ISSN: 0976-7126
Maliwal *et al.*, 16(8):21-26, 2025

International Journal of Pharmacy & Life Sciences
Open Access to Researcher

Formulation and Characterization of Phytosomes of Ginger Extract

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Article info

Received: 11/05/2025

Revised: 13/06/2025

Accepted: 15/06/2025

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Abstract

The present study focuses on the *Formulation and Characterization of Phytosomes of Ginger Extract* with the aim of enhancing its bioavailability and therapeutic efficacy. Ginger (*Zingiber officinale*) is well known for its potent antioxidant, anti-inflammatory, and gastroprotective properties, primarily attributed to its active constituents such as gingerols and shogaols. However, its clinical application is limited due to poor solubility and low bioavailability. To overcome these challenges, ginger extract was incorporated into phytosomes—a novel drug delivery system that forms a complex between phytoconstituents and phospholipids to improve absorption and stability. The phytosomes were prepared using the solvent evaporation method and were subjected to comprehensive physicochemical characterization including particle size analysis, polydispersity index (PDI), zeta potential, entrapment efficiency, and morphological studies using TEM.

The formulation exhibited nanosized particles with uniform distribution and good stability. The results indicated that phytosomal encapsulation significantly enhanced the solubility and potential for improved bioavailability of ginger phytoconstituents. This study supports the application of phytosome technology as a promising approach for the effective delivery of herbal bioactives like ginger extract.

Keywords: Ginger, Phytosomes, Anti-microbial activity

Introduction

Plant extracts play a crucial role in the preparation of phytosomes, serving as the primary source of bioactive phytoconstituents that confer therapeutic effects. These extracts, rich in flavonoids, phenolics, terpenoids, and other secondary metabolites, are selected for their known pharmacological activities such as antioxidant, anti-inflammatory, anticancer, or antimicrobial effects. In phytosome formulation, the active constituents from the plant extract interact with phospholipids—commonly phosphatidylcholine—through hydrogen bonding, forming a lipid-compatible molecular complex. This complex enhances the solubility, stability, and bioavailability of the phytoconstituents by facilitating their transport across biological membranes. The efficacy of phytosomes thus largely depends on the quality and concentration of the plant extract used, making it a vital component in the successful development of phytosome-based drug delivery systems. [1]

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Ginger (*Zingiber officinale*), a widely used medicinal and culinary herb, possesses a rich phytochemical and pharmacological profile that contributes to its broad therapeutic potential. The rhizome of ginger contains a variety of bioactive compounds, primarily gingerols, shogaols, paradols, and zingerone, which are responsible for its characteristic pungent flavor and medicinal properties. Among these, [6]-gingerol and [6]-shogaol are the most studied constituents, exhibiting potent antioxidant, anti-inflammatory, antiemetic, antimicrobial, and anticancer activities. Additionally, ginger contains essential oils, flavonoids, and phenolic acids that further enhance its biological efficacy. Pharmacologically, ginger has been shown to support gastrointestinal health, reduce nausea and vomiting, alleviate pain and inflammation, and modulate metabolic disorders such as diabetes and hyperlipidemia. Its diverse pharmacological actions make ginger a valuable candidate for incorporation into advanced delivery systems like phytosomes to improve its solubility and therapeutic impact. [2]

Material and Methods

Extraction of Ginger Extract (Maceration) [3]

- Clean the ginger rhizomes thoroughly to remove dirt and impurities.
- Slice and shade-dry the rhizomes (if using fresh ginger) for 5–7 days.
- Pulverize the dried ginger into a coarse powder using a mechanical grinder.
- Weigh an appropriate quantity of ginger powder (e.g., 100 g).
- Place the powder in a clean, dry glass container with a tight-fitting lid.
- Add distilled water in a ratio of 1:10 (w/v), i.e., 100 g of powder with 1000 mL of water.
- Stir the mixture well to ensure complete wetting of the powder.
- Cover the container and allow the mixture to stand at room temperature for **7** days**.**
- Stir the mixture once or twice daily to facilitate better extraction.
- After 7 days, filter the macerate using a muslin cloth followed by Whatman filter paper to obtain a clear aqueous extract.
- The filtrate can be concentrated by evaporating the water in a water bath at ≤40°C to obtain a semisolid or dry extract.

