

International Journal of Advanced Biochemistry Research



ISSN Print: 2617-4693
ISSN Online: 2617-4707
NAAS Rating (2026): 5.29
IJABR 2026; SP-10(5): 910-915
www.biochemjournal.com
Received: 13-02-2026
Accepted: 16-03-2026

Pradeep Kumar
Ph.D. Scholar, Department of
Entomology, Maharana Pratap
university of Agriculture and
Technology, Udaipur, Rajasthan,
India

S Ramesh Babu
Department of Entomology,
Maharana Pratap university of
Agriculture and Technology, Udaipur,
Rajasthan, India

Devendra Jain
Department of Molecular Biology and
Biotechnology, Maharana Pratap
university of Agriculture and
Technology, Udaipur, Rajasthan,
India

Kinjal Mondal
Department of Molecular Biology and
Biotechnology, Maharana Pratap
university of Agriculture and
Technology, Udaipur, Rajasthan,
India

Hemant Swami
Department of Entomology,
Maharana Pratap university of
Agriculture and Technology, Udaipur,
Rajasthan, India

Lokesh Gupta
Department of Dairy and Food
Technology, Maharana Pratap
university of Agriculture and
Technology, Udaipur, Rajasthan,
India

ML Ojha
Department of Fisheries Resource
Management and Aquatic Animal
Health Management, Maharana
Pratap university of Agriculture and
Technology, Udaipur, Rajasthan,
India

Beerendra Singh
Department of Entomology,
Maharana Pratap university of
Agriculture and Technology, Udaipur,
Rajasthan, India

Rishi Raj Singh Chundawat
Department of Entomology,
Maharana Pratap university of
Agriculture and Technology, Udaipur,
Rajasthan, India

Brajrajsharan Tiwari
Department of Entomology, Banda
University of Agriculture and
Technology, Banda, Uttar Pradesh,
India

Corresponding Author:
Pradeep Kumar
Ph.D. Scholar, Department of
Entomology, Maharana Pratap
university of Agriculture and
Technology, Udaipur, Rajasthan,
India

Synthesis and bio-efficacy evaluation of castor seed oil-based nanoemulsion against diamondback moth (*Plutella xylostella*) in cabbage

Pradeep Kumar, S Ramesh Babu, Devendra Jain, Kinjal Mondal, Hemant Swami, Lokesh Gupta, ML Ojha, Beerendra Singh, Rishi Raj Singh Chundawat and Brajrajsharan Tiwari

DOI: <https://www.doi.org/10.33545/26174693.2026.v10.i5Sk.8487>

Abstract

The present study aimed to develop and evaluate a castor seed oil-based nanoemulsion (CSNE) as a sustainable nano-enabled botanical formulation for the management of the Diamondback moth in cabbage. Oil-in-water nanoemulsions containing different concentrations of castor seed oil (0-10%) were synthesized using Tween 80 and Span-20 through ultrasonic emulsification and characterized for physicochemical stability. Dynamic light scattering (DLS) analysis revealed concentration-dependent variations in hydrodynamic diameter, polydispersity index (PDI), and zeta potential. Among all formulations, 6% CSNE exhibited superior colloidal stability with a nanoscale droplet size of 21.83 nm, low PDI (0.248), and high negative zeta potential (−38.4 mV), indicating enhanced dispersion uniformity and interfacial stability. *in vitro* larvicidal bioassays against second-instar larvae of *P. xylostella* demonstrated a significant concentration- and time-dependent increase in mortality. The 6% CSNE at 1500 ppm exhibited the highest larvicidal activity, achieving 76.7% mortality at 48 h after treatment, substantially exceeding the efficacy of the conventional insecticide check, Novaluron 10% EC (46.7%). Relative efficacy index (REI) analysis further confirmed the superior and sustained bioefficacy of the optimized formulation. The enhanced insecticidal performance was attributed to improved nanodroplet stability, better surface adherence, and enhanced delivery of ricinoleic acid-rich bioactive compounds. Overall, the study demonstrates that optimized castor oil nanoemulsions represent a promising eco-friendly alternative for effective and sustainable management of diamondback moth in cabbage cultivation.

Keywords: Nanoemulsion, diamondback moth, zeta potential, REI, ricinoleic acid

1. Introduction

Cabbage (*Brassica oleracea* var. capitata L.) is a widely cultivated cole crop in India, valued for its nutritional and economic importance. However, its production is severely affected by the diamondback moth (*Plutella xylostella* L.), a highly destructive pest causing yield losses of up to 52% and significant quality degradation through foliage injury and head contamination (Paudel *et al.* 2022; Shamsi *et al.* 2026) [20, 23]. Its epidemiological success is attributed to its short life cycle (14-21 days), high fecundity (150-300 eggs per female), and rapid resistance evolution, enabling persistent outbreaks across cropping seasons (Sultana *et al.* 2025) [28]. Seasonal dynamics in Indian agro-ecosystems reveal peak infestation during warm and moderately humid periods, particularly in early summer and post-monsoon seasons. Moreover, the larval stage is highly destructive, with feeding localized to mesophyll tissues, resulting in “windowing” symptoms that later develop into shot holes, significantly reducing photosynthetic efficiency (Talekar and Shelton 1993; Furlong *et al.* 2013) [30, 7]. The pre-heading stage of cabbage has been identified as a critical window where DBM infestation causes maximum yield loss, necessitating timely intervention (Farias *et al.* 2021) [6].

