



Pest Shield Granules and Grain Guardian Silica Beads Developed Using Extracts of *Decalepis hamiltonii*: A Safe Natural Agent for Pest Management

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Abstract

Decalepis hamiltonii, an endangered medicinal plant traditionally valued for its pest-resistant roots used in treating various ailments, has faced depletion due to excessive root use. To conserve the plant, this study shifted focus to the leaves, revealing their potential as bio-insecticide for stored food grains, thereby highlighting both conservation and scientific inquiry. The present study evaluates different solvent extracts of *Decalepis hamiltonii* for pesticidal activity against two pests, *Spodoptera litura* and *Callosobruchus macculatus*. GCMS study was carried out to understand the bioactive constituents present in the extract. Additionally, the fractions were screened for cytotoxic activity using the brine shrimp lethality bioassay. The brine shrimp lethality bioassay indicated the toxicity of the extracts, with methanol showing the lowest LC50 of 32 ppm. Remarkably significant pesticidal activity against *Spodoptera litura* (LC50 40.30 µg/ml) and *Callosobruchus macculatus* (LC50 21.2 mg/ml) were observed for methanolic extract of *Decalepis hamiltonii*. Innovative formulations, such as effervescent granules and drug-loaded silica beads for pesticidal purposes, yielded promising results. This study provides valuable insights into *Decalepis hamiltonii*, highlighting its potential as a natural pesticide due to the presence of many important compounds especially, Benzyl Benzoate, Docosane and Octadecane (identified by GCMS), and serving as a crucial reference for future research on its medicinal applications.

Keywords: *Decalepis Hamiltonii*; Effervescent Granules; GCMS; Natural Pesticide; Pest Control

Introduction

Pesticides are substances used to prevent, destroy, or control pests, including insects, fungi, rodents, and unwanted plants that harm crops. These include insecticides, herbicides, fungicides, and rodenticides, each targeting specific pests (Shekhar *et al.*, 2024). Stored product insect pests cause significant damage to grains, with losses ranging from 5-10% in temperate regions to 20–30% in tropical countries (Rajashekar *et al.*, 2025). Synthetic pesticides, such as DDT, began in the 1930s, gained popularity post-World War II, and significantly increased agricultural yields (Abubakar *et al.*, 2020). The current era transitioned to the use of organophosphates and carbamates in the latter half of the 20th century. Currently, global pesticide usage is around 5.2 billion pounds annually, utilized in homes to combat various pests (Saad *et al.*, 2025). Chemical insecticides, including fumigants, have been used for grain protection but have led to resistance and environmental problems, resulting in bans and restrictions on some pesticides. Synthetic pyrethroids, derived from the pyrethrum plant, are widely used, but there are few botanical equivalents for stored grain protection, highlighting the need for safer natural alternatives i.e neem plant which is very effective against pest control and antifeedant

via its bioactive compounds especially azadirachtin (Melanie et al., 2022). The adverse impacts of synthetic pesticides include disrupting biodiversity, affecting non-target species, and posing health risks to humans. These risks stem from improper safety practices during pesticide application, leading to poisoning, which causes a range of health issues from mild allergies to chronic diseases like cancer. Additionally, the development of pest resistance necessitates more frequent and higher doses of pesticides, exacerbating these problems (Sabarwal et al., 2018; Muñoz-Bautista et al., 2025). To address these challenges, there is a growing shift towards bio pesticides, which are environmentally friendly, target-specific, and decompose quickly. They are particularly effective in Integrated Pest Management (IPM) programs and help mitigate the negative effects of synthetic insecticides (Shrestha et al., 2024; Mahlangu and Sibisi 2026). The live organic movement, driven by health consciousness, has led to an increase in organic cultivation in India, with states like Uttarakhand and Sikkim declaring them organic (Singh et al., 2025). Historical use of plant-based bio-pesticides in India declined with the advent of cheaper and more effective synthetic insecticides. Nevertheless, in recent decades, there has been resurgence in the emphasis on plant-derived products to combat and reduce agricultural losses caused by pests and diseases. This study focuses on *Decalepis hamiltonii*, a plant from India's hilly forests traditionally used for its medicinal root properties. Recognized for its bioactive compounds with bio-pesticide properties, *D. hamiltonii* shows promise as a natural alternative to synthetic pesticides. While previous research centered on its roots, this investigation shifts to the leaves to explore their insecticidal activity. The aim is to provide a safer solution for grain protection and to conserve the plant by uncovering new pesticide properties.

Materials and Methods

Collection of plant material

Decalepis hamiltonii (DH) leaves were collected from the ICAR-Indian Institute of Horticultural Research, Bangalore, Karnataka. The plant taxonomy was authenticated by the Research Officer at the Department of Botany, Central Ayurvedic Research Institute, confirming it as DH *Wight & Arn* of the family Apocynaceae, with the voucher number RRCBI-8781.

Extraction of plant materials

Leaves were washed, shade-dried at room temperature for about ten days until brittle, then pulverized into powder and stored in an airtight container for future use. A successive solvent extraction method was carried out to obtain the extracts. 50 grams of powdered leaf material were placed into the thimble of a Soxhlet extractor and extracted sequentially with petroleum ether, chloroform, ethyl acetate, methanol, and water, in increasing order of polarity, for 8 hours each. The extract was collected, and before each successive extraction with the next solvent, the powdered material was air-dried. The obtained extracts were then concentrated under rotary evaporator at 35°C. The percentage yield was calculated based on the air-dried weight of the plant material, and the colour and consistency of the extracts were noted⁹. These extracts were then subjected to further analysis by GCMS. The analysis was performed using a Thermo GC-Trace Ultra (Version 5.0) coupled with a Thermo MS DSQ II system. Separation was achieved on a DB-35 MS capillary column (30 m × 0.25 mm i.d., 0.25 µm film thickness), with helium as the carrier gas at a constant flow rate of 1.0 ml/min. The injector temperature was maintained at 250 °C, and the oven program was set to an initial temperature of 60 °C (held for 15 min), followed by a gradual increase to 280 °C at 3 °C/min.

Evaluation of toxicity by Brine shrimp assay (Abubakar and Haque, 2020)

The cytotoxic activity of the plant was evaluated using the Brine shrimp lethality bioassay method. Freeze-dried cysts of *Artemia salina* were procured from PEQON, United States of America, through Flip kart in capsule form. One gram of *Artemia salina* cysts was added to separating funnel containing 0.9% NaCl solution with good aeration provided by an air pump, maintained at room temperature under continuous illumination for 48 hours. The cysts hatched and matured into nauplii after two days, which were then used for the bioassay. Extracts of *Decalepis hamiltonii* leaf were prepared in 0.9% NaCl solution at concentrations of 1 µg/ml, 10 µg/ml, 100 µg/ml, and 1000 µg/ml. 1

mL of each extract sample at the specified concentrations was added to 2 mL of isotonic solution (0.9% NaCl). Distilled water was used for the control group. Subsequently, 10 nauplii were added to each test tube, and the movement of the larvae was observed after 24 hours to determine their mortality. The % mortality of *Artemia salina* larvae was calculated using the formula,

$$\% M = [(LC-LT)/LC] \times 100$$

Where M is mortality; LC is the number of living nauplii in the control group after 24 hours; LT is the number of Living nauplii with the tested agent after 24 hours.

