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Pre-Formulation Characterization Of Nanophytosomes Loaded With Anti-Obesity Phytoconstituents

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Abstract:
Nanophytosomes (NPs) represent an innovative nanocarrier-based drug delivery system designed to enhance the bioavailability of phytochemical-derived nutraceuticals in both the food and pharmaceutical industries. These plant- or herb-based vesicular systems exhibit superior absorption in biological systems compared to conventional herbal extracts. Traditional herbal medicines, although widely used for their therapeutic benefits, often suffer from limitations such as poor lipid solubility, suboptimal molecular size, and low bioactivity—factors that hinder their absorption and overall bioavailability. Obesity is a significant global health challenge, with its prevalence and associated comorbidities steadily increasing worldwide. The rising incidence of obesity poses a serious public health concern, as it profoundly impacts individuals' quality of life and contributes to various non-communicable diseases. Conventional treatments for obesity typically involve synthetic pharmaceutical agents and surgical interventions, both of which carry the risk of adverse side effects and a high likelihood of relapse. As a result, there is growing interest in alternative, safer, and more sustainable approaches. The existing research on the use of medicinal plants as natural anti-obesity agents, highlighting their botanical sources, active phytochemical constituents, and proposed mechanisms of action. The findings underscore the potential of plant-based compounds in obesity management and emphasize the need for continued research into novel phytoconstituents for safer, more effective anti-obesity therapies.

BACKGROUND:
The emergence of phytosomes nanotechnology has a potential impact in the field of drug delivery and could revolutionize the current state of topical bioactive phytochemicals delivery. The main challenge facing the translation of the therapeutic activity of phytochemicals to a clinical setting is the extremely low absorption rate and poor penetration across biological barriers (i.e., the skin)[1]. Nano phytosomes are tailored in size, shape and composition to optimize the delivery of phytochemicals/phytocompounds through nanoscale size and surface modification for better physiological absorption. Nanophytosomes increase the stability of phytochemicals/phytocompounds and protect them from degradation due to heat or chemical reactions, leading to longer shelf life and improved therapeutic efficacy[1,2].
Nanophytosomes (NP) technology is a kind of nanocarrier for drug delivery mechanisms to improve the bioavailability of phytochemical-derived nutraceuticals in food and pharmaceutical industries. It is an herb/plant based vesicular delivery system that is more easily absorbed by the biological system than traditional extracts. Traditional herbal medicines used for various medicinal purposes result in poor bioactivity, lipid solubility and incorrect molecular size, resulting in poor absorption and bioavailability. Therefore, a novel drug delivery system is in high demand compared to conventional herbal medicine, which paved the way for the emerging field of nanotechnology[4]. As a rapidly evolving class of nanovesicles, NPs have received significant attention for phytochemical delivery. The compatible molecular structures of NPs are formulated by incorporating the phytoactive compounds with phospholipids, especially phosphatidylcholine (PC). The structural composition of PC is similar to the cell membrane composition, which is later used as a potential vehicle in NP preparation due to its dual solubility and carrier properties. Some examples include quercetin phytosomes, which enhance the delivery of doxorubicin, a chemotherapy drug, to cells with increased permeability in the MCF-7 breast cancer cell line[1,5].
The aim of our study was to evaluate ethanolic extracts Pre-formulation Parameters of nano-phytosomes of selected phytoconstituents having anti-obesity activity.

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MATERIALS AND METHODS

Solubility Studies

Solubility studies were performed by taking an excess of the sample resveratrol in various solvent viz. water, ethanol, DMSO by bottle shaking method. The order of solubility of Resveratrol was found to be Ethanol > Dimethyl Sulfoxide (DMSO) > Water. From the date it was inferred that the Resveratrol shows good solubility in Ethanol, DMSO and Phosphate Buffer (pH 6.8) + PEG 400 (1:1). The order of solubility of Gymnemic acid was found to be Acetate Buffer (pH 4.5) > Dimethyl Sulfoxide (DMSO) > Ethanol > Phosphate Buffer (pH 6.8) Water >Phosphate Buffer (pH 6.8) + PEG 400 (1:1). From the date it was inferred that the Gymnemic acid shows good solubility in all solvents except Acetate Buffer (pH 4.5). The order of solubility of Naringin was found to be Acetate Buffer (pH 4.5) > Dimethyl Sulfoxide (DMSO) > Ethanol > Phosphate Buffer (pH 6.8) >Phosphate Buffer (pH 6.8) + PEG 400 (1:1). Water. From the date it was inferred that the Gymnemic acid shows good solubility in all solvents. The order of solubility of Gymnemic acid was found to be Ethanol>DMSO> Phosphate Buffer (pH 6.8) + PEG 400 (1:1)> Phosphate Buffer (pH 6.8)>. Water. From the date it was inferred that the Gymnemic acid shows good solubility in all solvent except water[3].

