

## Research Article



## Pharmacognostical, Phytochemical Screening, Formulation and Evaluation of Nanophytosomes of Aqueous Extracts of *Trigonella foenum graecum*, (Fabaceae)

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### ABSTRACT

Traditional approach is the one of the emerging field of biomedical sciences. In this research work, we have evaluated the traditional approaches of the ancient healer plant *T. foenum graecum* belonging to fabaceae family. The leaves of *T. Foenum graecum* was subjected to extraction process, phytochemical analysis, preparation of phytosome and evaluation of its characterization. The scientific data shows that, leave extracts contain flavonoids, alkaloids and glycosides etc., Nanophytosome was prepared by thin layer lipid hydration method. The preparation was evaluated for DSC, FTIR, SEM, TEM and particle size analysis. Recently, Nanophytosome methodology has been fruitfully applied over several well-known natural drugs such as ginseng, green tea hawthorn, olive oil and grape seed etc. Indeed, phytosome are superior to conventional drug delivery systems in terms of the pharmacodynamic and pharmacokinetic properties. This research, we discusses the various aspects, components, methods of preparation of phytosome and their marketed formulations, therapeutic applications along with the recent research work reported on this technology.

**Keywords:** Nanophytosome, *T. foenum graecum*, herbal extract, phytosome.

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### INTRODUCTION

Ever since the birth of mankind there has been a relationship between disease and plants. There is no record that people in prehistoric times used synthetic medicines for their ailments. But they tried to make use of the things, which were easily available, i.e, the plants and animals. Plants have been used for medicinal purpose long before and recorded in the history. The term 'Phyto' means plant while 'Some' means cell-like. Phytosome is vesicular drug delivery system in which phytoconstituents of herb extract surround and bound by lipid. The Phytosomes process produces a little cell because of that the valuable components of the herbal extract are protected from destruction by digestive secretions and gut bacteria. Phytosomes are better able to transition from a hydrophilic environment into the lipid-friendly environment of the enterocyte cell membrane and from there into the cell finally reaching the blood. Phytosomes proves to be of significantly greater clinical benefit.<sup>1,2</sup>

*Trigonella foenum graecum* Linn., belonging to the family fabaceae and sub family papilionaeae is cultivated

throughout the country. It is nearly smooth erect annual plant. It is cultivated in India, Africa, Egypt, Morocco, and occasionally in England. *Hulba* is native to South Europe and Asia. The plant is widely cultivated in many Parts of India. It has been proven to be an excellent remedy for reducing level of bad Cholesterol levels from our body. It purifies blood and helps in flushing out the harmful toxins. They give strong mucilage, which is emollient and a decoction. Leaves to 1 pint water are used internally in inflamed conditions of the stomach and intestines. Externally it is used as a poultice for abscesses, boils, carbuncles, etc. It can be employed as a substitute for cod-liver oil in scrofula, rickets, anaemia, debility following infectious diseases.<sup>3,4</sup>

### MATERIALS AND METHODS

#### Collection Of Plant Material

The Leaves of the plant *T. foenum graecum* were collected locally authenticated, cleaned and then they are shadow dried for 2 days and they are made into powder for further studies.

#### Preparation of Extraction

The leaves were shade dried on the laboratory bench for 10 days. The dry sample was milled and ground into powder. Extraction involves the separation of a bioactive portion of the plant tissues from the inactive components by using selective solvents in standard extraction procedure. The dried coarse powder of leaves of *T. foenum-graecum* was extracted with 500ml of water by Cold maceration method. After 72 hours, the extract was



collected by filtration, evaporated to dryness. The collected extract was stored in desiccators at room temperature and used for further pharmacological study.

### Macroscopic Characters

The morphological studies were carried out for different parameters such as shape, size, color, odor and taste and fracture identification of the fenugreek seed.<sup>5</sup>

### Microscopic Characters

The dried seeds of *Trigonella foenum graecum* were taken and soaked overnight in tertiary butyl alcohol, pieces transfer in ethanol for 2 hours and the sections were stained with phloroglucinol and examined under microscope.<sup>6</sup>

### Physicochemical Parameters

The determination of various physicochemical parameters such as total ash, acid insoluble ash, water soluble ash, water soluble extractive value, alcohol soluble extractive value, swelling index, foaming index, moisture content, ash value, pH were calculated as per Indian Pharmacopoeia.<sup>7</sup>

### Preliminary Phytochemical Screening

For preliminary Phytochemical screening, 100 g of powder drug was extracted with Distilled water. The plant extract obtained from successive solvent extraction were then subjected to various qualitative chemical tests to determine the presence of various Phytoconstituent like glycosides, phytosterols, fixed oils and fats, alkaloids, proteins and amino acids, flavonoids, saponins, gums and mucilage etc.<sup>8</sup>