Formulation of Phytosomes from Ginger extract (Method-I [M-I])

- Ginger extract and PC (in1:1,~2g)mixture was added to100Ml ethanol,and heated at 60ºC for 2h.
- Evaporate the ethanolat40ºC.
- Dried the resulting solid at40ºC in an oven.
- Add 100 mL DW to the dried solid and stored until further use.

In this process perfom galic acid test with ginger extract and Pc . [4]

(Method-II [M-II])

- Ginger extract and PC (in 1:1, ~2g) mixture was added to 100 mL solvent (50 mL methanol and 50 mL Dichloro methane), and heated at 60ºC for 2h.
- Evaporate the solventat 40ºC upto5mL.
- Add 15 mL n-hexane.
- Stored until further use.

Characterization of Phytosomes [5]

Physical Appearance

The physical nature was noted down using organoleptic behaviors.

Particle size and polydispersity index (PDI)

The average particle size and polydispersity index (PDI) of phytosomes was determined by using Malvern Zeta sizer at 25℃. All samples were diluted 10 times with double distilled and filtered (0.2μm) water for measurement. Photon correlation spectroscopy (PCS) is a technique employed to determine the mean particle size and PDI.

Zeta potential

It is a parameter highly useful for the assessment of the physical stability of colloidal dispersions. The zeta potential of the phytosomes was carried out using Malvern Zetasizer. The ξ value for the phytosomes was measured for all batches immediately after preparation.

Entrapment efficiency

For this, Phytosomes dispersion was passed through the saturated sephadex G-50 column and elute was collected. Free drug was retained in the column, whereas phytosomes were eluted out. Eluted fraction was mixed with small amount of

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DMSO to dissolve lipidic fraction and then diluted with distilled water and analyzed using UV spectrophotometer at 282 nm to determine percentage drug entrapment.

Shape and surface morphology using transmission electron microscopy

TEM analysis of the phytosomes was carried out to understand the shape and surface morphology. For this, a drop of phytosomes with 0.01% of phosphotungstic acid was placed on a carbon film coated on a copper grid. Then copper grid was placed into sample holder and fixed in vacuum chamber and images were recorded at 80 kV using TEM.

Drug release studies

Cumulative *in-vitro* % drug release study of phytosomes was determined using dialysis bag diffusion technique with phosphate buffer pH 6.8 as release/diffusion medium (500ml). For this, phytosomes dispersion (equivalent to 10 mg of drug) was placed in dialysis bag which was previously wetted overnight in distilled water, cleaned and sealed from both ends. Then dialysis bag (already filled with phytosomes) was plunged in the receptor compartment containing the diffusion medium which was stirred at 50rpm at 37±0.5℃. Samples (5ml) from diffusion medium were withdrawn at regular time intervals and same volume was replaced with fresh medium to maintain the volume. Then samples were analyzed using U.V. visible spectrophotometer to determine the concentration of drug and cumulative in-vitro % drug release calculated and plotted against time.

Antimicrobial activity

Test Microorganisms

The antibacterial activity was evaluated against:

Gram-positive: *Staphylococcus aureus*

Gram-negative: *Escherichia coli*

Standard strains were obtained from a certified microbial culture collection (e.g., MTCC/ATCC).

Preparation of Phytosomal Formulation (As previously described)

Antibacterial Assay

a) Agar Well Diffusion Method

Mueller–Hinton agar plates were inoculated with 0.5 McFarland standard bacterial suspensions.

Wells were created and loaded with:

- Ginger extract (standardized)
- Ginger–phytosomes (equivalent dose)
- Phosphatidylcholine (control)
- Gallic acid (control)
- Ciprofloxacin (positive control)

Plates were incubated at 37°C for 24 hours.

Zone of inhibition (ZOI) was measured in mm.

b) Minimum Inhibitory Concentration (MIC)

Determined by broth microdilution in 96-well plates.

Concentration range: 100–6.25 µg/mL.

MIC determined as the lowest concentration with no visible turbidity.