Table 1: Solubility studies of Resveratrol in various solvents

Sample	Water	Ethanol	DMSO	Phosphate Buffer (pH 6.8)	Acetate Buffer (pH 4.5)	Phosphate Buffer (pH 6.8) + PEG 400 (1:1)
Resveratrol	0.08 ± 0.06	0.78 ± 0.08	0.67 ± 0.12	0.102 ± 0.11	0.052 ± 0.01	0.4228 ± 0.24
Bromeline	1.24 ± 0.06	0.96 ± 0.06	0.89 ± 0.06	1.02. ± 0.06	0.06 ± 0.02	2.46 ± 0.24
Naringin	1.02 ± 0.04	1.78 ± 0.08	1.67 ± 0.10	1.102 ± 0.02	0.92 ± 0.02	1.22 ± 0.02
Gymnemic acid	0.07 ± 0.02	1.82 ± 0.04	1.74 ± 0.02	1.02 ± 0.02	0.98 ± 0.02	1.04 ± 0.02

Figure 1: Solubility of Resveratrol in various solvents in various solvents

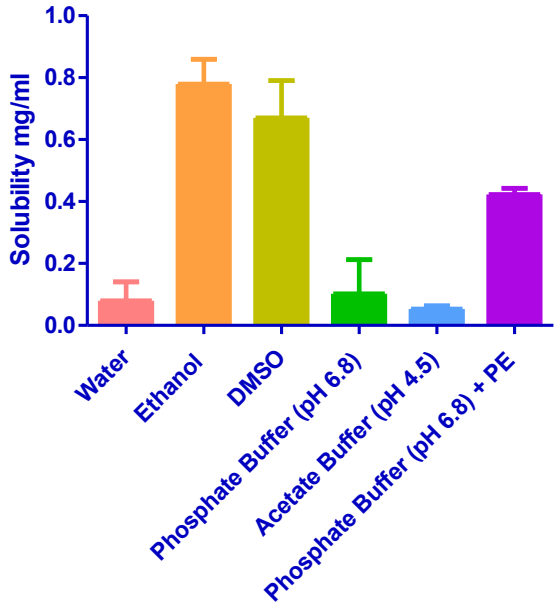


Figure 2: Solubility of Bromeline

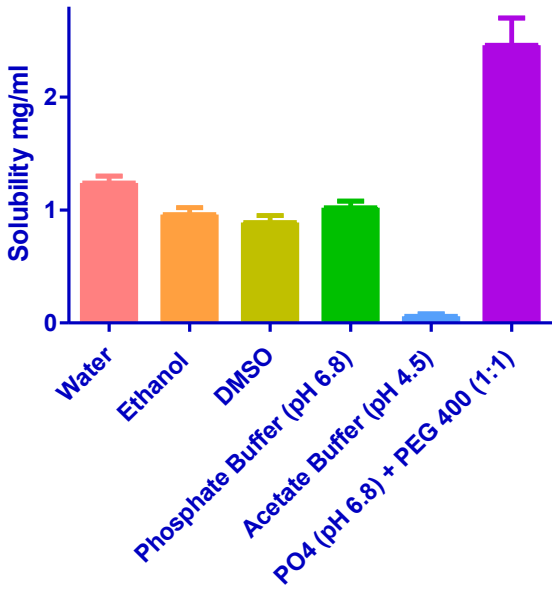


Figure 3: Solubility of Naringin in various solvents various solvents

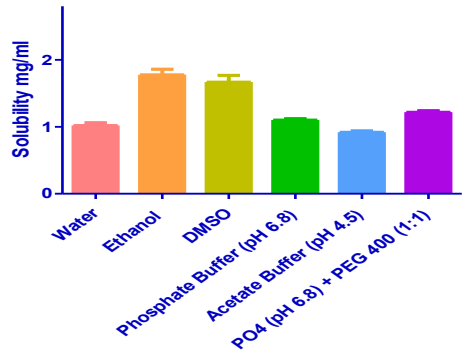
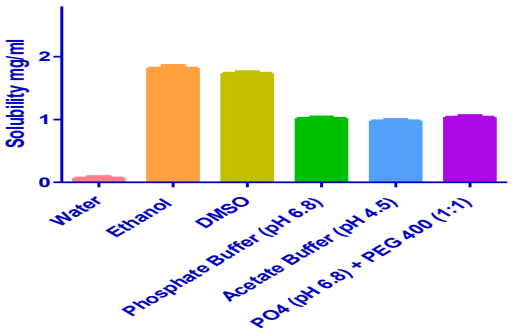


Figure 4: Solubility of Gymnemic acid in



Melting point by capillary method

The melting point of drugs are performed with the melting point apparatus and the same was tabulated below. From the data it was observed that, all the drugs melting point where within the acceptable criteria limits.