### Preparation of *Trigonella foenum graecum* nano-Phytosomes

#### Thin Layer Lipid Hydration Method

Accurately weighed quantity of phosphatidyl choline and cholesterol were dissolved in 10 ml of chloroform in round bottom flask (RBF) and sonicated for 10 min using bath sonicator. Organic solvent removal is done by Rotary evaporator (45-50°C). After complete removal of solvent thin layer of phospholipids mixture was formed. This film was hydrated with aqueous extract of *T. foenum graecum* leaves in rotary evaporator (37-40°C for 1 hour). After hydration, mixture of lipid and plant extract was sonicated for 20 minutes in presence of ice bath for heat dissipation. Then prepared *T. foenum graecum* Nano-Phytosomes were filled in amber coloured bottle and stored in freezer (2-8 °C) until used.<sup>9</sup>

### Evaluation of *Trigonella foenum graecum* Nano-Phytosomes

#### Fourier Transform – Infra Red Spectroscopic Analysis (FT-IR)

To investigate the interaction of *T.foenum-graecum* leaves extract and Phospholipids in *T. foenum graecum* nano-phytosome complex, FT-IR spectra were measured. FT-IR spectral data can be taken to determine the structure and

chemical stability of *T. foenum graecum aqueous* extract, physical mixture of phosphoditylcholine and cholesterol, nano-phyto *T. foenum graecum* some complex. The spectroscopic evaluation of the formed complex can be confirmed by FT-IR simply by comparing the spectrum of the complex and the individual components and that of the mechanical mixtures. Samples were mixed with dry crystalline KBr in a ratio of 1:100 and pellets were prepared. The mixture was grounded or triturated into fine powder using an agate mortar before compressing into KBr disc. Each KBr disc was scanned at 4 mm/s at a resolution of 2. FT-IR can also be considered as a valuable tool in confirming the stability of the nano-phytosomal complex. FT-IR spectra were obtained using a FT-IR spectrometer. Spectral scanning can be done in the range of 4000-400 cm<sup>-1</sup>.<sup>10</sup>

### Differential Scanning Calorimetry (DSC) Analysis

Thermodynamical techniques are applied for determining the thermal stress of medicinal compounds of the excipients as well as their interactions during the formulation process. The thermal analysis of the *T. foenum graecum* leaves extract, physical mixture of phospholipids and cholesterol, *T. foenum graecum* nano-phytosome complex were placed in the aluminium crimp cell and heated at 10 °C/min from 0 to 400°C in the atmosphere of nitrogen (TA Instruments, USA, model DSC Q10 V24.4 Build 116). Peak transition onset temperatures were recorded by means of an analyzer.<sup>11</sup>

### Particle Size and Zeta Potential

The particle size and Zeta potential of *T. foenum graecum* Nano-phytosome were assayed by dynamic light scattering (DLS) using a Malvern Zetasizer Nano-ZS (Malvern Instruments, Worcestershire, UK). Scattered light was detected at a 173° angle. All measurements were performed in the standard suitable conditions to validate the results of sample dilution and the constant temperature of 25 °C. Furthermore, the apparatus we used systematically and automatically adapts to the sample, the intensity of the laser as well as the attenuator. Therefore, the reproducibility of the experimental measurement conditions is ensured.<sup>12</sup>

### Scanning Electron Microscope (SEM) analysis

Scanning electron microscopy has been used to determine particle size distribution and surface morphology of the *T. foenum graecum* nano-phytosome complexes. Samples were studied using JEOL JSM-6360 Scanning microscope (Japan). Approximately 5 µL of the nano-phytosomal solution were suspended in absolute alcohol and transformed to a cover slip, which in turn was mounted on a specimen tab. The samples were allowed to dry at room temperature. Then the particle size of the formulation was viewed and photographed using Scanning Electron Microscope (Sigma, Carl Zeiss). Digital images of nano-phytosome complex of *T. foenum graecum* were taken by random scanning of the stub at 1000, 5000, 10000 and 30000 X magnifications.<sup>13</sup>



### Transmission Electron Microscope (TEM) Analysis

Vesicles morphology of *T. foenum graecum* nano-phytosome was observed visually Transmission Electron Microscopy (TEM; H-7500, Hitachi Scientific Instruments Ltd., Japan)). A total volume of 10 ml *T. foenum graecum* nano-phytosome was dispersed before the analysis. The mixture was then stirred and a drop of the sample was placed on a 400-mesh copper grid coated with carbon. After 20mins, the grid was tapped with filter paper to remove surface water and stained using a solution of phosphotungstic acid (2% w/v) for 20 mins. Then the stained sample was dried in air and the morphology was observed.<sup>14,15</sup>

