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## **RESEARCH PAPER**



## Synthesis of Eco-Friendly Nanostructured Lipid Carriers Decorated With Magnetic Nanoparticle Encapsulated *Sesbania sesban* Extract Against Vector Borne *Culex pipiens* (Diptera: Culicidae) and *Musca domestica* (Diptera: Muscidae) as Green Insecticides

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#### ABSTRACT

Mosquito-borne diseases remain a significant health concern amidst current microbial outbreaks. Phytochemicals offer environmentally safe, biodegradable, and targeted pest management. Nanostructure lipid carriers (NLCs), a second generation of solid lipid nanoparticles, are gaining attention as potential diagnostic and therapeutic tools. Sesbania leaves, rich in fatty acids, phenolics, and terpenes, were analyzed using gas chromatography-mass spectrometry. Magnetic nanoparticles (Se-NLC-MNPs) modified the surface of Sesbania extract, encapsulated in the NLC. The resulting nanoparticles were 129.2 and 218.5 nm in size, with zeta potentials of -6.20 and 43.9 mV, respectively. Transmission electron microscopy showed spherical and oval shapes. XRD patterns confirmed the successful decoration of the NLC with the magnetic nanoparticles. The Sesbania extract (Se) and its nanoparticle conjugates were tested for larvicidal efficacy against *Culex pipiens* and *Musca domestica* larvae, at doses ranging from 50 to 1500 ppm and 0.1 to 5 mg/mL. Se-NLC-MNPs showed higher larval mortality rates compared to their Se formulation extracts, achieving 100% mortality in third-instar larvae. Sesbania methanol extract contained more terpenes, fatty acids, and other organic compounds than the aqueous extract, making it more harmful to insect larvae. In terms of relative toxicity, Se-NLC-MNPs were more effective than Se-NLC. An in vitro cytotoxicity assay against the WI38 cell line indicated the cytotoxicity assay, suggesting the potential for these nanoparticles to develop into high-performance, environmentally acceptable therapeutics for mosquito-borne diseases.

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#### 1 | Introduction

Worldwide, vector-borne illnesses continue to be a major public health concern, particularly in tropical and subtropical regions. Over three billion people live in contaminated environments, further endangering public health (Cuthbert et al. 2023). Due to the wide range of harmful pathogens that arthropod vectors can disperse, both humans and animals can contract numerous infectious diseases (Burkett-Cadena and Vittor 2018; Socha et al. 2022). Specific conditions, such as interactions between hosts, viruses, vectors, and vulnerable populations, can lead to direct transmission of many illnesses from person to person (Gyawali 2018).

Mosquitoes are an important insect because they serve as a vector for disease transmission (Coetzee 2014). They can transmit a variety of diseases, including dengue, yellow fever, filariasis, malaria, and Japanese encephalitis. Infected female mosquitos, notably *Aedes aegypti* and *Aedes albopictus*, carry dengue viruses, which have become a severe public health concern globally in recent years (Conway et al. 2014). Also, *Culex pipiens* spread the West Nile virus (WNV), which has historically been enzootic across southern and central Europe, Asia, and Africa and is most important in Egypt.

The house fly is also an important mechanical vector for many diseases and could transmit nearly 100 diseases to both humans and animals, including bacteria such as *E. coli*, *Shigella* species, *Salmonella*, and viruses. This is because adult houseflies are attracted to human food, animal dung, garbage, and decaying animal waste (Issa 2019; Mohamed et al. 2024).

In order to mitigate the threats posed by mosquitoes and flies, a variety of vector control measures and monitoring techniques are regularly used. Over time, medical insect control programs have incorporated synthetic insecticides along with other pests. However, frequent and excessive use of insecticides can be extremely harmful, interfere with the food chain, and pollute the environment due to vectors' resistance to insecticides (Gyawali 2018). Therefore, the number of mosquitoes and flies is increasing in many developing countries, including Egypt, for several reasons, including operational and technological difficulties, increasing insecticide resistance in flies and mosquitoes, and lack of knowledge due to socioeconomic factors.

To eliminate these pests without endangering the environment, it was necessary to find a viable alternative to these synthetic insecticides. Phytochemicals are an important alternative to synthetic insecticides. These compounds are known as secondary metabolites in plants and serve a variety of functions, including response to environmental stresses, innate immunity, signaling defense responses, and defense against pests and diseases (Baz, Alfagham, et al. 2024). These chemicals also affect mosquitoes; they kill eggs, larvae, and adults; stop egg laying; stop growth; stop feeding; keep mosquitoes away; stop hatching; and stop mosquito emergence (Şengül Demirak and Canpolat 2022).

Natural insecticides, especially those transformed into nanoformulations, will undoubtedly serve as cost-effective, efficient, and environmentally friendly weapons to combat insect pests (Radwan, Baz, Khater, and Selim 2022; Radwan et al. 2024). Therefore, we aim to demonstrate the power of plant extracts containing bioactive chemical compounds, which, when transformed into nanoformulations, may increase their potency and effectiveness in killing and deterring insects, thus reducing the potential for disease spread.

Magnetic nanoparticles are eco-friendly and safe materials because their magnetic qualities allow them to be utilized and reassembled (Abdullah et al. 2023; Verma et al. 2024). Because it is simple to manufacture and handle, either chemically or physically, it can be developed and modified to maximize its biological advantage and effectiveness. Numerous studies have looked into the necessity of using ferrites, particularly magnetic nanoparticles (Nie et al. 2023). In addition to free magnetic nanoparticles, loaded magnetic nanoparticles with two advantages are also frequently utilized. By combining the essential oil of Volkameriana citrus (Volkamer lemon) with the magnetic nanoparticles, pesticides can be created (Alshallash et al. 2022). Thus, the combination of Volkameriana citrus oil with magnetic nanoparticles may be useful for suppressing insects. For instance, larvicides' ease of manufacture, nontoxicity, and high affinity help them control the yellow fever mosquito, A. aegypti.

Typically, magnetic NPs are used as insecticides or larvicides by keeping them in aqueous fluids and then activating them in the afflicted area. The most effective NPs for activation are easy to manage and have strong biocide qualities. When alternating magnetic fields of very low frequencies are present, these magnetic nanoparticles, known as magnetic nanostructure layered double hydroxide (MNP@LDH), can be used for bactericidal action because of their enormous magnetic moments (Lade and Gogle 2019). Particles move in response to the magnetic field, which enables researchers to get a wealth of information about their structure, anatomical features, and other sensory systems. But the biggest benefit is that nanoparticles probably will not affect plants' physiological or biochemical functions negatively (Singh et al. 2024). The MNP@LDH is a good candidate for effective protection on plants by providing multitargeted biopesticide activity, fast larvicidal kinetics, and stability in solution for long periods of time.

The second generation of solid lipid nanoparticles (SLNs) in the lipid NP family is called nanostructured lipid carriers (NLCs) (Esmaili et al. 2021). To overcome the drawbacks of pure SLNs, such as drug loading capacity, drug expulsion during storage, and undesirable modifications, NLCs use a binary blend of melted lipid materials, which includes solid lipids and liquid lipids (oils). They also provide a range of compositions to satisfy customized physicochemical properties of NLCs (Esmaili et al. 2021). The fact that NLCs are scalable and a profitable platform for loading medications and altering their properties, such as improved selective toxicity and solubility, is well acknowledged. Patients can safely use NLCs because of their biodegradable and biocompatible nature (Khan et al. 2022). NLCs are used to minimize adverse bodily damage and treat infected tissue selectively. With reduced medication dosages, quicker recovery times, and cheapr treatment expenses, this technology will enhance patients' lives. With reduced drug excipient toxicity and less allergic responses, NLCs help with controlled drug release (Nguyen et al. 2022). As a result, NLCs are in high demand for a variety of clinical

disorders. It is anticipated that biodegradable and biocompatible lipid nanoparticles, such as NLCs, may reduce the usual adverse effects of drugs like chemotherapeutics and enable more efficient treatment with lower quantities of medication.

## 2 | Materials and Methods

## 2.1 | Chemicals

Butyl alcohol, lauric acid, oleic acid, stearic acid, tween 20, ferrous sulfate (II) heptahydrate, ferric chloride (III) hexahydrate, sodium glycocholate, and ammonium hydroxide 50% hydrolyzed were purchased from Alfa Aesar Germany (Fisher Scientific). In our laboratory, two rounds of high-purity distillation were used to prepare distilled water and methyl alcohol from El-Gomhoria Company (Cairo, Egypt). All chemicals and solvents were utilized just as is, without any additional purification.

