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"DEVELOPMENT AND STANDARDIZATION OF NATURAL AND SAFE LIQUID ORAL ANTHELMINTIC FORMULATION COMPRISING LEAVES OF *DOLICHANDRONE FALCATA*"

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A B S T R A C T :

Background and Objective:

Helminthiasis, or parasitic worm infection, remains a major global health concern, emphasizing the need for effective, safe, and affordable anthelmintic agents from natural sources. *Dolichandrone falcata*, a traditionally used medicinal plant, was selected to scientifically evaluate its anthelmintic potential and develop a standardized phytosome-based liquid oral formulation.

Methods:

Comprehensive pharmacognostical studies were conducted, and successive solvent extraction of *D. falcata* leaves was performed using petroleum ether, chloroform, ethyl acetate, methanol, and water. Extracts were analyzed for phytochemical constituents, and the most active fraction was evaluated for Total Phenolic Content (TPC), Total Flavonoid Content (TFC), and Total Tannin Content (TTC). Anthelmintic efficacy was tested in-vitro using *Pheretima posthuma*, while toxicity was assessed via the Brine Shrimp Lethality Assay (BSLA). The extract showing maximum efficacy and minimum toxicity was used to prepare phytosomes, which were characterized for Entrapment Efficiency (EE), Particle Size (PS), and Zeta Potential (ZP), followed by formulation into a liquid oral suspension and evaluation of in-vitro drug release.

Results:

The ethyl acetate extract (EAE) showed the highest phytochemical content (TPC 21.80 ± 2.60 mg GAE/g, TFC 8.40 mg QE/g, TTC 15.18 mg TA/g) and superior HPTLC profile. EAE exhibited strong anthelmintic activity with paralysis and death times of 24.67 min and 33 min at 60 mg/mL, outperforming Albendazole (DT 38 ± 2.16 min, $p < 0.001$), and was least toxic (LC_{50} = 2666.7 µg/mL). The optimized phytosome (F2) showed high EE (82.56%), small PS (148 nm), and good stability (ZP –41.1 mV). The final suspension exhibited acceptable viscosity (130 cps) and sustained drug release (F2 = 75% after 8 h).

Conclusion:

The study validated *D. falcata*'s potent anthelmintic activity and developed a stable, non-toxic, and standardized phytosome-based liquid oral formulation. These findings support its potential as a natural therapeutic for helminthiasis, warranting further *In-vivo* and stability studies for future clinical and commercial applications.

Keywords: *Dolichandrone falcata*; Anthelmintic activity; HPTLC; Phytosome; Liquid Oral Formulation; Standardization.

1. INTRODUCTION

Helminthiasis, a disease caused by parasitic worms, remains one of the most prevalent infections affecting humans and animals worldwide, particularly in tropical and subtropical regions. These parasitic worms, collectively known as helminths, are classified into four main groups: roundworms (nematodes), flukes (trematodes), tapeworms (cestodes), and thorny-headed worms (acanthocephalans). While nematodes, trematodes, and cestodes are the primary infective groups in humans, thorny-headed worms mainly infect animals and only rarely affect humans. Transmission occurs through accidental ingestion of infective eggs or larvae, penetration of skin by larvae, vector bites, or consumption of infected hosts.

Helminth infections often go unnoticed in mild cases but can lead to serious clinical complications when untreated, including abdominal pain, diarrhea, blood loss, weight loss, malnutrition, cognitive impairment, and rectal prolapse. Severe infections are associated with intestinal obstruction, cholecystitis, pancreatitis, colonic inflammation, bile duct hyperplasia, blindness, and even death. Globally, soil-transmitted helminthiases such as ascariasis, trichuriasis, and hookworm infections affect over a billion people. Despite ongoing control programs, these diseases continue to pose major public health and socioeconomic challenges in developing nations.

Currently available synthetic anthelmintic drugs, including albendazole, mebendazole, pyrantel pamoate, levamisole, and ivermectin, are effective but associated with several drawbacks. Long-term use may cause gastrointestinal disturbances, nausea, dizziness, and other side effects. Additionally,

increasing drug resistance, high costs, and limited accessibility in low-resource areas emphasize the need for safe, effective, and affordable herbal alternatives. Hence, there has been growing interest in exploring medicinal plants as sources of novel anthelmintic agents.

Dolichandrone falcata (Family: Bignoniaceae), commonly known as Harwar (Hindi) and Vaddi (Kannada), is a traditionally used medicinal plant widely recognized in Ayurveda and folk medicine. It has been used to treat various ailments such as parasitic infections, inflammation, fever, and microbial diseases. Despite its ethnomedicinal importance, limited scientific validation exists regarding its pharmacological potential and formulation development. Phytochemical studies have reported that *D. falcata* leaves contain several bioactive constituents, including alkaloids, flavonoids, phenolics, and tannins, which are known to exhibit significant anthelmintic activity.

The mechanism of action of these compounds involves multiple biochemical pathways. Alkaloids are known to block the intake of acetylcholine at the neuromuscular junction of parasites, resulting in paralysis and expulsion from the host body. Tannins, being polyphenolic in nature, interfere with energy generation in helminths by uncoupling oxidative phosphorylation. They also form complexes with proteins and glycoproteins on the parasite's cuticle, leading to its death. Flavonoids exert anthelmintic effects by inhibiting key enzymes such as phosphodiesterase and Ca^{2+}-ATPase, thereby disrupting cellular homeostasis in parasites. These phytoconstituents collectively justify the plant's traditional use and support the scientific rationale for its pharmacological evaluation.

To enhance the therapeutic efficacy of herbal extracts, novel formulation approaches are increasingly being employed. One such advancement is **phytosome technology**, also known as the **phospholipid complex delivery system**, which bridges the gap between conventional herbal formulations and modern drug delivery systems. Developed by Indena, phytosomes are complexes of phytoconstituents with phospholipids that enhance the lipid compatibility, solubility, absorption, and bioavailability of water-soluble plant extracts. The interaction between the polar head of phospholipids (such as phosphatidylcholine) and the polar functional groups of plant molecules results in a stable complex that is better absorbed in biological membranes.

Phospholipids, especially soy-derived phosphatidylcholine, are biocompatible and efficiently absorbed in humans, achieving more than 90% absorption with no reported toxicity. They are integral components of cell membranes, improving fluidity and cellular uptake. Studies have shown that phytosomes significantly enhance pharmacokinetic and pharmacodynamic properties of herbal compounds compared to conventional extracts.

Phytosome technology offers numerous advantages, including enhanced bioavailability, improved therapeutic efficacy, reduced dose variability, and greater stability. Phosphatidylcholine acts not only as a carrier but also possesses hepatoprotective and membrane-stabilizing properties. The encapsulation of phytoconstituents within phospholipids protects them from enzymatic degradation and microbial metabolism in the gastrointestinal tract. Additionally, phytosomes can effectively transition between hydrophilic and lipophilic environments, enabling efficient systemic delivery and better tissue targeting. These properties make phytosomes an ideal vehicle for delivering plant-based molecules with poor solubility and limited absorption.

Considering these advantages, the present study focuses on the **development and evaluation of a phytosome-based drug delivery system using the leaves of *Dolichandrone falcata***. The research aims to scientifically validate the plant's **anthelmintic potential**, perform **standardized extraction and HPTLC profiling**, and develop a **stable liquid oral formulation** in the form of a **suspension**. The formulated phytosome suspension is further evaluated for physicochemical characteristics, entrapment efficiency, particle size, zeta potential, *In-vitro* anthelmintic activity, and drug release profile.

Ultimately, this study seeks to bridge the gap between traditional herbal knowledge and modern pharmaceutical science by developing an evidence-based, safe, and effective natural formulation for the management of helminthiasis. The successful formulation and standardization of *D. falcata* phytosomes could offer a cost-effective, biocompatible, and sustainable alternative to synthetic anthelmintic drugs, with significant implications for global health and herbal drug development.

2. METHODOLOGY

COLLECTION AND AUTHENTICATION OF LEAVES OF DOLICHANDRONE FALCATA.

Fresh leaves of *Dolichandrone falcata* were collected from local areas of Maharashtra Pune, Tornamal region in the month of December 2024. Identification and authentication of plant material was done at Central Ayurveda Research Institute, Bangalore by Dr. V. Rama Rao, Research Officer (Botany).

2.2 PHARMACOGNOSTICAL EVALUATION OF DOLICHANDRONE FALCATA LEAVES

2.2.1 Macroscopic Evaluation

Macroscopic characteristics such as size, shape, colour, odour, texture, and fracture of both fresh and dried *Dolichandrone falcata* leaves were studied as per WHO guidelines. The colour was examined under diffuse daylight, odour was determined by gentle inhalation, and size was measured using a graduated ruler in millimetres. The shape, surface characters, and texture were observed visually under natural light and confirmed by touch.