### X-Ray Diffractometer (XRD) Analysis

X-Ray diffractometer (XRD) is a unique method in determination of crystallinity of a compound and when properly interpreted, by comparison with drug XRD pattern before formulation, allows the identification of the drug crystalline changes. XRD was done on pure extract, physical mixtures of phosphatidyl choline and cholesterol, *T. foenum graecum* nano-phytosome complex to see the crystallinity in the substance. Sample was scanned in the angular range of 50 - 800 in a PHILIPS X-Pert Pro X-Ray Diffractometer. Dried powder sample was kept in sample holder (20 mm × 15mm × 2mm) which was fitted into the instrument and X-ray was passed through the sample.<sup>16</sup>

## RESULTS AND DISCUSSION

### Macroscopic Leaves of *Trigonella foenum graecum*



Figure 1: Aerial parts *T. foenum graecum*



Figure 2: leaves of *T. foenum graecum*

Colour: Yellowish green

Taste: Bitter

Odour: Agreeable

Shape: Ovate

Margin: Crenate

Apex: Obtuse

Size: 3.0-0 mm in length, 2.8-4.0 mm in width

### Microscopic Analysis

Leaves contains upper and lower epidermal cells with wavy anticlinal walls, striated cuticle and anomocytic stomata, more frequent on the lower epidermis; trichomas, more abundant on lower epidermis of two types; covering trichomas, uniseriate; glandular trichomas, slightly sunken, composed of a short, biseriate two or four-celled stalk and a biseriate head of four cells, around which the cuticle forms a bladder like covering. Transverse section of seed coat consists of an outer palisade layer. Parenchymatous layer is 1-2 cells thick followed by 2-3 layers of elongated cells of parenchyma. The aleuronic grains are oval to round in shape.



Figure 3: T.S of *T. foenum graecum*

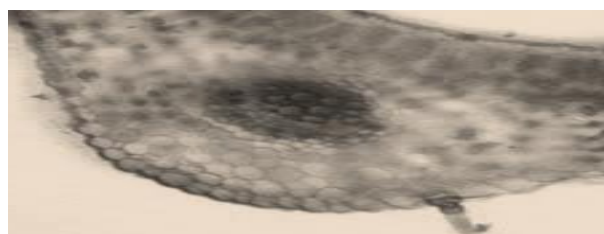


Figure 4: T.S of midrib of *T.foenum graecum*

### Preparation of Extraction

*T. foenum graecum* plant was extract by using aqueous solvent by cold maceration method. After extraction the decant subject evaporation and hot plate by rotator evaporator, until it get conc. macerate kept in desiccators for complete removal of moisture from the content The products were stored at -80 °c for further use. The percentage yield of extract of *T. foenum graecum* was found to be 14%w/w.

### Physicochemical Evaluation

The present work is helpful in determining the quality and purity of a crude drug. In this study the parameters used for the evaluation of *T. foenum-graecum* were moisture content, ash values (total ash, water soluble and acid insoluble ash). On incineration, drugs leave an ash which consists of carbonates, phosphates and silicates of sodium, potassium, calcium and magnesium. The determination of ash value is useful for detecting exhausted drugs, low-grade products and excess of sandy matter which is especially applicable to powdered drugs.

**Table 1:** Analytical parameters of *Trigonella foenum graecum*

S. No	Parameters	Values* expressed as %
1.	Moisture content	4.66± 0.46
2.	Ash values	
	Total ash	6.6 ± 0.25
	Acid insoluble ash	1.2 ± 0.17
	Water soluble ash	2.1±0.40

\*mean of three readings ± SEM

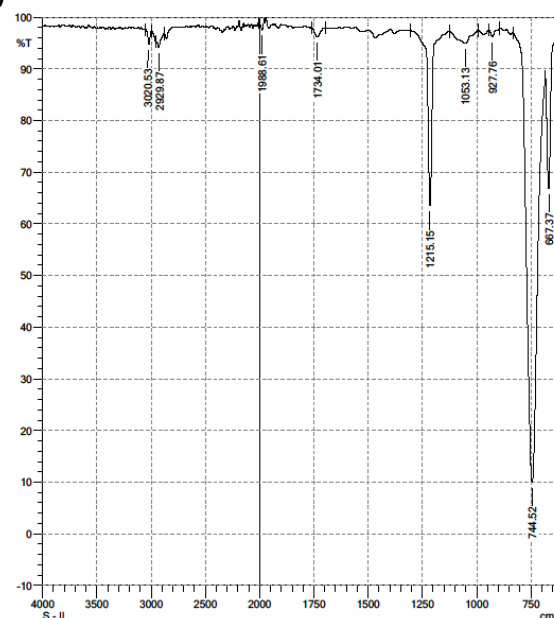
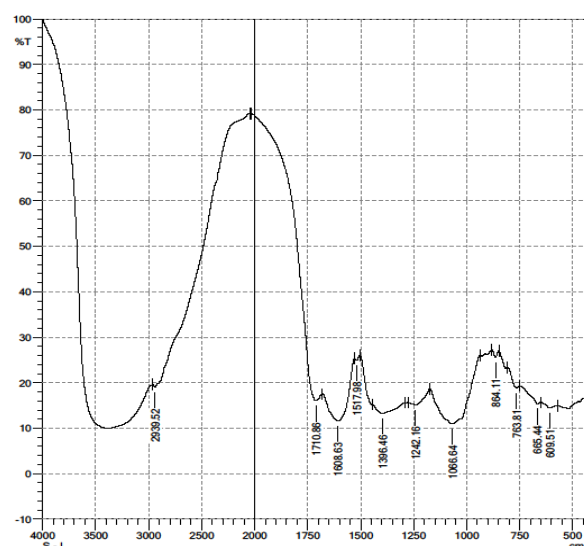
**Preliminary Phytochemical Studies**