## 2.2 | Plant Collection and Extraction

From August to October 2023, leaves of *Egyptian riverhemp* (*Fabaceae*), also known as *Sesbania sesban* L., were gathered from various locations in the Qalyubiya Governorate, Egypt.

One of the most common decorative and park plants is *S. sesban*. A voucher specimen with the code (PHG 512) was deposited in the herbarium after the gathered plant was recognized and verified by Dr. Rim Hamdy, a professor of plant taxonomy and flora in Cairo University's Botany and Microbiology Department.

After the gathered plants were allowed to air dry for 10 days at room temperature (27°C) in a shaded area until all of the water had been removed, the leaves were ground into a fine powder using a stainless-steel shredder. Exactly 50g of finely powdered leaves was added to 150 mL of 100% methanol in an airtight container that had been cleaned and dried. The mixture was then agitated for 48h at room temperature. To remove all debris and insoluble materials, the solid fibers and insoluble materials were filtered out using medical cotton and then Whatman cellulose filter paper (0.45 mm). The supernatant was then reconcentrated using a rotary evaporator for 10 min at 35°C under vacuum (Radwan, Baz, Khater, and Selim 2022). The residue that was left over was a 100% concentrated extract that was stored in opaque glasses at a lower temperature than  $-5^{\circ}$ C. The same procedure was used to create the aqueous extract, but instead of utilizing a rotary evaporator for the concentration step, lyophilization was used to freeze-dry the mixture at -55°C for 2 days. A similar method was used to preserve semisolid paste.

## 2.3 | Synthesis of Nanostructure Lipid Carrier Encapsulated *Sesbania* Extract

The synthesis of *Sesbania* extract loaded nanostructure lipid carrier (Se-NLC) was accomplished using hot homogenization method with little modification (Naseri et al. 2015; Gomaa et al. 2021; Radwan et al. 2024). A lipid mixture comprising 600 mg of each one of solid lipid components (600 mg of stearic acid and 600 mg of lauric acid) and 950 mg of liquid lipid (oleic

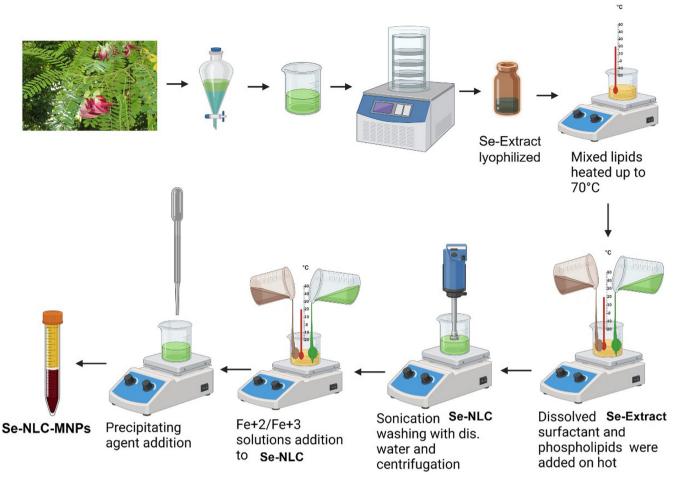
acid) was combined in a dry and clean beaker. The mixture was heated up to 70°C (melting point of steric acid), utilizing all the solid contents transformed into homogenous melted mixture. Exactly, 180 mg of the Sesbania extract paste was dissolved 3.5 mL in mixed solvents of butanol/chloroform, 3:1 (v/v). After complete dissolution, the extract was added to the melted lipids on hot. Stirring was processed for a few minutes (solvents should be removed completely), utilizing the addition of the aqueous phase containing phospholipids (0.05g of sodium glycocholate and dissolved in 5-mL distilled water) and 5.8g of tween 20 emulsifier. High shearing was introduced for 2 min followed by additional 10-mL precooled distilled water and sonication processed for additional time. To remove any nonencapsulated components, the resultant emulsion was centrifuged after 10 min of sonication. The filterate was removed and the triturate was then resuspended in 30 mL of fresh distilled water and 2.5g of tween 20 (to compensate what is disposed by filtration). At last, mixture dispersed using probe sonication for 10 min to produce (Se-NLC).

## 2.4 | Synthesis of Nanostructure Lipid Carrier Encapsulated Sesbania Extract and Decorated With Magnetic Nanoparticles (Se-NLC)

To apply magnetic nanoparticles by co-precipitation to the NLC's surface (Shen et al. 2014), approach with a few improvements, such as making a stock solution of ferrous sulfate heptahydrate (FeSO4.7H2O,  $5 \times 10^{-4}$ M) and ferric chloride hexahydrate (FeCl3.6H2O,  $5 \times 10^{-4}$  M), each of which is dissolved in 40 mL of distilled water. After thoroughly dispersing a sufficient amount of Se-NLC nanoparticles in a clean beaker, 7.5 mL of each iron di and trivalent salt (Fe [II]/Fe [III]) was added, and the mixture was stirred for 10 min. NH4OH (5% v/v), the precipitating agent, was added drop by drop while being vigorously stirred. Until all of the precipitating agent (5 mL) is used, the solution will tend to be darker because of the creation of magnetic nanoparticles. A 50mL Falcon tube was filled with the dispersion and centrifuged at 6000 rpm for 10 min at a lower temperature. Following filtration purification, thorough washing was carried out multiple times to ensure that all nonreacting iron salts had been removed. semisolid paste of Se-NLC-MNPs was obtained from the collected slurry (Figure 1). The dispersion was transferred into a 50-mL Falcone tube. Using the same procedure, free magnetic nanoparticles were created to verify that the decorated NPs had the same structure as free magnetite by comparing their XRD patterns.

## 2.5 | Particle Size (DLS) and Zeta Potential (ZP)

Dynamic light scattering (DLS) was used to quantify the hydrodynamic radius and the polydispersity index at room temperature and at an angle of 173° in order to assess the quality of the generated NPs. The DLS and PDI were tested three times to ensure the best results. The zeta potential (ZP) is measured using the frequency of scattered light shifting due to particle charge at scattering angle 12°. Measuring ZP is necessary to determine the stability of NPs. ZP describes the electrochemical equilibrium between particles and liquids, as in the case of NPs (Lunardi et al. 2020). The NP radius, ZP, and PDI are measured



**FIGURE 1** | Schematic illustration of the synthesis of both Se-NLC and Se-NLC-MNPs nanoparticles.

at the Egyptian Petroleum Research Institute (EPRI) utilizing Zeta sizer Nano series (HT), Nano ZS, and Malvern Instruments (United Kingdom). Each nanoformulation liquid contained 5–10 mg, which was dispersed and sonicated in 10 mL of distilled water. The sample was then placed in a quartz cell for analysis.

## 2.6 | Surface Morphology and Topography by Transmission electron Microscope (TEM)

Transmission electron microscopy (TEM) is a useful tool for defining the internal shape of NLCs, ensuring not only the regularity of NPs but also particle size. Thus, suggesting that aggregations are important properties of NPs, the high-resolution transmission electron microscope (HR-TEM, JSM-7100F) at the Egyptian Petroleum Research Institute (EPRI, Cairo) is used to examine the morphology of NLCs. Images were captured using JOEL JEM-2100F HR-TEM, a 200-kv FE (field emission) analytical electron microscope with multiple uses. After diluting 1 µL of NLCs with double distal water (1:200), it is put on a carboncoated grid (200 mesh) and left for 2 min. After adding two drops of 2% (w/w) phosphotungastic acid (PTA) to the grid for 10s, the extra liquid was gathered using filter paper. The filter paper absorbed the excess PTA that was added to generate the negative staining. After that, the grid was moved to a TEM, where highresolution pictures were taken and the morphology of NLCs was carefully examined.

## 2.7 | XRD

Powdered magnetic nanoparticles (MNPs) and powdered nanostructure lipid carriers decorated with magnetic nanoparticles (Se-NLC-MNPs) all had their power X-ray diffraction (PXED) patterns examined using X, pert PRO P analytical with Cu K $\alpha$ radiation ( $\lambda$  = 1.5406 Å). The diffraction pattern was at 2.4°/min scanning with a 22  $\theta$  range (4–80).