Evaluation of Pesticidal activity

A. Leaf dip bioassay method using *Spodoptera litura* (Waghulde et al., 2019)

Establishment of *Spodoptera litura*: *Ricinus communis* leaves, collected locally, were washed to remove dust and surface sterilized using 0.4% sodium hypochlorite for 30 seconds, and then rinsed with distilled water. A batch of 100 *Spodoptera litura* eggs from the National Bureau of Agricultural Insect Resources, Bangalore, was placed on these sterilized leaves under controlled conditions (25 ± 2°C, 70% RH). The eggs hatched within two days, and ten days later, the larvae reached the fourth instar stage, known for its voracious feeding behaviour, making it ideal for testing pesticidal activity.

Pesticidal activity (Abubakar and Haque, 2020)

Decalepis hamiltonii leaf extracts were dissolved in acetone at concentrations of 10, 20, 30, 40, and 50 µg/ml, with acetone alone as the control. Five sterilized leaves of *Ricinus communis* were cut into 2.5 cm diameter circles and immersed in the extracts for 30 seconds, then air-dried. The treated leaves were placed in petri dishes, each containing five larvae, while the control group had acetone-treated leaves. The petri dishes were covered with mesh or ventilated lids and maintained at 25 ± 2°C and 70% RH. Larval mortality was assessed at 24, 48, and 72-hour intervals to determine the percentage mortality rates for each concentration. The LC50, which represents the concentration of the extract at which 50% of the larvae are expected to die, was calculated using probit analysis method in Excel. Remaining live pests were frozen at 0-5°C for 24 hours before disposal.

B. Residual Bioassay Method using *Callosobruchus maculatus* (Bhoye and Makode, 2024)

Non-infested, untreated *Vigna radiata* (green gram) grains were procured from a local grocery store. Cultures of adult *Callosobruchus maculatus* were obtained from the National Bureau of Agricultural Insect Resources, Bangalore, and reared on green gram seeds. These cultures were maintained at 30 ± 2°C and 70% relative humidity. Adult insects (5-7 days old) were selected for the experiments. Various extracts of *Decalepis hamiltonii* leaves were dissolved in acetone at concentrations of 5, 10, 15, 20, and 25 mg/ml, with acetone alone serving as the control. Glass Petri dishes with specific dimensions (bottom internal diameter of 5 cm, rim height of 1.3 cm, and lid internal diameter of 5.7 cm) were used, providing a total surface area of 65.5 cm². One millilitre of each sample concentration was evenly applied to various sections of the Petri dish, covering the bottom, rim, and lid. In the control group, acetone without any extract was applied. The applied extracts were allowed to evaporate for 5 minutes. Next, 3 grams of green gram seeds were placed in each Petri dish to serve as food for the insects. Ten adult *Callosobruchus maculatus* insects were then introduced into each Petri dish. The experimental setup was observed over 24, 48, and 72 hours to monitor the effects on the insect population. After each exposure period, the number of surviving insects was counted. LC50 values, representing the concentration at which 50% of the insect population is killed, were calculated using "Probit analysis" method in excel after the specified exposure durations.

Development of two pesticidal formulations

A. Effervescent granules (Arunagiri and Sundararajan, 2024)

The most potent methanolic extract, based on LC50 values, was selected for formulation. The concentration was calculated to target approximately 1000 insects infesting 10 plants, ensuring rapid impact and extended prevention.

To prepare 50 grams of granules, the specified quantities of excipients and effervescent agents were weighed and blended for even distribution. Weighed quantity of the extract was dissolved in methanol and gradually added to the mixed powders, forming a wet mass. This mass was passed through a granulator to create granules, with the size controlled by the mesh size. The granules were then dried using suitable equipment to achieve the desired moisture content, ensuring they remained free of clumps and stable. Finally, the dried granules were sieved to obtain the desired particle size.

B. Drug loaded silica beads (Agrafioti et al., 2023)

The most potent methanolic extract, based on LC50 values, was selected for formulation. The concentration was calculated to target approximately 100 insects infesting 1 kg of green gram, resulting in a dose of 950 mg. This precise calculation ensures a rapid impact and extended prevention of insect infestation. Silica gel was dispersed in water to create a uniform suspension, continuously stirred to prevent clumping. A methanolic extract, containing 950 mg of the active ingredient, was added to the silica suspension and stirred gently to ensure proper mixing. Surfactants such as SLS were used to improve drug loading efficiency. Continuous stirring facilitated the adsorption of the drug onto the silica surface. The drug-loaded silica suspension was then filtered to separate the solid particles from the liquid. The collected solid material was dried in an oven to remove residual solvent and achieve the desired dryness. The dried material was passed through sieves to obtain uniformly sized beads. These sized silica beads were placed in a desiccator with a desiccant to remove any remaining moisture. The final drug-loaded silica beads were stored in a dry, airtight container until further use.

Evaluation Parameters for Pesticidal Formulations (Zhang et al., 2023)

Both the formulations, effervescent granules and drug loaded silica beads were evaluated using some common parameters such as organoleptic evaluation, uniformity of granule size, moisture content, pH, content uniformity and pesticidal activity. Additionally, the effervescent granules were specifically evaluated for effervescence time, disintegration time and dissolution rate. Whereas, drug loaded silica beads were specifically evaluated for release rate (Table-1).

Table 1: Formulae for one dose of effervescent granules

Sl. No.	Ingredients	Formula 1	Formula 2	Formula 3
1	Methanolic extract of DH	90 mg	85 mg	60 mg
2	Sodium bicarbonate	1600 mg	1700 mg	1900 mg
3	Citric acid	250 mg	275 mg	350 mg
4	Microcrystalline cellulose	350 mg	375 mg	400 mg
5	Lactose	2.3 g	2.2 g	1.745 g
6	Magnesium stearate	8 mg	8 mg	8 mg
7	Sodium starch glycolate	270 mg	300 mg	350 mg