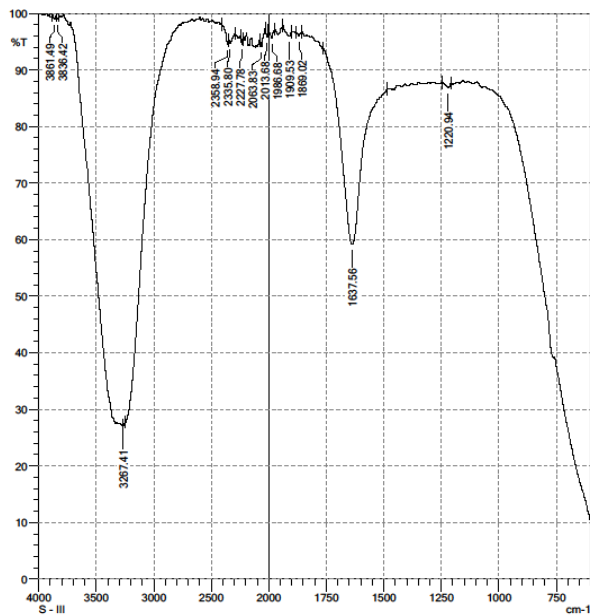
Phytochemical analysis was performed on the aqueous extracts of *T. foenum-graecum*. Aqueous extract contains carbohydrates, proteins and aminoacids, glycosides, alkaloids, flavonoids, phenolic compounds, phytosterols and tannins.

**Table 2:** Phyto-constituents of *Trigonella foenum graecum*

S.NO	TEST	RESULTS
<b>TESTS FOR CARBOHYDRATES</b>		
1.	a) Molisch's test	+
	b) Fehling's test	+
	c) Barfoed's test	+
	d) Benedict's test	+
<b>TESTS FOR ALKALOIDS</b>		
2.	a) Mayer's test	+
	b) Dragendroff's test	+
	c) Wagner's test	+
	d) Hager's test	+
<b>TEST FOR PHENOLIC COMPOUNDS AND TANNINS</b>		
3.	a) Braemer's test	+
<b>TEST FOR TERPENOIDS</b>		
4.		+
<b>TEST FOR FLAVONOIDS</b>		
5.	a) Shinoda test	+
<b>TEST FOR SAPONINS</b>		
6.	a) Foam test/ Frothing test	-
<b>TEST FOR VOLATILE OILS</b>		
7.		-
<b>TEST FOR GLYCOSIDES</b>		
8.	a) Legal's Test	+
	b) Borntrager's Test	+
<b>TEST FOR PHYTOSTEROLS</b>		
9.	a) Libermann-Burchard test	-
	b) Salkowski reactions	-

(+) indicates positive reaction (-) indicates negative reaction

**Evaluation of *Trigonella Foenum Graecum* Nano-Phytosomes****Fourier Transform Infra-Red Spectroscopic analysis(FT-IR)****Figure 5:** S-I represent FT-IR spectra of *Trigonella foenum-graecum* extract**Figure 6:** S-II FT-IR spectra of Physical mixture of phospholipids and cholesterol



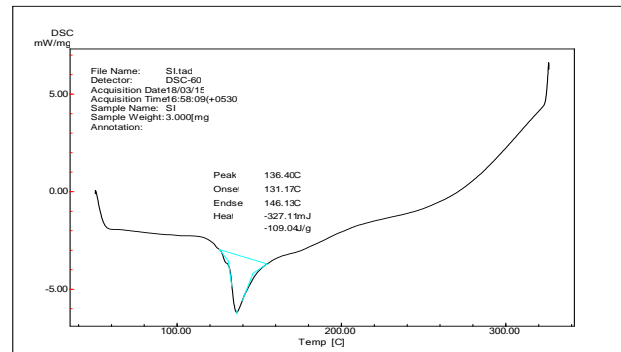
**Figure 7:** S-III FT-IR *Trigonella foenum-graecum* nano-phytosome