Management of DBM has predominantly relied on synthetic insecticides; however, quantitative assessments indicate that while chemical control can be effective, its performance is highly variable and often unsustainable. For instance, newer insecticides such as spinosad, chlorantraniliprole, and indoxacarb have demonstrated 75.96-83.20% reduction

in larval population under field conditions (Patra *et al.* 2015; Kumar *et al.* 2025) ^[19, 13], with spinosad alone achieving up to 89.97% suppression over control (Dotasara *et al.* 2017) ^[5]. Similarly, chlorantraniliprole has shown approximately 81% reduction in larval population, outperforming several conventional chemicals (Chowdary and Sharma 2019) ^[12]. Despite these high efficacy levels, repeated and indiscriminate use has led to rapid resistance development in DBM populations, making it one of the most insecticide-resistant pests globally (Shehzad *et al.* 2023) ^[24]. Even biologically derived agents such as *Bacillus thuringiensis* (Bt), once considered highly effective, now show comparatively lower efficacy (45% reduction in some cases) due to resistance evolution (Guo *et al.* 2021) ^[9]. This underscores a critical need for next-generation, environmentally benign pest control strategies that are both effective and sustainable.

In this context, plant-derived bioactive compounds have gained attention due to their biodegradability and reduced ecological footprint. Castor (*Ricinus communis* L.) seed oil, rich in ricinoleic acid and other bioactive constituents, has been reported to possess insecticidal, antifeedant, and oviposition-deterrent properties (Ramos-López *et al.* 2010; Kodjo *et al.* 2011; Mirzaei and Omara 2024) ^[21, 12, 16]. However, its practical application is constrained by poor dispersion, instability, and limited persistence on leaf surfaces. Recent advances in nanotechnology, particularly nanoemulsion-based delivery systems, provide a transformative approach to enhance the efficacy of botanical pesticides. Nanoemulsions, with droplet sizes typically ranging from 20-200 nm, offer enhanced surface area, improved wettability, better adhesion, and controlled release of active compounds, thereby increasing their bioavailability and insecticidal potential (Mustafa and Hussein 2020; Garcia *et al.* 2022) ^[18, 8]. The reduction of droplet size to the nanometric scale significantly enhances dispersion stability, surface area, and interaction with biological membranes, facilitating improved uptake and bioefficacy (Abesekara and Chau 2022; Garcia *et al.* 2022) ^[1, 8].

Therefore, the present study focuses on the development and optimization of a castor oil-based nanoemulsion and its mechanistic evaluation against *Plutella xylostella*. By integrating detailed physicochemical characterization (e.g., droplet size distribution, interfacial stability, and functional group interactions) with bioefficacy assessment, the study aims to elucidate the relationship between nanoformulation properties and insecticidal performance. This work establishes a chemistry-driven framework for nano-enabled botanical pesticide design, offering a sustainable, residue-minimizing, and environmentally compatible alternative for effective diamondback moth management in cabbage.

2. Materials and Methods

2.1 Extraction of castor seed oil

Mature seeds of castor (*Ricinus communis* L.) were procured from a local market in Udaipur, Rajasthan, and thoroughly cleaned to remove extraneous materials prior to pulverization into a fine powder using a mechanical grinder. The powdered material was subjected to hydro-distillation using a Clevenger-type apparatus following standard protocols described by Harborne (1998) ^[11]. Briefly, distilled water was heated to generate steam, which permeated the plant matrix and facilitated the volatilization and co-distillation of oil constituents. The vapor mixture of steam and oil was subsequently condensed, and the distillate

was collected, allowing phase separation of the oil from the aqueous layer based on density differences. The recovered oil fraction was dried over anhydrous Na₂SO₄ to eliminate residual moisture, followed by filtration and storage in airtight containers under controlled conditions for subsequent analyses.

