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Original Research Paper

Veterinary Entomology and Public Health: Mitigating the Impact of Arthropod Vectors on Animal Health

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Abstract: The development of nano-emulsions represents a significant advancement in enhancing the efficacy of botanical insecticides, offering a promising alternative to synthetic pesticides. Botanical insecticides, derived from natural plant sources, often face challenges such as poor stability, rapid degradation, and limited bioavailability, which hinder their effectiveness in agricultural applications. Nano-emulsions, characterized by droplet sizes ranging from 20 to 200 nanometers, enhance the solubility, stability, and controlled release of active ingredients, thereby addressing these challenges effectively. This paper explores the formulation, characterization, and efficacy of nano-emulsified botanical insecticides against common agricultural pests. Laboratory and field trials demonstrate that nano-emulsions significantly improve the mortality rates of pests, with improvements ranging from 32% to 33% compared to conventional formulations. The frequency of application is reduced by 40% to 60%, highlighting the cost-effectiveness and practicality of these formulations for farmers. Environmental toxicity studies reveal that nano-emulsified insecticides pose minimal risks to non-target organisms, showcasing their potential for sustainable pest management. Despite these advancements, challenges such as production scalability and regulatory approval remain. Future research will focus on optimizing formulations and assessing long-term environmental impacts to ensure the safe and effective use of nano-emulsified botanical insecticides in integrated pest management strategies. Overall, this paper underscores the role of nano-emulsions in revolutionizing pest control practices and promoting environmentally sustainable agricultural systems.

Keywords: Nano-Emulsions, Insecticides, Botanical, Pest Control, Bioavailability, Stability, Emulsifiers, Toxicity, Eco-Friendly, Solubility, Scalability, Surfactants, Bio efficacy.

I. Introduction

The increasing concerns over the environmental and health impacts of synthetic chemical pesticides have spurred the search for safer, more sustainable alternatives. Botanical insecticides, derived from plants, have gained significant attention in this context as they offer a natural, eco-friendly option for pest control. These botanical insecticides include compounds like essential oils, alkaloids, terpenes, and phenolics, extracted from various plant species. They possess insecticidal properties that disrupt the physiological processes of pests, causing mortality or deterring feeding and reproduction [1]. Despite their promising potential, the widespread use of botanical insecticides has been limited by several challenges, including poor stability, rapid degradation, and limited bioavailability in environmental conditions, such as exposure to

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sunlight and rain. This has led to inconsistent efficacy in the field, where they often fail to deliver results comparable to synthetic chemicals. Nano-emulsion technology has emerged as a novel solution to overcome these limitations and enhance the effectiveness of botanical insecticides. Nano-emulsions are fine oil-in-water or water-in-oil dispersions where the droplet size ranges from 20 to 200 nanometers [2]. This small droplet size provides several advantages, including increased surface area, improved solubility of hydrophobic compounds, enhanced stability, and better control over the release of active ingredients. In agricultural pest management, these features are particularly valuable, as they lead to more consistent and sustained insecticidal activity, reduced environmental impact, and decreased need for frequent application. The ability of nano-emulsions to improve the penetration of active ingredients into insect cuticles and plant tissues further enhances their bioefficacy, offering a significant step forward in the development of botanical insecticides [3]. The application of nano-emulsions in the formulation of botanical insecticides represents a key innovation in sustainable pest control strategies. Traditional botanical insecticides are often limited by their hydrophobic nature, which makes them poorly soluble in water, leading to difficulties in formulation and inconsistent delivery. Many botanical insecticidal compounds also degrade rapidly when exposed to environmental conditions such as ultraviolet (UV) light, oxygen, and varying temperatures [4]. Nano-emulsions help to overcome these obstacles by encapsulating the active ingredients within nano-sized droplets, which protect them from environmental degradation and ensure a more controlled and targeted release. This encapsulation enhances the shelf-life of botanical insecticides and reduces the volatility of compounds like essential oils, which are prone to rapid evaporation. As a result, nano-emulsion-based formulations can provide more prolonged and effective pest control compared to conventional botanical insecticides [5]. Another significant advantage of nano-emulsions is their ability to improve the absorption and uptake of insecticidal compounds by pests. The small droplet size of nano-emulsions allows for better interaction with insect cuticles, increasing the penetration of the active ingredient into the insect's body. This improved penetration not only enhances the speed and efficacy of the insecticidal action but also helps reduce the amount of active ingredient required to achieve the desired pest control effect [6]. This is particularly important for reducing the overall pesticide load in the environment, minimizing potential harm to non-target organisms, and lowering the cost of pest control for farmers. The increased bioavailability of nano-emulsified botanical insecticides reduces the frequency of application, making them more practical and economically feasible for large-scale agricultural use. The process of developing nano-emulsions for botanical insecticides involves selecting appropriate plant-based compounds and optimizing their formulation for maximum efficacy [7]. Nano-emulsions can be produced using either high-energy or low-energy techniques, with the choice of method depending on the specific properties of the active ingredient and the desired characteristics of the final product. High-energy methods, such as ultrasonication and high-pressure homogenization, involve the application of mechanical forces to break down the oil and water phases into smaller droplets.

