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**Investigation on Standardization Parameters of leaves of  
*Thespesia populnea* Linn. with reference to Formulation and  
Evaluation of Phytosomes**

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**Abstract**

Phytosomes are complex compounds of flavonoid with phospholipids, characterized by high lipophilic and improved bioavailability and therapeutic properties as compare with free, not complex flavonoids. The present paper deals with the evaluating various standardization parameters of the selected plant to set the quality, safety and efficacy for the authentication of the plant. Further the phytosomes were formulated and evaluated.

Key-Words: Phytosomes, Standardization, Leaves, *Thespesia populnea*

**Introduction**

Phytosomes are produced by a patent process in which individual component of herbal extract like flavonoides and terpenoids are bound to the phospholipids like phasphatidylcholin through a polar end. Phytosomes are also called as Phytolipids delivery system. These are advanced form of herbal extract that are better absorbed which results better than conventional herbal extract. Phytosomes have improved pharmacokinetic and pharmacological parameters, which in result can advantageously be used in treatment of acute liver diseases, either metabolic or infective origin.

*Thespesia populnea* is a small evergreen tree with a short, often crooked, stem and a broad, dense crown. It has glossy green heart-shaped simple leaves, 6 to 22 cm long, that are alternately arranged. It produces yellow, mostly solitary, many-stamened hibiscus-like axillary flowers. The flowers are 4 to 7 cm long with 5 broad overlapping petals. The petals are pale yellow with a maroon spot at the base of each petal and star-shaped hairs on the outer surface. Individual flowers open and close on the same day and the yellow flowers turn dark red, purple, or pink as the day progresses.

The dry, brittle fruits are woody or papery seed capsules, rounded and flattened, containing 5 cells and several seeds. The brown or gray capsules are around 2.5 to 5 cm in diameter and 2 cm tall. Mature fruits are present year-round. They are indehiscent but eventually disintegrate, releasing the seeds. Bark and fruits possess more curative properties. The plant is astringent, cooling, depurative, anti-inflammatory, haemostatic, anti-diarrhoeal and anti-bacterial. It is useful in dermatopathy such as scabies, psoriasis, ringworm, leprosy, arthritis, haemorrhages wounds, ulcer, cholera, diabetes, as cites, dyspsia, cough, asthma, catarrh.

The plant was used traditionally and so far, no any systematic work was carried out in setting the standardization parameters of the species, also, no any novel formulation were reported, Hence, the present work was conceived.<sup>1-2</sup>

**Material and Methods**

**Selection, collection and authentication of plant/plant material**

The leaves of the selected plant were collected in the months of July 2013 from the Medicinal Plant Garden of Ujjain Institute of Pharmaceutical Sciences, Ujjain, M.P. and identified & authenticated by Dr. S.N. Dwivedi, Prof. and Head, Department of Botany, Janata PG College, A.P.S. University, Rewa, M.P. and was deposited in our Laboratory, Voucher specimen No. PCog/TP/042. The leaves were then, dried under shade, powdered and stored in an air-tight container for further use.

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**Physiochemical Constant Determinations**

The coarsely powdered leaves of *Thespesia populnea* were subjected to standard procedure for the determination of various physiochemical parameters.<sup>3</sup>

**Preparation of Extract**

Sample were shattered and screened with 40 mesh. The shade dried coarsely powdered leaves (250gms) was loaded in Soxhlet apparatus and was extracted with petroleum ether (60-62°C), Chloroform, ethanol and water until the extraction was completed. After completion of extraction, the solvent was removed by distillation. The extracts were dried using rotator evaporator. The residue was then stored in dessicator and percentage yield were determined.<sup>4</sup>

**Preliminary Phytochemical Screening**

The various extract obtained were evaluated for the presence of active phytoconstitents as per suggested standard protocol.<sup>5</sup>

**Formulation of Phytosomes**

In general phytosomes are prepared by mixing of drug/extract with phospholipids such as phasphatidylcholin in the solvent by hand rolling method. In the present investigation firstly cholesterol and lecithin were mixed in the chloroform and filtered. This solution was poured in sterilized round bottom flask, and role in one direction, until the film was not appearing; the flask was role in one direction for one hour. On the other hand solution of drug/extract was prepared by dissolving 100mg of drug/extract in the 10ml of distilled water. Poured the solution in to RB F, and roled in apposite direction.<sup>6</sup>

**Evaluation of Phytosomes<sup>6</sup>****Drug entrapment efficiency**

The ability of phytosomes to efficiently entrapt drug was measured by centrifugation technique. The vasicles (2 ml) were centrifuged at 4°C with 14000 rpm for three hrs. The supernatant and sediment obtained were then analysed for drug content using UV. The entrapment efficiency was calculated by the following equation.

$$\text{Entrapment efficiency(\%)} = (D/DT) \times 100\%$$

Where,

D = Amount of drug in sediment.