Spectroscopic analysis was used in order to identify and diagnose of complex formation between phospholipid and *Trigonella foenum-graecum* extract. In FT-IR spectroscopy, functional groups and their numbers were identified from the frequency of radiation that absorbs infrared spectra which showed the main chemical groups in extract and PC as well as the formation of new interactions between them in the nano-phytosomes preparation process (Table 3). The FT-IR spectroscopy of *T. foenum-graecum* leaf extract (S-I), mixture of Cholesterol and phospholipids(S II), *T. foenum-graecum* nano-phytosome (S-III) are shown figure 9, 10, 11. *T. foenum-graecum* leaf extract (S-I) shows that the characteristic C-H peak at  $2939.52\text{ cm}^{-1}$ , C=O peak at  $1710.86\text{ cm}^{-1}$ , C-C peak at  $1608.63\text{ cm}^{-1}$ , C-C peak at  $1517.98\text{ cm}^{-1}$ , C-H peak at  $1396.46\text{ cm}^{-1}$ , C-N peak at  $1242.16\text{ cm}^{-1}$ , C-N peak at  $1066.64\text{ cm}^{-1}$ . *T. foenum-graecum* nano-phytosomes (S-III) shows that the characteristic peak at O-H peak at  $3861.49\text{ cm}^{-1}$ , O-H peak at  $3836.42\text{ cm}^{-1}$ , N-H peak at  $3267.41\text{ cm}^{-1}$ , C≡ N peak at  $2358.94\text{ cm}^{-1}$ , C≡ N peak at  $2335.80\text{ cm}^{-1}$ , C≡ N peak at  $2227.78\text{ cm}^{-1}$ , C=C peak at  $2013.68\text{ cm}^{-1}$ , C=C peak at  $1986.68\text{ cm}^{-1}$ , C=C peak at  $1909.53\text{ cm}^{-1}$ , C=C peak at  $1869.02\text{ cm}^{-1}$ , N-H peak at  $1637.56\text{ cm}^{-1}$ , C-H peak at  $1453.16\text{ cm}^{-1}$ , C-C peak at  $1412.23\text{ cm}^{-1}$ , C-C peak at  $1398.76\text{ cm}^{-1}$ , C-H peak at  $1357.17\text{ cm}^{-1}$ , N-O peak at  $1346.78\text{ cm}^{-1}$ , C-N peak at  $1220.94\text{ cm}^{-1}$ . Some of the peaks of both *T. foenum-graecum* extract and phospholipid were shown at their physical mixture spectrum indicating no chemical interaction was occurred in this physical mixture.

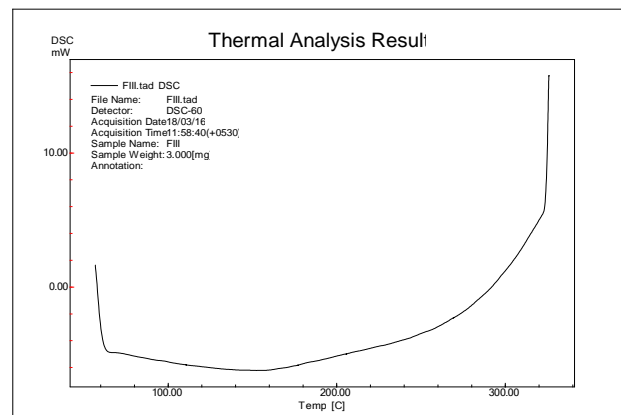
#### Differential scanning calorimetry analysis (DSC)

Differential scanning calorimetry was carried out to ensure formation of complex between *T. foenum-graecum* extract, cholesterol and egg lecithin, to confirm the presence of *T. foenum-graecum* extract in the phytosome.

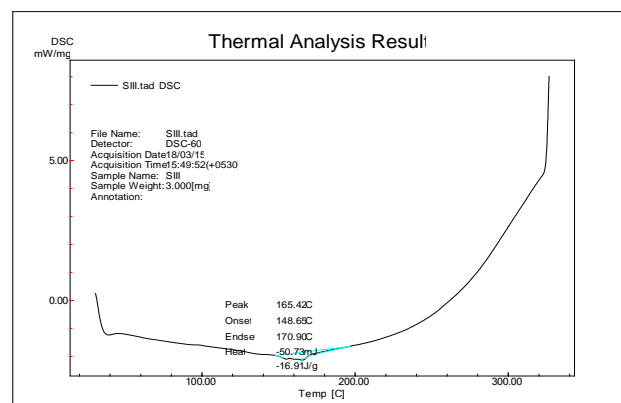
DSC thermograms of the pure *T. foenum-graecum* extract, cholesterol and egg lecithin, physical mixture of them are shown in Figure 12,13,14 . DSC thermogram of Pure *T. foenum-graecum* extract (S I) showed an endothermic peak at  $136.40^{\circ}\text{C}$  and its beginning melting point was  $131.17^{\circ}\text{C}$ , it is attributed to its melting point. Phospholipids and Cholesterol (S II) showed an endothermic peak at  $156.40^{\circ}\text{C}$ . DSC thermogram of physical mixture of *T. foenum-graecum* extract, cholesterol, phospholipids(S III) showed a endothermic peak at  $165.42^{\circ}\text{C}$ .