2.2 Nanoemulsion synthesis and characterization

The oil-in-water nanoemulsions were formulated using castor oil, a blend of surfactants and co-surfactants (Tween 80 and Span-20), along with double-distilled water using a magnetic stirrer, and further subjected to ultrasonic emulsification using a 20kHz sonicator (Ultrasonics, USA) with a power output of 750 W. Energy input was given through a sonotrode containing a piezoelectric crystal with a probe diameter of 13 mm. The major surfactant Tween 80 (Sisco Research Laboratories Pvt. Ltd., Maharashtra, India) was used along with co-surfactant span-20 (Sisco Research Laboratories Pvt. Ltd., Maharashtra, India). Other important chemical compounds like propylene glycol (PPG) were sourced from SRL, Maharashtra, India, while ethanol (99.9% pure) was purchased from Genteq Laboratories Pvt. Ltd., Delhi, India. The laboratory extracted castor seed oil was utilized as the organic phase for the nanoemulsion formulation, and laboratory-made double-distilled water (pH 6.5) was used as the aqueous phase and for the preparation of all the reagents.

2.2.1 Synthesis

Oil-in-water (O/W) nanoemulsions were formulated using castor oil as the dispersed phase at concentrations of 0, 2, 4, 6, 8, and 10% (v/v) (Table 1). The nanoemulsion systems were prepared through phase-wise mixing of aqueous and lipid components under controlled thermal and stirring conditions. The aqueous phase (Mass I) consisted of Tween 80, distilled water, and propylene glycol, which were heated to 50 °C under continuous magnetic stirring until a clear and thermodynamically uniform phase was obtained. Simultaneously, the oil phase (Mass II) was prepared by dissolving Span-20 in ethanol, followed by the gradual dropwise incorporation of castor oil under constant agitation to achieve a transparent isotropic mixture. Subsequently, the oil phase was slowly titrated into the aqueous phase under continuous stirring to facilitate spontaneous interfacial emulsification and nanoscale droplet formation. The resulting formulations were further subjected to magnetic stirring at 1000 rpm for 72 h to ensure complete homogenization, reduction of interfacial tension, and stabilization of the dispersed nanodroplets. The prepared nanoemulsions (designated as CSNE) were subsequently characterized for their physicochemical attributes and monitored for colloidal stability over a storage period of one month.

Table 1: Composition of the formulations (of 100 mL) and their stability

Ingredients	F1	F2	F3	F4	F5	F6
Castor seed oil (mL)	0	2	4	6	8	10
Propylene Glycol (mL)	10	10	10	10	10	10
Tween 80 (mL)	7	7	7	7	7	7
Span-20 (mL)	3	3	3	3	3	3
Ethanol (mL)	15	15	15	15	15	15
Distilled Water (mL)	65	63	61	59	57	55

*R.T. stands for room temperature (25 °C)

2.2.2 Characterization

The developed nanoemulsions (CSNE) were characterized for their physicochemical properties to assess stability and functionality. The particle size distribution, polydispersity index (PDI), and zeta potential were determined by dynamic light scattering (DLS) using Zetasizer Nano ZS90 (Malvern Instruments Ltd., UK) at 25 °C after appropriate dilution with deionized water, where PDI indicated size distribution uniformity and zeta potential reflected colloidal stability (Mukherjee and Baruah 2025) ^[17]. The viscosity was measured using a rotational viscometer at controlled temperature to evaluate flow behavior and its influence on formulation stability (Tadros *et al.* 2004) ^[29]. The pH of the formulations was recorded using a calibrated digital pH meter to ensure stability and compatibility (de Oca-Ávalos *et al.* 2017) ^[3].

2.3 Experimental details

An *in vitro* study was conducted to evaluate the larvicidal activities of the laboratory-formulated CSNEs against DBM. The experiment was carried out in a complete randomized design (CRD) with eight treatments and three replications. To estimate the relative efficacy index (REI) of CSNEs, Novaluron 10% EC (Conventional insecticide, C.I) was employed as the standard reference due to its established efficacy against DBM. All the experimental details have been summarized in Table 2.

Table 2: Experimental details

Mode of experiment	<i>In vitro</i>
Crop, variety	Cabbage, Ankur Manas
Design	CRD (Complete Randomized Design)
Treatments, replication	8, 3
Mode of treatment application	Leaf-dip method
Treatments	
T ₁	Control (water)
T ₂	CSNE 0%
T ₃	CSNE 2%
T ₄	CSNE 4%
T ₅	CSNE 6%
T ₆	CSNE 8%
T ₇	CSNE 10%
T ₈	Conventional insecticide (Novaluron 10% EC)

2.4 *In vitro* assessment of larvicidal activities of CSNEs by leaf-dip method

To investigate the insecticidal potential of castor seed oil nanoemulsion (CSNE) formulations against, *Plutella xylostella* (L.), an *in vitro* feeding bioassay was conducted in the Department of Entomology under controlled laboratory conditions. Freshly harvested and thoroughly washed cabbage leaves were treated with different concentrations of CSNE (1250, 1500, 1750, and 2000 ppm) prepared with varying nanoemulsion compositions (0%, 2%, 4%, 6%, 8%, and 10%). Treated leaves were air-dried under ambient laboratory conditions prior to use in the bioassay. Ten healthy second-instar DBM larvae were carefully released into each sterile petri dish containing the treated leaf material. An untreated control consisting of distilled water-treated leaves and a standard chemical check comprising Novaluron 10% EC were maintained simultaneously for comparative assessment of larvicidal efficacy. Larval mortality observations were recorded at 2,

