RESEARCH PAPER

Phytosome-Enhanced Coriandrum sativum Leaf Extract for Improved Bioavailability and Antidiabetic Activity

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ABSTRACT
Water-soluble phytoconstituents have poor oral bioavailability and so restrict the therapeutic potential of Coriandrum sativum leaves in diabetes control. The present work was carried out to prepare phytosomes of Coriandrum sativum leaf extract (HECSL) for improved absorption and antidiabetic activity. Phytosomes were made by solvent evaporation method using phosphatidylcholine and optimised for particle size, zeta potential and entrapment efficiency and in vitro quercetin release was determined. STZ induced diabetic rats were treated with HECSL, HECSL phytosomes (100-200 mg/kg) and Glibenclamide for 28 days and blood glucose levels were measured on day 1, 7 and 21. Phytosomes of HECSL were more effective in decreasing glucose and 200 mg/kg resulted in near normal serum glucose (96.18 mg/dL). The enhanced efficacy than simple extract was attributed to better bioavailability and sustained release.
Conclusion: Phytosomal encapsulation of Coriandrum sativum significantly increases oral bioavailability and antidiabetic potential and is promising as a natural therapy for Type II diabetes.
Keywords: Coriandrum sativum, Phytosome, Bioavailability, Antidiabetic, STZ-diabetes, Herbal extract
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1. INTRODUCTION

Throughout ancient history, individuals have used herbal healers and phytotherapy in various contexts to maintain their wellness. Over the past decade, a large number of plant extracts have been chemically and pharmaceutically analysed to determine their core chemical components and demonstrate their potential medicinal efficacy. Most of the active ingredients in phytomedicines are water-soluble chemicals (e.g., phenols, glycosides, and flavonoids). Water-soluble phytoconstituents are not effective, whether applied orally or topically, due to their poor absorption. To improve oral bioavailability, several approaches have been developed, such as incorporation of solubility and bioavailability enhancers, structural modification, and entrapment with lipophilic carriers. Phytosomes are made by binding certain components of herbal extract to phosphatidylcholine to create a product that is more absorbable and more effective than regular herbal extracts. Phytosome phospholipids have a proven health-providing impact.

One of the most important contributions of nanotechnology to patients with diabetes is the creation of new nano-sensors for the easy, accurate, and sensitive monitoring of glucose levels. However, with breakthroughs in nanotechnology, the engineering of vehicles for insulin administration becomes viable, which will not need daily injections of subcutaneous insulin as they do not encounter the acid environment of the stomach. So, nanotechnology is used to develop nanodrugs and bio-functional foods for treating prediabetes (Sonaje et al, 2010; Tonda-Turo et al, 2018). Type 2 diabetes is complicated – and patients can be split into five groups Often described as a state of severe insulin insufficiency or resistance, depending on the individual, type 2 diabetes is currently the most common form of diabetes. Classification into five subgroups shows that more than 60% of persons with T2DM are not linked to insulin. Different types of nanocarriers such as liposomes, phytosomes, polymeric nanoparticles and dendrimers can be used for the treatment of diabetes. So far, the nanocarrier phytosome has showed promising results as a diabetic medicine (Ghalandarlaki et al, 2014; Sarma et al, 2021).

The proposed study planned to formulate a phytosome from Coriadrum sativum leaf for enhancement of bioavailability of herbal extract and better efficacy to treat Type II diabetes.

2. MATERIAL METHODS

Materials

Various materials from standard suppliers have been procured and used. All modern instrumentation was part of the study. The analytical grades chemicals were used in the experimental work. Fresh leaf of Coriander (Coriandrum sativum L.), were collected from the local market of Sagar (M.P.). The plant specimen was identified and authenticated by the Department of botany Dr. H. S. Gour Central University Sagar (M.P.) (No: BOT/H/03/79/18).

Preparation of extract

We used a 70:30 v/v ratio of ethanol to water to perform a 72-hour Soxhlet extraction on powdered, shade-dried leaves of Coriandrum sativum. A rotary evaporator was used to filter and concentrate the ethanolic extract. The unrefined mixture was kept at 40C until needed again. In the end, the percentage yields of the dried extracts were determined by applying the formula that is presented further below (Bajpai

et al, 2012). The hydroalcoholic extract of Coriandrum sativum also named as HECL.

Phytochemical Screening

Standard qualitative methods were used for preliminary phytochemical screening of ethanolic extract for the presence of flavonoids, polyphenols, alkaloids, tannins and terpenoids.

Preparation of Phytosomes

Phytosomes were produced by solvent evaporation method: (figure 1)

Step 1: The leaf extract of Coriandrum sativum and phosphatidylcholine (molar ratio 1:1) were dissolved in dichloromethane.