2.2.2 Microscopic Evaluation

Materials and Method:

Microscopic studies were performed using a compound microscope, watch glass, razor blade, coverslip, and reagents such as chloral hydrate, phloroglucinol, glycerine, and concentrated hydrochloric acid.

2.2.2.1 Transverse Section (T.S.):

Thin sections of *Dolichandrone falcata* leaves were cleared with chloral hydrate, stained with phloroglucinol and concentrated HCl, mounted in glycerine, and examined under 10x and 45x magnifications to study internal tissue organization.

2.2.2.2 Powder Microscopy:

Powdered leaf samples were treated with chloral hydrate and stained with phloroglucinol and concentrated HCl. Slides were mounted with glycerine and examined microscopically to identify characteristic components such as trichomes, stomata, xylem vessels, and calcium oxalate crystals.

2.2.2.3 Proximate Analysis of Leaf Powder

a. Determination of Moisture Content:

Accurately weighed powdered leaves were dried in a hot air oven at 100–105°C until a constant weight was achieved to determine the percentage of moisture content.

b. Determination of Total Ash:

The air-dried powdered drug was incinerated in a muffle furnace at 450°C to obtain the total ash, representing the total inorganic content.

c.Determination of Acid-Insoluble Ash:
The total ash was boiled with dilute hydrochloric acid and filtered. The residue was collected, ignited, and weighed to determine the acid-insoluble ash.

d.Determination of Water-Soluble Ash:
The total ash was boiled with distilled water and filtered. The difference between the total ash and the insoluble residue was used to calculate the water-soluble ash.

2.2.2.4. Determination of Extractive Values

a. Alcohol-Soluble Extractive:
A known weight of powdered leaves was macerated with ethanol for 24 hours with occasional shaking. The extract was filtered, evaporated to dryness, and weighed to determine the alcohol-soluble extractive value.
3. Water-Soluble Extractive:
Similarly, the powdered leaves were macerated with distilled water for 24 hours, filtered, evaporated, and weighed to obtain the water-soluble extractive value.

EXTRACTION

The dried and coarsely powdered leaves of *Dolichandrone falcata* were subjected to successive Soxhlet extraction using solvents in the order of increasing polarity — petroleum ether, chloroform, ethyl acetate, and methanol. A known quantity of powdered material was accurately weighed and packed into a thimble, which was then placed inside the Soxhlet extractor. The extraction process was first carried out with petroleum ether to remove non-polar compounds such as fats, waxes, and chlorophyll. The process was continued until the solvent in the siphon tube became colorless, indicating the completion of extraction. The petroleum ether extract was collected and concentrated by evaporating the solvent on a water bath, and the dried extract was stored in a desiccator for further analysis.

The marc obtained after petroleum ether extraction was air-dried to remove any residual solvent and then subjected to extraction with chloroform using the same procedure. The chloroform extract was similarly concentrated, dried, and stored. The remaining marc was further extracted with ethyl acetate, followed by methanol, maintaining the same extraction conditions for each solvent. Each extraction cycle continued until the siphoning solvent became clear. After completion of the successive extractions, all solvent extracts were concentrated to dryness using a water bath and kept in airtight containers to prevent moisture absorption. The percentage yield of each extract was calculated based on the weight of the dried extract obtained relative to the initial weight of the plant material.

This sequential extraction method ensures the systematic separation of phytoconstituents based on their solubility and polarity, allowing for effective isolation of both non-polar and polar bioactive compounds present in *Dolichandrone falcata* leaves.

Preliminary phytochemical tests

Standard color reactions were run on all extracts: carbohydrates (Molisch, Benedict, Fehling), proteins/amino acids (Biuret, Millon, Ninhydrin), fixed oils (filter paper), alkaloids (Dragendorff's, Wagner, Mayer, Hager), glycosides (pre/post-hydrolysis Fehling), flavonoids (alkali, lead acetate, Shinoda), tannins/polyphenols (FeCl3, gelatin, lead acetate), saponins (foam), steroids/triterpenoids (Liebermann–Burchard).

2.5 EVALUATION OF TOXICITY OF DOLICHANDRONE FALCATA LEAVES EXTRCT BY BRINE SHRIMP LETHALITY ASSAY (BSLA)

The toxicity of *Dolichandrone falcata* leaf extracts was evaluated using the Brine Shrimp Lethality Assay (BSLA), a simple and reliable method to assess the cytotoxic potential of plant extracts. Brine shrimp eggs (*Artemia salina*) were hatched in 0.9% sodium chloride (NaCl) solution with continuous aeration and illumination for 48 hours at room temperature to obtain active nauplii. Different concentrations (500, 1000, and 2000 µg/ml) of each extract—petroleum ether, chloroform, ethyl acetate, methanol, and aqueous—were prepared by dissolving the respective extract in a small amount of its solvent and diluting with 0.9% NaCl solution.

For the assay, 1 ml of each extract solution was added to 2 ml of 0.9% NaCl solution in separate test tubes. Ten brine shrimp nauplii were transferred into each test tube, while the control group contained only distilled water. All test tubes were maintained under light at room temperature, and the number of surviving nauplii was counted after 24 hours using a magnifying glass or microscope. Mortality was determined based on the absence of forward movement of larvae during 30 seconds of observation.

The percentage mortality of the brine shrimp larvae was calculated by comparing the number of living nauplii in the test samples with those in the control group after 24 hours. The LC_{50} value, representing the concentration that kills 50% of the larvae, was determined from the dose–response relationship. This assay provided a preliminary indication of the cytotoxicity and safety profile of the *D. falcata* extracts, helping to identify the least toxic and most suitable extract for further pharmacological evaluation.

2.6 TO SCREEN THE SUCCESSIVE EXTRACTS FOR IN VITRO ANTHELMINTIC ACTIVITY USING PHERETIMA POSTHUMA MODEL.

The in vitro anthelmintic activity of successive extracts of *Dolichandrone falcata* leaves was evaluated using adult earthworms (*Pheretima posthuma*), which are physiologically similar to intestinal roundworms of humans. Earthworms measuring approximately 8–10 cm in length and 0.2–0.3 cm in width were selected for the study. A total of 54 groups were prepared, each containing six worms. The groups included a control (2% v/v Tween 80 in warm

distilled water), a standard (Albendazole 20 mg/ml in 2% Tween 80), and test groups treated with petroleum ether, chloroform, ethyl acetate, methanol, and aqueous extracts of *D. falcata* leaves at concentrations of 20, 40, and 60 mg/ml. All extract and standard solutions were freshly prepared on the day of the experiment.

Before testing, the earthworms were washed thoroughly with distilled water to remove any adhering dirt. They were then transferred into Petri dishes containing 20 ml of the respective test or standard solutions. The worms were observed continuously, and the time taken for paralysis and death was recorded for each concentration. Paralysis was noted when the worms showed no movement except upon vigorous shaking, while death was confirmed when there was complete cessation of movement and loss of body color.

The concentrations used were selected based on previous toxicity studies indicating that the plant extract was safe up to 2000 mg/kg, with no observed mortality, ensuring that all test concentrations were within the non-toxic range. The data collected were analyzed using one-way ANOVA followed by Dunnett's test in GraphPad Prism (version 10.1), and results were expressed as mean values to determine the statistical significance of the extracts compared to the control and standard groups.

Total flavonoid content (TFC) : AlCl3 colorimetry at 415 nm with quercetin standards (10–80 µg/mL); extracts at defined dilutions; TFC expressed as quercetin equivalents using calibration linearity; triplicate determinations. $T=C\times V/M$

2.8 Total phenolic content (TPC): Folin–Ciocalteu assay at 725 nm with gallic acid standards (10–50 µg/mL), 20% Na2CO3, 40-min room-temperature development; TPC expressed as gallic acid equivalents via T=C×V/M; triplicates

2.9 Total Tannin Content (TTC): Folin–Ciocalteu method at 700 nm with tannic acid standards (20–100 µg/mL); extracts prepared at 1 mg/mL concentration; TTC expressed as tannic acid equivalents using calibration linearity; triplicate determinations

3.0 HPTLC Fingerprinting Analysis of Successive Solvent Extracts of *Dolichandrone falcata*

High-Performance Thin-Layer Chromatography (HPTLC) is a sophisticated chromatographic technique that provides high resolution and sensitivity for separating complex mixtures. It enhances the traditional thin-layer chromatography (TLC) by incorporating modern technological improvements, enabling precise and reproducible analysis. HPTLC is widely applied in the qualitative and quantitative analysis of phytochemicals, pharmaceuticals, and other complex substances.