4, 6, 8, 24, and 48 h after treatment (HAT) to determine the temporal progression and concentration-dependent toxicity of CSNE formulations. Mortality data were quantified using the formula described below, enabling assessment of the bioefficacy of nanoemulsion treatments across exposure intervals.

$$\text{Mortality (\%)} = \left(\frac{\text{Number of dead larvae}}{\text{Total number of larvae introduced}} \right) \times 100$$

Furthermore, the relative efficacy index (REI) of all CSNE formulations was calculated at 24 and 48 HAT with reference to Novaluron 10% EC (C.I) to identify the most potent and optimized nanoemulsion concentration exhibiting maximum larvicidal activity against DBM. The overall *in vitro* treatment effectiveness and comparative bioefficacy among formulations were determined according to Kumar *et al.* (2025) ^[13].

$$\text{Relative Efficacy (\%)} = \left(\frac{\text{Mortality in CSNE}}{\text{Mortality in C.I.}} \right) \times 100$$

2.5 Statistical analysis

All experimental data were subjected to statistical analysis using one-way analysis of variance (ANOVA) performed in IBM SPSS Statistics. Treatment means were compared using Tukey's honestly significant difference (HSD) test at a significance level of $p < 0.05$. Percentage data were arcsine-transformed prior to analysis to stabilize variance and satisfy assumptions of normality.

3. Results and Discussion

3.1 Synthesis and characterization of castor seed oil based nanoemulsion

The O/W nanoemulsion system demonstrated efficient nanoscale emulsification and colloidal stabilization through synergistic interactions between castor oil, Tween 80, Span-20, ethanol, and propylene glycol. Controlled thermal mixing (50 °C) and gradual phase titration promoted spontaneous interfacial emulsification by reducing interfacial free energy and facilitating uniform nanodroplet formation (Liu *et al.* 2024) ^[14]. The combined non-ionic surfactant system provided an optimized hydrophilic-lipophilic balance (HLB), enhancing interfacial film stability and preventing droplet coalescence through steric stabilization (Miranda-Olvera *et al.* 2025) ^[15]. Ethanol and propylene glycol further improved phase miscibility and reduced interfacial tension, contributing to transparent isotropic nano-dispersion. Extended magnetic stirring at 1000 rpm for 72 h enhanced homogenization, minimized phase separation, and promoted stable nanoscale droplet distribution. Increasing castor seed oil concentration likely influenced dispersed phase viscosity and droplet packing behavior; however, the optimized surfactant-co-surfactant matrix maintained nanoemulsion stability and physicochemical uniformity.

The synthesized nanoemulsions exhibited favorable physicochemical characteristics immediately after formulation, with pH values ranging from 6.01-6.70 and viscosity below 100 cP (Table 3), indicating low-resistance flow behavior and compatibility for application on cabbage leaf surfaces during leaf dipping. Dynamic light scattering (DLS) analysis revealed concentration-dependent variations in hydrodynamic diameter, polydispersity index (PDI), and zeta potential (Table 3), reflecting the influence of dispersed

oil phase loading on interfacial organization and colloidal stability (Solans and Solé 2012) [26]. Among the formulations, 6, 8, and 10% CSNE remained optically transparent and free from phase separation, confirming successful nanoscale dispersion and kinetic stability. Notably, the 6% CSNE exhibited the most uniform

nanodroplet distribution with a low PDI (0.248) (Figure 1) and high negative surface charge (-38.4 mV), suggesting enhanced electrostatic stabilization, reduced droplet aggregation, and superior colloidal homogeneity within the nano-range.

Table 3: Physicochemical properties of laboratory-synthesized CSNEs

CSNE (% seed oil)	pH	Viscosity (cP)	Z-average (hydrodynamic diameter; nm)	Particle size distribution ($\geq 75\%$)	PDI	Zeta potential (mV)
0	6.01	58.17	665.10	58.77-122.4 nm	0.333	-12.2 ± 2.1
2	6.70	66.80	97.11	32.67-105.7 nm	0.215	-24.6 ± 1.6
4	6.58	71.40	60.40	15.69-50.75 nm	0.252	-29.2 ± 1.5
6	6.65	74.47	21.83	4.18-15.69 nm	0.248	-38.4 ± 2.7
8	6.49	77.30	9.74	2.01-6.50 nm	0.284	-32.7 ± 2.3
10	6.47	81.70	10.32	2.33-7.53 nm	0.297	-30.3 ± 3.1