## 2.8 | Mosquito Larvicidal Assay

## 2.8.1 | Rearing Culex pipiens

Culex pipiens larvae were reared under laboratory conditions at  $27 \pm 2^{\circ}$ C and 75%-80% relative humidity under a 14:10h (light/dark) photoperiod in the Entomology Department of the Department of Medical and Molecular Entomology, Faculty of Science, Benha University, Egypt, according to Baz et al. (2022) for several generations. The mosquito larvae were reared in enamel plates of  $25 \times 20 \times 10$  cm and filled with 2L of dechlorinated tap water. They were fed Tetramin fish food and powdered dog biscuits every 2 days. Following pupation, *Cx. pipiens* was placed in insectary cages measuring  $30 \times 30 \times 30$  cm adult mosquitoes fed on an 8%-10% sugar solution. The larvae of both species were consistently available for studies and were housed in the same laboratory conditions.

#### 2.8.2 | Rearing Musca domestica

Adult houseflies were collected from the vegetable market in Benha, Qalyubiya, Egypt. They were then housed at room temperature  $(30^{\circ}C-32^{\circ}C)$  in solid wooden cages measuring  $30 \times 30 \times 30 \text{ cm}^3$ . The cages were covered with wire mesh covers. The diet consisted of 300g of mackerel in a plastic dish measuring  $18 \times 25 \times 9 \text{ cm}^3$ , in addition to a mixture of 10% syrup and 10% milk absorbed on cotton wool. Placing the mackerel on top of a mixture of dry bread and seaweed provides the perfect location for houseflies to breed and lay their eggs.

#### 2.8.3 | Larvicidal Activity

The larvicidal effectiveness of the S. sesban extracts and their nanoparticles against Cx. pipiens larvae in their third instar was assessed. Six concentrations of plant materials were administered to Cx. pipiens larvae: 50, 100, 200, 500, 1000, and 1500 ppm. Five repetitions (100 larvae) were utilized for each concentration, with 20 mosquito larvae used for each replicate. Deaths were noted 24 and 48h after treatments (PT) (WHO 2005); third-instar larvae of M. domestica were conducted using ingestion and contact methods (feeding technique) for evaluation of the larvicidal activity of plant extracts and their nanoformulations. Fifteen larvae in their early third instar were put in little paper cups with 10g of rearing medium (5cm in diameter and 7cm in height). The cups were then treated with 0.1, 0.5, 1.0, 2.5, and 5.0 mg/mL of plant materials. The untreated groups were treated with water only. The treated and untreated cups were covered with a cotton cloth tied to prevent the larvae from escaping. The dead larvae were counted after 24 h, and 10 g of sawdust was added to new cups to record the larval duration and pupation. The experiment was repeated five times.

## 2.9 | GC/MS Analysis

For the biochemical analyses, Thermo Scientific Trace GC Ultra/ISQ Single Quadrupole MS, TG-5MS-fused silica capillary columns with thicknesses of 0.1 mm, 0.251 mm, and 30 m were employed (Agilent Technologies, Santa Clara, CA, United States). The procedure involved the use of an electronic ionizer with an ionization energy of 70 eV. Helium gas was used as a carrier gas (flow rate: 1 mL/min). Both the injector and the MS gearbox line had been set at 280°C. The oven was preheated at 35°C. After that, the temperature was increased by 7°C/min to 150°C, then by 5°C/min to 270°C (with a 2min pause), and finally by 3.5°C/min to 310°C (this was done for 10 min). A relative peak area was used to investigate and quantify each component that was discovered. The chemicals were at least substantially identified by matching their mass spectra and retention times with NIST and Willy Library data from the GC/MS instrument. Identification was done using the whole spectrum of user-generated reference libraries. Peak uniformity was evaluated using reconstructions from single-ion chromatography. Cochromatographic analysis of reference materials was used whenever possible to validate GC retention times (Abd El-Kareem et al. 2024).

#### 2.10 | Statistical Analysis

The software SPSS V23 (IBM, United States) was used to analyze the data in order to do the one-way analysis of variance (ANOVA) (post hoc/Turkey's HSD test) and Probit analyses to determine the lethal concentration (LC) values. The threshold for significance was established at p < 0.05.

## 3 | Results

#### 3.1 | Drug Delivery System

#### 3.1.1 | Particle Size and Stability (ZP)

The particle size measurements and homogeneity index (polydispersity index) of the synthesized NPs were measured. According to Figure 2a,b, the particle size and polydispersity index were 129.3 nm, 0.35, and 218.5 nm, 0.42, respectively, for *Sesbania* extract encapsulated nanostructure lipid carrier (Se-NLC) and its surface modified analog (Se-NLC-MNPs). And ZP and stability were found to be -6.20 and 43 mV, respectively as described at Figure 3a,b.

#### 3.1.2 | Transmission Electron Microscope and Internal Morphology

According to Figures 4a,b and 5a,b, transmission electron microscope (TEM) visualization of the synthesized Se-NLC and Se-NLC-MNPs presented divers regular and irregular spherical shapes with different dimensions assorted from sizes less than 300 nm.

## 3.1.3 | XRD

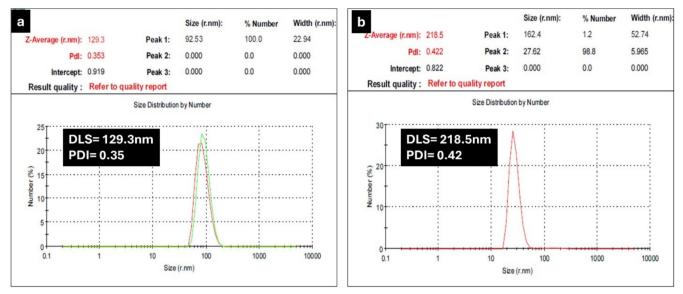
The MNPs free sample exhibits broad peaks around  $30.1^{\circ}$ ,  $35.5^{\circ}$ ,  $43.1^{\circ}$ ,  $53.5^{\circ}$ ,  $57.1^{\circ}$ , and  $62.7^{\circ}$ , characteristic of the spinel structure of magnetite (Fe<sub>3</sub>O<sub>4</sub>) in both samples correspond to the spinel structure of magnetite (Figure 6).

## 3.2 | In Vitro Assessment of Drug Cytotoxicity

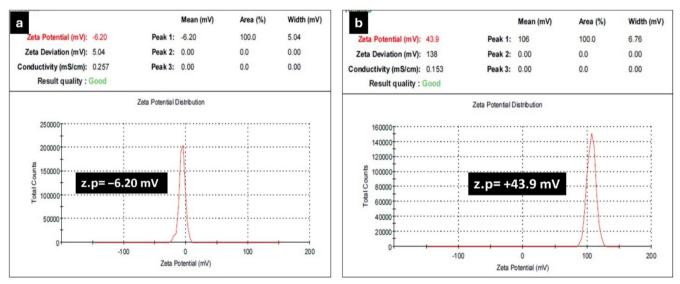
Normal human lung fibroblast WI38 was used for the cytotoxicity evaluation, and Table 1 shows the optical densities, cell viability, and Ic50 values. According to the evaluation, the IC50 values for Po-NLC and Po-NLC-MNPs were 151.23 and  $183.75 \,\mu$ g/mL, respectively.

## 3.3 | Mosquitoes Larvicidal Activity

This work assessed the larvicidal effects of extracts from *S. sesban* (Se) and their nanoparticles against the third larva, *Cx. pipiens*, suggesting possible insecticidal action. The findings demonstrated that whereas larval mortality rates were low following treatment with Free-NLC and Free NLC-MNPs, they were higher following posttreatment (PT) with nanoparticles as opposed to their respective plant extracts.



**FIGURE 2** | Average particle size and polydispersity index of the synthesized nanoparticles: (a) *Sesbania* extract encapsulated nanostructure lipid carrier (Se-NLC); (b) *Sesbania* extract encapsulated nanostructure lipid carrier surface modified with magnetic nanoparticles (Se-NLC-MNPs).



**FIGURE 3** | Zeta potential of the synthesized nanoparticles: (a) *Sesbania* extract encapsulated nanostructure lipid carrier (Se-NLC); (b) *Sesbania* extract encapsulated nanostructure lipid carrier surface modified with magnetic nanoparticles (Se-NLC-MNPs).

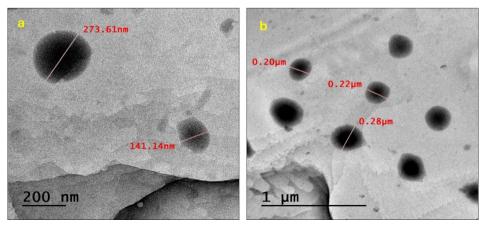


FIGURE 4 | Internal morphology manifestation by transmission electron microscope at different field (a, b) of the examined Se-NLC nanoparticles.