For the analysis, a CAMAG HPTLC system was used along with pre-coated silica gel 60 F254 aluminium plates. The chemicals employed included toluene, ethyl acetate, formic acid, and methanol, all sourced from reputed suppliers. Standards such as quercetin, rutin, and gallic acid were used to identify phytochemical components in the extracts.

The standard solutions were prepared by weighing 10 mg of each reference compound, dissolving them in methanol with sonication to ensure complete dissolution, and making up the volume to 10 mL to achieve a concentration of 1 mg/mL. Similarly, each successive solvent extract of *Dolichandrone falcata* leaves (petroleum ether, chloroform, methanol, and aqueous extracts) was prepared at the same concentration. The solutions were filtered to remove any undissolved particles before analysis.

The mobile phase was optimized based on literature reports and consisted of toluene, ethyl acetate, formic acid, and methanol in the ratio of 3:4:0.8:0.7. This solvent system provided efficient separation of quercetin, rutin, and gallic acid and was employed for the chromatographic analysis of the leaf extracts.

Samples and standards were applied as 6 mm wide bands on silica gel plates using a CAMAG Linomat 5 applicator with a microlitre syringe. Plates were pre-washed with the mobile phase to ensure uniform migration, and a 5 mm distance was maintained between adjacent bands to prevent overlap. The plates were developed in a CAMAG Twin Trough chamber with a saturation time of 20 minutes. After development, densitometric scanning was performed at 254 nm using a CAMAG TLC Scanner III under absorbance measurement mode, with detection supported by deuterium and white lamps.

The analytical conditions including instrumentation and procedural parameters such as application volumes (2 µL for standards and 5 µL for samples), band width, development distance, and software version (WinCATS 1.4.6) were standardized to ensure reproducibility and accuracy of the fingerprinting analysis.

4.0 Development of *Dolichandrone falcata* Extract-Loaded Phytosome by Rotary Evaporation Method

The phytosome formulation of *Dolichandrone falcata* leaf extract was developed using the rotary evaporation technique. This method involves the complexation of the plant extract with soya lecithin to enhance its bioavailability and therapeutic potential. Two different formulations were prepared by varying the amount of soya lecithin while keeping the extract quantity consistent or varied accordingly.

In the first formulation, 1200 mg of soya lecithin was used with 1200 mg of the ethyl acetate extract. For the second formulation, the amount of soya lecithin was increased to 2400 mg while the extract quantity was scaled up to 12,000 mg to investigate the effect of phospholipid concentration on the phytosome characteristics. Both formulations employed a solvent system composed of 30 mL methanol and 20 mL dichloromethane to dissolve the components effectively.

After dissolving the extract and soya lecithin in the respective solvents, the mixture was subjected to rotary evaporation, which facilitates the removal of solvents under reduced pressure, resulting in the formation of a thin film. This film was then hydrated with 25 mL of phosphate buffer (pH 7.4) to obtain

the phytosome suspension, which is expected to improve the solubility and bioavailability of the *Dolichandrone falcata* extract for potential therapeutic applications.

4.1 Evaluation of Developed Phytosome

The entrapment efficiency of the *Dolichandrone falcata* phytosome was assessed by diluting 1 mL of the phyto-phospholipid complex to 10 mL with phosphate buffer (pH 7.4). The solution was then centrifuged at 5000 rpm for 30 minutes using an ultracentrifuge. The supernatant was collected and filtered through a 0.45 µm Whatman filter paper. The absorbance of the filtrate was measured at 256 nm using a UV spectrophotometer. Entrapment efficiency was calculated by comparing the actual amount of drug present to the theoretical amount, with the results summarized in Table 25.
Particle size distribution of the phytosome formulation was analyzed using the Malvern Zetasizer instrument, which provides accurate measurement of nanoparticle size to ensure optimal delivery properties. Additionally, the zeta potential, indicative of the surface charge and stability of the phytosome particles, was measured using the Horiba SZ-100 instrument.
Fourier Transform Infrared (FTIR) spectroscopy was employed to characterize the phytosome and confirm the interaction between the ethyl acetate extract of *Dolichandrone falcata* leaves and soya lecithin phospholipid. Analysis was conducted using a Shimadzu FTIR-8400S spectrophotometer, where samples were prepared as potassium bromide (KBr) pellets. Spectra were recorded in the range of 4000 to 400 cm^{-1} under dry air purge conditions, allowing identification of functional groups involved in complex formation. Instrumental parameters included a spectral resolution of 4 cm^{-1} with transmission scan mode and an average of 20 scans per sample.

4.2 Development of Oral Formulation for Anthelmintic Use

An oral suspension of the ethyl acetate extract-loaded phytosome was formulated using sodium carboxymethyl cellulose (NaCMC) as the suspending agent in varying concentrations (0.5%, 1.0%, and 1.5% w/v). Tween 80 (0.1% v/v) was used as a wetting agent, methylparaben (0.04% w/v) as a preservative, stevia (0.2% w/v) as a sweetening agent, and lemon oil (0.1% w/v) for flavoring. The total volume of the suspension was adjusted to 20 ml. The formulation process involved triturating the loaded phytosome with Tween 80 and distilled water to create a uniform mixture. Separately, stevia and methylparaben were dissolved in distilled water, followed by the gradual addition of NaCMC with continuous stirring to ensure complete dissolution. The triturated phytosome mixture was then added dropwise into the NaCMC solution while stirring, after which lemon oil was incorporated for flavor. Finally, the volume was made up to 20 ml with distilled water under constant stirring to achieve a homogenous oral suspension.

4.3 Pharmaceutical Evaluation of Suspension : The prepared oral suspensions were evaluated for organoleptic properties such as color and odor, along with physicochemical parameters including sedimentation volume, redispersibility, flow rate, pH, viscosity, particle size, and *in vitro* drug release. Their anthelmintic potential was also tested using an earthworm motility assay.

***a. In-vitro* Anthelmintic Activity:** Adult earthworms were exposed to the test suspension and an excipient control without extract. Observations were recorded for the time taken to cause paralysis and death of the worms.

b. Sedimentation Volume: Suspension samples were transferred into graduated cylinders and left undisturbed at controlled room temperature. Sediment heights were recorded at fixed intervals over three hours to calculate sedimentation ratio, indicating suspension stability.

c. Redispersibility: After sedimentation, suspensions were inverted repeatedly until uniform mixing was achieved. The number of inversions required was noted to assess ease of redispersion and suspension quality.

d. Flow Rate: The time taken for 10 ml of suspension to flow through a pipette under gravity was measured. The mean flow rate was calculated to assess suspension pourability.

e. pH Measurement: pH was measured using a calibrated pH meter at controlled room temperature, with multiple readings averaged for accuracy.

f. Viscosity: Using a Brookfield viscometer, viscosity was measured at a set spindle speed. Values were averaged to determine the flow characteristics of the suspension.

g.*In-vitro* Drug Release: Using a Franz diffusion cell with a phosphate buffer receptor medium maintained at body temperature, drug release from the formulations was monitored over eight hours. Samples were withdrawn periodically and analyzed spectrophotometrically to determine release profiles.

5. RESULTS

5.1 COLLECTION AND AUTHENTICATION OF LEAVES OF DOLICHANDRONE FALCATA.

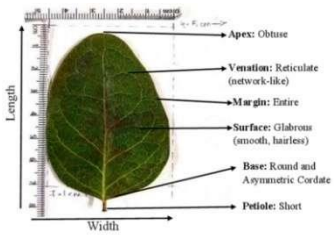
Leaves of *Dolichandrone falcata* were collected from local areas of Maharashtra, Pune region Tornamal, in the month of December 2025 from its natural habitat.