**Figure 8:** S-I DSC thermogram of extract *Trigonella foenum-graecum*



**Figure 9:** S-II DSC Thermogram of phospholipids and Cholesterol



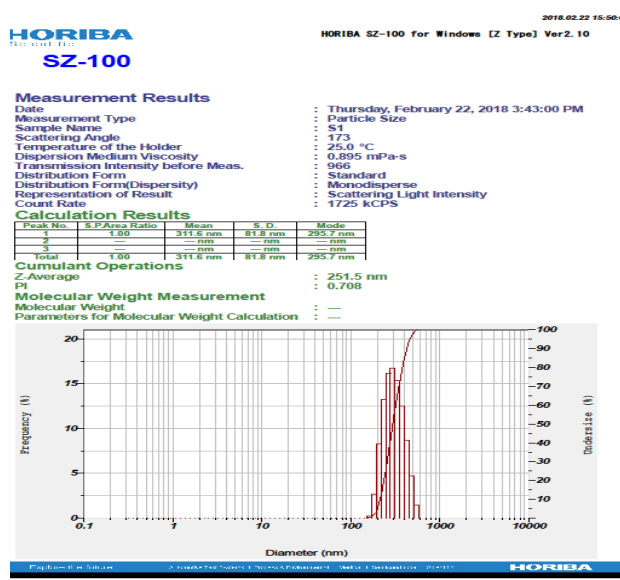
**Figure 10:** S-III DSC thermogram of physical mixtures of *T. foenum-graecum* extract, phospholipids, cholesterol

**Table 3:** Interpretation of FT-IR studies

S. No.	Functional Group	Wave number (cm <sup>-1</sup> )			
		Reference	<i>Trigonella foenum-graecum</i> Extract	Phospholipid	<i>Euphorbia hirta</i> nano-phytosome
1	O-H (stretch) Free hydroxyl, alcohols, phenols	3640-3610	-	-	3861.49
2	O-H(stretch) Free hydroxyl, alcohols, phenols	3640-3610	-	-	3836.42
3	N-H(stretch) 1 <sup>o</sup> ,2 <sup>o</sup> amines, amides	3400-3250	-	-	3267.41
4	C-H(stretch) aromatics	3100-3000	-	3020.53	-
5	C-H(stretch) alkanes	3000-2850	2939.52	2929.87	-
6	C≡N(stretch) nitriles	2260-2210	-	-	2358.94
7	C≡N(stretch) nitriles	2260-2210	-	-	2335.80
8	C≡N(stretch) nitriles	2260-2210	-	-	2227.78
9	C=C(stretch) alkenes	1900-2000	-	-	2013.68
10	C=C(stretch) alkanes	1900-2000	-	1988.61	1986.68
11	C=C(stretch) alkenes	1900-2000	-	-	1909.53
12	C=C(stretch) alkenes	1900-2000	-	-	1869.02
13	C=O(stretch) aldehyde, ketones, saturated aliphatic	1665-1740	1710.86	1734.01	-
14	C-C(stretch) aromatics	1600-1585	1608.63	-	1637.56
15	C-C(stretch) aromatics	1500-1400	1517.98	-	1398.76
16	C-H(Bend) alkanes	1470-1450	-	-	1453.16
17	C-C(stretch) aromatics	1500-1400	-	-	1412.23
18	C-H(rocking) alkanes	1370-1350	1396.46	-	1357.17
19	N-O Symmetric (stretch) nitro compounds	1360-1290	-	-	1346.78
20	C-N(stretch) aliphatic amines	1250-1020	1242.16	1215.15	1220.94
21	C-N(stretch) aliphatic amines	1250-1020	1066.64	1053.13	-

The carbon-hydrogen chain in soya lecithin perhaps happened to be melt, isomeric or the crystal changes. The physical mixture of *T. foenum-graecum* extract, Cholesterol and Phospholipids showed a broad endothermic peak at 165.42°C. When compare the *T. foenum-graecum* extract (136.40°C) with Physical mixture (165.42°C) it showed that there is no wide variation between the peaks and the difference is within ±20°C. This slight variation in the peaks may be due to the physical interact by Hydrogen bonding between-OH group of *T. foenum-graecum* extract and polar part of phosphatidylcholine. On the other hand, in the *T. foenum-graecum* extract-cholesterol-phospholipids complex, the major endothermic peak was observed at 165.42°C, which was different from the peaks of the individual components of the complex. Thermogram of Physical mixtures of *T. foenum-graecum* extract, phospholipids and cholesterol interestingly showed disappearance of the endothermic melting peak of *T. foenum-graecum* extract, indicating that *T. foenum-graecum* was completely embedded inside the matrix of physical mixture which had different thermal properties.