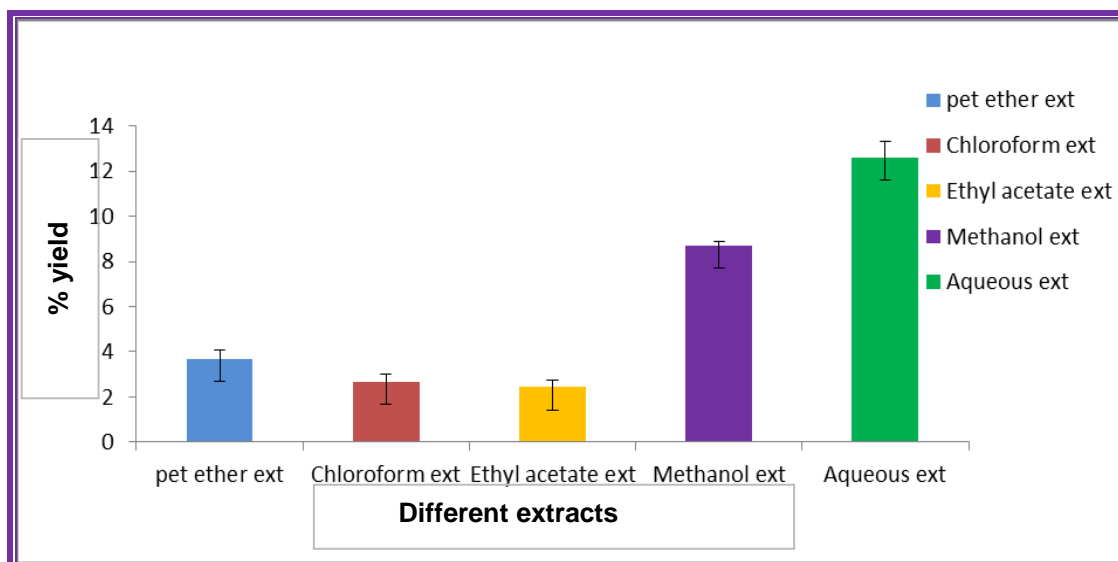
Results

Extraction of plant material

Successive solvent extraction of *Decalepis hamiltonii* leaves using the Soxhlet apparatus revealed that the aqueous extract had the highest yield (12.6±0.71%) and the ethyl acetate extract had the lowest yield (2.43±0.32%), with the methanol extract yielding 8.693±0.21%. The yields and colours of the extracts are summarized in Table 2, with a graphical representation in Graph 1.

Table 2: Properties and percentage yield of successive solvent extracts

Extract	% Yield	Color
Pet ether	3.67±0.405	Yellowish brown
Chloroform	2.67±0.33	Blackish green
Ethyl acetate	2.43±0.32%	Green
Methanol	8.693±0.21%	Blackish green
Aqueous	12.6±0.71%	Light brown



Graph 1: Percentage Yield of *Decalepis hamiltonii* Leaf Extracts by Successive Solvent Extraction

GSMS study:

Various bioactive compounds were identified by GCMS study and Identification of chemical constituents was carried out by comparing mass spectra and retention indices with reference data from the NIST and Wiley libraries. The compounds detected were confirmed through spectral matching with the GC–MS computer library, and the results were tabulated accordingly (listed in table-3 and showed in Figure-1).

Table-3: Important Phytochemicals of *Decalepis hamiltonii* by GCMS

Rt	Compound name	CAS#	Formula	Component area	Match factor
12.5732	Methyl salicylate	119-36-8	C8H8O3	97992319.5	98.2
12.6493	Dodecane	112-40-3	C12H26	72877563.9	97.4
13.9317	D-Carvone	2244-16-8	C10H14O	4250763.3	81.8
21.2796	Diethylcyanamide	617-83-4	C5H10N2	2598753.1	60.3
21.3570	Diethyl Phthalate	84-66-2	C12H14O4	507103794.6	95.2
21.4172	4-Cyanoimidazole	57090-88-7	C4H3N3	69454348.9	73.7
21.4575	Isoflavone	574-12-9	C15H10O2	16467785.9	61.5
23.5208	Tetradecanoic acid	544-63-8	C14H28O2	15273072.0	77.9
23.5999	Benzyl Benzoate	120-51-4	C14H12O2	31739261.7	95.7
23.8522	Octadecane	593-45-3	C18H38	26948800.4	97.0
23.9546	Dodecylcyclohexane	1795-17-1	C18H36	2884780.1	78.3
24.2971	Neophytadiene	504-96-1	C20H38	6728865.9	92.1
24.7558	Neophytadiene	504-96-1	C20H38	15384874.0	71.1
25.7017	Dibutyl phthalate	84-74-2	C16H22O4	17532120.0	92.1
25.7194	n-Hexadecanoic acid	57-10-3	C16H32O2	315659867.7	96.0
26.2407	Dodecylcyclohexane	1795-17-1	C18H36	4675004.4	74.0
26.2937	2,6-diphenyl-3,4-dimethylpyridine	1000379-00-8	C19H17N	522671.0	61.0
26.5366	Heptadecane	629-78-7	C17H36	2310085.6	64.0
27.2309	Heneicosane	629-94-7	C21H44	2417406.4	62.4
27.4170	Phytol	150-86-7	C20H40O	63146392.6	96.8
27.5549	Methyl stearate	112-61-8	C19H38O2	844205.1	60.3
28.3057	Docosane	629-97-0	C22H46	12615111.8	77.7
28.6859	Tetracosane	646-31-1	C24H50	6696846.1	77.3
29.3033	Tricosane	638-67-5	C23H48	26479853.5	94.6