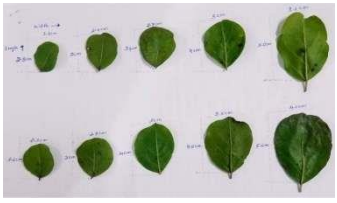
a) Dolichandrone falcata plant in its natural habitat ***b) Leaf twig of Dolichandrone falcata***

5.2 PHARMACOGNOSTICAL EVALUATION OF DOLICHANDRONE FALCATA LEAVES

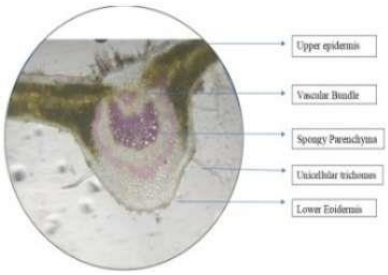
5.2.1 Macroscopic Evaluation: -

Morphological character **Measurement of *Dolichandrone falcata* leaf Size** **Assessment of Length and Width**

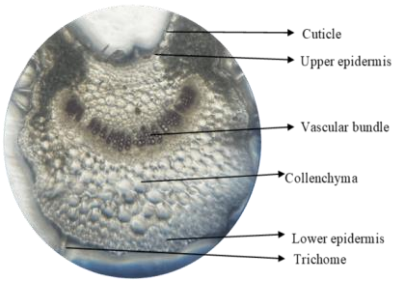




5.2.2 Transverse section of the *Dolichandrone falcata* leaf observed in 10x and 45x microscope

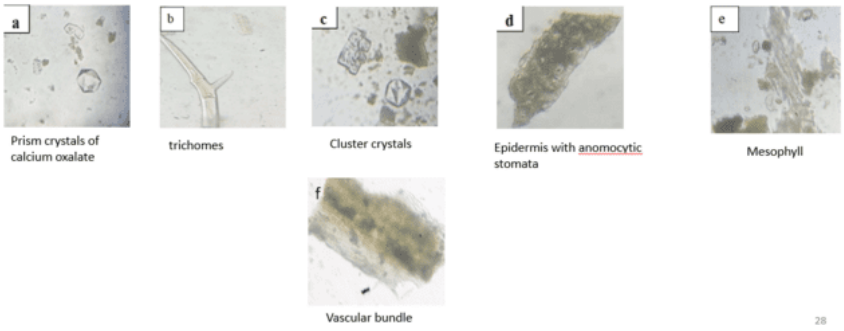


10x



45x

5.2.3 Microscopic Characteristics of *Dolichandrone falcata* Leaf Powder



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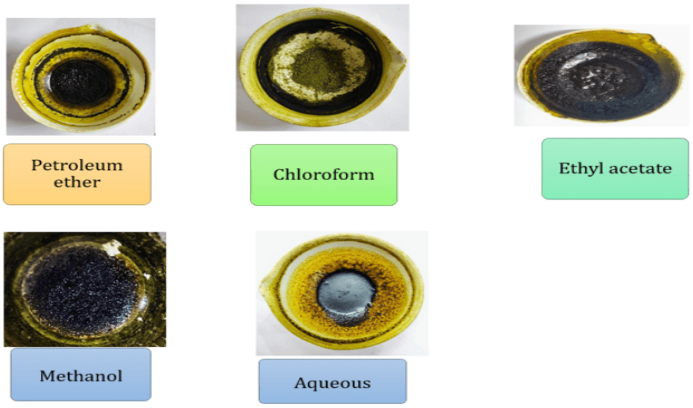
PROXIMATE ANALYSIS OF DOLICHANDRONE FALCATA LEAVES POWDER

Sl.no.	Parameters	Results
1	Moisture content	9.25±0.42%w/w
2	Total Ash	7.69±0.22%w/w
3	Acid insoluble ash	3.72±0.41%w/w

4	Water soluble ash	4.47±0.23%w/w
5	Alcohol soluble extractive	4.93±0.49%w/w
6	Water soluble extractive	9±0.42%w/w

*Values are expressed in terms of mean± SD (%w/w) of results done in triplicate

5.5 SUCCESSIVE EXTRACTION OF LEAVES OF DOLICHANDRONE FALCATA WITH VARIOUS SOLVENTS IN THE INCREASING ORDER OF POLARITY (PETROLEUM ETHER, CHLOROFORM, ETHYL ACETATE, METHANOL).



Extract	**%yield**	**Nature of the extract**	
		Colour	Consistency
Petroleum Ether	2.05±0.21%w/w	Yellowish brown	Sticky
Chloroform	2.84±0.35%w/w	Blackish green	Powder
Ethyl acetate	8.88±0.21%w/w	Green	Semisolid
Methanol	10.78±1.48%w/w	Reddish brown	Sticky
Aqueous	1.61±0.23 %w/w	Blackish red	Sticky

5.6 PRELIMINARY PHYTOCHEMICAL SCREENING OF THE SUCCESSIVE EXTRACTS OF DOLICHANDRONE FALCATA LEAVES

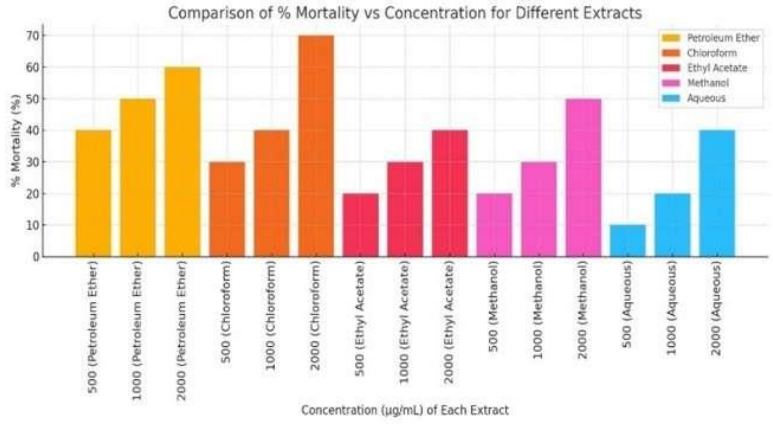
Phytoconstituents	**Petroleum ether extract**	**Chloroform extract**	**Ethyl acetate extract**	**Methanolic extract**	**Aqueous extract**
Carbohydrates	-	-	-	+	+
Proteins and amino acids	-	-	-	+	-
Fixed oils and fats	+	-	-	-	–
Alkaloids	-	-	+	+	-
Glycosides	-	+	-	-	–
Flavonoids	-	-	+	+	-
Tannins	-	-	+	+	+
Saponins	-	-	+	+	+

Steroids	+	-	-	-	-
Triterpenoids	+	-	-	-	_

EVALUATION OF TOXICITY OF SUCCESSIVE SOLVENT EXTRACTS OF DOLICHANDRONE FALCATA LEAVES BY BRINE SHRIMP LETHALITY ASSAY (BSLA)

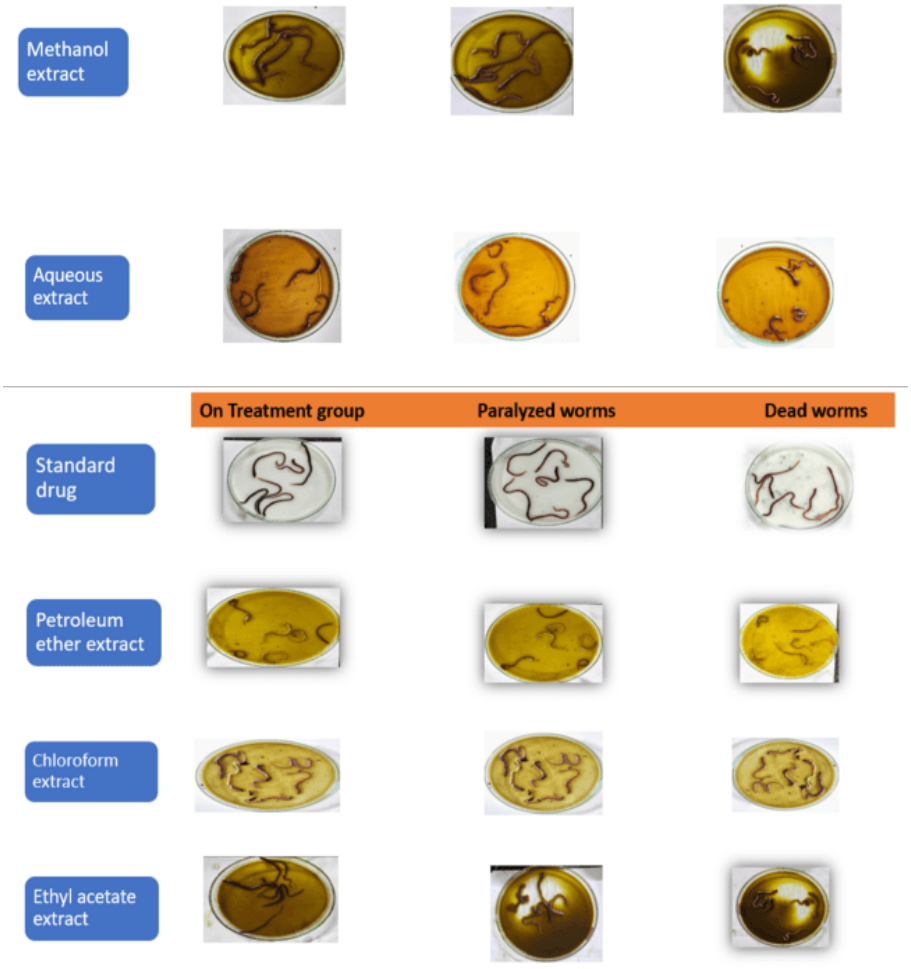
Sl. No	Extracts	Concentration (µg/ml)	No. of Nauplii Used	No. of Nauplii Dead	No. of Nauplii Survived	% Mortality	Probit value	Log Conc	**LC50**
1	Control group	-	10	0	10	0%	0	0	
2	Petroleum Ether	500	10	4	6	40%	4.76	2.69897	**1166.67**
3		1000	10	5	5	50%	5	3.00000	
4		2000	10	6	4	60%	5.25	3.30103	
5	Chloroform	500	10	3	7	30%	4.48	2.69897	**1288.46**
6		1000	10	4	6	40%	4.76	3.00000	
7		2000	10	7	3	70%	5.52	3.30103	
8	Ethyl Acetate	500	10	2	8	20%	4.16	2.69897	**2666.67**
9		1000	10	3	7	30%	4.48	3.00000	
10		2000	10	4	6	40%	4.76	3.30103	
11	Methanol	500	10	2	8	20%	4.16	2.69897	**2000.00**
12		1000	10	3	7	30%	4.48	3.00000	
13		2000	10	5	5	50%	5	3.30103	
14	Aqueous	500	10	1	9	10%	3.72	2.69897	**2500.00**
15		1000	10	2	8	20%	4.16	3.00000	
16		2000	10	4	6	40%	4.76	3.30103	