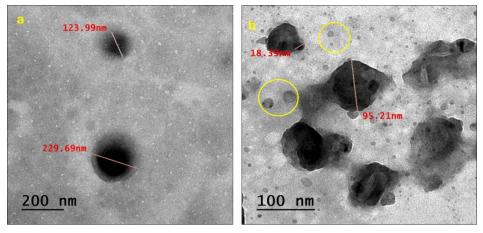
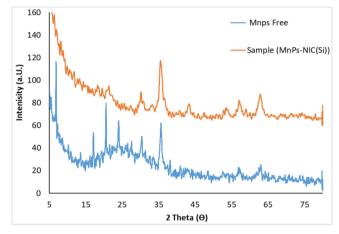


FIGURE 5 | Internal morphology manifestation by transmission electron microscope at different field (a, b) of the examined Se-NLC-MNPs nanoparticles.



**FIGURE 6** | XRD pattern of both free MNPs and Se-NLC-MNPs.

According to this investigation, at the maximum concentration of 1500 ppm, the whole (100 MO%) larvicidal effects of Ss extracts and their nanoparticles were observed 24-h posttreatment. The data indicated that at 500 ppm, the methanol MO% of Se, Se-NLC, and Se-NLC-MNPs was 82%, 100%, and 100%, respectively, at 24-h posttreatment (Table 2). The methanol extracts' LC50 (50%, median lethal concentration) was 161.10, 101.80, and 88.68 ppm, whereas the aqueous extracts were 65%, 92%, and 99% (MO%), with LC50 values = 262.81, 134.96, and 114.23 ppm, respectively (Table 3).

Additionally, the negative control (199 mL of distilled water with 1 mL of solvent) was not toxic to all larvae, whereas the positive control (temephos) had a significant effect on the mosquito larvae 96% (MO%). Free Se-NLC and Se-NLC-MNPs caused 32 and 47 MO% of the treated third-instar larvae, respectively. After 48 h of PT, the maximum larval mortality was observed, with death levels reaching 96%, 100%, and 100% in Se methanol, Se-NLC, and Se-NLC-MNPs and 85%, 100%, and 100% in aqueous extracts at 500 ppm, respectively (Table 2). *S. sesban* NLC-decorated MNPs (Se-NLC-MNPs) showed the most fatal quantities (LC<sub>50</sub>=69.24 ppm) against *Cx. pipiens* larvae, followed by *S. sesban* NLC nano (LC<sub>50</sub>=77.55 ppm) and *S. sesban* extract (LC<sub>50</sub>=130.23 ppm).

#### 3.4 | Housefly Larvicidal Activity

All tested plant materials had significantly higher mortality rates than the controls. The percentage of dead larvae in plant-treated feeding technique at the high concentration (5%) was 100% compared to 0.0% in control groups. At a concentration of 5 mg/mL, Se, Se-NLC, and Se-NLC-MNPs methanol resulted in a 100% mortality rate (Table 4), whereas those of aqueous extracts were 80%, 96%, and 100% (MO%), respectively, after 24-h exposure. The LC<sub>50</sub> were 0.65%, 0.42%, 0.38%, and 1.76, 0.81, 9% (Table 5). Se-NLC-MNPs and free Se-NLC caused 24 and 32 MO% of the treated third-instar housefly larvae, respectively. The positive control (temephos) had a significant effect on the mosquito larvae 75% (MO%), whereas the negative control (199 mL of distilled water with 1 mL of solvent) was not toxic to any of the larvae.

#### 3.5 | Metabolomic Analysis of Four Plant Extracts

#### 3.5.1 | GC-MS Data Analysis

The plant extracts underwent metabolic analysis, and GC-MS analysis was used to compare the methanol and aqueous extracts. Using two distinct solvents (methanol and aqueous), the results of our study's GC-MS analysis allowed us to identify a number of compounds in the *S. sesban* leaves, including terpenes, fatty acids, esters, cyclohexane, alkane, and steroids (Table 6). Table 6 lists the 18 chemicals found in the *S. sesban* methanol leaf extract, whereas in the aqueous leaf extract, six compounds were found (Table 7). There was a significant amount of octadecanoic acid (17.86%), phytol (15.35%), 1-Naphthalenepropanol, alp (9.53%), and 17-pentatriacontene (8.8%) in the *S. sesban* methanol extract. Erucic acid (57.04%), 1(z)-(z)-docos-13-en-1-yl icos-11-enoate (20.50%), and 9-Octadecenoic acid (z)- (9.13%) were all present in the aqueous extract.

#### 4 | Discussion

As an inevitable outcome of the increase in drug resistance, insects' self-defense makes the newer insecticidal agents become nonefficient. The nanotechnology breakthrough raises as

 TABLE 1
 In vitro cytotoxicity assessment of Se-NLC and Se-NLC-MNPs against wi38.

ID	Conc	R1 (OD)	R2 (OD)	R3 (OD)	Mean OD	SE	Viability %	Toxicity %	IC <sub>50</sub>
Wi38	_	0.579	0.566	0.571	0.572	0.003786	100	0	$151.23 \pm 0.55$
Se-NLC	1000	0.031	0.036	0.03	0.032333	0.001856	5.652681	94.34732	
	500	0.059	0.057	0.055	0.057	0.001155	9.965035	90.03497	
	250	0.129	0.139	0.132	0.133333	0.002963	23.31002	76.68998	
	125	0.367	0.369	0.373	0.369667	0.001764	64.62704	35.37296	
	62.5	0.505	0.519	0.501	0.508333	0.005457	88.86946	11.13054	
	31.25	0.553	0.566	0.562	0.560333	0.003844	97.96037	2.039627	
Se-NLC-	1000	0.039	0.029	0.033	0.033667	0.002906	5.885781	94.11422	$183.75 \pm 0.81$
MNPs	500	0.042	0.047	0.049	0.046	0.002082	8.041958	91.95804	
	250	0.204	0.21	0.195	0.203	0.004359	35.48951	64.51049	
	125	0.388	0.396	0.391	0.391667	0.002333	68.47319	31.52681	
	62.5	0.495	0.51	0.501	0.502	0.004359	87.76224	12.23776	
	31.25	0.54	0.551	0.566	0.552333	0.007535	96.56177	3.438228	

Abbreviations: Se-NLC, Sesbania nanolipid particle; Se-NLC-MNPs, Sesbania nanolipid particle magnetic nanoparticles.

a gateway that helps humans to get rid of the epicenter of the drug resistance. The nanostructure lipid carrier is one of safer (Radwan et al. 2024) nanoparticles, as it contains lipids surrounded by phospholipids, which is not much different from the basic components of *Homo sapiens* cells. The surface modified nanostructure lipid carrier was used to encapsulate *Sesbania* extract and protect its active ingredients to be used as greener insecticide, trying to achieve maximum utility by introducing magnetic iron nanoparticles to benefit from their high energy in fighting insects (Kaur et al. 2014).

The motility of colloidal system is governorates with the Brownian motion, which confirms that the lighter particles move faster and their speed directly proportional to its particle size. Particle radii of the nanostructured lipid-based NPs revealed the average particle size is between 100 and 300nm. The intense efforts in synthesis of the nanostructured lipid carriers and its applications as insecticides depicted that its particle size in may extended to few micrometers. The particle size measurements are consistent with the results obtained by Tortorici et al. (2022). Sesbania extract encapsulated to the vicinity of NLC (Se-NLC) and its analogy surface modified NLC (Se-NLC-MNPs) showed higher poly dispersity index of 0.34 and 0.42. As increasing PDI value as indicting more heterogenous solution and more varied size distribution (Danaei et al. 2018), the incorporation of the magnetic NPs at the surface of the synthesized NLC makes sever heterogeneity changes (larger PDI) of the prepared Se-NLC-MNPs; this is due to the introduction of a smaller range of nanometric particles represented by magnetic iron particles, which are characterized by their relative small size (usually less than 100 nm) compared to NLC microparticles, which widens the differences in sizes and consequently leads to more heterogeneity.