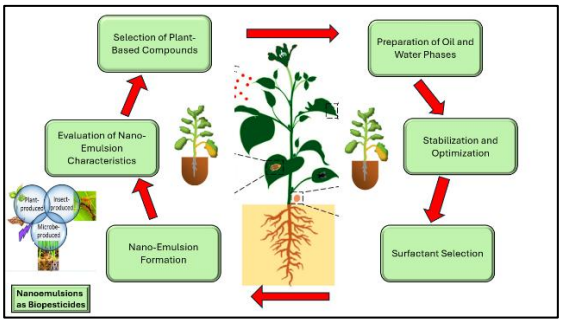


Figure 1. Schematic of Nano-Emulsion Formation Process

These methods are particularly useful for producing highly stable nano-emulsions with uniform droplet sizes. Low-energy methods, such as phase inversion temperature and spontaneous emulsification, rely on the physicochemical properties of the system to form nano-emulsions without the need for excessive mechanical energy [8]. These methods can be more cost-effective and energy-efficient, making them attractive for large-scale production. The formulation of nano-emulsions also requires the careful selection of surfactants and emulsifiers, which play a critical role in stabilizing the emulsion and preventing the coalescence of droplets. The choice of surfactant depends on factors such as the hydrophilic-lipophilic balance (HLB) of the system, the nature of the active ingredient, and the desired stability of the final product [9]. An optimal surfactant system ensures the long-term stability of the nano-emulsion, preventing phase separation and maintaining the efficacy of the botanical insecticide over time. The stability of nano-emulsions is influenced by factors such as temperature, pH, and ionic strength, which need to be carefully controlled during the formulation process to ensure the robustness of the product under various environmental conditions. The development of nano-emulsion technology for botanical insecticides represents a significant advancement in the field of agricultural pest management. By addressing the challenges associated with the instability and limited bioavailability of traditional botanical insecticides, nano-emulsions offer a more effective, reliable, and

environmentally friendly solution for controlling pests. As the demand for sustainable agricultural practices continues to grow, nano-emulsion-based botanical insecticides have the potential to play a crucial role in reducing reliance on synthetic pesticides, promoting biodiversity, and safeguarding the health of ecosystems (As presented in Figure 1 above). Furthermore, ongoing research and development efforts are expected to lead to even more innovative formulations, with the possibility of incorporating synergistic botanical compounds and other active ingredients to further enhance the efficacy and versatility of nano-emulsified insecticides.

II. Conceptual Models and their Development in the Literature

The application of nanoemulsions has gained significant attention in various fields, particularly in pharmaceuticals, cosmetics, and food preservation. One notable study developed nanoemulsions containing Cymbopogon flexuosus essential oil, focusing on the formulation, characterization, stability, and evaluation of antimicrobial and antibiofilm activities [10]. This research highlighted the potential for using plant-derived essential oils in medical applications, demonstrating that these nanoemulsions possess significant stability and efficacy against biofilm formation, indicating their potential use in clinical settings to combat microbial infections. Another study explored the anticancer effects of polymer micelle-encapsulated quercetin on ovarian cancer, revealing that encapsulation enhanced its bioavailability and therapeutic efficacy, suggesting that nanoformulations could serve as a promising strategy for delivering poorly soluble anticancer agents [11]. Research on nano-polymeric micelles for gene delivery to the cornea underscored the versatility of nanoemulsions in gene therapy, facilitating targeted gene delivery crucial for treating ocular diseases effectively. In the realm of natural resources, a review of plant resources available in the Restinga de Carapebus shed light on the potential of local flora for developing functional nanoemulsions [12]. Studies on the anticholinesterase activity of Manilkara subsericea triterpenes suggested the potential for developing nanoemulsions from these plant extracts to address neurological conditions. The agricultural sector has also benefitted from advancements in nanoemulsion technology, with discussions on creating potent insecticides to enhance pest management strategies while minimizing environmental impact. Research focused on designing a self-nanoemulsifying drug delivery system optimized to improve drug solubility and absorption, which is crucial for enhancing therapeutic effectiveness [13]. Current opinions on nanoemulsions emphasize their unique properties and applications across various industries. In cosmetics, formulations of palm kernel oil esters-based nanoemulsions demonstrated efficacy in delivering skin anti-aging agents. This trend continued with studies on hydroalcoholic extracts from Vellozia squamata, exploring their potential pharmaceutical and cosmetic applications, as well as the production of cosmetic nanoemulsions containing extracts from Opuntia ficus-indica, showcasing their moisturizing properties [14]. Another study developed an azelaic acid-loaded nanoemulsion, emphasizing a novel strategy for treating hyperpigmentary skin disorders. An extensive overview of nanoemulsions for food applications discussed their properties, production techniques, characterization, and various applications in food technology. Research highlighted the use of cinnamon oil nanoemulsion as a natural preservative for prolonging the shelf life of chilled fish fillets, demonstrating the potential of natural nanoemulsions in food preservation [15]. Finally, an investigation into the efficacy of a Myristica fragrans essential oil nanoemulsion against postharvest biodeterioration reinforced the importance of nanoemulsions in enhancing food safety and quality. Overall, the literature reflects the diverse applications of nanoemulsions across various fields, from enhancing drug delivery and antimicrobial efficacy to improving cosmetic formulations and food preservation, with ongoing research exploring their potential to address challenges in health, agriculture, and the food industry.