% mortality of nauplii cells tested against the extracts of Dolichandrone falcata

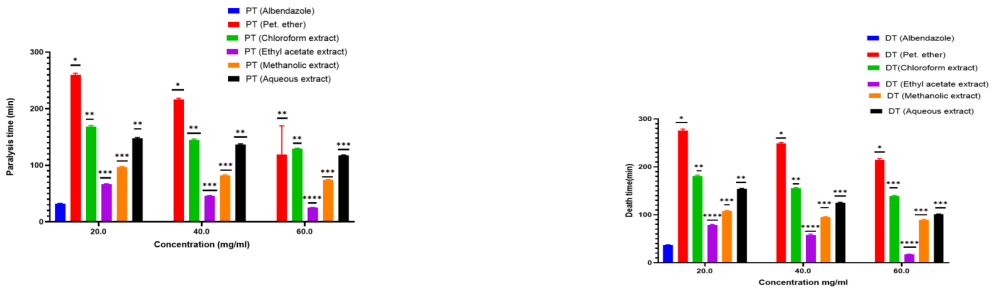


5.8 Evaluation of in-vitro anthelmintic activity by using standard drug albendazole and successive solvent extracts of Dolichandrone falcata leaves

Concentr-ation	Standard drug albendazole		Petroleum ether extract		Chloroform extract		Ethyl acetate extract		Methanolic extract		Aqueous extract	
	PT (min)	**DT (min)**	**PT (min)**	**DT (min)**	**PT (min)**	**DT (min)**	**PT (min)**	**DT (min)**	**PT (min)**	**DT (min)**	**PT (min)**	**DT (min)**
20 mg/ml (400mg)	26.66± 1.69	38±2.16	261.67± 3.86	277± 2.83	168.47±3.81	180.0.8	**66.33± 3.86**	**78.33± 5.31**	97.38±2.45	107.32±0.82	147.3±1.82	153.21±3.12
40 mg/ml (800mg)	N/A	N/A	232.33± 2.49	248.67± 4.19	145.33±2.05	155.33±2.03	**45.78± 4.89**	**63.67± 4.11**	82.33± 2.05	95.00± 2.05	136.58±1.63	124.39±2.82
60 mg/ml (1200mg)	N/A	N/A	254.67± 4.92	271.67± 4.92	127.33± 4.92	139± 3.74	**24.67± 3.29**	**33± 1.63**	74.12.33 ± 2.05	89.33± 2.05	117.43±2.32	101.37±3.44



Statistical graph showing the effect of Standard Albendazole and various concentrations of Dolichandrone falcata extracts on paralysis time of worms and death time of worms



5.9.0 Estimation of total phenolic content for different solvent extract of *Dolichandrone falcata*

The total phenolic content of successive solvent extracts of *Dolichandrone falcata* leaves was quantified using the Folin-Ciocalteu method with gallic acid as the standard. Among the extracts, the *ethyl acetate extract exhibited the highest phenolic content* at *21.80 ± 2.60 mg GAE/g*, followed by the methanolic extract (*15.97 ± 0.23 mg GAE/g*), chloroform extract (*11.30 ± 0.36 mg GAE/g*), aqueous extract (*7.45 ± 0.97 mg GAE/g*), and petroleum ether extract (*4.55 ± 1.06 mg GAE/g*). These results indicate that phenolic compounds are predominantly concentrated in the ethyl acetate and methanolic extracts.

5.9.1 Estimation of Total Flavonoid Content (TFC)

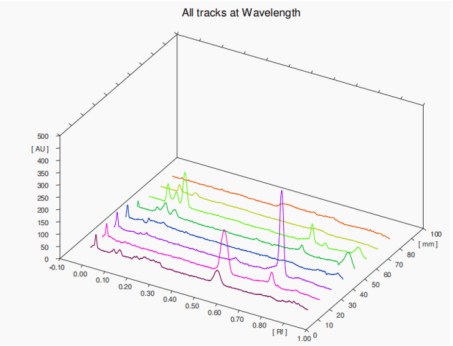
Total flavonoid content was determined by the aluminum chloride colorimetric method using quercetin as the standard. The *ethyl acetate extract showed the highest flavonoid content* at *8.40 mg QE/g*, followed by methanolic extract (*6.00 mg QE/g*), chloroform extract (*4.97 mg QE/g*), aqueous extract (*3.80 mg QE/g*), and petroleum ether extract (*2.48 mg QE/g*). This suggests that flavonoids are also mainly concentrated in the ethyl acetate and methanolic extracts.

5.9.2 Estimation of Total Tannin Content (TTC)

Total tannin content was measured using the Folin-Ciocalteu method with tannic acid as the standard. The *ethyl acetate extract had the highest tannin content* at *15.18 mg TAE/g*, followed by methanolic extract (*7.86 mg TAE/g*), aqueous extract (*6.03 mg TAE/g*), chloroform extract (*4.54 mg TAE/g*), and petroleum ether extract (*2.30 mg TAE/g*). This confirms that tannins are predominantly present in the ethyl acetate and methanolic extracts.

5.10 HPTLC FINGERPRINTING ANALYSIS OF SUCCESSIVE SOLVENT EXTRACTS OF *DOLICHANDRONE FALCATA*

❖ ***HPTLC profiling of successive solvent extracts of Dolichandrone falcata leaf.***

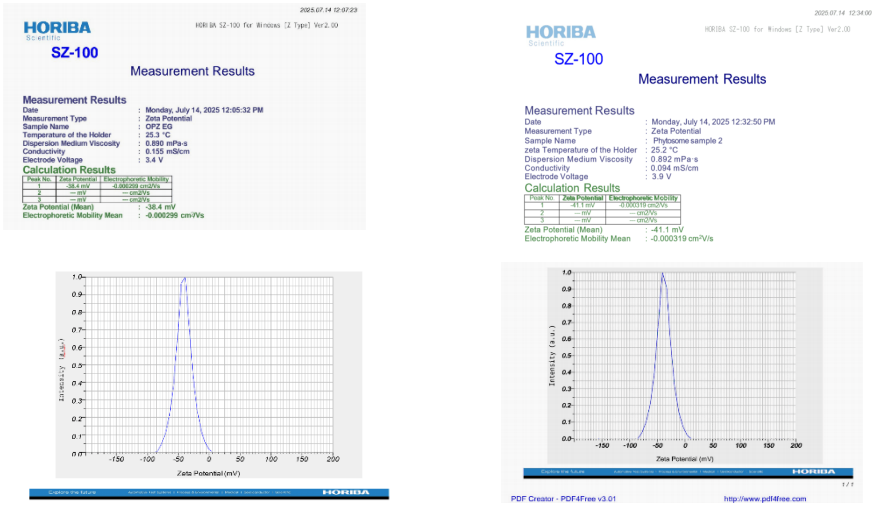


- ***Plate Preparation**: A 10 x 10 cm HPTLC plate was prepared for analysis.*
- ***Spots Applied**:*
  - ***Track 1**: Rutin (standard)*
  - ***Track 2**: Gallic acid (standard)*
  - ***Track 3**: Quercetin (standard)*
  - ***Track 4**: Petroleum ether extract of Dolichandrone falcata (PE-DCF)*
  - ***Track 5**: Chloroform extract of Dolichandrone falcata (C- DCF)*
  - ***Track 6**: Ethyl acetate extract of Dolichandrone falcata (EA- DCF)*
  - ***Track 7**: Methanolic extract of Dolichandrone falcata (MET- DCF)*
  - ***Track 8:** Aqueous extract of Dolichandrone falcata (AQ- DCF)*