The stability profile and shelf lifetime of any synthesized nanoparticles are controlled by the electrokinetic potential or electric potential at the slipping plane in colloidal dispersions. The net electrical charge stored inside the area bordered by the sliding plane creates the ZP, an electrical potential at the interface that differentiates fluid that is mobile from fluid that stays stuck to the surface. In general, a ZP that is positive or negative and away from zero indicates that the system is more stable (Azevedo et al. 2020). The synthesized Se-NLC presented a negatively charged zp of -6 mV on the contrary Se-NLC-MNPs presented positively charged zp of 42 mV. Perhaps the reason for the discrepancy between the two values can be attributed to the fact that NLC is usually negatively charged due to the fatty acids they contain as well as phospholipids, which stimulates the formation of a negative charge on the separating surface. When their surface is modified to be decorated with magnetic NPs, which are characterized by their high positive charge (Dabagh et al. 2023), perhaps what confirms the process of decorating as well as stability or surface modification the fatty material by magnetic materials is the qualitative transformation in the charge from negative to positive.

Along with ZP, the internal morphology by TEM shares the same importance in the particle stability checklist. The morphology of the Se-NLC NPs depicted regular spherical, oval, and semispherical with wide range of sizes extended from 140 to 280 nm, which is consistent with the results obtained by the DLS. The particle morphology of Se-NLC-MNPs showed regular and irregular spherical in most of examined fields in range of sizes do not differ from Se-NLC. Having no one else present, Se-NLC-MNPs presented very small debris (less than 50 nm) attached to the surface of the NLC confirming the decoration of surface of NLC with MNPs. The presence of unloaded MNPs contributes in heterogeneity of Se-NLC-MNPs keeping PDI larger than those in Se-NLC nanoparticles.

Significant variations in crystallinity and peak intensity were seen in the X-ray diffraction (XRD) patterns for the free magnetite nanoparticles, MNPs Free, and magnetite nanoparticles loaded with Sesbania extract Se-NLC-MNPs. The MNPs Free sample exhibits broad peaks around 30.1°, 35.5°, 43.1°, 53.5°, 57.1°, and 62.7°, characteristic of the spinel structure of magnetite

**TABLE 2** | The larvicidal effects of *Sesbania sesban* extracts and its nanoformulations against *Culex pipiens*, 24 and 48 h post- treatment.

SolventTime (h)Methanol24	Treatment	0	50	100	000	200	1000	1 500
			20	TOOT	2002	nnc	TUUU	OUCL
	Sesbania extract	$0\pm0^{\mathrm{aG}}$	$15\pm2.24^{\mathrm{bF}}$	$27\pm2.00^{\mathrm{cE}}$	$60 \pm 3.54^{cD}$	$82 \pm 2.45^{bC}$	$98 \pm 2.00^{\mathrm{bB}}$	$100\pm0.00^{\mathrm{aA}}$
	Se-NLC	$0\pm0^{\mathrm{aE}}$	$23 \pm 1.22^{aD}$	$46 \pm 1.87^{\mathrm{bC}}$	$75 \pm 4.18^{\mathrm{bB}}$	$100\pm0.00^{\mathrm{aA}}$	$100 \pm 0.00^{\mathrm{aA}}$	$100\pm0.00^{\mathrm{aA}}$
	Se-NLC-MNPs	$0\pm0^{\mathrm{aE}}$	$25\pm1.58^{\mathrm{aD}}$	$55 \pm 2.74^{aC}$	$82 \pm 3.00^{\mathrm{aB}}$	$100\pm0.00^{\mathrm{aA}}$	$100 \pm 0.00^{\mathrm{aA}}$	$100\pm0.00^{\mathrm{aA}}$
	Free- NLC	$0\pm0^{\mathrm{aG}}$	$2\pm1.22^{\mathrm{cF}}$	$3\pm1.22^{\mathrm{dE}}$	$8\pm1.22^{\mathrm{dD}}$	$13 \pm 2.00^{\circ C}$	$20\pm5.24^{\mathrm{dB}}$	$32 \pm 2.55^{cA}$
	Free_NLC-MNPs	$0\pm0^{\mathrm{aG}}$	$2\pm1.22^{\mathrm{cF}}$	$4\pm1.87^{ m dE}$	$10\pm0.00^{\mathrm{dD}}$	$15\pm0.00^{\rm cC}$	$27 \pm 1.22^{\mathrm{cB}}$	$47 \pm 2.55^{bA}$
48	Sesbania extract	$1\pm1.00^{\mathrm{aF}}$	$19\pm2.92^{\mathrm{cE}}$	$40\pm1.87^{\mathrm{cD}}$	$72 \pm 2.92^{\circ C}$	$96 \pm 2.45^{\mathrm{bB}}$	$100\pm0.00^{\mathrm{aA}}$	$100\pm0.00^{\mathrm{aA}}$
	Se-NLC	$1\pm1.00^{\mathrm{aE}}$	$28 \pm 2.00^{bD}$	62±4.64 <sup>bC</sup>	$96\pm1.87^{\mathrm{bB}}$	$100\pm0.00^{\mathrm{aA}}$	$100\pm0.00^{\mathrm{aA}}$	$100\pm0.00^{\mathrm{aA}}$
	Se-NLC-MNPs	$1\pm1.00^{\mathrm{aE}}$	$31 \pm 2.92^{aD}$	$66 \pm 2.92^{\mathrm{aC}}$	$99\pm1.00^{\mathrm{aB}}$	$100\pm0.00^{\mathrm{aA}}$	$100\pm0.00^{\mathrm{aA}}$	$100\pm0.00^{\mathrm{aA}}$
	Free- NLC	$0\pm0^{\mathrm{aF}}$	$2\pm1.22^{\mathrm{dF}}$	$6.\pm1.00^{\mathrm{eE}}$	$13 \pm 2.00^{\mathrm{eD}}$	$20 \pm 2.24^{\rm dC}$	$30\pm5.24^{\mathrm{cB}}$	$44\pm1.87^{\mathrm{cA}}$
	Free_NLC-MNPs	$0\pm0^{\mathrm{aG}}$	$3\pm1.22^{\mathrm{dF}}$	$9\pm1.00^{\mathrm{dE}}$	$18 \pm 2.55^{\mathrm{dD}}$	$26 \pm 3.67^{cC}$	$35 \pm 3.16^{\mathrm{bB}}$	$48\pm3.74^{\mathrm{bA}}$
Water 24	Sesbania extract	$0\pm0^{\mathrm{aG}}$	$10\pm0.00^{\mathrm{bF}}$	$20\pm1.58^{\mathrm{cE}}$	$44\pm1.87^{\mathrm{cD}}$	$65 \pm 2.55^{bC}$	$85 \pm 2.74^{\text{bB}}$	$100\pm0.00^{\mathrm{aA}}$
	Se-NLC	$0\pm0^{\mathrm{aE}}$	$18\pm1.58^{\mathrm{aD}}$	$33 \pm 2.00^{bC}$	$64 \pm 3.67^{\mathrm{bB}}$	$92\pm0.00^{\mathrm{aA}}$	$100 \pm 0.00^{\mathrm{aA}}$	$100\pm0.00^{\mathrm{aA}}$
	Se-NLC-MNPs	$0\pm0^{aE}$	$20 \pm 1.22^{\mathrm{aD}}$	$40\pm4.30^{aC}$	$70 \pm 3.54^{\mathrm{aB}}$	$99 \pm 1.00^{\mathrm{aA}}$	$100\pm0.00^{\mathrm{aA}}$	$100\pm0.00^{\mathrm{aA}}$
	Free- NLC	$0\pm0^{\mathrm{aF}}$	$2\pm1.22^{\mathrm{cF}}$	$3\pm1.22^{\mathrm{dE}}$	$8\pm1.22^{\mathrm{dD}}$	$13 \pm 2.00^{\circ C}$	$20\pm5.24^{\mathrm{dB}}$	$32\pm2.55^{cA}$
	Free_NLC-MNPs	$0\pm0^{\mathrm{aF}}$	$2\pm1.22^{\mathrm{cF}}$	$4\pm1.87^{ m dE}$	$10\pm0.00^{\mathrm{dD}}$	$15\pm0.00^{\rm cC}$	$27 \pm 1.22^{\mathrm{cB}}$	$47 \pm 2.55^{bA}$
48	Sesbania extract	$0\pm0^{\mathrm{aG}}$	$14\pm1.58^{\mathrm{cF}}$	$30 \pm 2.92^{\mathrm{cE}}$	$60 \pm 2.24^{cD}$	$85 \pm 4.47^{\rm bC}$	$96\pm2.45^{\mathrm{bB}}$	$100\pm0.00^{\mathrm{aA}}$
	Se-NLC	$0\pm0^{aE}$	$21 \pm 2.45^{bD}$	$45 \pm 2.55^{bC}$	$80\pm1.58^{ m bB}$	$100\pm0.00^{\mathrm{aA}}$	$100\pm0.00^{\mathrm{aA}}$	$100\pm0.00^{\mathrm{aA}}$
	Se-NLC-MNPs	$0\pm0^{aE}$	$25 \pm 2.45^{aD}$	$49 \pm 4.00^{aC}$	$88\pm2.55^{\mathrm{aB}}$	$100\pm0.00^{\mathrm{aA}}$	$100\pm0.00^{\mathrm{aA}}$	$100\pm0.00^{\mathrm{aA}}$
	Free- NLC	$0\pm0^{\mathrm{aG}}$	$2\pm1.22^{\mathrm{dF}}$	$6\pm1.00^{\mathrm{eE}}$	$13 \pm 2.00^{eD}$	$20 \pm 2.24^{\rm dC}$	$30\pm5.24^{\mathrm{dB}}$	$44\pm1.87^{\mathrm{cA}}$
	Free_NLC-MNPs	$0\pm0^{\mathrm{aG}}$	$3\pm1.22^{\mathrm{dF}}$	$9\pm1.00^{\mathrm{dE}}$	$18 \pm 2.55^{\mathrm{dD}}$	$26 \pm 3.67^{cC}$	$35 \pm 3.16^{cB}$	$48 \pm 3.74^{\text{bA}}$
Temphos (1 mg/L)		96						I