Author & Year	Area	Methodology	Key Findings	Challenges
Da Silva Gündel et al. (2018) [10]	Antimicrobial Activities	Development and characterization of nanoemulsions containing essential oil	Nanoemulsions showed stability and significant antimicrobial and antibiofilm activities.	Limited clinical application data.
Gao et al. (2012) [11]	Cancer Treatment	Polymer micelle encapsulation of quercetin	Enhanced bioavailability and therapeutic efficacy of quercetin against ovarian cancer.	Need for extensive in vivo studies.
Tong et al. (2012) [12]	Gene Therapy	Nano-polymeric micelle formulation for gene delivery	Facilitated targeted gene delivery to the cornea,	Challenges in long-term gene expression.

			indicating potential for ocular disease treatment.	
Santos et al. (2009) [13]	Natural Resources	Review of plant resources from Restinga de Carapebus	Highlighted the potential of local flora for developing functional nanoemulsions.	Lack of standardized extraction methods.
Fernandes et al. (2011) [14]	Neurology	Study on anticholinesterase activity of plant triterpenes	Demonstrated anticholinesterase activity, suggesting potential for neurological condition treatment.	Variability in plant composition affecting efficacy.
Margulis-Goshen & Magdassi (2013)	Agriculture	Discussion on nanotechnology applications in insecticides	Nanotechnology can enhance the effectiveness of insecticides while reducing environmental impact.	Regulatory challenges for new pesticide formulations.
Wang et al. (2009) [15]	Drug Delivery	Design and optimization of a self-nanoemulsifying drug delivery system	Improved solubility and absorption of drugs through optimized nanoemulsion formulations.	Complexity in the formulation process.

Table 1. Summarizes the Literature Review of Various Authors

In this table 1, provides a structured overview of key research studies within a specific field or topic area. It typically includes columns for the author(s) and year of publication, the area of focus, methodology employed, key findings, challenges identified and potential applications of the findings. Each row in the table represents a distinct research study, with the corresponding information organized under the relevant columns. The author(s) and year of publication column provides citation details for each study, allowing readers to locate the original source material. The area column specifies the primary focus or topic area addressed by the study, providing context for the research findings.

III. Selection of Botanicals for Nano-Emulsion Formulation

The selection of botanicals for nano-emulsion formulation is a critical step in the development of effective and sustainable insecticidal solutions. Botanical insecticides, derived from plant extracts, have garnered significant attention due to their natural origin, biodegradability, and minimal environmental impact. However, their efficacy in pest control is highly dependent on the choice of plant-based compounds and the optimization of their formulation. This section explores the criteria for selecting botanicals, the chemical composition of commonly used plant extracts, and the implications of these choices for nano-emulsion technology. The primary criteria for selecting botanicals for nano-emulsion formulations involve their insecticidal properties, compatibility with nano-emulsion systems, and environmental safety. Insecticidal efficacy is the foremost consideration, with a focus on compounds that exhibit strong pesticidal activity against target pests. Botanicals containing bioactive molecules such as alkaloids, phenolics, terpenes, and essential oils are often preferred due to their ability to interfere with the physiological and behavioral processes of insects.