Table 2: Melting point of drugs

Sl.No	Drug	Melting point (^{0}C)	Acceptance criteria (^{0}C)
1	Resveratrol	257	*254*
2	Bromeline	164	164 - 170
3	Naringin	173	166-174
4	Gymnemic acid	175	150 - 175

Compatibility Studies

The drugs (Resveratrol, Gymnemic acid, Naringin, Gymnemic acid) and excipients (Phospholipid) compatibility studies were carried out by FT-IR.

The results shows the drug and excipients used in the formulations were found to be compatible with each other.

FT-IR Studies

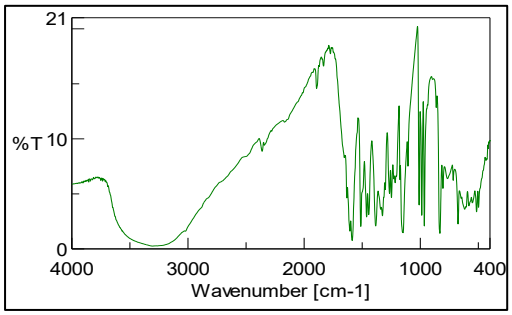


Figure 5: FTIR spectrum of Resveratrol Standard

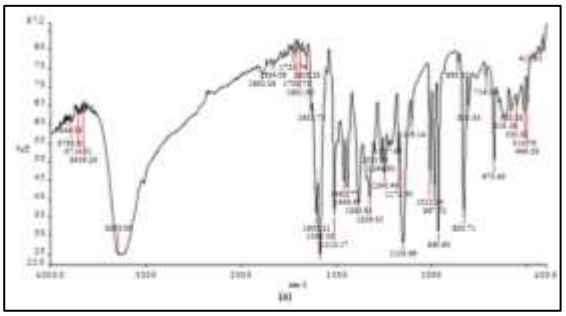


Figure 6: FTIR spectrum of Resveratrol extracted

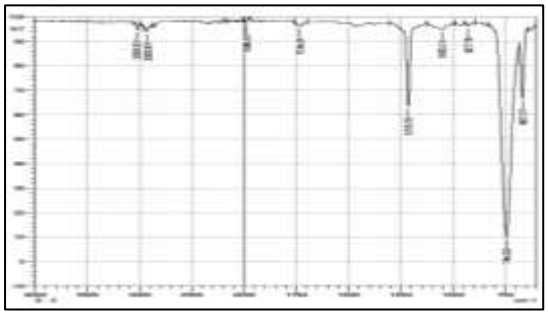


Figure 7: FTIR spectrum of Phosphodityl-choline with Cholesterol

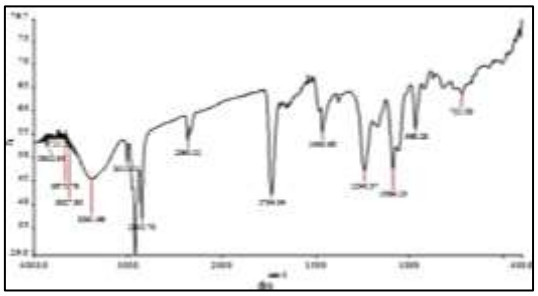


Figure 8: FTIR spectrum of Resveratrol extracted Phosphodityl-choline

Infra-red spectrum of resveratrol showed a typical trans olefinic band at 965.6 cm-1 and narrow band of O-H stretching at 3293 cm^{-1}. Three characteristic intense bands at 1383.85, 1586.53 and 1606.21 cm^{-1} correspond to C–O stretching, C–C olefinic stretching and C–C aromatic double-bond stretching. On the other side, infra-red spectrum of phospholipid showed O-H stretching at 3391 cm^{-1} and characteristic P=O stretching band at 1218 cm^{-1} and P-O-C stretching band at 1089 cm^{-1} and $N(CH_3)_3$ stretching at 744 cm^{-1}. With reference to the standard IR the sample procured is having similarities with the band ranges in Resveratrol and PC[6].