Step 2: The mixture was sonicated for 30 min and then rotary evaporated at 40 °C to achieve a thin layer.

Step 3: The film was hydrated with phosphate buffer (pH 7.4) and agitated for 2 h.

Step 4: The suspension was sonicated to decrease the particle size and kept at 4°C.

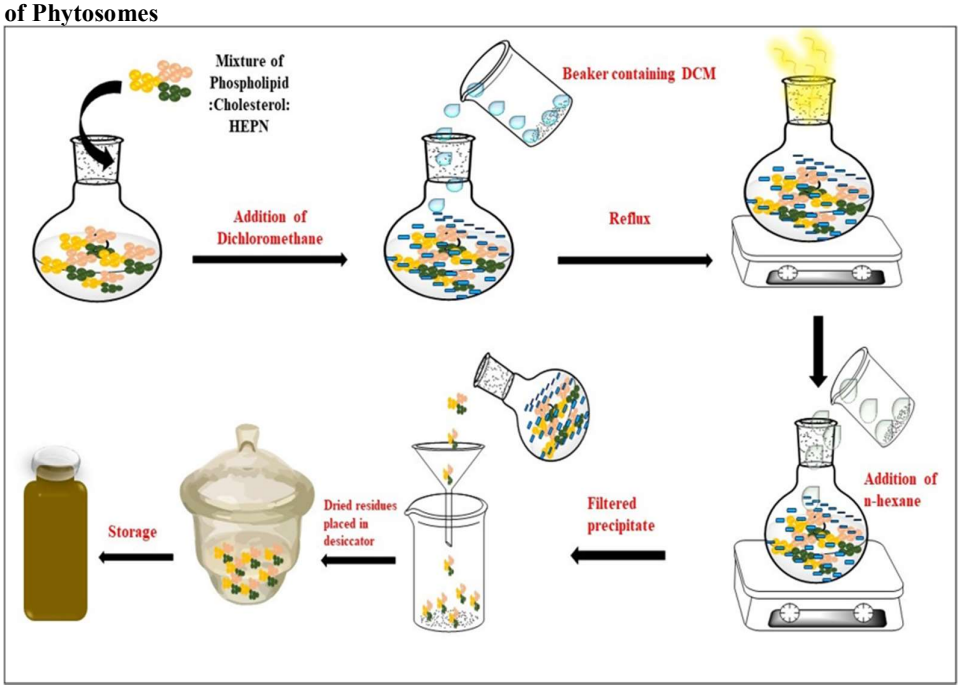

Fig. 1: Steps involved in the preparation of *HECSL* loaded phytosomes

Characterization of HECSL loaded Phytosome

Determination of Entrapment Efficiency

HPLC was utilised so that the entrapment efficiency (EE), which is the amount of quercetin that was successfully incorporated into phytosomes, could be determined. The efficiency of entrapping phytosomal vesicles was determined by the use of the centrifugal technique. It became necessary to remove the transparent supernatant in order to extract the flavonoids (quercetin) which were neither bound in the supernatant. After that the solution was processed for centrifugation at the speed of 12,000 rpm under 4 °C. In order to lyse the vesicles, 1 mL of 0.1 percent Triton X 100 was incorporated with the mixture and further required dilution was done by using phosphate buffer saline (pH 7.4). HPLC was used to determine the amount of quercetin present in the HECSL phytosome at 280 nm (Singh et al, 2011).

% EE was computed by using formula:

$$\%EE = \frac{\text{Quantity of drug added} - \text{Quantity of free drug}}{\text{Total amount of drug added}} \times 100$$

Size, zeta potential and polydispersity index

The phytosomes formulation was evaluated for particle size, PDI and zeta potential using a computerised system employing dynamic light scattering (DLS) and particle size analyser (Malvern Zeta master ZEM 5002, Malvern, UK) respectively. The backscattered laser light is detected at 25 ºC by a laser diffraction technique called zeta sizer. ZP of produced nanoformulations were evaluated by zeta sizer (Malvern Zeta master ZEM 5002, Malvern, UK). It was determined by measuring the speed of the nanoparticles and their direction of motion as they moved in the supplied electrical field. The ZP calculations were made by subtracting the standard deviation from the mean value. All the measures were done in triplicates (n=3) to avoid mistake (Anwar et al, 2018; Wan et al, 2019).