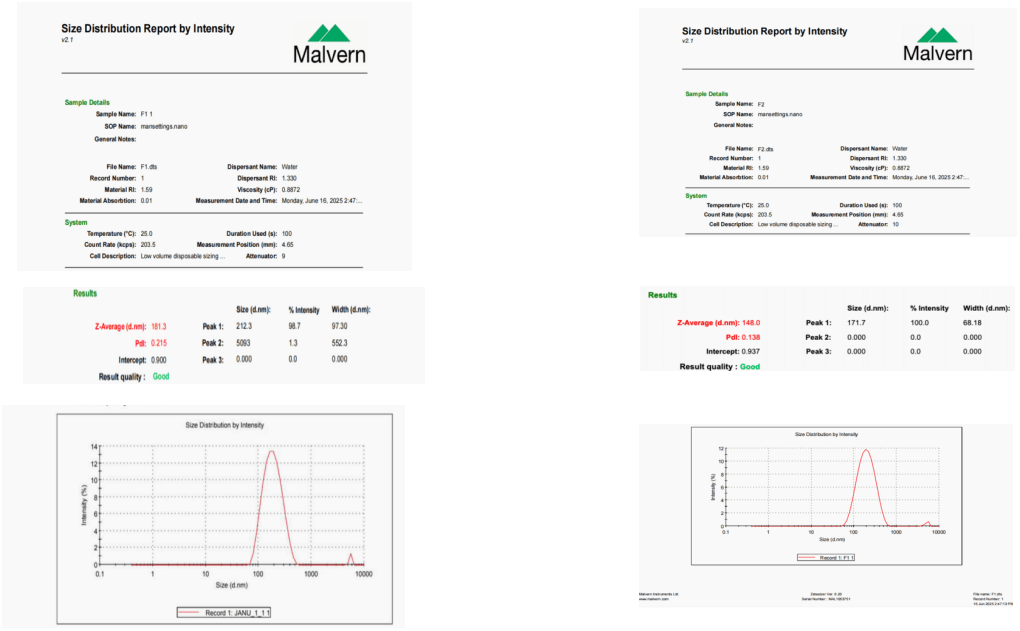
Overall, the results indicated that the ethyl acetate extract was richest in phytoconstituents, showing bands similar to quercetin, while the chloroform and aqueous extracts also demonstrated the presence of polyphenolic compounds. These findings confirmed the presence of flavonoids, phenolics, and related phytoconstituents in the successive extracts of *Dolichandrone falcata*.

5.11 EVALUATION OF PHYTOSOME FORMULATION:

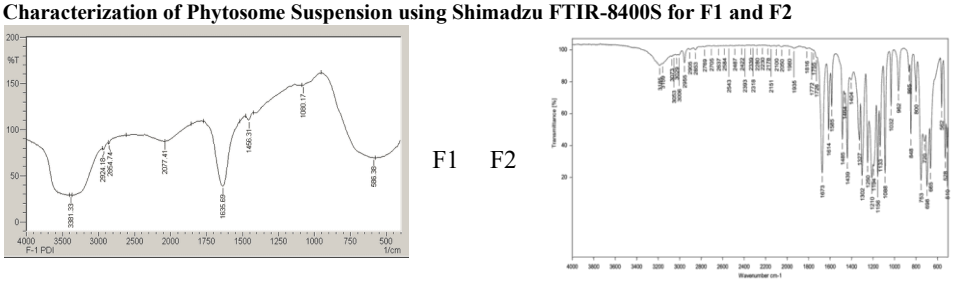
a. HORIBA SZ-100 instrument analyser used for zeta potential for F1 and F2



b. Malvern zetasizer instrument used for particle size distribution of F1 and F2



c. FTIR Characterization of Phytosome Suspension using Shimadzu FTIR-8400S for F1 and F2



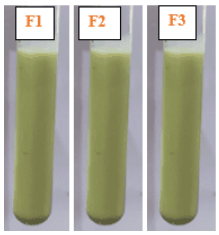
F1 F2

The FTIR analysis of the formulated phytosomes F1 and F2 reveals notable differences in their spectral features, indicating variations in the interaction between the plant extract and phospholipids. Both formulations exhibit characteristic hydroxyl peaks (O–H) at 3381 cm^{-1} for F1 and 3455 cm^{-1} for F2, confirming the presence of phenolic groups from the plant extract. However, the carbonyl stretching peaks are sharper and more intense in F2 (1736–1728 cm^{-1}), suggesting a stronger and more effective complexation with phospholipids compared to the weaker and broader peak observed in F1 at 1635 cm^{-1}. Additionally, phospholipid-related absorption bands, specifically the P=O and C–O–C stretching vibrations, are more pronounced and clearly defined in F2 within the range of 1250–1032 cm^{-1}, whereas F1 only shows a less distinct peak at 1080 cm^{-1}. The overall spectrum of F2 displays more distinct and multiple peaks, indicative of stronger molecular interactions and bonding between the plant extract and phosphatidylcholine, while the comparatively fewer and less intense peaks in F1 suggest weaker interactions and a less stable complex formation.

d. ENTRAPMENT EFFICACY:

Entrapment efficiency was evaluated for the two formulated phytosome systems, F1 and F2, with distinct results observed for each. Formulation F1 demonstrated an entrapment efficacy of 72.14% In contrast, Formulation F2 achieved a considerably higher entrapment efficacy of 82.56%, indicating that F2 was more successful at incorporating the active phytoconstituents into the liposomal vesicles than F1.

5.12 DEVELOPMENT OF FORMULATION FOR ANTHELMINTIC USE

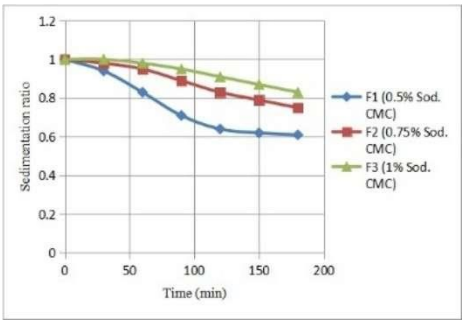


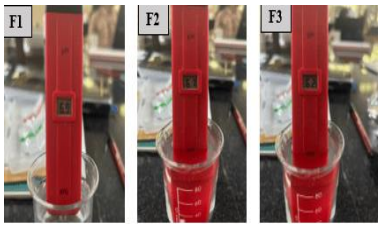
a. Results of rate of sedimentation of the oral suspensions

Time (min)	**Height of the total suspension (H_i) (ml)**	**F1**		**F2**		**F3**	
		Height of the sediment (H_f) (ml)	Sedimentation ratio (H_f/ H_i)	Height of the sediment (H_f) (ml)	Sedimentation ratio (H_f/ H_i)	Height of the sediment (H_f) (ml)	Sedimentation ratio (H_f/ H_i)
0	10	10	1	10	1	10	1
30	10	9.4	0.94	9.8	0.98	10	1
60	10	8.3	0.83	9.5	0.95	9.8	0.98
90	10	7.1	0.71	8.9	0.89	9.5	0.95
120	10	6.4	0.64	8.3	0.83	9.1	0.91
150	10	6.2	0.62	7.9	0.79	8.7	0.87
180	10	6.1	0.61	7.5	0.75	8.3	0.83

Evaluation parameter results of Oral Suspensions

Evaluation Parameter	**F1**	**F2**	**F3**
Nature	Liquid	Liquid	Liquid
Colour	Pale yellow	Pale yellow	Pale yellow
Odour	Characteristic	Characteristic	Characteristic
Redispersibility	3 strokes	3 strokes	2 strokes
Flow rate	20 ml/21 sec	20 ml/50 sec	20 ml/117 sec
pH	6.9	6.7	6.2
Viscosity	75 cps	130 cps	192 cps



