [108]. However, there is widespread concern over negative impact of insecticides on environmental and human health due to accumulation of insecticide residues as well as emergence of pesticide resistance in the pests [5,9]. Due to this reason, many researchers focused on alternative control methods. Botanicals are effective against a variety of insect pests; they are easily degradable [15].

Besides crop pests, mosquitoes represent the major arthropod vectors of human disease worldwide, transmitting malaria, lymphatic filariasis, and arboviruses such as dengue fever and Zika virus [10,11,47,77]. Dengue, mainly vectored by the bites of infected *Aedes* mosquitoes, has the greatest epidemic potential worldwide, with huge negative impact on the economy and health of the population in urban areas [12–14]. Dengue can be divided into four serotypes (DENV 1, 2, 3, and 4), each of which confers partial cross-protective immunity to the other serotypes in humans [99]. Dengue is affecting more than 128 countries, and the results of biometric analysis of dengue burden in Arabian countries have been recently revealed [110]. Every year about 390 million people are infected by dengue virus, among which 96 million become severe and results in about 21,000 deaths [16]. About half of the world's population is now at risk.

The main transmission cycle is identified for dengue, which is largely caused by the urban adapted *Aedes aegypti* mosquitoes, and along with some other species such as *Aedes albopictus* [12,74]. Currently, a global alert has been issued for Zika, given the increase in congenital abnormalities, Guillain-Barré syndrome, and other autoimmune manifestations, as well as the increase in chronic joint diseases due to chikungunya [62].

Since dengue and Zika virus are currently not vaccine preventable communicable diseases, vector control remains the only way to prevent arbovirus transmission [13,20,34]. As such, vector control and personal protection from the bites of infected mosquitoes are necessary [104]. Effective dengue control requires the community's participation. The community's health knowledge, attitudes and practices (KAP) will determine their participation in community-based programs. Taken together, these scenarios highlighted the need for effective and sustainable vector control strategies [9]. Very recently it is reported that a particular strain of *Wolbachia* can reduce the transmission of Zika virus by *A. aegypti* [2]. Overall, new drugs with unique structures and mechanisms of action are urgently required to treat drug-resistant strains of dengue [12]. Natural products and their derivatives from plants are continued to play an important role in the development of drugs for the treatment of human diseases as well as mosquitoicides [64,112].

Mangroves are a rich source of biologically active and pharmacologically valuable natural products [40,80]. Therefore the present research presents recent advances in order to develop insecticidal compounds from mangrove plant extracts, with special reference to the green synthesis of silver nanoparticles. Mangrove forest ecosystems are characterized by facultative halophytic species of trees

and shrubs that fringe the intertidal zone along sheltered coastal, estuarine and riverine areas in tropical and subtropical latitudes [8,37]. Mangroves are biochemically unique vegetation that produces a wide array of natural products with immense medicinal potential [68,69]. They have been traditionally used in fisher-folk medicine to treat several diseases.

It has been reported that the mostly halophytic genus *Suaeda* consists of 110 species worldwide covering the coastal areas of tropical and subtropical regions [29]. The distribution and zonation of different mangrove species also depends on physico-chemical variations of salinity and available nutrients [48,98]. Overall, mangroves have an immense ecological role in the coastal and marine environment [97].

*Suaeda maritima* (L.) Dumort (Chenopodiaceae) is a salt marsh-mangrove annual herb that grows in very alkaline and saline moist soils [78,80]. This plant is distributed throughout the east-west coast mangroves in India, i.e. Sunderbans in West Bengal, Mahanadi and Bitharkanika in Orissa, Coringa, Krishna and Godavari in Andhra Pradesh, Karangadu and Pichavaram in Tamil Nadu. Leaf extracts of *S. maritima* have been used as traditional remedies for hepatitis [41], viral [61,71] and bacterial infections [45].

Nanotechnology is a promising field of interdisciplinary research. It opens up a wide array of opportunities in various fields including pest control, pharmaceuticals, electronics and parasitology [52–54,73]. Nowadays, the green synthesis of insecticidal nanoparticles is an interesting issue of nanoscience [11,25,46,57,75,77,81,84]. Synthesis of silver nanoparticles using mangroves scarcely analyzed the potential of nano-insecticides for insect pest management [58].

Therefore, in this research, a selected mangrove species (*S. maritima*) was used for biosynthesis of silver nanoparticles (AgNP) effective against insect pests of medical and agricultural relevance. *S. maritima*-fabricated AgNP were characterized by UV–vis spectroscopy, Fourier transform infrared (FTIR) spectroscopy, scanning electron microscopy (SEM), energy-dispersive X-ray spectroscopy (EDX), and X-ray diffraction (XRD) analysis. We investigated the toxicity of the aqueous extract of *S. maritima* and *S. maritima*-synthesized AgNP in laboratory conditions against larvae and pupae of the dengue vector *A. aegypti* and the tobacco cutworm *S. litura*. We also evaluated the impact of *S. maritima* extract and AgNP as ovicides on *A. aegypti*. The smoke toxicity of herbal coils prepared using different parts of *S. maritima* on *A. aegypti* adults was studied. Both insecticides were validated in the field against *A. aegypti*. Finally, we also assessed antibacterial properties of AgNP against *Bacillus subtilis*, *Klebsiella pneumoniae*, and *Salmonella typhi*.

## 2. Materials and methods

### 2.1. *Suaeda maritima* collection and extraction

*S. maritima* leaves were collected from coastal areas of Pichavaram (11°25'47.9"N 79°48'08.5"E, Cuddalore district), Tamil Nadu, India. Specimens were washed with tap water and shade-dried at room temperature. Dried leaves were powdered using an electrical blender; 500 g of the powdered plant material were extracted using 1.5 L of ethanol for 72 h. The crude plant extract was concentrated at reduced temperature using a rotary evaporator, and stored at 22 °C. One gram of the residue was dissolved in 100 mL of acetone (fixative agent to separate the aqueous impurities altering the chemical composition of plant crude extract) and considered as 1% stock solution. From this stock solution, experimental concentrations were prepared [52,55].

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