The polydispersity index was determined with the following formula:

$$\text{Polydispersity index} = \frac{\text{Standard deviation}}{\text{Average particle size}}$$

In vitro drug release study

The in vitro release profile of plain HECSL solution and

HECSL loaded phytosomes was evaluated using the dialysis method. Sufficient quantity of phytosomal preparation (HECSL content 1 mg/ml) was placed across a dialysis membrane in a glass beaker containing 200 ml of 0.1 N HCl of pH 1.0. The glass beaker was presumably heated to 37±0.5 degrees Celsius and maintained at the temperature by continuous spinning at 75 revolutions per minute. Aliquots of two millilitres were collected at different time points such as one hr, two hrs, four hrs, six hrs, eight hrs, ten hrs and twelve hrs and the volume was immediately supplied with 0.1 N hydrochloric acid (pH 1.0). The extracted specimens were filtered through Whatmann filter paper and submitted to HPLC analysis at 280 nm to measure the amount of quercetin present in HECSL phytosomes. All steps were performed in triplicates. This ensures the correct dissolving of the medication, so that the correct description of the release profile of the drug with a range of various preparations can be achieved. (Karole et al, 2019).

***In Vivo* Anti-Diabetic Activity**

Antidiabetic activity of polyherbal formulation

The rodents have been subdivided into 7 groups of six rats each, totalling 30 rats. The animals were treated with the polyherbal formulation and reference medication for a period of 28 days. Group I was designated as normal control and was given only drinking water, Group II consisted of diabetic control rodents, Group III was treated with reference drug Glibenclamide (0.5 mg/kg), while Group IV to Group VII were administered with Coriadrum sativum extract (100 mg/kg/day p.o.), Coriadrum sativum extract (200 mg/kg/day p.o.), optimised phytosome formulation (100 mg/kg/day p.o.) and optimised phytosome formulation (200 mg/kg/day p.o.), respectively for a period of 28 days. Blood glucose levels were measured on the first, seventh, and twenty-eighth days following medication administration. The rodents were weighed periodically during the whole study and the mean difference in body mass was calculated (Chaudhuri et al, 2016).

3. RESULT AND DISCUSSION

Extraction of plant material

Fresh Coriandrum sativum leaves were shade-dried, powdered, and Soxhlet-extracted with 70% ethanol. The ethanolic extract was filtered and concentrated using a rotary evaporator. The crude extract was stored at 4°C for further use. (Figure 2)

The yield was approximately 19.78 w/w.

Figure 2: Fresh leaf and powder of Coriander

Phytochemical screening

Phytochemical screening confirmed the presence of flavonoids, polyphenols, tannins, alkaloids, and saponins, as shown in Table 1

Table 1: Phytochemical analysis of hydroalcoholic extract of *Coriandrum sativum*

Phytochemical	Presence (+/-)
Flavonoids	++
Phenolics	++
Alkaloids	+
Saponins	+
Tannins	++

Characterization of HECSL loaded Phytosome **Determination of Entrapment Efficiency Size,**

The efficiency of entrapping phytosomal vesicles was determined by the use of the centrifugal technique. (Table 2)

Table 2: Particle size and %EE of HECSL loaded phytosomes

Variables	Ratio of Phospholipid and cholesterol (%)	Temperature (°C)	Sonication time (hr)	Vesicle size (nm)	Entrapment Efficiency (%)
Predicted Results	0.082	46	36	183.8	65.3
Observed results				177.5±5.47	72.76 ± 1.77
Percentage Prediction error (%)				3.48 %	1.15 %

In vitro drug release study

In *vitro* drug release studies of formulations were performed using dialysis tube method. (Table 3)

Table 3: *In-vitro* dissolution study of HECSL loaded phytosomes and HECSL

Time (hrs)	Sq. rt. of Time	Log Time	%Cumulative drug release of HECSL loaded phytosomes	%Cumulative drug release of HECSL
1	1	0.00	19.47±0.53	12.32±0.37
2	1.41	0.30	29.08±0.68	20.21±0.43
4	2	0.60	42.51±0.32	32.62±0.18
6	2.44	0.77	57.28±0.71	45.87±0.52
8	2.82	0.90	69.71±0.48	53.18±0.84
10	3.16	1.00	81.41±0.27	62.93±0.63
12	3.46	1.07	94.79±0.93	65.23±0.57

In Vivo Anti-Diabetic Activity

The oral glucose tolerance test revealed significant variations in serum glucose levels among the groups. The diabetic control group exhibited markedly elevated glucose levels throughout the study period (Day 1: 296.15 mg/dL, Day 7: 383.23 mg/dL, Day 21: 410.12 mg/dL), indicating persistent hyperglycemia. Treatment with Glibenclamide significantly reduced glucose levels by Day 7 and Day 21 (132.10 and 92.34 mg/dL, respectively), demonstrating its anti-hyperglycemic efficacy. HECSL at 100 and 200 mg/kg doses also lowered glucose levels compared to diabetic controls, with higher doses showing greater efficacy (Day 21: 137.20 mg/dL for 100 mg/kg, 119.16 mg/dL for 200 mg/kg). Notably, HECSL-loaded phytosomes exhibited enhanced glucose-lowering effects, especially at 200 mg/kg, achieving near-normal glucose levels by Day 21 (96.18 mg/dL), (Table 4 and figure 3)suggesting improved bioavailability and antidiabetic potential of the phytosome formulation.