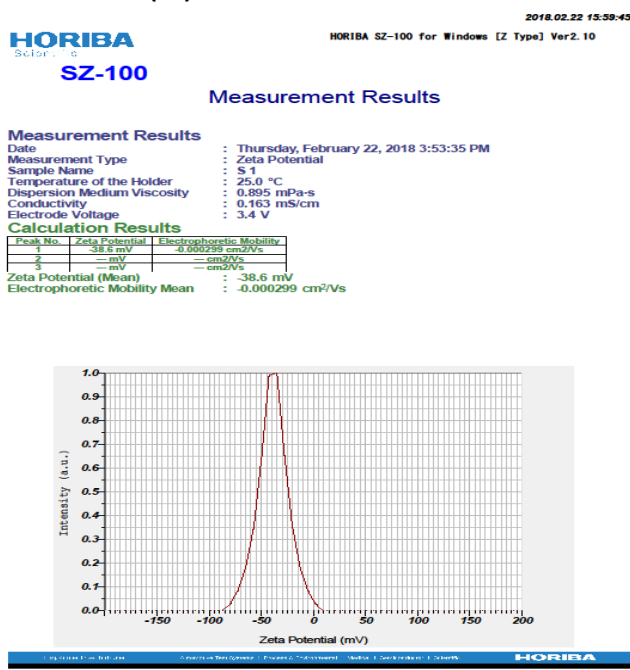
### Particle size (PS)



**Figure 11:** Particle sizes of *Trigonella foenum-graecum* nano-phytosomes

Particle size plays an important role in the stability, availability and organoleptic properties of the solution and the particles with smaller size are desirable. Results of particle size analysis indicated that nano-phytosomes prepared with *T. foenum-graecum* extract and PC possess the particle size in the average range of 251nm. Vesicle size tends to increase with increasing concentration of the complex. When the concentration of particles is too high, physical interaction either collision or electrostatic between vesicles is more pronoun. These interactions alter the movement of the particles and produce vesicles with a larger size. The high lipid composition in the formulation also increases the tendency for the formation of agglomerates, resulting in the bigger size of the vesicles. Polydispersity Index is a measure of the heterogeneity of sizes of molecules or particles in a mixture. *T. foenum-graecum* nano-phytosome prepared show polydispersity index value of 0.7 (Figure 15). This indicates the uniformity and homogeneity of the size of vesicles in the system. A relative high polydispersity index was observed in *T. foenum-graecum* nano-phytosomes which may be attributed to high phospholipid content.

### Zeta Potential (ZP)



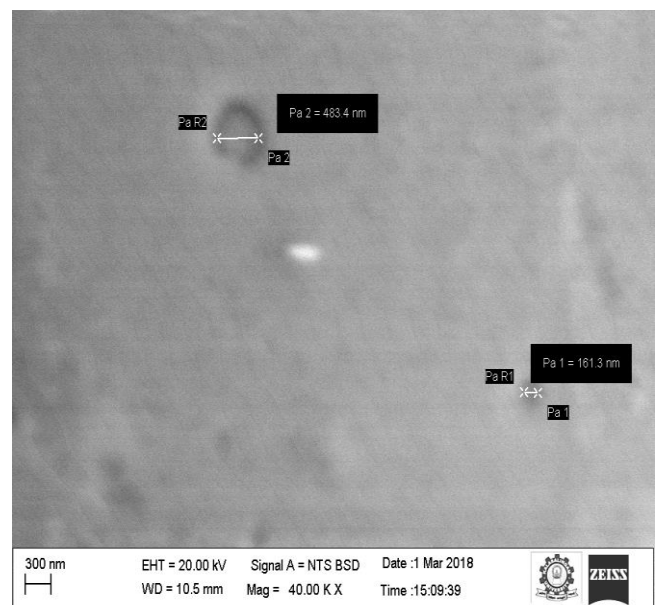
**Figure 12:** Zeta potential of *Trigonella foenum-graecum* nano-phytosomes

The Zeta potential is the electric potential in the interface or particle surface and is used to predict the stability of colloidal systems. The magnitude of the Zeta potential indicates the degree of electrostatic repulsion between adjacent, similarly charged particles which affects the stability of colloidal system. Colloids with high absolute Zeta potential values (normally  $\pm 30$  mV), regardless of their positivity or negativity, are electrically stabilized and those with low Zeta potential values are not stable and tend to coagulate or flocculate. In general, higher Zeta potential values indicate a higher and longer-term stability of the particles. Several factors such as pH, ionic strength,

type and concentration of the used biopolymers are effective on the Zeta potential of the particles. The surface charge analysis results (-38.6 mV) are shown in (Fig 16) and point to the physical stability of *T.foenum-graecum* nano-phytosomes.