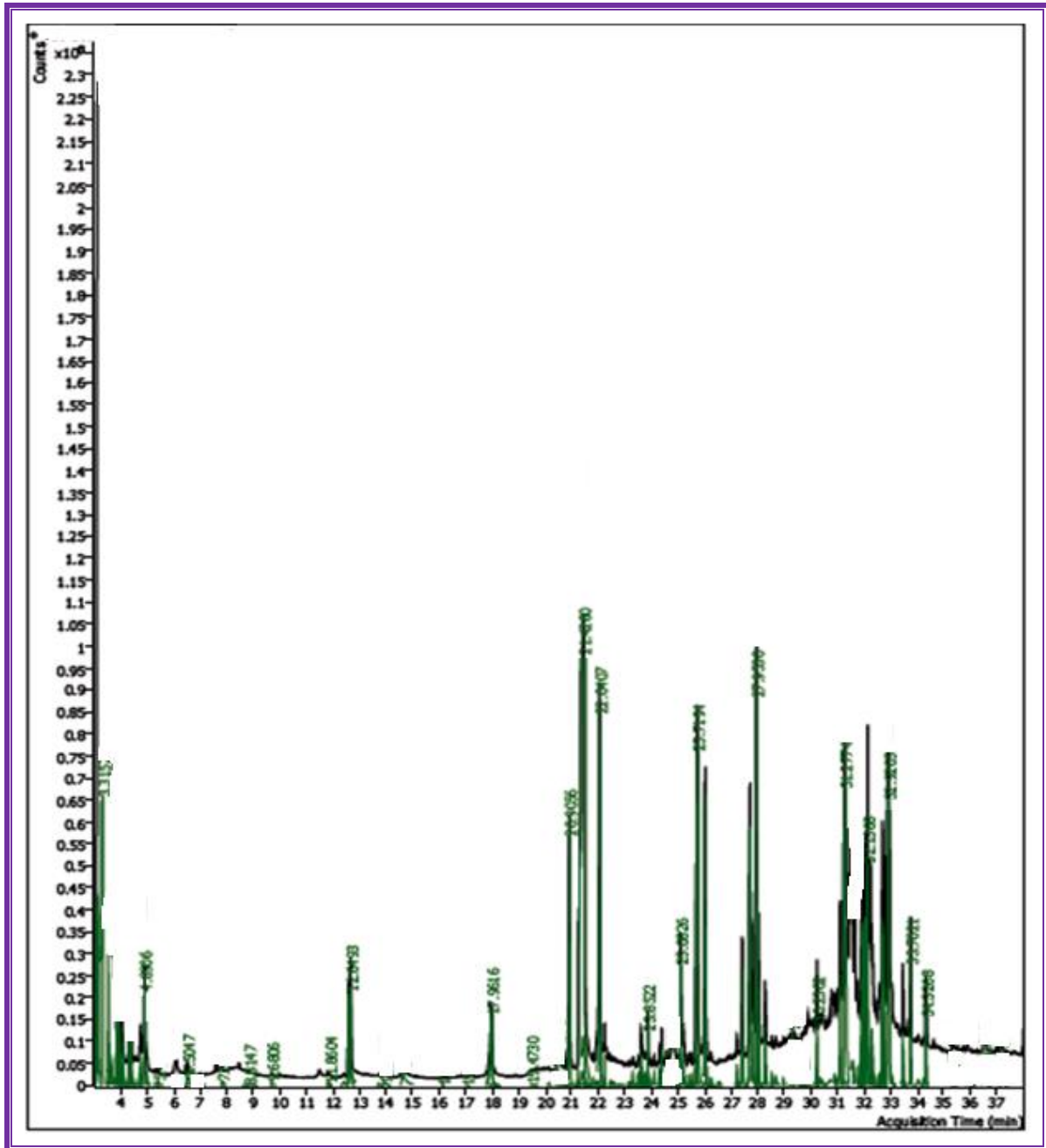


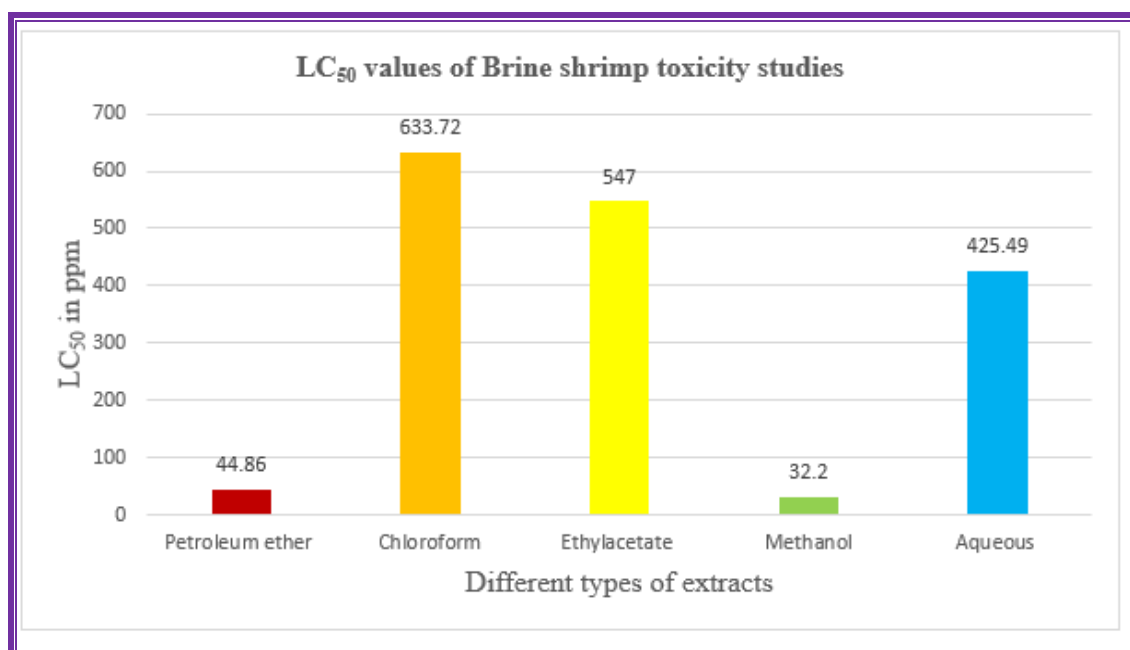
Figure 1: GCMS of *Decalepis hamiltonii* methanol leaves extract

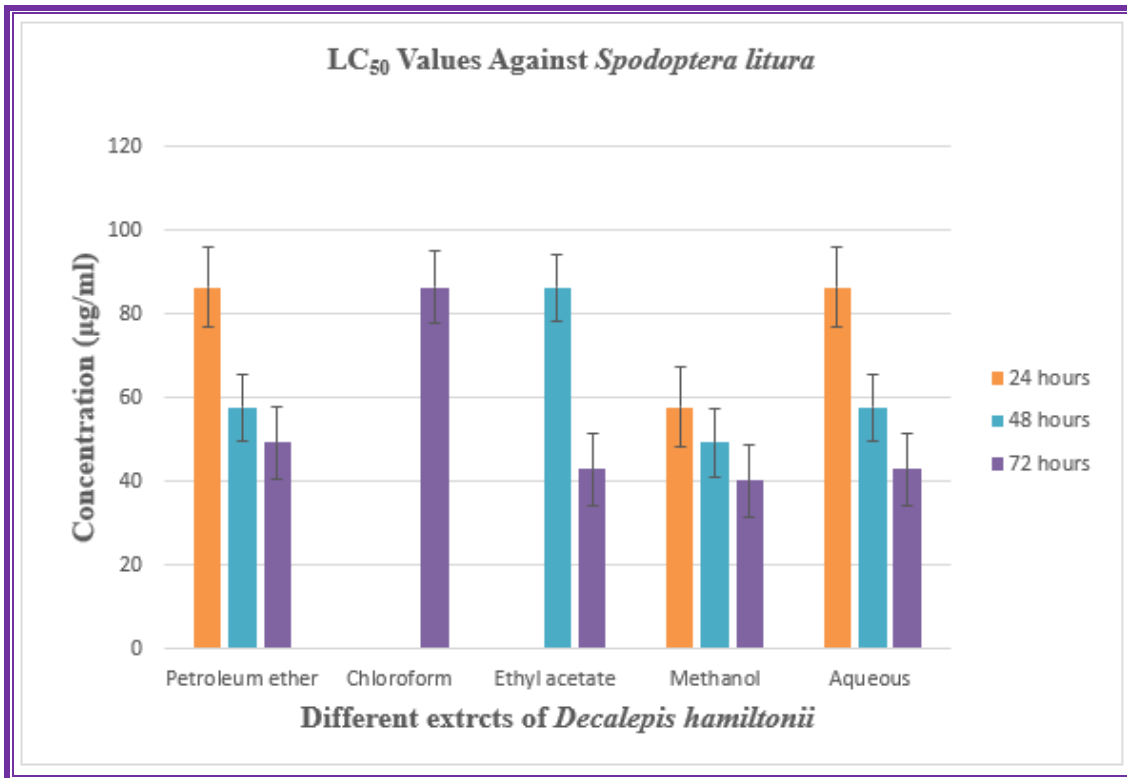
Evaluation of toxicity with brine shrimp assay

The brine shrimp assay is a dependable tool for assessing the cytotoxicity of phytochemicals. Our investigation focused on solvent extracts from *Decalepis hamiltonii* leaves, evaluating their lethality on brine shrimp nauplii as potential bioactive agents. Results indicated mortality increased with increase in extract concentration, with the methanolic extract demonstrating the highest mortality rate (8.3%) and a lower LC₅₀ (32.2029), highlighting its potent pesticidal efficacy. This LC₅₀ value guided pesticide formulation, ensuring effectiveness while maintaining safety standards. Detailed results are given in Table-4 maintaining safety standards. Detailed results are given in Table 2, and visual representation is given in Graphs 2, 3, and 4.