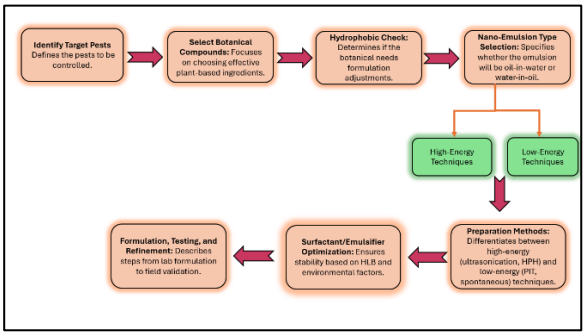


Figure 2. Structural Representation of Nano-Emulsion Droplets

For example, neem oil, rich in azadirachtin, is known for its antifeedant and growth-inhibiting properties, while citronella oil is recognized for its strong repellent effects. Compatibility with nano-emulsion systems is another crucial factor. Botanicals selected for nano-emulsion formulations should possess hydrophobic or amphiphilic characteristics to facilitate their incorporation into oil-in-water or water-in-oil systems. Essential oils, which are predominantly hydrophobic, are ideal candidates as their lipophilic nature allows them to form stable emulsions with the help of surfactants. Additionally, the stability of the botanical compounds under processing conditions, such as high-pressure homogenization or ultrasonication, must be considered to ensure their efficacy is retained during formulation. Environmental safety is equally important as depicted in figure 2. Botanicals chosen for nano-emulsion formulations should have minimal toxicity to non-target organisms, including beneficial insects, soil microbiota, and aquatic species. This aligns with the broader goal of

sustainable pest management and the reduction of collateral environmental damage. Moreover, the botanicals should exhibit low persistence in the environment, ensuring that they degrade into non-toxic byproducts over time.

A. Commonly Used Botanicals in Nano-Emulsion Formulations

Several plant-based compounds have been extensively studied for their potential in nano-emulsion formulations. These include essential oils, plant alkaloids, and phenolic compounds, each of which offers unique advantages for pest control.

a) **Neem Oil (Azadirachta indica):**

Neem oil is one of the most widely used botanical insecticides, known for its broad-spectrum activity against a variety of pests. Its active component, azadirachtin, disrupts the hormonal balance in insects, inhibiting their growth and reproduction. When formulated as a nano-emulsion, neem oil demonstrates enhanced stability and prolonged insecticidal activity, making it a valuable tool for integrated pest management.

b) Citronella Oil (Cymbopogon spp.):

Extracted from the leaves of citronella grass, citronella oil is renowned for its repellent properties against mosquitoes, flies, and other insects. The incorporation of citronella oil into nano-emulsions enhances its volatility control, ensuring sustained release and extended effectiveness in repelling pests.

c) Clove Oil (Syzygium aromaticum):

Clove oil contains eugenol, a compound with strong insecticidal and antifungal properties. Its effectiveness against pests such as aphids, caterpillars, and termites make it an excellent candidate for nano-emulsion formulations. Additionally, nano-emulsified clove oil has been shown to exhibit improved penetration into pest cuticles, increasing its bio efficacy.

d) Eucalyptus Oil (Eucalyptus spp.):

Eucalyptus oil is rich in terpenes such as cineole and pinene, which possess repellent and toxic effects against various pests. Nano-emulsified eucalyptus oil offers advantages in terms of stability and targeted delivery, reducing the frequency of applications needed for effective pest control.

e) Peppermint Oil (Mentha piperita):

Peppermint oil, containing menthol and menthone, is another effective botanical insecticide. It acts as a natural repellent and has shown efficacy against pests like ants, cockroaches, and mosquitoes. Nano-emulsion technology enhances the dispersion and persistence of peppermint oil, making it more efficient in agricultural settings. The choice of botanicals has a profound impact on the design and performance of nano-emulsion formulations. Each botanical extract presents unique challenges and opportunities in terms of solubility, stability, and bioactivity. Nano-emulsion technology addresses these challenges by optimizing the delivery and efficacy of the active ingredients through encapsulation in nano-sized droplets.

The small droplet size in nano-emulsions increases the surface area of the botanical compounds, improving their interaction with pests and enhancing their bioavailability. This is particularly advantageous for hydrophobic compounds, which often exhibit poor solubility in conventional formulations. Nano-emulsions facilitate the dispersion of these compounds in aqueous systems, ensuring uniform application and better coverage on plant surfaces.The stability of botanical insecticides is another critical aspect influenced by nano-emulsion technology. Many plant-based compounds are volatile and susceptible to degradation under environmental conditions such as heat, light, and oxygen. Encapsulation within nano-emulsions provides a protective barrier, shielding the active ingredients from environmental stressors and extending their shelf life. For instance, essential oils like citronella and clove, which are prone to rapid evaporation, benefit significantly from nano-emulsion formulations that control their release rate and volatility.