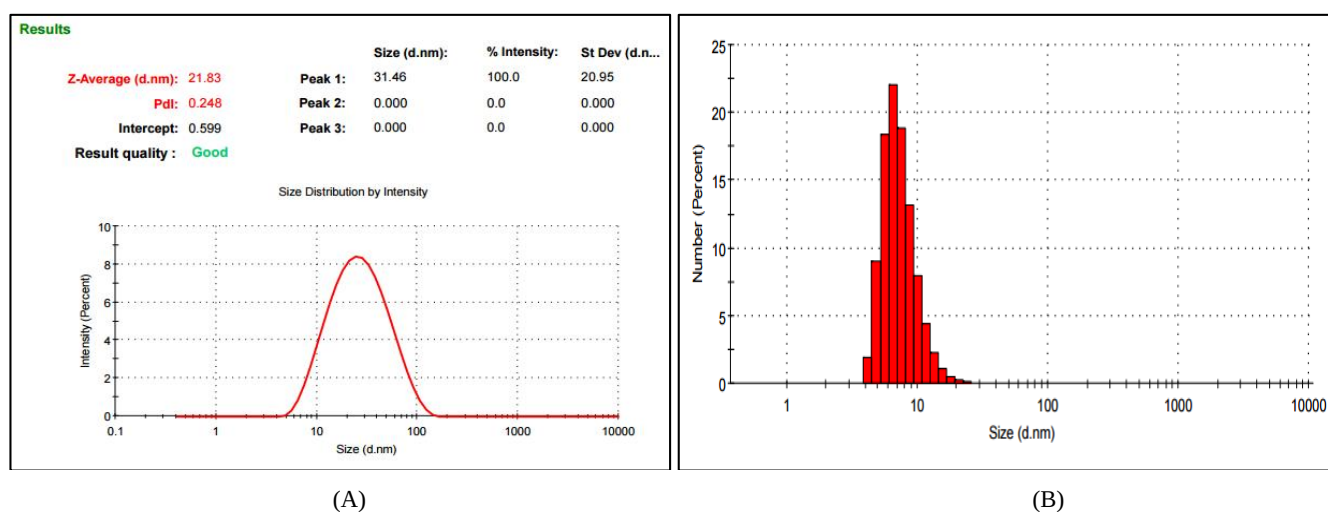


Fig 1: DLS analysis of 6% CSNE is showing (A) particle hydrodynamic diameter (Z-average) and PDI, (B) Particle size distribution

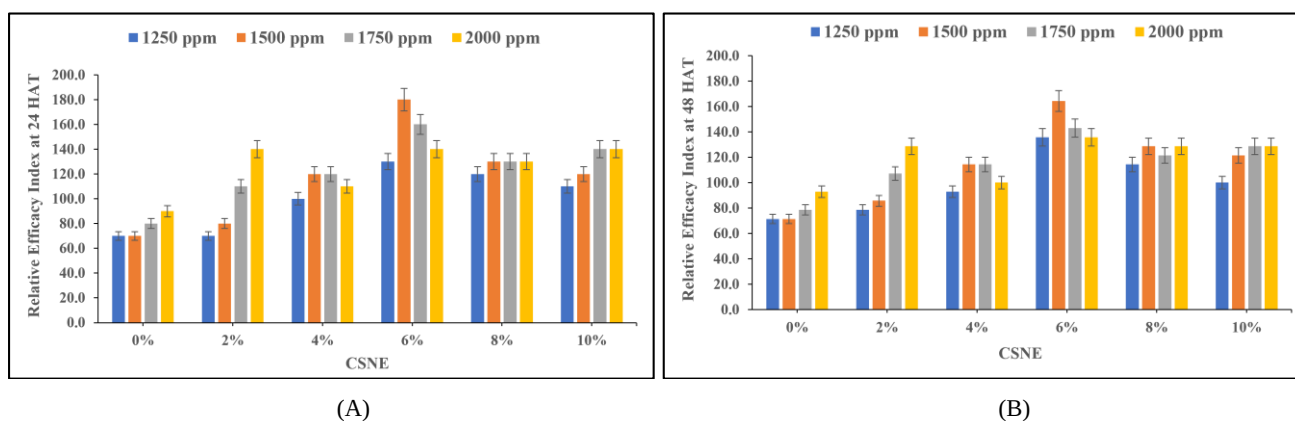
Larval mortality progressively increased with both concentration and exposure duration across all CSNE formulations, demonstrating a pronounced time-dependent toxicological response (Table 4). Among the tested nanoemulsions, 6% CSNE emerged as the most potent formulation, particularly at 1500 ppm, where mortality escalated from 3.3% at 2 HAT (hours after treatment) to 76.7% at 48 HAT, substantially outperforming the commercial standard Novaluron 10% EC (46.7%). The enhanced efficacy of 6% CSNE may be attributed to the optimized surfactant-oil balance, which likely improved nano-droplet stability, dispersion uniformity, and penetration of bioactive ricinoleic acid-rich constituents through the larval cuticle (Shekade *et al.* 2023) [25]. In contrast, the 0% CSNE formulation exhibited comparatively poor larvicidal activity, indicating that the castor seed oil phase played a critical role in toxicity enhancement. Interestingly, increasing oil loading beyond 6% (8-10% CSNE) did not proportionally improve mortality, suggesting possible droplet aggregation, interfacial saturation, or reduced physicochemical stability at higher oil concentrations, ultimately limiting bioavailability. The delayed but steep rise in mortality between 24 and 48 HAT further indicates a sustained-release or cumulative mode of action of the nanoemulsion system rather than immediate