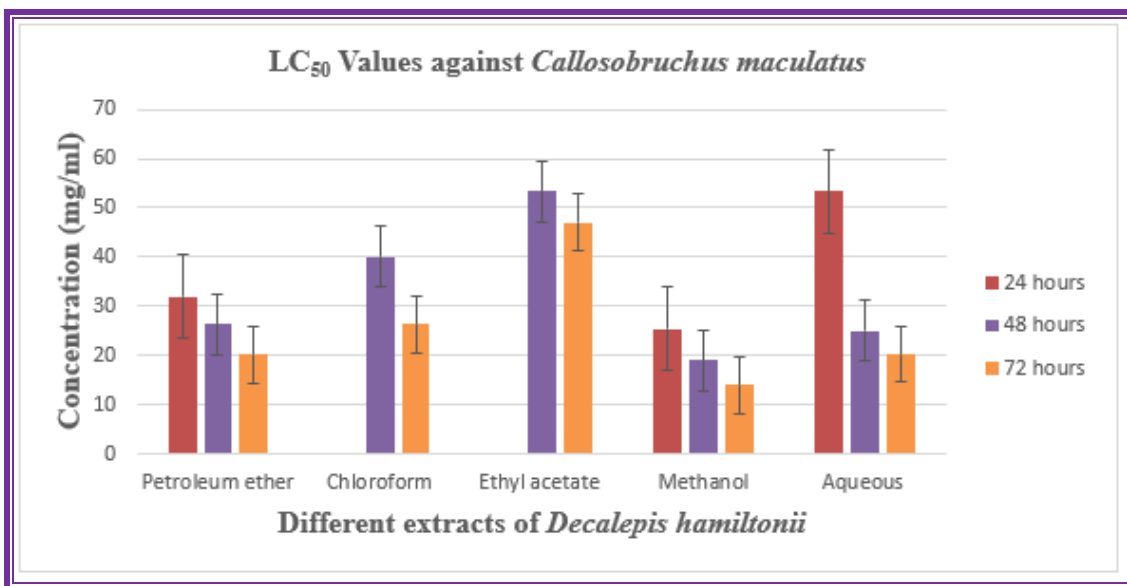
Table 4: Brine Shrimp Toxicity Studies of *Decalepis hamiltonii* Extracts

Extracts	Concentration (ppm)	No. of Nauplii Tested	No. of Nauplii Survived	No. of Nauplii Dead	% Mortality	LC ₅₀ (ppm)
Control	Distilled water	30	30	0	0	-
Petroleum ether	1	30	30	0	0	44.86
	10	30	29	1	3.33	
	100	30	24	6	20	
	1000	30	22	8	26.6	
Chloroform	1	30	29	1	3.33	633.72
	10	30	27	3	10	
	100	30	25	5	16.66	
	1000	30	20	10	33.3	
Ethyl acetate	1	30	30	0	0	547.00
	10	30	30	0	0	
	100	30	30	0	0	
	1000	30	29	1	3.33	
Methanol	1	30	26	4	13.33	32.20
	10	30	19	11	36.67	
	100	30	12	18	60	
	1000	30	5	25	83.33	
Aqueous	1	30	25	5	16.6	425.49
	10	30	20	10	33.3	
	100	30	14	16	53.6	
	1000	30	6	24	80	

**Graph 2:** LC₅₀ values of various extracts of *Decalepis hamiltonii* on Brine shrimp nauplii cells



Graph 3: LC₅₀ of *Decalepis hamiltonii* extracts against *Spodoptera litura*



Graph 4: LC₅₀ of *Decalepis hamiltonii* extracts on *Callosobruchus maculatus*

Evaluation of Pesticidal Activity

A. Leaf dip bioassay method using *Spodoptera litura*

Based on the results of the leaf dip bioassay, the methanolic extract exhibited the highest pesticidal activity, achieving 60% mortality with LC₅₀ 40.30 µg/ml at 50 µg/ml after 72 hours, with significant activity even at lower concentrations, highlighting its efficacy. The aqueous extract showed moderate efficacy, with up to 40% mortality with LC₅₀ 43.04 µg/ml, at 50 µg/ml after 72 hours and consistent activity at 40 µg/ml. The ethyl acetate extract displayed low activity, reaching 40% mortality with LC₅₀ 43.04 µg/ml only at higher concentrations, and minimal impact at lower concentrations. Both the petroleum ether and chloroform extracts exhibited negligible pesticidal activity, with mortality rates

peaking at 40% and 20% respectively at 50 µg/ml after 72 hours, and little to no effect at lower concentrations (Table-5). Overall, the methanolic extract was the most potent, followed by the aqueous extract, while the petroleum ether, chloroform, and ethyl acetate extracts showed minimal efficacy.

Table 5: Pesticidal Activity of *Decalepis hamiltonii* extracts on *Spodoptera litura*

Extract	Concentration (µg/ml)	Mortality Rate (%) after			LC ₅₀ (µg/ml) after		
		24 hours	48 hours	72 hours	24 hours	48 hours	72 hours
Petroleum ether	10	0	0	0	86.38	57.73	49.43
	20	0	0	0			
	30	0	20	20			
	40	20	20	40			
	50	20	20	40			
Chloroform	10	0	0	0	N/A	N/A	86.38
	20	0	0	0			
	30	0	0	0			
	40	0	0	20			
	50	0	0	20			
Ethyl acetate	10	0	0	0	N/A	86.38	43.04
	20	0	0	20			
	30	0	0	20			
	40	0	20	40			
	50	0	20	40			
Methanol	10	0	0	0	57.73	49.43	40.30
	20	0	0	20			
	30	20	20	20			
	40	20	40	40			
	50	20	40	60			
Aqueous	10	0	0	0	86.38	57.73	43.04
	20	0	0	20			
	30	0	20	20			
	40	20	20	40			
	50	20	20	40			