DT = Total amount of drug in supernatant and sediment

**pH measurement**

The pH of phytosomes formulation was measured by using the pH meter. The electrode was dipped into the vesicles as long as covered by the vesicles.

**Microscopy**

The microscopy of phytosomes was carried out and it was visualised under projection microscope.

**SEM (Scanning Electron Microscopy)**

The SEM test of phytosomes was carried out and it was visualised under instrument.

**Results and Discussion**

The morphological features of leaves of the plant *Thespesia populnea* were studied. The results were given in table 1. Various physico-chemical constant were carried out and were reported in table 2. The percentage yield were reported and presented in table 3. Preliminary phytochemical studies were performed for revealed the presence of active phytoconstituents (Table 4).

**Table 1: Morphology of the leaves of *Thespesia populnea***

S/No.	Characters	Appearance
1.	Colour	Dark green
2.	Odour	Characteristics
3.	Taste	Astringent.
4.	Shape	Heart-shaped
5.	Surface	6 -22 cm long

**Table 2: Physico-chemical constant of leaves of *Thespesia populnea***

S/No.	Parameters	Results (%w/w)
1.	Total value	17.3
2.	Water soluble ash	9.2
3.	Acid insoluble ash	4.5
4.	Foreign organic matter	1.7
5.	Loss on drying	7.2
6.	Water soluble extractive value	0.7
7.	Alcohol insoluble extractive value	0.3

**Table 3: Percentage Yield of extract of leaves of *Thespesia populnea***

S/No.	Extract	% Yield (w/w)
1.	Pet. ether Extract	
2.	Chloroform Extract	
3.	Ethanolic Extract	
4.	Aqueous Extract	

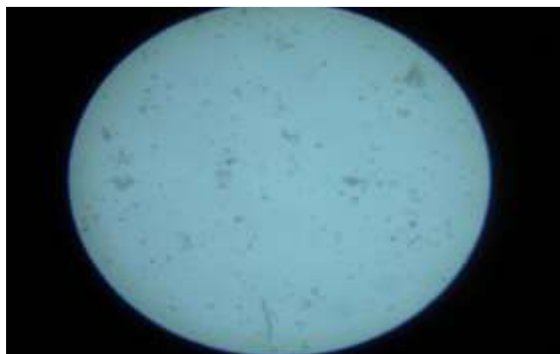
**Table 4: Phytochemicals of extract of leaves of *Thespesia populnea***

S/No.	Name of Test	Observation
1.	Gelatin test	+ ve
2.	Ferric Chloric test	+ ve
3.	Zinc hydrochloride test	+ ve
4.	Shinoda test	+ ve
5.	Alkaline reagent test	+ ve

The present study is directed towards the development of stable phytosomes from medicinal plant *Thespesia populnea*. Categorized as remedy for skin ailments, Antioxidant activity and Wound healing activity. Skin disease is severe chronic disease which affects all the ages and peoples. For this many allopathic drugs are available but still for herbal drugs are popularizing among the world so smart delivery system (phytosomes) was formulated using well documented plant with bibliographic traditional support. The major part of study was development of ethosomes by ethanolic extract of *Thespesia populnea*. The prepared phytosomes were subjected to determination of physical and chemical evaluation and result complies as per standards (Table 5, Fig. 1&2).

**Table 5: Evaluation parameters of Phytosomes**

S/No.	Parameters	Observation
1.	DEE	78.3%
2.	pH	6.9±0.3



**Fig. 1: Image of phytosomes**



**Fig. 2: Evaluation of Phytosomes of *T. populnea* ethanolic extract by SEM**

### Conclusion

Many herbal formulations are available for skin disease but scientific validation and standardization with better bioavailability and efficacy is very difficult for the formulation. Phytochemical evaluation of the crude drug such as Ash value, Extractive value were carried out and complies the limit given by quality standards of medicinal plants (ICMR). The Phytosomes is could be a promising new therapeutic approach for the treatment of clinical skin disease. The stability studies and clinical trials to be carried out in the future.

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