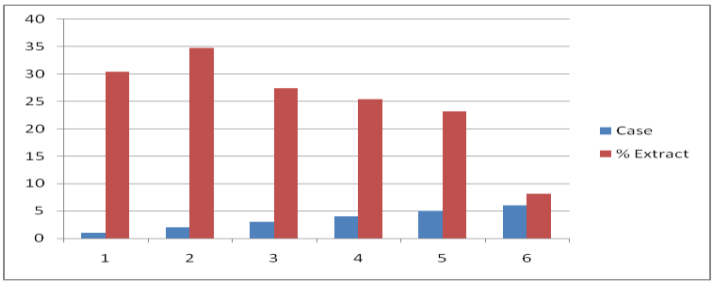
Results and Discussion

Extraction of Ginger

The percentage extract obtained after maceration process using different solvents were mentioned below:

Table 1: Percentage of Extract

Case	% Extract obtained
1	30.46
2	34.72
3	27.41
4	25.39
5	23.18
6	8.11



Characterization of Phytosomes

Physical Appearance

The formulated ginger phytosomes (M-I & M-II) were evaluated for their physical appearance, and the results indicated that the phytosomal dispersion appeared as a smooth, yellowish-brown colloidal suspension without visible aggregates depending on the concentration of the ginger extract used. The texture was uniform with no visible signs of aggregation, phase separation, or crystallization. When dispersed in water, the formulation produced a stable colloidal suspension without sedimentation or floating particles, indicating good dispersibility. The phytosomes exhibited no offensive odor**,** retaining a mild characteristic aroma of ginger. These observations confirm that the physical

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characteristics of the phytosomal formulation were acceptable and suitable for further pharmaceutical development.

Particle size and polydispersity index (PDI)

The particle size and PDI of the Phytosome formulation (M-I & M-II) was recorded and given in table. Particle size distribution image of formulation is shown in figure.

Table 2: Particle size and PDI values of Ginger Phytosomes

Batch No.	Particle Size (Z avg in nm ± SEM)	PDI
M-I	185.4 ± 3.5	0.238 ± 0.02
M-II	184.2 ± 2.8	0.216 ± 0.02

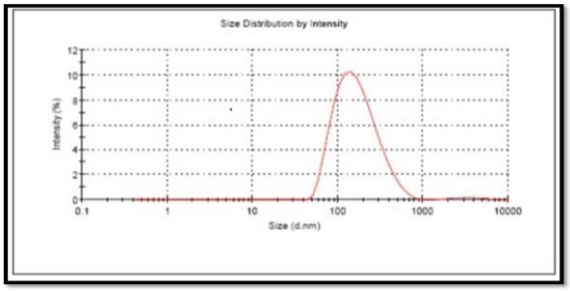

Fig. 1: Particle size of ginger phytosomes

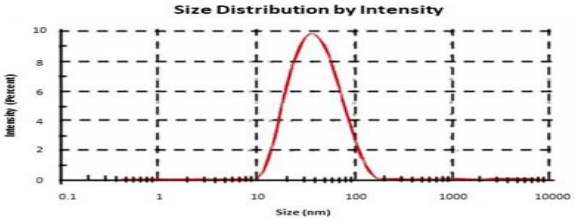

Fig. 2: PDI of Ginger phytosomes

Zeta potential

The zeta potential of the optimized formulation was found to be −32.4 ± 1.7 mV mV.

Table 3: Zeta Potential of Ginger Phytosomes

Batch No.	Zeta Potential (mV)
M-I	−32.4 ± 1.7
M-II	−32.2 ± 1.9

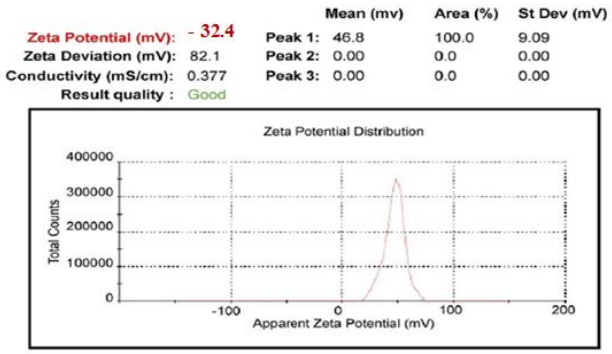

Fig. 3: Zeta potential of ginger phytosome

Entrapment efficiency

The percentage drug entrapment of the phytosome formulation was found to be 78.6 ± 2.3% indicating effective entrapment of the active compound within the phytosomal matrix.