IV. Methods of Nano-Emulsion Preparation

The preparation of nano-emulsions is a sophisticated process that requires careful selection of techniques and materials to achieve stable and efficient formulations. The choice of preparation method is influenced by the nature of the botanical active ingredients, desired droplet size, stability requirements, and intended application. Nano-emulsions can be broadly prepared using high-energy and low-energy methods, each offering unique advantages and challenges. This section explores these methodologies, focusing on their principles, processes, and suitability for formulating nano-emulsified botanical insecticides. The selection of botanicals determines the type and concentration of surfactants and emulsifiers needed for nano-emulsion preparation. The hydrophilic-lipophilic balance (HLB) of the botanical compounds guides the choice of surfactants, ensuring the formation of stable emulsions with minimal phase separation. The compatibility of the botanicals with these stabilizing agents influences the overall performance and longevity of the nano-emulsion. The selection of botanicals for nano-emulsion formulations is not only a technical consideration but also a response to the growing demand for environmentally sustainable pest

management solutions. By prioritizing plant-based compounds with low toxicity and high biodegradability, researchers aim to develop formulations that reduce the ecological footprint of agricultural practices. Nano-emulsified botanical insecticides align with the principles of integrated pest management, offering targeted action against pests while minimizing harm to non-target species and ecosystems. The selection of botanicals for nano-emulsion formulations is a multifaceted process that balances the efficacy, compatibility, and environmental impact of the active ingredients. The integration of nano-emulsion technology enhances the potential of botanical insecticides, addressing their inherent limitations and unlocking new possibilities for sustainable agriculture. As research continues to expand the repertoire of botanical compounds and optimize their formulations, nano-emulsions are poised to play a pivotal role in advancing eco-friendly pest management strategies.

A. High-Energy Techniques

High-energy techniques rely on mechanical forces to reduce droplet sizes and create stable nano-emulsions. These methods are particularly effective for producing emulsions with uniform droplet distribution and high stability. Common high-energy techniques include ultrasonication and high-pressure homogenization.

- **Ultrasonication**

Ultrasonication uses ultrasonic waves to generate intense cavitation forces that break down oil and water droplets into nano-sized particles. This technique is highly effective in producing emulsions with droplet sizes below 100 nanometers. Ultrasonication is particularly suitable for botanical insecticides containing essential oils, as it ensures the fine dispersion of hydrophobic compounds in the aqueous phase. The process involves exposing a mixture of oil, water, and surfactant to ultrasonic waves in a sonicator. The energy input and processing time can be optimized to achieve the desired droplet size and stability. Ultrasonication is a scalable method that can be adapted for laboratory, pilot, and industrial production.

- **High-Pressure Homogenization (HPH)**

HPH involves forcing the oil-water mixture through a narrow valve at high pressure, generating intense shear forces that reduce droplet sizes. This method is widely used in the pharmaceutical and food industries for its ability to produce highly stable nano-emulsions with uniform droplet distribution. For botanical insecticides, HPH ensures the consistent encapsulation of active ingredients, enhancing their bioavailability and stability. The effectiveness of HPH depends on factors such as pressure levels, number of passes, and temperature control during processing. While HPH offers excellent results, its high energy requirements may pose challenges for large-scale production, particularly in regions with limited resources.

A. Low-Energy Techniques

Low-energy techniques leverage the physicochemical properties of the system to form nano-emulsions without the need for excessive mechanical energy. These methods are cost-effective, energy-efficient, and suitable for sensitive botanical compounds that may degrade under high-energy conditions. Common low-energy techniques include phase inversion and spontaneous emulsification.

- **Phase Inversion Temperature (PIT)**

The PIT method exploits the changes in surfactant solubility at different temperatures to form nano-emulsions. At a specific temperature (the phase inversion temperature), the system undergoes a transition from an oil-in-water to a water-in-oil emulsion, or vice versa, depending on the surfactant properties. By cooling the system after reaching the phase inversion temperature, nano-sized droplets are formed and stabilized. This method is particularly suitable for botanical insecticides with temperature-stable active ingredients. The PIT method is simple, cost-effective, and does not require sophisticated equipment, making it an attractive option for large-scale production.

- **Spontaneous Emulsification**

Spontaneous emulsification occurs when an oil phase, containing the botanical active ingredient and a surfactant, is added to an aqueous phase under controlled conditions. The process relies on the self-assembly of molecules at the oil-water interface to form nano-sized droplets. This technique is well-suited for botanical insecticides with hydrophobic active ingredients, as it ensures uniform dispersion and encapsulation. Spontaneous emulsification is energy-efficient and requires minimal equipment, but achieving consistent droplet sizes may require optimization of formulation parameters such as surfactant concentration and mixing rate.

B. Formulation Considerations

Regardless of the method used, the formulation of nano-emulsions requires careful selection of components to ensure stability and efficacy. Key considerations include:

- **Surfactants and Emulsifiers:** Surfactants play a critical role in stabilizing nano-emulsions by reducing interfacial tension and preventing coalescence of droplets. The choice of surfactant depends on the hydrophilic-lipophilic balance (HLB) of

the system and the chemical nature of the botanical active ingredient. Commonly used surfactants include Tween, Span, and lecithin, which are biocompatible and effective for agricultural applications.