Solvent	Time (h)	Treatment	LC <sub>50</sub> (low-up)	LC <sub>90</sub> (low-up)	LC <sub>95</sub> (low-up)	Slope±SE	$X^2$ (sign.)
Methanol	24	Sesbania extract	161.10(140.80 - 183.43)	567.83 (474.59–707.40)	811.45 (656.58–1057.93)	$2.343 \pm 0.160$	4.613 (0.329)
		Se-NLC	101.8(89.53 - 114.82)	290.2 (245.39–360.54)	$390.66(319.64{-}509.51)$	$2.816 \pm 0.234$	5.217 (0.265)
		Se-NLC-MNPs	88.68 (77.87–100.42)	248.57 (210.35–309.51)	332.59(272.09-436.36)	$2.872 \pm 0.256$	2.306 (0.679)
		Free- NLC	4994.61 (2857.60–13,015.58)	79,831.64(25,448.06-617,345.98)	175,139.79 (47,016.16 $-1,854,634.93$ )	$1.064 \pm 0.156$	1.445(0.836)
		Free_ NLC-MNPs	2318.07 (1652.16–3814.09)	22,610.64(10,971.10-70,632.43)	$43,124.19(18,619.92{-}162,810.78)$	$1.295 \pm 0.152$	4.647 (0.325)
	48	Sesbania extract	130.23(116.66 - 146.65)	399.11 (346.48-471.61)	547.05(463.66-668.16)	$2.653 \pm 0.161$	5.855(0.210)
		Se-NLC	77.55 (68.56–86.67)	183.74 (156.39–230.35)	234.64 (193.06–311.03)	$3.421 \pm 0.361$	0.791(0.939)
		Se-NLC-MNPs	69.24 (63.63–78.72)	$151.02\ (132.33-180.37)$	$186.87\ (159.50-232.97)$	$3.927 \pm 0.387$	3.863 (0.424)
		Free- NLC	2366.34 (1632.77–4112.35)	$30,538.21(13,534.04{-}111,770.90)$	63,054.58(24,443.11-287,435.57)	$1.153 \pm 0.139$	$1.774\ (0.777)$
		Free_NLC-MNPs	1863.41 (1313.58 - 3091.21)	28,623.42(12,789.41-101,570.91)	62,092.43(24,128.78-276,182.60)	$1.080 \pm 0.126$	2.131 (0.711)
Water	24	Sesbania extract	262.81 (225.37–305.87)	$1298.38(1023.14{-}1754.15)$	2042.00(1535.87 - 2943.88)	$1.847 \pm 0.132$	1.660(0.797)
		Se-NLC	134.96 (118.36–152.97)	433.59 $(364.45 - 538.01)$	603.63 (491.52–783.73)	$2.528 \pm 0.183$	4.048(0.399)
		Se-NLC-MNPs	114.23 (100.80–128.67)	327.27(276.80-405.00)	441.04(361.54-571.47)	$2.803 \pm 0.221$	5.585 (0.232)
		Free- NLC	4994.61 (2857.60–13,015.58)	79,831.64(25,448.06-617,345.98)	175,139.79 (47,016.16 $-1,854,634.93$ )	$1.064 \pm 0.156$	1.445(0.836)
		Free_NLC-MNPs	2318.07 (1652.16–3814.09)	22,610.64(10,971.10-70,632.43)	$43,124.19(18,619.92{-}162,810.78)$	$1.295 \pm 0.152$	4.647 (0.325)
	48	Sesbania extract	160.13 (139.43 - 182.73)	586.88(488.74-734.54)	848.11 (682.95–1112.74)	$2.272 \pm 0.155$	2.070 (0.722)
		Se-NLC	101.16(89.55 - 113.46)	269.67 (229.15–333.23)	356.08 (293.26-461.23)	$3.009 \pm 0.255$	3.606(0.461)
		Se-NLC-MNPs	89.56 (7920–100.29)	229.32(195.54 - 283.14)	299.36 (247.31–388.18)	$3.138 \pm 0.284$	5.364(0.251)
		Free- NLC	2366.34(1632.77 - 4112.35)	$30,538.21(13,534.04{-}111,770.90)$	63,054.58(24,443.11-287,435.57)	$1.153 \pm 0.139$	1.774(0.777)
		Free_NLC-MNPs	1863.41 (1313.58 - 3091.21)	28,623.42(12,789.41-101,570.91)	62,092.43(24,128.78-276,182.60)	$1.080 \pm 0.126$	2.131(0.711)

**TABLE 3** | Lethal concentrations (ppm) of *Sesbania sesban* extracts and its nanoformulations against *Culex pipiens*, 24 and 48 h post-treatment.

				Concentrat	Concentration (mg/mL)		
Solvent	Treatment	Control	0.1	0.5	1.0	2.0	5.0
Methanol	Sesbania extract	$0.00\pm0.00^{\mathrm{aF}}$	$8.00 \pm 2.49^{cE}$	$26.67 \pm 2.98^{cD}$	68.00±2.49 <sup>cC</sup>	$88.00 \pm 1.33^{cB}$	$100.00 \pm 0.00^{aA}$
	Se-NLC	$0.00\pm0.00^{\mathrm{aF}}$	$14.66\pm1.33^{\mathrm{aE}}$	$44.00 \pm 3.4^{bD}$	$76.00 \pm 2.67^{bC}$	$97.33 \pm 1.63^{bB}$	$100.00 \pm 0.00^{aA}$
	Se-NLC-MNPs	$0.00\pm0.00^{\mathrm{aE}}$	$16.00\pm1.63^{\mathrm{aD}}$	$48.00 \pm 2.49^{aC}$	$84.00 \pm 4.00^{\mathrm{aB}}$	$100.00 \pm 0.00^{aA}$	$100.00 \pm 0.00^{aA}$
	Free- NLC	$0.00\pm0.00^{\mathrm{aE}}$	$0.00\pm0.00^{\mathrm{dE}}$	$4.00\pm1.63^{dD}$	$9.33 \pm 1.63^{dC}$	$14.67\pm2.49^{\mathrm{eB}}$	$24.00 \pm 1.63^{cA}$
	Free_NLC-MNPs	$0.00\pm0.00^{\mathrm{aE}}$	$0.00\pm0.00^{\mathrm{dE}}$	$5.33 \pm 2.49^{dD}$	$10.67 \pm 1.63^{dC}$	$21.33 \pm 3.89^{\mathrm{dB}}$	$32.00 \pm 2.49^{bA}$
Water	Sesbania extract	$0.00\pm0.00^{\mathrm{aF}}$	$5.33 \pm 2.49^{bE}$	$16.00 \pm 2.67^{\mathrm{bD}}$	$28.00 \pm 1.33$ <sup>cC</sup>	$56.00 \pm 2.67^{\mathrm{cB}}$	$80.00\pm2.11^{\mathrm{cA}}$
	Se-NLC	$0.00\pm0.00^{\mathrm{aF}}$	$8.00\pm1.33^{\rm aE}$	$28.00 \pm 2.49^{aD}$	$48.00 \pm 2.49^{bC}$	$84.00 \pm 2.67^{bB}$	$96.00 \pm 1.63^{\mathrm{bA}}$
	Se-NLC-MNPs	$0.00\pm0.00^{\mathrm{aF}}$	$9.33\pm1.63^{aE}$	$30.67 \pm 3.4^{aD}$	$60.00 \pm 2.98^{\mathrm{aC}}$	$88.00 \pm 2.49^{aB}$	$100.00 \pm 0.00^{aA}$
	Free- NLC	$0.00\pm0.00^{\mathrm{aE}}$	$0.00\pm0.00^{\mathrm{cE}}$	$4.00\pm1.63^{\mathrm{cD}}$	$9.33 \pm 1.63^{dC}$	$14.67\pm2.49^{\mathrm{eB}}$	$24.00 \pm 1.63^{\mathrm{eA}}$
	Free_NLC-MNPs	$0.00\pm0.00^{\mathrm{aE}}$	$0.00\pm0.00^{\mathrm{cE}}$	$5.33 \pm 2.49^{cD}$	$10.67 \pm 1.63^{dC}$	$21.33 \pm 3.89^{\mathrm{dB}}$	$32.00 \pm 2.49^{dA}$
	Temphos (1 mg/L)	75					

**TABLE 4**The larvicidal effects of Sesbania sesban extracts and its nanoformulations Musca domestica, 24 h post-treatment.