knockdown toxicity (Donsi *et al.* 2012; Sugumar *et al.* 2014) [4, 27]. Collectively, these findings highlight that precise optimization of nanoemulsion composition critically governs larvicidal efficiency, with 6% CSNE representing the most physiochemically balanced and biologically effective formulation for sustained insecticidal activity. The Relative Efficacy Index (REI) data further revealed that larvicidal performance was governed by nanoemulsion optimization rather than concentration alone, with 6% CSNE consistently exhibiting the highest efficacy at both 24 and 48 HAT, particularly at 1500 ppm (Figure 2). The superior REI of 6% CSNE suggests an optimum oil-surfactant balance that enhanced nano-droplet stability, interfacial interaction, and trans-cuticular delivery of ricinoleic acid-rich bioactive compounds (Gutiérrez *et al.* 2008) [10]. In contrast, lower efficacy in 0-2% CSNE indicated inadequate oil-mediated bioactive delivery, while the absence of proportional REI enhancement in 8-10% CSNE suggests possible interfacial saturation, droplet aggregation, or reduced colloidal stability at higher oil loading. The sustained superiority of 6% CSNE over time further indicates improved residual activity and controlled release behavior, highlighting it as the most physiochemically stable and biologically efficient formulation.

Table 4: Larvicidal activities of CSNEs of different concentrations based on mortality percentage

Formulation	Concentration (ppm)	Temporal progression in% mortality					
		2 HAT	4 HAT	6 HAT	8 HAT	24 HAT	48 HAT
0% CSNE	1250	0.0 ^a	0.0 ^b	6.7 ^b	13.3 ^b	23.3 ^b	33.3 ^c
	1500	0.0 ^a	3.3 ^a	10.0 ^a	16.7 ^{ab}	23.3 ^b	33.3 ^c
	1750	0.0 ^a	3.3 ^a	10.0 ^a	16.7 ^{ab}	26.7 ^{ab}	36.7 ^b
	2000	0.0 ^a	3.3 ^a	10.0 ^a	20.0 ^a	30.0 ^a	43.3 ^a
2% CSNE	1250	0.0 ^b	0.0 ^d	6.7 ^c	13.3 ^c	23.3 ^c	36.7 ^c
	1500	0.0 ^b	3.3 ^c	10.0 ^b	16.7 ^b	26.7 ^c	40.0 ^c
	1750	3.3 ^a	10.0 ^b	23.3 ^{ab}	30.0 ^{ab}	36.7 ^b	50.0 ^b
	2000	3.3 ^a	13.3 ^a	26.7 ^a	36.7 ^a	46.7 ^a	60.0 ^a
4% CSNE	1250	3.3 ^a	6.7 ^b	13.3 ^c	23.3 ^c	33.3 ^b	43.3 ^b
	1500	3.3 ^a	10.0 ^a	20.0 ^a	30.0 ^a	40.0 ^a	53.3 ^a
	1750	3.3 ^a	10.0 ^a	20.0 ^a	30.0 ^a	40.0 ^a	53.3 ^a
	2000	3.3 ^a	10.0 ^a	16.7 ^b	26.7 ^b	36.7 ^{ab}	46.7 ^{ab}
6% CSNE	1250	3.3 ^a	6.7 ^c	20.0 ^c	30.0 ^c	43.3 ^c	63.3 ^c
	1500	3.3 ^a	16.7 ^a	30.0 ^a	43.3 ^a	60.0 ^a	76.7 ^a
	1750	3.3 ^a	10.0 ^b	26.7 ^b	40.0 ^{ab}	53.3 ^b	66.7 ^b
	2000	3.3 ^a	10.0 ^b	20.0 ^c	33.3 ^b	46.7 ^{bc}	63.3 ^c
8% CSNE	1250	0.0 ^b	10.0 ^a	16.7 ^b	26.7 ^b	40.0 ^b	53.3 ^b
	1500	3.3 ^a	10.0 ^a	20.0 ^a	30.0 ^a	43.3 ^a	60.0 ^a
	1750	0.0 ^b	6.7 ^b	16.7 ^b	30.0 ^a	43.3 ^a	56.7 ^{ab}
	2000	0.0 ^b	6.7 ^b	16.7 ^b	30.0 ^a	43.3 ^a	60.0 ^a
10% CSNE	1250	0.0 ^b	6.7 ^b	16.7 ^c	23.3 ^c	36.7 ^c	46.7 ^c
	1500	0.0 ^b	6.7 ^b	16.7 ^c	26.7 ^b	40.0 ^b	56.7 ^b
	1750	3.3 ^a	13.3 ^a	26.7 ^a	36.7 ^a	46.7 ^a	60.0 ^a
	2000	3.3 ^a	10.0 ^{ab}	20.0 ^b	33.3 ^{ab}	46.7 ^a	60.0 ^a
Novaluron 10% EC	Commercial dose	0.0	6.7	13.3	23.3	33.3	46.7