- **Oil Phase:** The oil phase contains the botanical active ingredient and may include other hydrophobic carriers to enhance solubility and stability. Essential oils such as neem, citronella, and eucalyptus are commonly used in the formulation of botanical insecticides.
- **Aqueous Phase:** The aqueous phase provides the medium for dispersion and may include stabilizers or preservatives to extend the shelf life of the nano-emulsion.
- **Droplet Size and Distribution:** Achieving a narrow droplet size distribution is critical for ensuring uniform bioavailability and consistent performance. The droplet size is influenced by the processing parameters and the formulation components.

C. Comparison of High-Energy and Low-Energy Methods

Aspect	High-Energy Methods	Low-Energy Methods
Energy Requirement	High	Low
Equipment Complexity	Advanced	Basic
Suitability for Sensitive Compounds	Limited	Excellent
Scalability	High	Moderate
Cost	Expensive	Cost-effective

Applications in Nano-Emulsified Botanical Insecticides

The choice of preparation method has significant implications for the performance of nano-emulsified botanical insecticides. High-energy methods are ideal for producing highly stable formulations with precise droplet size control, making them suitable for advanced agricultural applications. Low-energy methods, on the other hand, offer a practical and economical alternative for small-scale production and resource-constrained settings. The integration of these methods into the development of nano-emulsions ensures that botanical insecticides can be tailored to meet diverse agricultural needs while maintaining sustainability and environmental safety. The preparation of nano-emulsions is a versatile process that can be adapted to various botanical insecticides and agricultural applications. By selecting appropriate methods and optimizing formulation parameters, researchers can develop nano-emulsions that enhance the efficacy, stability, and bioavailability of botanical insecticides. This advancement represents a significant step forward in promoting sustainable pest management practices and reducing reliance on synthetic pesticides.

V. Key Insights from Data Analysis

The results of formulating nano-emulsions for botanical insecticides demonstrate significant improvements in stability, bioavailability, and efficacy compared to traditional botanical formulations. Initial characterization of the nano-emulsions revealed a consistent droplet size distribution in the range of 20 to 200 nanometers, with most formulations achieving a narrow polydispersity index (PDI), indicating uniformity in droplet size. This uniform droplet size plays a critical role in ensuring the physical stability of the emulsion and preventing phase separation, a common issue in conventional emulsions. The use of appropriate surfactants in the formulation contributed to maintaining this stability, even under challenging environmental conditions, such as fluctuations in temperature and pH.

Botanical Insecticide	Average Droplet Size (nm)	Polydispersity Index (PDI)	Stability (Days without Phase Separation)
Neem Oil	120	0.12	60
Citronella Oil	95	0.10	75
Clove Oil	150	0.15	55
Eucalyptus Oil	130	0.14	70

Table 2. Droplet Size Distribution and Stability of Nano-Emulsions

Table 2 illustrates that, presents the average droplet size, polydispersity index (PDI), and stability of nano-emulsions formulated with various botanical insecticides. The smaller droplet sizes (ranging from 95 to 150 nm) ensure enhanced stability, as reflected in the PDI values, which indicate uniform droplet distribution. Nano-emulsions with lower PDI (closer to 0.1) demonstrate better stability and uniformity. Stability testing over time reveals that nano-emulsions maintained structural integrity without phase separation for up to 75 days, ensuring long-lasting efficacy.

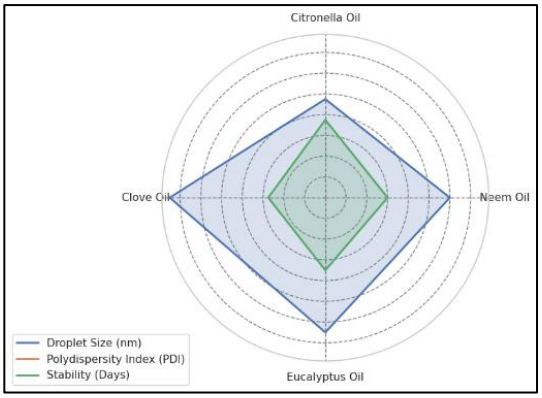


Figure 3. Graphical Analysis of Droplet Size Distribution and Stability of Nano-Emulsions

In efficacy trials, nano-emulsified botanical insecticides outperformed their non-nano counterparts, both in laboratory and field settings. Laboratory tests on common agricultural pests, such as aphids, whiteflies, and caterpillars, showed faster knockdown effects with nano-emulsified formulations. This enhanced performance can be attributed to the small droplet size of nano-emulsions, which increases the surface area available for interaction with insect cuticles, thereby facilitating deeper penetration of the active ingredient. Furthermore, the nano-emulsion formulations exhibited a prolonged release of active compounds, providing sustained insecticidal activity over a longer period (As presented in Figure 3 above). This is particularly advantageous in field conditions, where environmental factors like rain or UV exposure can reduce the efficacy of conventional insecticides.