 $({\rm Fe_3O_4}).$  These broad peaks are indicative of nanoscale particles, as smaller particle sizes typically result in peak broadening due to the reduced crystallite size.

In comparison, the XRD pattern of the Se-NLC-MNPs sample showed similar peaks corresponding to magnetite, confirming that the crystalline structure is retained even after loading with *Sesbania* extract. However, the Se-NLC-MNPs exhibit broader peaks compared to the free nanoparticles, which indicate a smaller crystallite size. The broader peaks suggest that the nanoparticles in the loaded sample have reduced particle size. However, the peaks in this sample are more intense than those of the free nanoparticles, suggesting enhanced crystallinity and reduced surface defects.

This may indicate that the *Sesbania* extract and other NLC components act as stabilizing agents, leading to more ordered crystallite structures. Additionally, the increase in peak intensity may be due to the improved dispersion of the nanoparticles within the extract, reducing particle aggregation and promoting crystallite growth. The successful incorporation of Sesbania extract into the magnetite nanoparticles without disrupting the crystalline structure is promising for applications such as insecticidal, where the magnetic properties of magnetite play a key role. The enhanced crystallinity in the Se-NLC-MNPs sample could result in improved magnetic performance and stability, which is critical for its effectiveness in its biological importance as insect (Farooq et al. 2022).

Research on the application of botanicals, such as essential oils and plant extract, against pests is an encouraging field (Ebeed et al. 2024; Baz et al. 2022). This study looked at how *S. sesban* extract and its nanoparticles killed third *Cx. pipiens* larvae. All formulations, including aqueous plant extracts, methanol, and nanoparticles, exhibited the highest rates of larval mortality. But when used in small amounts, Se-NLC and Se-NLC-MNP nanoparticles were better at killing mosquito larvae than Se-extract, whether the solution was methanol or water. Furthermore, our studies show that Free-NLC is not hazardous to mosquito larvae.

What we found was similar to what Radwan et al. (2024) showed that peppermint and jasmine nanoparticles worked better than essential oils against *Cx. pipiens* larvae in their second and fourth instars when used at 2000 ppm. This study showed that Se-NLC and Se-NLC-MNP nanoparticles had strong insecticidal activity. This was similar to what other studies had found the nanoparticles killed mosquito larvae just as well as plant extracts or essential oils by themselves (Radwan, Baz, Khater, and Selim 2022; Radwan, Baz, Khater, Alkhaibari, and Selim 2022). Hikal et al. (2017) evaluated the effectiveness of essential oils and their nanoparticles in a study that was like ours. They showed that patchouli ( $LC_{50} = 93.05$  ppm) and honeysuckle ( $LC_{50} = 88.30$  ppm) nanoparticles were much more effective at killing mosquito larvae than the bulk oils ( $LC_{50} = 276.29$  ppm and 247.72 ppm, respectively).

within the same row that have the same superscript letter.

Aphis gossypii, the cotton aphid, was very hurt by Mentha piperita nanoparticles ( $LC_{50} = 3879.5 \pm 16.2 \mu L/L$ ) (Heydari et al. 2019). According to a study, jasmine nanoformulations effectively kill *Tetranychus urticae*, a two-spotted spider mite, with a death rate of 68.50% and a decrease rate of 49.03% (Farouk et al. 2021).

TABLE 5 | Lethal concentrations (ppm) of Sesbania sesban extracts and its nanoformulations against Musca domestica, 24h post-treatment.

Solvent	Treatment	LC <sub>50</sub> (low-up)	LC <sub>90</sub> (low-up)	LC <sub>95</sub> (low-up)	Slope ± SE	X <sup>2</sup> (sign.)
Methanol	Sesbania extract	0.65 (0.28–1.13)	2.39 (1.90-9.23)	3.47 (3.08–17.65)	$2.260 \pm 0.177$	16.640 (0.000)
	Se-NLC	0.42 (0.18-0.71)	1.71 (1.21–5.43)	2.53 (1.97–10.17)	$2.118 \pm 0.163$	14.419 (0.002)
	Se-NLC-MNPs	0.38 (0.19-0.64)	1.78 (1.25-5.38)	2.74 (2.01–10.53)	$1.939 \pm 0.166$	9.871 (0.019)
	Free- NLC	19.91 (10.05–76.47)	300.12 (77.72–4770.97)	647.56 (138.00–15,485.93)	$1.087 \pm 0.193$	0.935 (0.816)
	Free_NLC-MNPs	10.57 (6.59–23.66)	116.67 (43.87–683.18)	230.46 (74.55–1784.59)	$1.228 \pm 0.184$	1.205 (0.751)
Water	Sesbania extract	1.76 (1.46–2.16)	11.27 (7.76–18.93)	19.08 (12.22–35.76)	$1.587 \pm 0.147$	7.407 (0.060)
	Se-NLC	0.81 (0.44–1.35)	3.72 (2.73–10.98)	5.73 (4.36-20.8)	$1.939 \pm 0.146$	11.632 (0.008)
	Se-NLC-MNPs	0.65 (0.30-1.13)	2.64 (2.03-9.73)	3.92 (3.33–18.88)	$2.111 \pm 0.165$	15.228 (0.001)
	Free- NLC	19.91 (10.05–76.47)	300.12 (77.72–47770.97)	647.56 (138.00–15485.93)	$1.087 \pm 0.193$	0.935 (0.816)
	Free_NLC-MNPs	10.57 (6.59–23.66)	116.67 (43.87–683.18)	230.46 (74.55–1784.59)	$1.228 \pm 0.184$	1.205 (0.751)

The *Sesbania* extract is poisonous to mosquito and fly larvae, but it also kills other insects very well, such as the stored grain pest *Sitophilus oryzae* (SaM and Al-Moajel 2005), the *Trogoderma granarium* betlees (Sahi 2020), the red flour beetle *Tribolium castaneum* (Taha Al-Abdalli et al. 2024), and the diamondback moth *Plutella xylostella* (Suripto et al. 2017). Egyptian riverhemp, *S. sesban* extract nanoparticles may be a viable way to manage mosquito larvae. In addition to its toxicity towards insects and parasitic organisms, the *Sesbania* extract also effectively regulates a variety of microbes.

Combining oil, water, and surfactants creates colloidal systems known as nanoemulsions, where droplets of one liquid disperse within another at a nanoscale. Their better stability, larger surface area for interacting with target species, and increased bioavailability of active chemicals are just a few of the benefits they offer for pesticide administration. Because of their high delivery and low cost, target action, chemical stability, water dispersal, and low ecological toxicity, these Se-nanoparticles make excellent insecticides. It is possible to draw the conclusion that nanopesticides will usher in a new era of environmentally acceptable, high-performing mosquito-borne illness control solutions. For further research, we recommend applying the applicable nanoparticles in the field and conducting ecotoxicological analyses.

Researchers have studied many natural plant compounds as insecticides against mosquitoes and houseflies (Arivoli et al. 2016; Mohamed et al. 2024) because they are safe for mammals and have the potential to replace conventional pesticides (Şengül Demirak and Canpolat 2022). Most of these investigations have reported the larvicidal activity of the extracts and occasionally discovered the presence of a variety of compounds, but only a few have actually identified the responsible compounds and their structures (Lim et al. 2023). Different types of plants in the *Solanaceae*, *Asteraceae*, *Fabaceae*, *Labiatae*, *Miliaceae*, *Oocystaceae*, and *Rutaceae* families can kill, repel, or kill mosquito and housefly larvae in different ways (Ebadollahi et al. 2020; Singh et al. 2023). Currently, numerous researchers are searching for locally accessible plant materials to identify environmentally friendly products that can effectively control various mosquito species (Aminu et al. 2024).