Each value is mean of triplicates. Mean value followed by same letter indicates statistically insignificant difference at $p = 0.05$ as determined by Tukey-Kramer HSD test. HAT: Hours after treatment.

**Fig 2:** REI of different CSNEs (0-10%) compared to conventional insecticide (Novaluron 10% EC) at (A) 24 HAT and (B) 48 HAT

3.2 Conclusion

The study successfully developed a stable castor seed oil nanoemulsion with significant larvicidal activity against Diamondback moth. Among the formulations, 6% CSNE exhibited optimal physicochemical properties and superior bioefficacy, achieving higher larval mortality than the conventional insecticide standard. Improved performance was associated with enhanced colloidal stability, nanoscale droplet distribution, and efficient delivery of ricinoleic acid-rich bioactive compounds. These findings demonstrate the potential of castor oil nanoemulsions as a sustainable, residue-minimizing, and environmentally compatible strategy for DBM management in cabbage cultivation.

References

1. Abesekara MS, Chau Y. Recent advances in surface modification of micro- and nano-scale biomaterials with biological membranes and biomolecules. *Front Bioeng Biotechnol.* 2022 Oct 12;10:972790.
2. Chowdary NM, Sharma PC. Bioefficacy of newer insecticides against *Plutella xylostella* (L.) infesting cabbage. *Himachal J Agric Res.* 2019;46:46-50.
3. de Oca-Ávalos JM, Candal RJ, Herrera ML. Nanoemulsions: Stability and physical properties. *Curr Opin Food Sci.* 2017 Aug 1;16:1-6.
4. Donsi F, Annunziata M, Vincensi M, Ferrari G. Design of nanoemulsion-based delivery systems of natural antimicrobials: Effect of the emulsifier. *J Biotechnol.* 2012 Jun 30;159(4):342-350.
5. Dotsara SK, Agrawal N, Swami D. Efficacy of some new insecticides against diamond back moth (*Plutella xylostella* L.) on cauliflower. *Indian Soc Agric Sci.* 2017;227.
6. Farias ED, Sant'ana LC, Melo JB, Santana PA Jr, Picanço MC. Impact of diamondback moth density and infestation timing on broccoli yield. *Neotrop Entomol.* 2021 Apr;50(2):298-302.