B. Residual bioassay method using *Callosobruchus maculatus*

Based on the results of the residual bioassay method, the methanolic extract exhibited the highest pesticidal activity, achieving 90% mortality with LC₅₀ 14.03 mg/ml at 25 mg/ml after 72 hours, with significant activity even at lower concentrations, highlighting its efficacy. Both petroleum ether and aqueous extract showed moderate efficacy, with up to 50% mortality with LC₅₀ 20.22 and 20.27 mg/ml respectively, at 25 mg/ml after 72 hours. Chloroform extract exhibited moderate mortality rates at 25 mg/ml after 72 hours of exposure, reaching 40% with LC₅₀ 26.29 mg/ml. Ethyl acetate extract exhibited low mortality rate of 20% with LC₅₀ 47.05 mg/ml at the highest concentration even after 72 hours of exposure. Overall, the methanolic extract was the most potent, followed by the petroleum ether and aqueous extract, while the chloroform, and ethyl acetate extracts showed minimal efficacy (Table-6). The pesticidal activity of *Decalepis hamiltonii* extracts against *Callosobruchus maculatus* revealed clear differences in efficacy among solvents. Abbott's correction confirmed that mortality values remained unchanged, as control mortality was zero. Among the extracts tested, methanol exhibited the strongest activity, producing up to 90% mortality at 25 mg/ml after 72 hours, with the lowest LC₅₀ values across all time points. Petroleum ether and aqueous extracts showed moderate activity, while chloroform and ethyl acetate extracts were comparatively less effective. The dose-dependent increase in mortality highlights the potential of methanolic extracts as a promising biopesticide candidate. These findings suggest that solvent choice significantly influences the extraction of active phytochemicals, and methanol may be most effective in isolating compounds with insecticidal properties. Further phytochemical characterization and mechanism-based studies are warranted to validate these results and explore their practical application in sustainable pest management.

Table 6: Pesticidal Activity of *Decalepis hamiltonii* Extracts on *Callosobruchus maculatus* (Abbott's Corrected Mortality)

Extract	Concentration (mg/ml)	Mortality Rate (%) after 24h	48h	72h	Corrected Mortality (%) after 24h	48h	72h	LC ₅₀ (mg/ml) after 24h	48h	72h
Petroleum ether	5	0	0	0	0	0	0	31.91	26.42	20.22
	10	0	0	10	0	0	10			
	15	10	20	40	10	20	40			
	20	10	30	40	10	30	40			
	25	20	30	50	20	30	50			
Chloroform	5	0	0	0	0	0	0	N/A	40.06	26.29
	10	0	0	0	0	0	0			
	15	0	0	10	0	0	10			
	20	0	20	30	0	20	30			
	25	0	30	40	0	30	40			
Ethyl acetate	5	0	0	0	0	0	0	N/A	53.42	47.05
	10	0	0	0	0	0	0			
	15	0	0	0	0	0	0			
	20	0	10	10	0	10	10			
	25	0	10	20	0	10	20			
Methanol	5	0	0	10	0	0	10	25.41	19.06	14.03
	10	0	20	30	0	20	30			
	15	10	30	40	10	30	40			
	20	30	40	60	30	40	60			
	25	50	70	90	50	70	90			
Aqueous	5	0	0	0	0	0	0	53.42	25.09	20.27
	10	0	10	20	0	10	20			
	15	0	20	30	0	20	30			
	20	10	30	40	10	30	40			
	25	10	20	50	10	20	50			

Evaluation of Pesticidal Formulations

A. Effervescent Granules

Three different formulas were utilized to prepare effervescent granules, with the third formula, which included *Decalepis hamiltonii* methanolic extract at a concentration of 60 mg per dose, demonstrating superior activity and stability compared to the first and second formulas. The concentration was calculated for approximately 1000 insects infested in 10 plants, based on the LC₅₀, ensuring rapid impact and extended prevention of pest infestation.

The effervescent granules incorporating the methanolic extract exhibited significant pesticidal activity. Standardization of the granules showed that they were spherical, sweet warm in flavour, and pleasant in odour. The particle size was uniform, around 1.18 mm, and the disintegration time was approximately 2 minutes and 43 seconds. Dissolution rate studies indicated that dissolution was higher between 5 to 15 minutes and also prepared the label for the safety use (Figure-2 and 3). HPLC quantification revealed that the disintegrated granules contained approximately 4.36% vanillin content (Graph-5). The results of the optimized formulation have been represented in table 7 to 8.



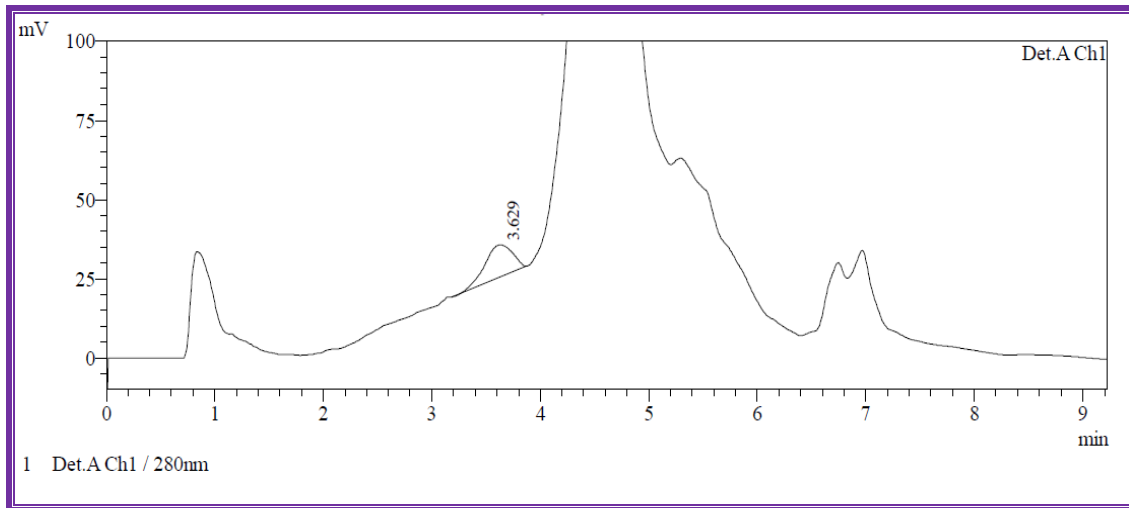
Figure 2: Drug incorporated Effervescent granules

<p>Warnings:</p> <ul style="list-style-type: none"> • For Agricultural use only. Not for human or animal consumption. Keep out of reach of children and pets. • Avoid contact with eyes and skin. In case of contact, rinse thoroughly with water. • Do not ingest. If swallowed, seek medical attention immediately. • Use appropriate protective clothing and equipment during handling and application. • Wash hands thoroughly after use. <p>Storage: Store in a cool, dry place. Keep the single dose packet sealed until use. Keep away from food, feed and seeds.</p>	<p align="center"><u>Pest Shield Granules</u></p> <p align="center">Herbal Effervescent Granules for plant infestation control</p> <p align="center">Net wt.: 10 packets*5g</p> <p>Each packet contains 5g of effervescent granules with 60 mg of DH methanol extract.</p> <p>Direction for use: Dissolve 1 dose (5g) in 500 ml of water completely. Spray the solution on the leaves above a infested plants.</p> <p>Repeat the application as needed, following plant specific guidelines.</p>	<p>Manufactured date: Oct 2023</p> <p>Expiry date: Nov 2025</p> <p>Manufactured at: Government College of Pharmacy, Bangalore.</p> <p>Batch No: PG23DLSB01</p> <p>Caution: Please read the instructions carefully before use</p> <p><i>This unique formulation, enriched with Decalepis hamiltonii, is designed to protect your grains effectively. Use as directed for best results.</i></p>
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Figure 3: Label for the package of Effervescent Granules