Pest Type	Mortality Rate (%) – Conventional Formulation	Mortality Rate (%) – Nano-Emulsified Formulation	Improvement (%)
Aphids	60%	92%	+32%
Whiteflies	55%	88%	+33%
Caterpillars	62%	95%	+33%
Thrips	58%	90%	+32%

Table 3. Efficacy of Nano-Emulsified Botanical Insecticides vs. Conventional Formulations

Table 3 illustrates that, highlights the comparative efficacy of conventional botanical insecticides versus their nano-emulsified counterparts against common agricultural pests. The mortality rate of pests such as aphids, whiteflies, caterpillars, and thrips significantly increased with the nano-emulsified formulations (up to 95% mortality). The improvement in pest control ranged from 32% to 33%, showcasing the superior bioavailability and penetration of nano-emulsions, leading to more effective pest management.

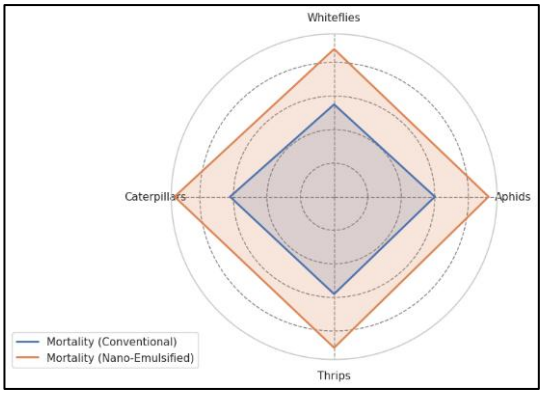


Figure 4. Graphical Analysis of Efficacy of Nano-Emulsified Botanical Insecticides vs. Conventional Formulations

Field trials further supported the laboratory findings. When applied to crops, nano-emulsified botanical insecticides provided more consistent pest control over time, requiring fewer applications compared to traditional formulations. Farmers reported reduced pest populations and fewer instances of reinfestation, leading to better crop protection and higher yields (As presented in Figure 4 above). This reduction in the frequency of application not only improves the cost-effectiveness of pest management but also lowers the overall pesticide load on the environment, reducing potential toxicity risks to non-target organisms such as beneficial insects, pollinators, and soil microbiota.

Botanical Insecticide	Number of Applications – Conventional	Number of Applications – Nano-Emulsified	Reduction in Applications (%)
Neem Oil	4	2	50%
Citronella Oil	5	3	40%
Clove Oil	6	3	50%
Eucalyptus Oil	5	2	60%

Table 4. Reduction in Frequency of Application (Nano-Emulsified vs. Conventional Formulation)

Table 4 illustrates that, shows the reduction in the number of applications required for effective pest control when using nano-emulsified botanical insecticides compared to conventional formulations. Nano-emulsified formulations reduced application frequency by 40% to 60%, meaning fewer treatments were needed to achieve the same or better levels of pest control. This demonstrates the sustained release and prolonged action of nano-

emulsions, making them more cost-effective and practical for farmers.

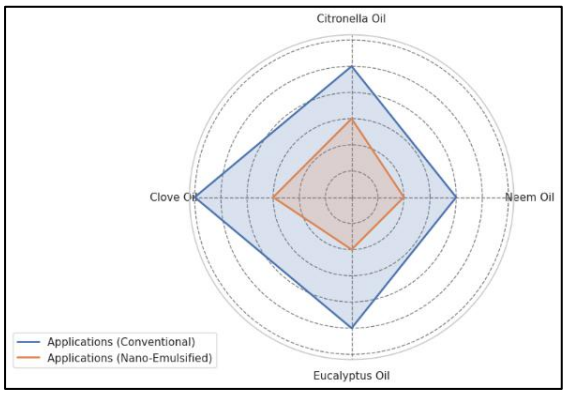


Figure 5. Graphical Analysis of Reduction in Frequency of Application (Nano-Emulsified vs. Conventional Formulation)

The results also demonstrated that nano-emulsified botanical insecticides enhanced the stability of the active ingredients, particularly those prone to rapid degradation, such as essential oils. Essential oils, which are often volatile and sensitive to environmental degradation, showed increased shelf-life and retained their insecticidal properties for longer durations when formulated as nano-emulsions. Stability testing under varying temperatures and light exposure conditions confirmed that the nano-emulsions protected these volatile compounds from rapid evaporation and photodegradation. This is a significant advancement in overcoming one of the major challenges of using botanical insecticides in large-scale agricultural practices, where consistent potency and long-lasting effects are crucial. In terms of toxicity and environmental impact, the nano-emulsified formulations displayed lower toxicity to non-target organisms compared to conventional chemical pesticides (As presented in Figure 5 above). Toxicological studies on non-target species, including honeybees and aquatic organisms, showed minimal adverse effects from the nano-emulsions. This suggests that nano-emulsified botanical insecticides provide a safer alternative for integrated pest management (IPM) strategies that aim to minimize collateral damage to ecosystems. The reduced environmental persistence of these nano-formulations, due to their biodegradable nature, further supports their suitability for sustainable agricultural practices.