Table 4: Entrapment Efficiency of Ginger Phytosomes

Batch No.	% EE
M-I	78.6 ± 2.3
M-II	77.9± 2.1

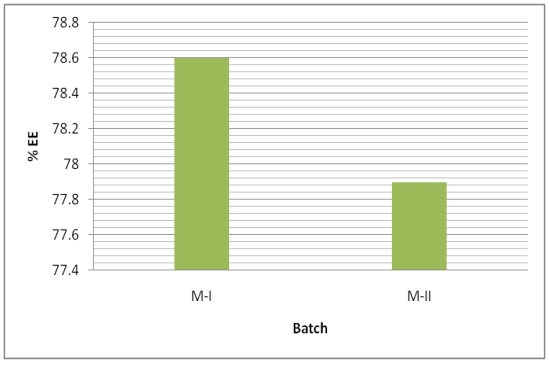

Fig. 4: % EEE of Ginger phytosome

Shape and surface morphology using transmission electron microscopy

TEM image of ginger phytosomes shown in figure. It indicated the smooth surface and spherical shape of the particle with desirable size range.

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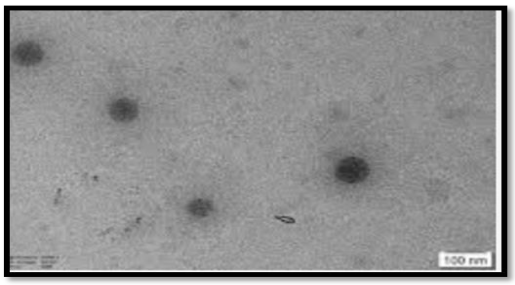

Fig. 5: TEM of Ginger phytosome

Drug release studies

The ginger extract released 98.4% of its drug content within 12 hours, whereas the phytosomal formulation exhibited a sustained release of 91.5 ± 1.7**%** over 24 hours, showing improved release kinetics.

Table 5: % Drug release of ginger phytosome

Time (hr)	% Drug release	
	Ginger Extract	Ginger Phytosome
0	0	0
4	36.7	20.3
8	43.9	33.4
12	98.4	40.6
16		70.2
20		88.3
24		91.5

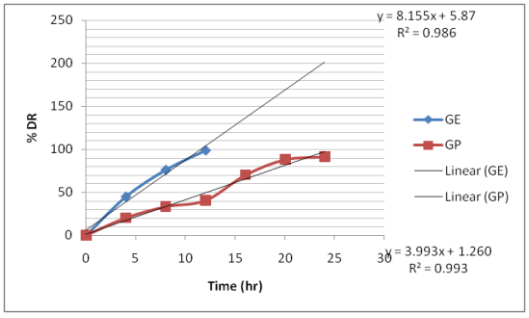

Fig. 6: % Drug release of ginger phytosome

Antimicrobial activity

The antibacterial activity of phytosomes containing ginger extract was performed by well diffusion method against pathogenic bacteria and results were given in table. MIC values were also reported in table.

Table 6: Antimicrobial activity

Sample	*S. aureus* (mm)	*E. coli* (mm)
Ginger Extract	14.2 ± 0.5	12.1 ± 0.4
Phytosomes	19.3 ± 0.7	16.5 ± 0.6
Gallic Acid (control)	10.5 ± 0.4	9.3 ± 0.3
Phosphatidylcholine	No activity	No activity
Ciprofloxacin (10 µg)	24.6 ± 0.6	23.8 ± 0.5

Table 7: MIC values

Sample	MIC – *S. aureus* (µg/mL)	MIC – *E. coli* (µg/mL)
Ginger Extract	50	75
Phytosomes	**25**	**37.5**
Gallic Acid	100	100

Conclusion

The results confirm that ginger extract-loaded phytosomes exhibit enhanced antibacterial activity compared to crude ginger extract. The zone of inhibition increased significantly for both *S. aureus* and *E. coli* when the extract was delivered in phytosomal form. The MIC values support this observation, showing that the phytosome formulation required nearly half the concentration of the crude extract to inhibit bacterial growth.

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Cite this article as:
Maliwal K., Chasta P. and Ali Md. Z. (2025). Formulation and Characterization of Phytosomes of Ginger Extract. *Int. J. of Pharm. & Life Sci.,* 16(8):21-26.

Source of Support: Nil
Conflict of Interest: Not declared
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