Table 4: Serum Glucose Levels in STZ-treated Rodents (Mean ± SEM)

Group	Treatment	Serum glucose levels (mg/dl)		
		1st Day	7th Day	21st Day
I	Normal Control	75.25 ± 4.18	95.30 ± 5.20	111.10 ± 6.22
II	Diabetic Control	296.15 ± 5.40	383.23 ± 6.05#	410.12 ± 8.35[a]
III	Diabetic + Glibenclamide	264.73 ± 6.55	132.10 ± 6.30*	92.34 ± 6.41*
IV	Diabetic + HECSL (100 mg/kg)	269.12 ± 5.36	146.20 ± 4.74	137.20 ± 7.14*
V	Diabetic + HECSL (200 mg/kg)	264.03 ± 4.29	141.10 ± 7.90*	119.16 ± 5.23*
VI	Diabetic + HECSL loaded Phytosomes (100 mg/kg)	274.50 ± 6.87	136.00± 7.61	122.52 ± 6.19*
VII	Diabetic + HECSL loaded Phytosomes (200 mg/kg)	269.40± 6.32	133.10 ± 5.25*	96.18 ± 4.72*

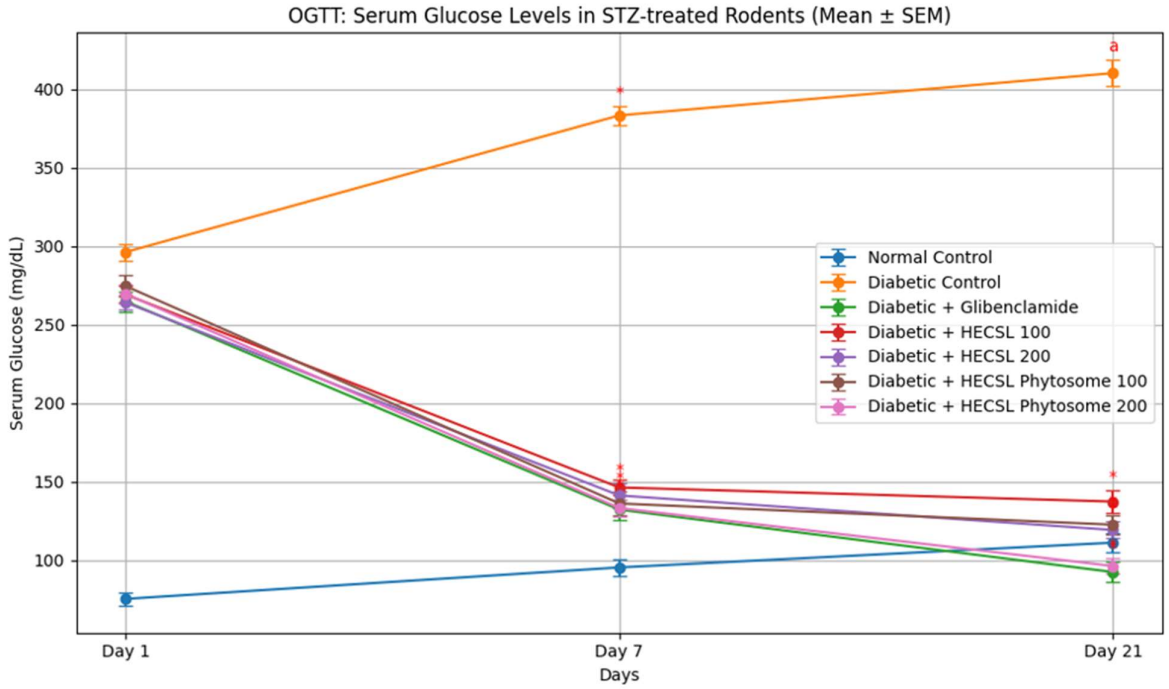

Figure 3: Serum Glucose Levels in STZ-treated Rodents (Mean ± SEM)

Discussion

The greater anti-diabetic effect seen in phytosome group may be due to increased bioavailability and better membrane permeability of active components. Phytosome technology overcomes the hurdles of poor water solubility and limited gastrointestinal absorption of herbal ingredients including polyphenols and flavonoids present in Coriandrum sativum.

4. CONCLUSION

Phytosomal encapsulation considerably enhances the bioavailability and anti-diabetic efficacy of Coriandrum sativum leaf extract. The unique formulation may be a possible natural treatment method for control of Type II diabetes.

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Disclosure

The authors report no conflicts of interest in this work.

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