Thus from the FTIR studies it was inferred that the main function group as in Resveratrol standard and extracted resveratrol was found to be reproducible. It confirms that the extracted having polyphenol i.e., resveratrol which is responsible for anti-obesity activity. The main functional group of resveratrol was found to be reproducible in the physical mixture i.e. Resveratrol and phospholipid which is used for preparation of phytosomes. From the data it was confirmed that the resveratrol and phospholipids are compatible to each other, and selected phospholipid was suitable for formulation of phytosomes[7].

Compatibility studies by FTIR analysis – Gymnemic acid and its Mixture of excipients formulation

Figure 9: FTIR spectra for Pure drug Gymnemic acid

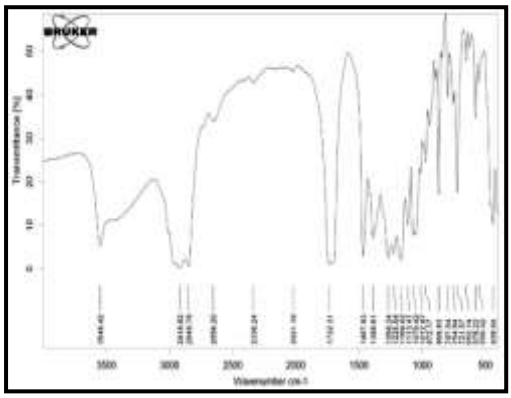


Figure 10: FTIR spectra for Gymnemic acid and excipients mixture

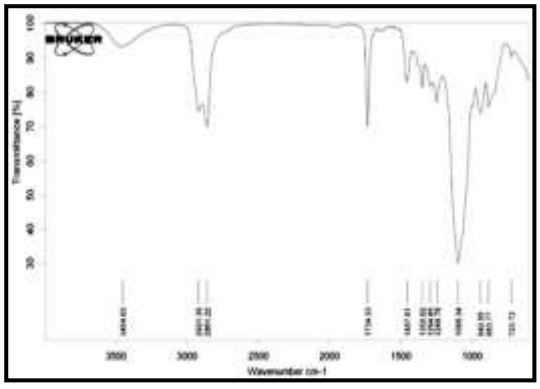


Table 3: Interpretation data of FTIR spectra showing possible functional groups for Gymnemic acid and its excipients

Wavenumbers in cm^{-1}	Origin	Wavenumbers in cm^{-1}	
		Gymnemic acid	Gymnemic acid and excipients
3600 – 3200	O – H alcohol(S)	3549.42	3454.03
3200 – 2500	O – H Carboxylic (S)	2918.62	2921.35
3000 - 2500	O – H Carboxylic (S)	2849.78	2861.22
1725 – 1705	C=O (S)	1732.31	1734.33
1470 – 1430	CH2	1467.93	1457.01
1390 – 1370	CH3	1388.61	1350.02
1150 – 1070	C-O (S)	1113.41	1248.78

The main functional groups with their wave numbers are O-H alcohol (symmetric) of 3549.42cm-1, O–H Carboxylic (symmetric) of 2918.62cm-1, O-H Carboxylic 2849.78cm-1 stretching, C=O stretching of 1732.31cm-1, CH2 of 1467.93cm-1, CH3 of 1388.61cm-1, C–O stretching 1113.41cm-1in the Gymnemic acid drug; The main functional groups with their wave numbers are O-H alcohol (symmetric) of 3454.03cm-1, O–H Carboxylic (symmetric) of 2921.35cm-1, O-H Carboxylic 2861.22cm-1 stretching, C=O stretching of 1734.33cm-1, CH2 of 1457.01cm-1, CH3 of 1350.02cm-1, C–O stretching 1248.78cm-1 in the Gymnemic acid with excipients respectively[9].

Compatibility studies by FTIR analysis – Naringin and its Excipients used in formulation

The main functional groups with their wave numbers are –OH stretching of 3337.85 cm^{-1}, 2922.84 cm^{-1}, -NH Stretching of 3337.85 cm^{-1}, C-H Stretching of 1450.20 cm^{-1},-OH group of 1333.33 cm^{-1}, C-O Stretching of 1256.02 cm^{-1},-C-H bending of 851.29 cm^{-1} in the Naringin drug; –OH stretching of

3615.92cm^{-1}, 3394.03 cm^{-1},-N-H stretching of 3350.52 cm^{-1}, 1508.99 cm^{-1},-O-H group of 1342.64 cm^{-1},–C-O stretching of 1239.05 cm^{-1},-C-H bending of 842.38 cm^{-1} in the Naringin with excipients respectively[8].