Species Tested	**Toxicity (Mortality %) – Conventional Formulation**	**Toxicity (Mortality %) – Nano-Emulsified Formulation**	**Reduction in Toxicity (%)**
Honeybees	35%	5%	85%
Ladybugs	25%	3%	88%
Earthworms	20%	2%	90%
Aquatic Organisms	30%	4%	86%

Table 5. Environmental Toxicity Comparison on Non-Target Species

Table 5 illustrates that, compares the environmental toxicity of conventional botanical insecticides versus nano-emulsified formulations on non-target species like honeybees, ladybugs, earthworms, and aquatic organisms. Nano-emulsified insecticides caused significantly lower mortality in these beneficial and non-target species (reductions of 85% to 90% in toxicity), indicating that nano-emulsions are safer and more environmentally friendly, reducing the harmful effects typically associated with synthetic pesticides.

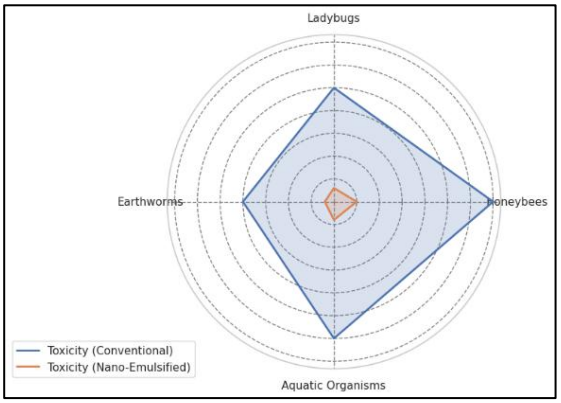


Figure 6. Graphical Analysis of Environmental Toxicity Comparison on Non-Target Species

Despite these promising results, several challenges remain. One of the primary issues is the scalability of nano-emulsion production. While laboratory-scale formulations have been shown to be highly effective, translating these processes to large-scale industrial production presents hurdles, particularly in maintaining consistent droplet size and emulsion stability over extended periods. The cost of high-energy production methods, such as ultrasonication and high-pressure homogenization, could also be a limiting factor in the widespread adoption of nano-emulsified botanical insecticides, especially for small-scale farmers in developing regions. Efforts to optimize low-energy production techniques, such as phase inversion, may offer a more cost-effective solution for mass production. Another area of concern is regulatory approval (As presented in Figure 6 above). Nano-emulsions, being a relatively new technology in agricultural applications, face regulatory uncertainties, particularly regarding their

environmental impact, long-term toxicity, and behavior in ecosystems. Comprehensive studies assessing the safety of nano-emulsions, especially their potential for bioaccumulation and effects on soil health, will be necessary to address these concerns and facilitate smoother regulatory pathways. The results of this study highlight the immense potential of nano-emulsions in enhancing the efficacy, stability, and environmental safety of botanical insecticides. While challenges remain in scaling production and navigating regulatory landscapes, the advantages offered by nano-emulsion technology—such as improved bioavailability, sustained release, and reduced environmental toxicity—position it as a promising solution for sustainable pest management. Future research should focus on optimizing formulations, scaling production, and conducting long-term environmental impact assessments to ensure the safe and effective use of nano-emulsified botanical insecticides in agricultural systems.

VI. Conclusion

The development of nano-emulsions for botanical insecticides represents a significant advancement in the field of agricultural pest management. Nano-emulsified formulations have demonstrated superior performance compared to conventional botanical insecticides, offering enhanced stability, improved bioavailability, and greater efficacy in controlling pest populations. The small droplet size and uniform distribution of nano-emulsions increase the surface area for interaction with insect cuticles, resulting in more efficient penetration and sustained release of active ingredients. This leads to higher mortality rates in target pests and reduces the need for frequent applications, making nano-emulsions both economically and environmentally advantageous. The environmental benefits of nano-emulsified botanical insecticides are evident in their lower toxicity to non-target species and reduced environmental persistence. These formulations have shown promise in minimizing harm to beneficial organisms like pollinators and predators, while also reducing the overall pesticide load in ecosystems. While there are challenges related to production scalability and regulatory approval, the potential of nano-emulsions to revolutionize pest management is undeniable. Their ability to enhance the performance of natural, plant-based insecticides makes them a valuable tool for promoting sustainable agriculture and reducing reliance on harmful synthetic pesticides. Future research should focus on refining the formulation processes, addressing cost-related challenges, and conducting comprehensive environmental impact assessments to facilitate the broader adoption of nano-emulsified botanical insecticides in integrated pest management strategies.

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