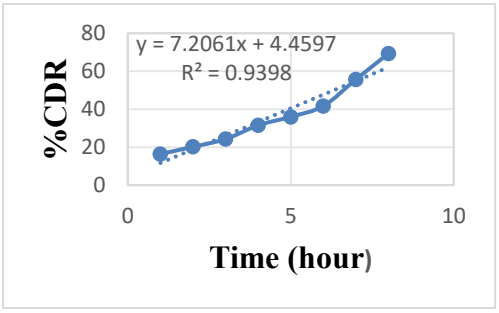


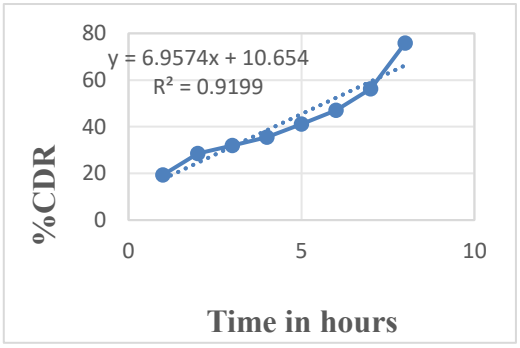
***c. In-vitro* drug release profile of F1 formulation**

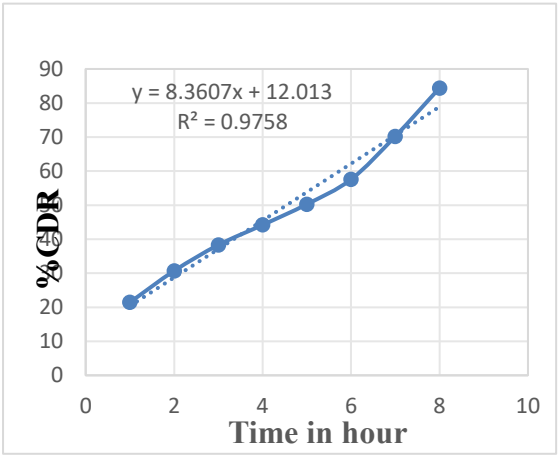
***Invitro* drug release profile of F2 formulation**

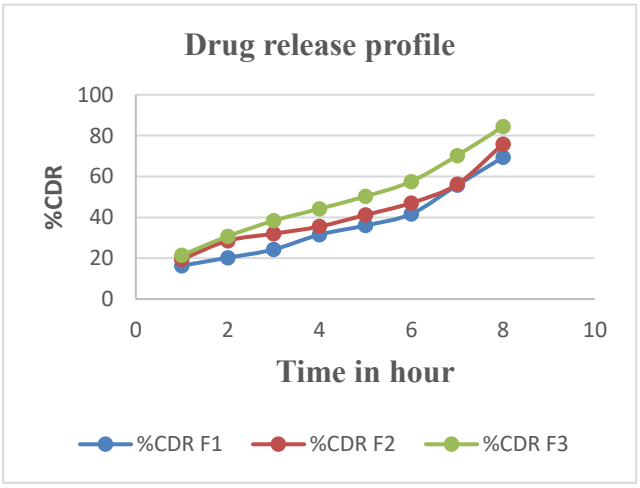
In-vitro drug release profile of F3 formulation

Comparison of in-vitro drug release profile for F1, F2, F3



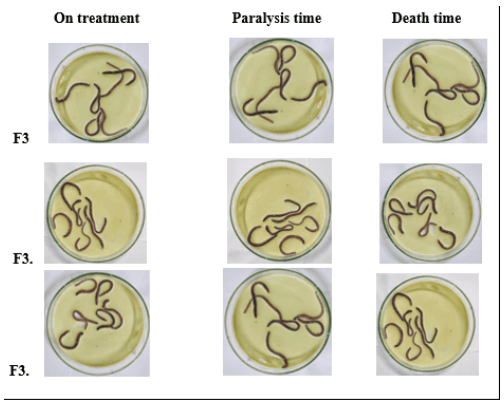






Drug release profile of each formulation was studied over an 8-hour period. The results showed a sustained release pattern for all three formulations. Formulation F3 consistently released the highest percentage of the drug, reaching approximately 84% cumulative drug release after 8 hours. F2 exhibited an intermediate release profile, with a final release of about 76%, while F1 demonstrated the lowest drug release, with approximately 70% of the drug released at the 8-hour mark. The data indicated that the drug release rate was directly proportional to the viscosity of the formulations, with the most viscous formulation (F3) providing the highest and most sustained release

***d. In-vitro* Anthelmintic activity for formulated Oral Suspensions**



Excipient control group (a) F1 control group; (b) F2 control group; (c) F3 control group

***In-vitro* Anthelmintic activity of the Oral Suspensions**

Formulation	Paralysis time (min)	Death time (min)
F3	12.66±1.69	16.33±1.69

6. DISCUSSION

Fresh leaves of *Dolichandrone falcata* were collected from the local areas of Toranmal region, Maharashtra (Pune), during December 2024, when the plant was in its optimal growth stage. DCF plant was identified and authenticated at the Central Ayurveda Research Institute, Bangalore, by Dr. V. Rama Rao, Research Officer (Botany), certificate given in Annexure 1. Proper collection and authentication ensured the genuineness and reliability of the plant used for pharmacognostical and phytochemical investigations.

Macroscopic and microscopic studies of *Dolichandrone falcata* leaves confirmed their diagnostic characters. Microscopically, leaf was dorsiventral with an upper epidermis, palisade and spongy parenchyma, and a midrib vascular bundle. Diagnostic features included unicellular trichomes, anomocytic stomata, and calcium oxalate crystals (both prism and cluster forms). Powder microscopy further confirmed these features, shown prism crystals, cluster crystals, mesophyll fragments, trichomes, and vascular elements. These characters were consistent with descriptions of Bignoniaceae family and authenticated the *Dolichandrone falcata* leaves for pharmacognostic evalution purposes.

Proximate analysis revealed a total ash value of 7.69±0.22% w/w, acid-insoluble ash 3.72±0.41% w/w, and water-soluble ash 4.47±0.23% w/w, moisture content 9.25±0.42 w/w, alcohol soluble extractive 4.93±0.49 w/w,water soluble extractive 9±0.42 w/w which were within the acceptable limits, indicating low contamination and good quality of the raw material. Successive solvent extraction demonstrated that polarity strongly influenced yield. Among the extracts, methanolic (10.78 ± 1.48% w/w) and ethyl acetate (8.88 ± 0.21% w/w) fractions recorded the highest yields, whereas petroleum ether (2.05 ± 0.21%), chloroform (2.84 ± 0.35%), and aqueous (1.61 ± 0.23%) extracts yielded comparatively less. High yield of methanolic and ethyl acetate extracts correlates with their ability to solubilize polar to semi-polar phytoconstituents such as flavonoids, tannins, and phenolic compounds.

Phytochemical screening confirmed that methanol and ethyl acetate extracts were rich in alkaloids, flavonoids, tannins, and polyphenols. These bioactive groups are well documented for their anthelmintic activities. Tannins, for instance, bind to glycoproteins on parasite cuticles, leading to death, while alkaloids disrupt neurotransmission by blocking acetylcholine receptors. The presence of flavonoids adds additional value, as they inhibit phosphodiesterase and Ca^{2+}-ATPase, mechanisms that compromise parasite survival.

Comparative literature also highlights similar trends. For example, methanolic extracts of *Azadirachta indica* and *Terminalia chebula* have shown higher phenolic contents correlating with stronger anthelmintic activity. Thus, *D. falcata* follows the established paradigm of phenolic-rich extracts being more pharmacologically potent.

Brine shrimp lethality assay revealed LC_{50} values of 1050 µg/mL for petroleum ether extract, 1333.3 µg/mL for chloroform extract, 2666.7 µg/mL for ethyl acetate extract, 1583.3 µg/mL for methanol extract, and 1816.6 µg/mL for aqueous extract. Among these, the ethyl acetate extract showed the least toxicity, indicating a favorable safety margin for pharmacological application.

In vitro anthelmintic assay demonstrated a dose-dependent response. Albendazole (20 mg/mL) showed a PT of 26.66 ± 1.69 min and DT of 38 ± 2.16 min. At the same concentration, the ethyl acetate extract showed PT 66.33 ± 3.86 min and DT 78.33 ± 5.31 min, while methanol showed PT 97.38 ± 2.45 min and DT 107.32 ± 0.82 min. At 60 mg/mL, the ethyl acetate extract exhibited the most potent effect with PT 24.67 ± 3.29 min and DT 33 ± 1.63 min, which was highly significant ($p < 0.001$) compared to other extracts. These results strongly indicated that the ethyl acetate fraction possessed superior anthelmintic activity, attributable to its higher content of phenolics, flavonoids, and tannins.