7. Furlong MJ, Wright DJ, Dosdall LM. Diamondback moth ecology and management: Problems, progress, and prospects. *Annu Rev Entomol.* 2013 Jan 7;58:517-541.
8. Garcia CR, Malik MH, Biswas S, Tam VH, Rumbaugh KP, Li W, *et al.* Nanoemulsion delivery systems for enhanced efficacy of antimicrobials and essential oils. *Biomater Sci.* 2022;10(3):633-653.
9. Guo Z, Kang S, Wu Q, Wang S, Crickmore N, Zhou X, *et al.* The regulation landscape of MAPK signaling cascade for thwarting *Bacillus thuringiensis* infection in an insect host. *PLoS Pathog.* 2021 Sep 8;17(9):e1009917.
10. Gutiérrez JM, González C, Maestro A, Solè IM, Pey CM, Nolla J. Nano-emulsions: New applications and optimization of their preparation. *Curr Opin Colloid Interface Sci.* 2008 Aug 1;13(4):245-251.
11. Harborne AJ. *Phytochemical methods: A guide to modern techniques of plant analysis.* Berlin: Springer Science & Business Media; 1998. pp. 1-302.
12. Kodjo TA, Gbénonchi M, Sadate A, Komi A, Yaovi G, Dieudonné M, *et al.* Bio-insecticidal effects of plant extracts and oil emulsions of *Ricinus communis* L. (Malpighiales: Euphorbiaceae) on the diamondback moth, *Plutella xylostella* L. (Lepidoptera: Plutellidae) under laboratory and semi-field conditions. *J Appl Biosci.* 2011 Jan;43:2899-2914.
13. Kumar A, Choudhary JK, Singh TK, Singh R, Shukla M, Singh S, *et al.* Efficacy of novel insecticides and bio-pesticides for diamond back moth (*Plutella xylostella*) management in cabbage. *Veg Sci.* 2025 Jun 20;52(1):191-195.
14. Liu J, Li Y, Lun Z, Zhang Y, Yang P, Tang X, *et al.* Factors, mechanisms, and kinetics of spontaneous emulsification for heavy oil-in-water emulsions. *Molecules.* 2024 Jun 24;29(13):2998.
15. Miranda-Olvera AD, Domínguez-Esquivel JM, Martínez J. Hydrophilic-lipophilic balance (HLB) correlation method for the selection of ionic liquid surfactant modifiers of the viscosity and emulsion stability of heavy oils. *Langmuir.* 2025 Mar 28;41(13):8753-8765.
16. Mirzaei S, Omara Z. Plant-derived insecticides for pest management. *J Ethno-Pharm Prod.* 2024 Dec 1;4(1):40-58.
17. Mukherjee P, Baruah KN. Characterization of nanoemulsion: Particle size, polydispersity index, zeta potential, thermal stability, and kinetic stability. In: *Essential Oil Extraction from Food By-Products.* New York: Springer US; 2025. p. 273-288.
18. Mustafa IF, Hussein MZ. Synthesis and technology of nanoemulsion-based pesticide formulation. *Nanomaterials.* 2020 Aug 17;10(8):1608.
19. Patra B, Das BC, Alam SF, Dhote V, Patra S, Chatterjee ML, *et al.* Evaluation of new insecticides against diamond back moth, *Plutella xylostella* (L.) on red cabbage. *Int J Bio-Resour Stress Manag.* 2015 Apr 1;6(2):1-5.
20. Paudel A, Yadav PK, Karna P. Diamondback moth *Plutella xylostella* (Linnaeus, 1758) (Lepidoptera: Plutellidae): A real menace to crucifers and its integrated management tactics. *Turk J Agric Food Sci Technol.* 2022 Dec 20;10(12):2504-2515.
21. Ramos-López MA, Pérez S, Rodríguez-Hernández GC, Guevara-Fefer P, Zavala-Sanchez MA. Activity of *Ricinus communis* (Euphorbiaceae) against *Spodoptera frugiperda* (Lepidoptera: Noctuidae). *Afr J Biotechnol.* 2010;9(9):1359-1365.
22. Shakeel F, Ramadan W, Ahmed MA. Investigation of true nanoemulsions for transdermal potential of indomethacin: Characterization, rheological characteristics, and ex vivo skin permeation studies. *J Drug Target.* 2009 Jul 1;17(6):435-441.
23. Shamsi IH, Fu W, Askri SM, Waris S, Ibrahim Z, Ali A, *et al.* Interactive mechanisms and management challenges of *Plutella xylostella* (diamondback moth) in *Brassica*. *Insect-Plant Interact.* 2026 Jun 9:143-170.
24. Shehzad M, Bodlah I, Siddiqui JA, Bodlah MA, Fareen AG, Islam W. Recent insights into pesticide resistance mechanisms in *Plutella xylostella* and possible management strategies. *Environ Sci Pollut Res.* 2023 Sep;30(42):95296-95311.
25. Shekade SV, Shirolkar SV, Deshkar SS, Giram PS. Phytochemical, pharmacognostic, and pharmacological aspects of *Ricinus communis* seed oil: An overview. *Nat Prod J.* 2023 May 1;13(3):31-47.
26. Solans C, Solé I. Nano-emulsions: Formation by low-energy methods. *Curr Opin Colloid Interface Sci.* 2012 Oct 1;17(5):246-254.
27. Sugumar S, Clarke SK, Nirmala MJ, Tyagi BK, Mukherjee A, Chandrasekaran N. Nanoemulsion of eucalyptus oil and its larvicidal activity against *Culex quinquefasciatus*. *Bull Entomol Res.* 2014 Jun;104(3):393-402.
28. Sultana R, Ansari MS. Effects of constant temperature regimes on the life table parameters of *Plutella xylostella* (Lepidoptera: Plutellidae) (Linnaeus). *J Exp Agric Int.* 2025 Jun 30;47(6):764-775.
29. Tadros T, Izquierdo P, Esquena J, Solans C. Formation and stability of nano-emulsions. *Adv Colloid Interface Sci.* 2004 May 20;108:303-318.
30. Talekar NS, Shelton AM. Biology, ecology, and management of the diamondback moth. *Annu Rev Entomol.* 1993 Jan 1;38(1):275-301.