Table 7: Evaluation of Effervescent Granules and Drug Loaded Silica Beads

Sl. No.	Parameters	Effervescent Granules	Drug Loaded Silica Beads
1.	Organoleptic evaluation		
	Colour	Off-white, buff	Greenish yellow
	Shape	Irregular granules	Spherical granules
	Odour	Pleasant	No odour
	Flavour	Slightly sweet	No flavour
	Texture	Smooth, coarse	Smooth, coarse
2.	Uniformity of Granule size	Approximately 80% of the granules were retained by sieve number 16 (particle size > 1.18 mm). The remaining 20% had particle sizes ranging from 1.17 mm to 0.7 mm	95% of the silica gel beads were in the range of 1-3 mm
3.	Moisture content	0.2%	0.103%
4.	Effervescent Time	2.43 mins	-
5.	Disintegration Time	6.7 mins	-
6.	pH of the solution	6.63	5.7
7.	Dissolution rate	Higher below 15 mins, indicating faster dissolution and release of the extract into the solution	Higher at around 30 minutes and lower below 15 minutes, indicating that the drug-loaded silica beads do not leach out the extract onto the food grains
8.	Content uniformity of active substance	HPLC analysis showed that the effervescent granules contain 4.36% vanillin content	The % vanillin content was found to be 0.092%.
9.	Pesticidal activity	5 g of effervescent granules dissolved in 500 ml of water resulted in 80% mortality	8 insects were dead among 10 insects in a period of 15 days.



Graph 5: HPLC chromatogram of effervescent granules

Table 8: Dissolution studies of Effervescent granules

Sl. No.	Time	Absorbance	Dissolution Rate (g/L/min)
1	15 mins	0.986	0.0656
2	30 mins	1.174	0.0125
3	60 mins	1.513	0.0113

B. Drug Loaded Silica Beads

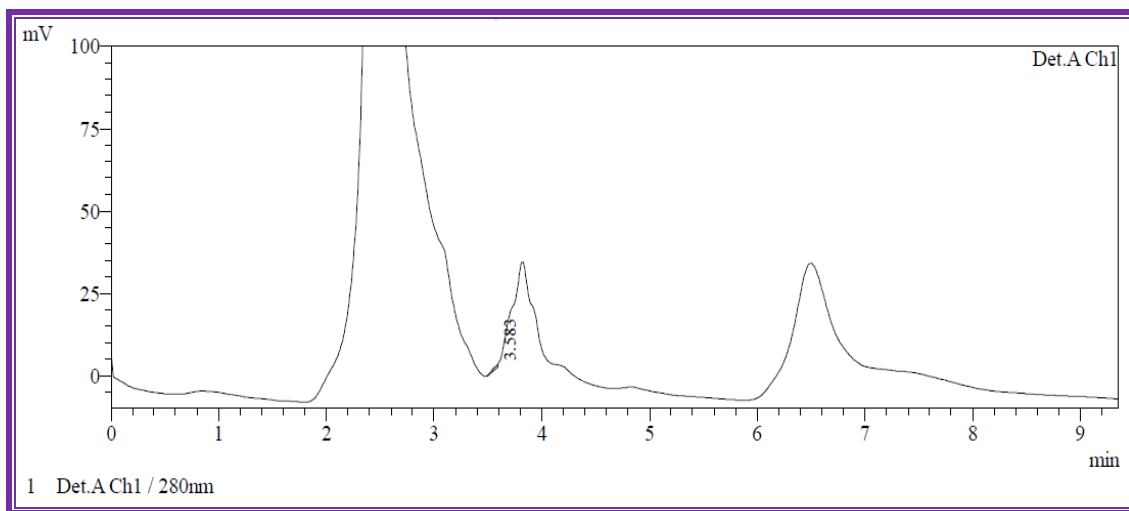
Silica beads loaded with *Decalepis hamiltonii* methanolic extract at a concentration of 951.5 mg were prepared to combat food grain pests, demonstrating significant effectiveness with an 80% mortality rate among approximately 100 insects infesting 1 kg of green gram, based on the LC₅₀ at 48 hours. The beads were thoroughly standardized, displaying a slight green color, smooth texture, and no discernible odour or flavor and also prepared the label for the safety use (Figure-4 and 5). High-Performance Liquid Chromatography (HPLC) quantification revealed a release rate of approximately 0.092%, indicating minimal deposition of the drug on the grain, with a low release rate of 0.02% further supporting efficient drug delivery while minimizing its impact on the grain (Graph-6). This meticulous preparation ensures rapid pest control and extended prevention of infestations, maintaining grain quality (Table-9).



Figure 4: Drug incorporated silica beads and single dose packed

<p>Warnings:</p> <p>For Agricultural use only keep out of reach of children and pets.</p> <p>Do not open, consume or inhale.</p> <p>Wear gloves and a mask while handling the product.</p> <p>Remove the packet before using the grains.</p> <p>Storage: Store in cool and dry place. Keep the single dose packet sealed until use. Keep away from food, feed and seeds.</p>	<p>Grain Guardian Silica Beads</p> <p>Net weight: 25 packets*3g</p> <p>Each single dose package contains 950mg of <i>Decalepis hamiltonii</i> methanolic extract.</p> <p>Direction for use:</p> <ol style="list-style-type: none"> 1. Place one unopen single dose packet directly in between every 1 kg of infested grains. 2. Do not open or remove the packet before using the grains. 3. Seal the infested grain container tightly 4. Store in a cool, dry place away from direct sunlight. 	<p>Manufactured date: Oct 2023</p> <p>Expiry date: Nov 2025</p> <p>Manufactured at: Government College of Pharmacy, Bangalore.</p> <p>Batch No: PG23DLSB01</p> <p>Caution: Please read the instructions carefully before use</p> <p><i>This unique formulation, enriched with Decalepis hamiltonii, is designed to protect your grains effectively. Use as directed for best results.</i></p>
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Figure 5: Label of the final package of Drug loaded silica beads



Graph 6: HPLC chromatogram of Drug loaded silica beads

Table 9: Release rate studies of Drug loaded silica beads

Sl. No.	Time	Absorbance	Dissolution Rate (µg/ml/min)
1	15 mins	0.986	0.0023 ± 1.231
2	30 mins	1.174	0.0043 ± 2.103
3	60 mins	1.513	0.0011± 2.042

Mean ± SD (n=3)

Discussion

Decalepis hamiltonii, a longstanding Ayurvedic remedy, is valued for its therapeutic uses, including appetite stimulation, relief from flatulence, and as a general vitalizer. Its root has been extensively employed in treating various ailments such as fever, skin diseases, diarrhoea, and CNS disorders. However, *Decalepis hamiltonii* faces the threat of overexploitation due to the mortality valuable medicinal properties found in its roots (Sun et al., 2019). Rampant root harvesting, devoid of sustainable practices and awareness about later-generation sources, has resulted in widespread plant destruction. Environmental changes in its habitat further exacerbate this issue, leading to the endangerment of this valuable species. Therefore, it urgently requires conservation efforts.