### Scanning Electron Microscope analysis (SEM)

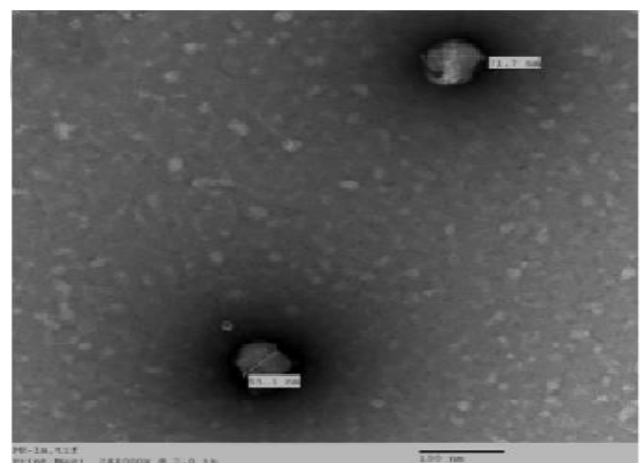
Scanning Electron Microscopy give important insight into the solid-state properties, surface morphology of drug and drug complexes. SEM analysis as given in confirms the vesicle size measured by size analyzer i.e., around 100-500 nm. The drug particles are associated with the phospholipids forming complexes with spherical shape, uniform and rigid vesicles.



**Figure 13:** SEM of Nano-phytosome *Trigonella foenum-graecum*

### Transmission Electron Microscope (TEM)

TEM analysis as presented in confirmed the spherical shape of the vesicle. The aggregation between vesicles was observed in single particle shape, forming irregular large size particles.

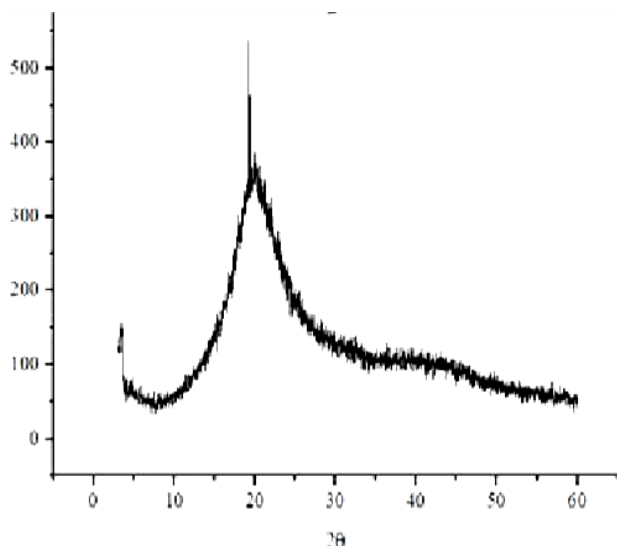


**Figure 14:** TEM analysis of *Trigonella foenum-graecum* nano-phytosomes

TEM pictures depicted that the nanophytosomes are discretely distributed. Nanophytosomes are nearly spherical in shape with smooth surfaces. Tem pictures shows the internal architecture of the nanophytosomes

#### X-Ray diffraction studies of nanophytosomes

Presence of single peak at  $20^{\circ}$  of  $2\theta$  indicates lack of crystallinity of the nanophytosomes and the absence of sharp peaks inferes the drug become amorphous after nanonization process



**Figure 15:** X-rd Analysis of *Trigonella foenum-graecum*nano-phytosomes

**Acknowledgment:** I sincerely thank P.Arjunkumar sir for giving me valuable suggestion for this project work.

#### CONCLUSION

In the present study, it is concluded that the plant *T. foenum-graecum* contains variety of phytoconstituents. Nanophytosomes of the aqueous extract of the leaves of the plant *Trigonella* were successfully developed. Phytochemical investigation gave the report of presence of flavonoids and tannins majorly. Preformulation studies confirmed the absence of drug excipient interactions. Characterization studies supported that the formed nanophytosomes are having spherical shapes with smooth surfaces. Zeta size and potential studies revealed that the developed phytosomes are nanometer in size with high macroscopic stability. Hence the preparation of nano phytosomes of *Trigonella* is an alternative method to increase the therapeutic ability of the plant.

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