Quantitative estimation of phytoconstituents from the successive extracts of *Dolichandrone falcata* leaves revealed distinct variations depending on solvent polarity. The total phenolic content (TPC), expressed as mg gallic acid equivalent per gram of extract, was found to be highest in the ethyl acetate extract (21.80 ± 2.60 mg GAE/g), followed by methanol (15.97 ± 0.23 mg GAE/g), chloroform (11.30 ± 0.36 mg GAE/g), aqueous (7.45 ± 0.97 mg GAE/g), and petroleum ether (4.55 ± 1.06 mg GAE/g). A similar trend was observed in total flavonoid content (TFC), estimated as mg quercetin equivalent per gram of extract, where the ethyl acetate extract again showed the maximum value (8.40 mg QE/g), followed by methanol (6.00 mg QE/g), chloroform (4.97 mg QE/g), aqueous (3.80 mg QE/g), and petroleum ether (2.48 mg QE/g). The total tannin content (TTC), expressed as mg tannic acid equivalent per gram, was also highest in the ethyl acetate extract (15.18 mg TA/g), with progressively lower values in methanol (7.86 mg TA/g), aqueous (6.03 mg TA/g), chloroform (4.54 mg TA/g), and petroleum ether (2.30 mg TA/g).

HPTLC profiling of different solvent extracts of *Dolichandrone falcata* leaves (petroleum ether, chloroform, ethyl acetate, methanol, and aqueous) was performed to establish characteristic chemical fingerprints and to compare them with standard markers (gallic acid, rutin, and quercetin).

Chromatographic analysis revealed distinct peak patterns for each extract, confirming that solvent polarity significantly influenced the phytochemical composition.

Petroleum ether extract showed a limited number of peaks, mainly at higher Rf values (0.72, 0.81, and 0.88) with relatively low peak areas (<10% each), indicating the presence of non-polar phytoconstituents such as terpenoids or fatty components. No significant correlation with the phenolic or flavonoid standards was observed in this extract.

Chloroform extract displayed peaks at Rf 0.52 (12.1%), 0.61 (10.2%), and 0.79 (9.3%), along with minor bands at higher Rf values, suggesting the presence of moderately polar constituents. Although some faint peaks overlapped near the rutin region (Rf ~0.35), no strong evidence of marker compounds was confirmed in this extract.

Ethyl acetate extract demonstrated the richest chemical profile with eight distinct peaks. Prominent peaks were observed at Rf 0.28 (12.35%), 0.35 (17.62%), and 0.42 (21.48%), which corresponded exactly with the standards gallic acid, rutin, and quercetin, respectively. Additional peaks at 0.52, 0.61, 0.71, 0.79, and 0.88 accounted for 6–14% area each, suggesting the presence of other polyphenols or flavonoids. This extract clearly showed the strongest match with standards and was phytochemically the most diverse.

Methanolic extract produced six visible peaks, with two moderate peaks at Rf 0.35 (11.3%) and 0.42 (15.8%), aligning with rutin and quercetin. A weaker band at Rf 0.28 (8.7%) also indicated the presence of gallic acid, although in lesser amounts than the ethyl acetate extract. This supported the quantitative estimation data where methanol ranked second highest for total phenolics, flavonoids, and tannins.

Aqueous extract displayed four primary peaks, with moderate intensity near Rf 0.28 (gallic acid region, 10.4%) and weaker bands at Rf 0.35 (rutin region, 7.8%) and 0.42 (quercetin region, 9.6%). While the marker compounds were present, their relative abundance was lower than in methanol and ethyl acetate, which corresponded with the lower values obtained in the estimation assays.

When compared with the standards, gallic acid (Rf 0.28), rutin (Rf 0.35), and quercetin (Rf 0.42) showed consistent and reproducible bands in the methanolic, aqueous, and ethyl acetate extracts, with the ethyl acetate fraction having the most intense and well-defined peaks. Petroleum ether and chloroform extracts showed negligible alignment with these standards, indicating that non-polar solvents extracted fewer phenolic or flavonoid constituents.

Phytosome technology was successfully employed to enhance delivery of the active extracts. FTIR studies confirmed complexation between phospholipids and phytoconstituents, evidenced by characteristic carbonyl stretching peaks at 1736–1728 cm^{-1}. Particle size analysis revealed nanoscale dispersion with acceptable polydispersity, while zeta potential analysis indicated stable formulations with negative surface charge.

Such nanoscale phytosomes are known to significantly improve solubility and membrane permeability of poorly absorbed phytochemicals. This is particularly important for flavonoids and tannins, which often face bioavailability limitations in conventional formulations. Hence, the phytosome approach adds technological value by transforming a crude extract into a standardized, bioavailable dosage form.

Phytosome formulations were developed using the ethyl acetate extract to enhance bioavailability. Entrapment efficiency was 76.42% for F1 and 82.56% for F2, indicating better drug loading in F2. Particle size analysis revealed that F1 had a Z-average of 181.3 nm with PDI 0.215, while F2 had a smaller size of 148.0 nm with PDI 0.138, indicating superior homogeneity. Zeta potential values of –38.4 mV (F1) and –41.1 mV (F2) suggested both were stable, with F2 exhibiting comparatively higher stability. FTIR spectra confirmed stronger interaction of phytoconstituents with phospholipids in F2, ensuring successful complexation.

Oral suspensions were prepared using phytosome-loaded ethyl acetate extracts. The pH values were 6.9 (F1), 6.7 (F2), and 6.2 (F3). Viscosity values were 75 cps (F1), 130 cps (F2), and 192 cps (F3), with corresponding flow times for 20 ml being 21 s (F1), 50 s (F2), and 117 s (F3). Sedimentation ratios after 180 min were 0.61 (F1), 0.75 (F2), and 0.83 (F3). Redispersibility was easy, with 3 shakes (F1, F2) and 2 shakes (F3) required. In-vitro drug release at 8 h was 69.36% (F1), 75.89% (F2), and 84.42% (F3). Anthelmintic evaluation of the formulated suspensions showed that F3 had the strongest activity with PT 12.66 ± 1.69 min and DT 16.33 ± 1.69 min, while F2 provided the best balance of stability, redispersibility, and drug release.

Taken together, these results demonstrated that the ethyl acetate extract of *Dolichandrone falcata* leaves exhibited the most significant anthelmintic activity among all extracts, which was strongly correlated with its high content of phenolic, flavonoid, and tannin compounds. The phytosome formulation F2, with smaller particle size, higher entrapment efficiency, and better stability, proved to be the most promising delivery system for enhancing the bioefficacy of the extract

The present study successfully demonstrated the development of a *Dolichandrone falcata* phytosome-based oral suspension, establishing a strong foundation for future research and advancement toward clinical and commercial application. Future work should focus on several key areas. Firstly, the isolation and elucidation of active compounds is essential to identify and characterize the specific phytoconstituents, such as flavonoids and tannins, responsible for the observed anthelmintic activity. Comprehensive structural elucidation of these isolated compounds using advanced spectroscopic techniques will further clarify their pharmacological potential. Secondly, *In-vivo* anthelmintic efficacy studies should be conducted using suitable animal models infected with target parasites to confirm therapeutic activity, determine the dose-response relationship, and evaluate the safety and therapeutic index of both the standardized extract and optimized phytosome formulation. Thirdly, advanced pharmacokinetic and bioavailability studies are necessary to quantify the enhanced absorption and systemic availability of active markers delivered via the phytosome system compared to the crude extract, along with investigations into tissue distribution and elimination profiles. Lastly, long-term stability and scale-up studies should be undertaken following ICH guidelines to establish the formulation's shelf life under various storage conditions. Additionally, efforts should be directed toward developing a cost-effective, reproducible, and scalable manufacturing process to enable successful commercial production of the *Dolichandrone falcata* phytosome formulation.

7. CONCLUSION

This study successfully demonstrated the extraction, phytochemical profiling, formulation, and evaluation of *Dolichandrone falcata* leaves for their anthelmintic potential using a phytosome-based drug delivery approach. Research study findings confirmed that the plant possessed significant

pharmacological potential, supported by its rich phytochemical content, including flavonoids, tannins, and phenolic compounds with known anthelmintic activity. HPTLC analysis validated the presence of key bioactive markers, ensuring consistency and standardization of the extracts. Toxicity studies established the safety of the plant extracts, while in vitro evaluations showed strong anthelmintic activity comparable to standard drugs. The application of phytosome technology enhanced the stability, bioavailability, and therapeutic performance of the active constituents, resulting in an effective oral formulation. Overall, *Dolichandrone falcata* was established as a safe and effective natural source of anthelmintic agents, with the phytosome formulation offering a promising approach for future herbal drug development. Future studies suggests for focus on isolating the specific active compounds, performing comprehensive *In-vivo* anthelmintic trials in animal models, and conducting long-term stability studies to transition this promising formulation toward commercial development**.**

8.REFERENCES

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