To address this challenge and promote sustainability, researchers are exploring the medicinal properties of other parts of the plant, such as the leaves. Shifting the focus to leaves is a proactive

approach to diversify the utilization of the plant, reducing the pressure on its endangered root part and facilitating the conservation of this valuable species for future generations.

Decalepis hamiltonii was selected for its broad range of potential pharmacological activities, notably its insecticidal properties. This choice aligns with traditional knowledge, which indicates that storing *Decalepis hamiltonii* roots for extended periods renders them unaffected by microorganisms and insects due to the plant's unique phytochemical composition and bacteriostatic properties (Mustapha et al., 2022). Despite these indications, scientific documentation on the phytochemicals of *Decalepis hamiltonii* is limited. To bridge this gap, we initiated this study to assess its potential pesticidal properties.

The methanolic extract of *Decalepis hamiltonii* was collected from IIHR, where it was cultivated specifically for preservation and scientific research. Initially, to assess the safety of this extract for potential pharmacological activities (Ashalatha et al., 2019), a brine shrimp lethality assay was performed. The methanolic extract demonstrated an LC₅₀ of very low, which falls within the acceptable limit range for toxicity, indicating that the extracts are safe for further evaluation of pesticidal and insecticidal activities.

Excessive pesticide use, with its high toxicity and non-biodegradability, poses a pressing concern. Addressing this issue requires a dual strategy: intensive research to discover highly selective and biodegradable pesticides and exploring eco-friendly pesticides and organic techniques to reduce synthetic pesticide usage while maintaining optimal crop yields. "Green Pesticides," utilizing natural products, offer a promising alternative by effectively controlling pests while being safe and eco-friendly (Gitanjali et al., 2023). In this context, *Decalepis hamiltonii* has emerged as a successful herbal pesticide, addressing the need for environmentally compatible alternatives.

Pesticidal evaluation was conducted using the leaf dip bioassay method against *Spodoptera litura*, a common and threatening pest in agriculture known for voraciously feeding on leaves and causing significant plant damage. This study revealed a dose-dependent increase in pesticidal activity, with the highest activity observed in the methanol extract (LC₅₀ 40.30 µg/ml) of *Decalepis hamiltonii*. In contrast, chloroform extracts exhibited the lowest activity, followed by petroleum ether and ethyl acetate extracts. This variation in activity could be attributed to the higher concentration of phytochemicals found in methanol extract compared to other extracts.

The Pesticidal activity was also studied through the Residual Contact Toxicity method against *Callosobruchus maculatus*, a dangerous pest that infests and spreads in stored food grains. This method demonstrated a significant increase in mortality rates corresponding to higher plant extract concentrations and longer exposure times. Methanolic and aqueous extracts of *Decalepis hamiltonii* displayed superior efficacy (LC₅₀ 14.03 mg/ml), while ethyl acetate extracts exhibited the least activity. This suggests that the extract possesses insecticidal properties at an average rate, exhibiting maximum activity at mean exposure, without requiring either long or immediate exposure.

To further explore the practical application of this useful plant, we attempted to make formulations using the methanolic extract. Considering the LC₅₀ value, the dosage for formulating treatments can be determined to minimize toxicity to non-target pests. Effervescent granules and drug-loaded silica beads particularly for pesticidal use, were formulated. Effervescent granules, an innovative formulation for pesticidal use, offer advantages such as easy application, biodegradability, and economic feasibility (Souto et al., 2021). The faster dissolution property of the effervescent granules is primarily responsible for paralyzing the insects due to its froth, thereby creating a way for the drug to exhibit its pesticidal activity.

Silica gels were incorporated into formulations to combine pesticidal properties with desiccant properties. The use of porous materials as the drug-loading core leverages their high surface area. Drug-loaded silica beads were formulated for pesticidal use against food grain storage pests. These scientific details serve as a valuable reference for future research on the same plant and reveal

promising results, establishing *Decalepis hamiltonii* as a potential herb with pesticidal properties that could be applied in practical settings.

Finally, GCMS study revealed the presence of acids, acid esters, aliphatic alcohols and other terpenoids in higher amount. Among them some are important and have their pesticidal activities namely Benzyl Benzoate, Docosane and Octadecane. In one study it was resulted that Docosane acts as pesticidal activity against the cowpea beetle *Callosobruchus maculatus* (Huynh et al., 2023). Earlier study revealed that benzyl benzoate had a significant insecticidal effect, with the LC50 at a lower concentration in the direct-contact toxicity assay (Dimetry et al., 2015). Recently, Octadecane was isolated from *Spodoptera frugiperda* and identified as potent pesticidal activity (Aboelhadid et al., 2023; Sharanappa et al., 2024).

Limitations

The present research on *Decalepis hamiltonii* highlights its strong potential as a natural pesticide, but certain limitations must be acknowledged. The experiments were largely confined to laboratory bioassays, which may not fully replicate field conditions where environmental variables influence efficacy. Toxicity evaluation was restricted to brine shrimp assays, leaving scope for broader safety assessments on non-target organisms and humans. Moreover, the formulations developed—effervescent granules and drug-loaded silica beads—were tested only on a small scale, and their long-term stability and performance in real storage environments remain to be validated.

Future Scope

Looking ahead, future studies should focus on large-scale field trials under diverse agro-climatic conditions to confirm laboratory findings. Advanced formulation strategies, such as nano-delivery systems, could be explored to enhance controlled release and efficacy. Detailed phytochemical characterization and mechanism-based studies will help identify the active compounds responsible for insecticidal activity. Importantly, integrating *D. hamiltonii* extracts into sustainable pest management programs could reduce dependence on synthetic pesticides while supporting biodiversity conservation. By shifting focus from roots to leaves, this research also contributes to conservation efforts, ensuring the long-term survival of this endangered medicinal plant while promoting eco-friendly pest control solutions.

Conclusion

Natural molecules hold greater potential compared to their synthetic counterparts, primarily due to their low toxicity and favourable tolerance responses. The preference for natural remedies is often rooted in the belief that they are safer and have fewer side effects than synthetic options. This study represents a significant step forward in pest management research by highlighting the outstanding pesticidal activity of methanolic and aqueous extracts from *Decalepis hamiltonii*, a traditional medicinal plant as effective as neem, pyrethrum, and tobacco plants. These extracts demonstrated high efficacy against pests like *Spodoptera litura* and *Callosobruchus maculatus* without posing toxic side effects to humans, animals, or the environment and constituents like Benzyl Benzoate, Docosane and Octadecane which may involved for the said activity. This non-toxic nature positions them as promising alternatives to synthetic pesticides, offering a safer and eco-friendly solution for effective pest control.

Conflict of interest

The authors declare that they have no conflicts of interest among the authors.

Acknowledgement

The authors are thankful to institutional authority for the infrastructural support.

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