The pharmacological effects of medicinal plants are caused by a variety of compounds called secondary metabolites, which are produced by living things called plants. These chemicals include alkaloids, polysaccharides, flavonoids, saponins, tannins, and terpenoids. At the insect level, pests or animals that consume or come into contact with any of the many diverse groups of bioactive chemicals present in or derived from botanical insecticides may suffer adverse consequences (Roy et al. 2022).

Several additional plant families, including the Fabaceae family, have shown adverse effects on different species of mosquitoes (Baeshen and Baz 2023). Researchers have experimented with a variety of botanical remedies as one of the most successful vector control strategies, including mosquitoes, before the development of chemical insecticides (Samada and Tambunan 2020). In addition, the positive reception of phytochemicals from the public, their increasing popularity, and their nontoxic properties create new opportunities for research and development in the field of botanical insecticides.

This study investigated the organic components in methanol and aqueous extracts of *S. sesban*. The results showed a higher concentration of fatty acids and terpenes and a wider range of different organic components in the methanol extract, such as phytosteroid, alkane, and phenols. On the other hand, most phytochemical compounds in the aqueous extract were fatty acids.

Some studies have shown that the polarity of extraction solvents affects the content of phytochemicals and extraction yield (Nawaz et al. 2020). Therefore, differences in solvent polarity explain the variation in the amount of biologically active

No.	RT	Area %	Compound name	MF	MW
			Phenols (3.42)		
2	14.51	0.65	Benzene, 1,2-dimethoxy-4-(1-propenyl)—	$C_{11}H_{14}O_2$	178
4	16.42	2.77	3-Buten-2-one, 4-(2,6,6-trimethyl-1-cyclohexen-1-yl)—	$C_{13}H_{20}O$	192
			Cyclohexane (2.13)		
15	24.50	2.13	Mome inositol	$C_7 H_{14} O_6$	194
			Faty acids and esters (35.79)		
7	19.98	2.36	2 h-Pyran, 2-(7-heptadecynyloxy)tetrahydro—	$C_{22}H_{40}O_2$	336
11	22.65	2.06	13,16-Octadecadiynoic acid, methyl ester	$C_{19}H_{30}O_2$	290
18	26.95	7.63	Hexadecanoic acid	$C_{16}H_{32}O_{2}$	256
22	29.46	3.09	10-Octadecenoic acid, methyl ester	$C_{19}H_{36}O_2$	296
26	30.65	17.86	Octadecanoic acid	$C_{18}H_{36}O_2$	284
28	35.06	2.79	Ethanol, 2-(9-octadecenyloxy)-, (z)—	$C_{20}H_{40}O_2$	312
			Glycoside (0.66)		
32	36.62	0.66	Rhamnetin-3-O-glucoside	$C_{27}H_{30}O_{16}$	610
			Acylaminosugars (0.75)		
12	22.88	0.75	á-d-Glucopyranose, 4-o-á-d-galactopyranosyl—	$C_{12}H_{22}O_{11}$	342
			Phytosteroid (14.92)		
33	38.82	1.42	Pseduosarsasapogenin-5,20-dien	$C_{27}H_{42}O_{3}$	414
36	39.53	5.50	Ethyl iso-allocholate	$C_{26}H_{44}O_5$	436
38	39.81	8.00	Squalene	$C_{30}H_{50}$	410
			Alkane (8.80)		
34	38.98	8.80	17-Pentatriacontene	$C_{35}H_{70}$	490
			Terpene (monoterpene and sesquiterpene) (33.44)		
5	19.11	3.26	1,1,4,7-Tetramethyldecahydro-1 h-cyclopropa[e]azulen-4-ol	$C_{15}H_{26}O$	222
19	28.54	9.53	1-Naphthalenepropanol,.alp	$C_{20}H_{34}O$	290
23	29.65	15.35	Phytol	$C_{20}H_{40}$	296
42	40.75	5.30	3-(acetyloxy)-14à,15à-epoxy-5à-cholest-8-en-7-one Polyketides	$C_{29}H_{44}O_{4}$	456

**TABLE 7** I
 The major chemical constituents of Sesbania sesban aqueous extract.

No.	RT	Area %	Compound name	MF	MW
			Fatty acid and esters		
1	25.67	0.51	Palmitic acid methyl ester	$C_{17}H_{34}O_2$	270
2	28.68	5.27	9,12-Octadecadienoic acid (z,z)-, methyl ester	$C_{19}H_{34}O_2$	294
3	28.86	5.40	9-Octadecenoic acid, methyl ester	$C_{19}H_{36}O_2$	296
4	29.42	2.15	Methyl stearate	$C_{19}H_{38}O_2$	298
5	42.65	9.13	9-Octadecenoic acid (z)—	$C_{18}H_{34}O_2$	282
6	44.19	57.04	Erucic acid	$\mathrm{C_{22}H_{42}O_2}$	338
7	45.11	20.50	(z)-(z)-docos-13-en-1-yl icos-11-enoate	$C_{42}H_{80}O_{2}$	616
		100			

compounds in the sesban extract. This is due to the fact that plant components contain large amounts of polar molecules, which dissolve in highly polar liquids. In a different study parallel to our present work, we found that acetone extract contains more phenols, alkanes, flavonoids, terpenes, and ketones compared to the aqueous extract (Baz, Mostafa, et al. 2024).

Plant extracts contain many physiologically active, and these compounds have important environmental activities in nature, such as insect attractants, insect repellents, insect growth regulators, nematicides, fungicides, antiparasitic, and allelopathic agents. They also serve as a promising source of new pest control agents or biopesticides, so the plants are still an indispensable treasure of nature in our daily lives (Kumar et al. 2021; Muhammed et al. 2022; Chauhan et al. 2024).

Using gas chromatography and mass spectrometry, scientists found that myo-inositol, 4-C-methyl, and neophytadiene are chemicals in *S. sesban* plants that help fight malaria. Our results are consistent with previous studies that support methanol as the best solvent for obtaining more potent compounds from several medicinal plants (Mohanty et al. 2014). Furthermore, the results showed that methanol extracts of plant materials were more harmful to *Cx. pipiens* larvae than water-based extracts (Abo El-Kassem Bosly 2022). Some phytochemicals, like flavonoids, alkaloids, terpenoids, steroids, tannins, glycosides, and others, were found in the ethanolic extracts of *Sesbania* bark and *Carica papaya* leaves (Fatmawaty et al. 2017).

Phytochemicals compounds could interfere with a variety of processes or pathways, including the insect nervous system. If this occurs, it can disrupt nerve function, making it difficult for insects to move, eat, or reproduce (Ağuş 2021); prevent them from feeding (Larson et al. 2020); prevent them from making chitin, an important part of their exoskeleton; prevent them from inhibiting cholinesterase activity (Liu et al. 2021); disrupt cell membranes, which can cause cell leakage and death (Srivastava and Singh 2019); or act as a repellent, changing their behavior and scaring them away. Pani et al. (2020) said that myo-inositol and 4-C-methyl can help with hair loss, liver disease, nerve pain, cancer, high cholesterol, fatty effects, and improving the taste of things.

## 5 | Conclusion

The need for environmentally sustainable and creative vector control techniques is growing. The current insecticidal properties could be enhanced via nanotechnology. Enhancing plant extracts, essential oils, and other materials' physical, chemical, and biological characteristics improves them. Utilizing lesser doses and lowering the concentration of pesticides improves their effectiveness and lessens their negative environmental consequences. The results of the present investigation showed that polonium and its nanoformulations have significant insecticidal effects on *Cx. pipiens* and *M. domestica* larvae. It is intriguing to note that Se-NLC and Se-NLC-MNPs nanoparticles have encouraging insecticidal properties. Because they are cheap, easily to be dispersed in water, have a targeted effect, are highly conductive, and are not harmful to the environment, these nanoparticles are great insecticides that are also safe for

the environment. We conclude that nanopesticides will usher in a new era of environmentally acceptable and high-performance solutions for the control and suppression of mosquito-borne diseases. We recommend continued field testing and ecotoxicological investigations.

#### Consent

The authors have nothing to report.

#### **Conflicts of Interest**

The authors declare no conflicts of interest.

#### Data Availability Statement

The datasets used and/or analyzed during the current study are available from the corresponding author upon reasonable request.

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