Figure 11: FTIR spectra of Naringin pure drug

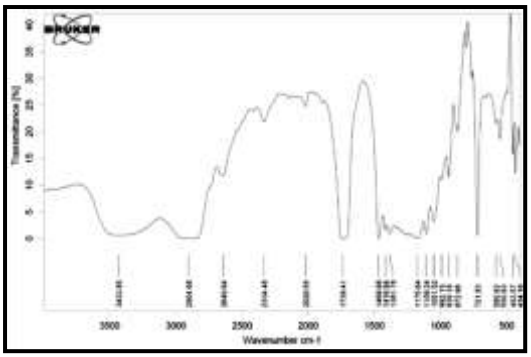


Figure 12.: FTIR spectra of Naringin with excipients

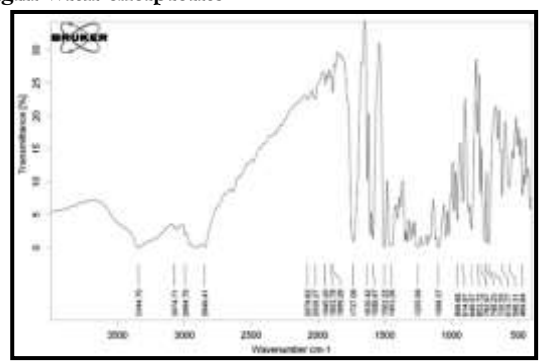


Table 4: Interpretation data of FTIR spectra showing possible functional groups for Naringin with excipients

Wave numbers in cm^{-1}	Origin	Wavenumbers in cm^{-1}	
		Naringin	Naringin with excipients
3600 - 3200	O–H alcohol(S)	3337.85	3344.70
3200 - 2500	O–H Carboxylic (S)	2922.84	2994.70
1750 - 1735	C=O (S)	1739.41	1737.09
3000 - 2500	O–H Carboxylic (S)	2922.84	2994.70
3350 - 3050	N-H (S)	3337.85	3344.70
1580 - 1490	-NH	1505.86	1503.22
1470 - 1430	C- H (S)	1450.20	1453.26
1410 - 1260	-OH	1333.33	1381.78
1300 - 1050	C-O (S)	1256.02	1099.37
900 – 800	-C-H (B)	851.29	849.57

Drug and Excipient compatibility studies:

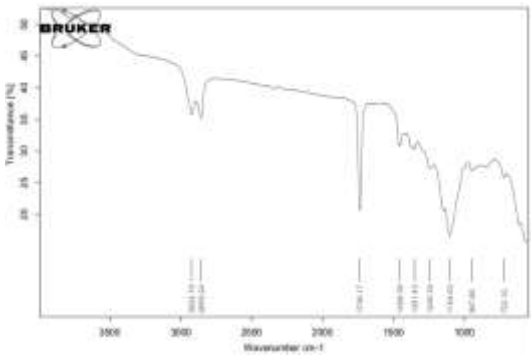


Figure 13: FTIR spectra of Bromeline

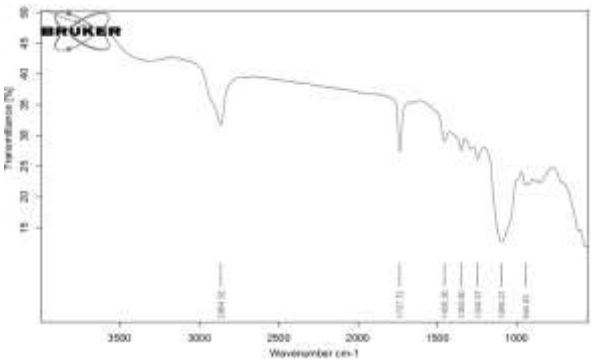


Figure 14: FTIR spectra of Bromeline with excipients

Table 5: Interpretation data of FTIR spectra showing possible functional groups for Bromeline with excipients

Range	Bromeline Wave length (cm^{-1})	Bromeline with excipients Wave length (cm^{-1})	Characterization
2850-2970	2864.52cm^{-1}	2924.70cm^{-1}	C–H stretching
1690-1760	1737.72cm^{-1}	1738.17cm^{-1}	C=O stretching
1340-1470	1456.36cm^{-1}	1458.36cm^{-1}	C–H bending

1340-1470	1350.80cm^{-1}	1351.43cm^{-1}	C=H stretching
1050-1300	1095.21cm^{-1}	1245.35cm^{-1}	C–O bending
675-995	944.55cm^{-1}	722.10cm^{-1}	C=H bending

DISCUSSION

The Bromeline formulations shown same spectra as isolated drug which demonstrates that the chemical structure of the drug doesn't change after converting to formulation and shows there is no interaction between the drug and excipients. The Gymnemic acid with excipients shown same spectra as standard Gymnemic acid drug which demonstrates that the chemical structure of the drug doesn't change after mixing with excipients and shows that there is no interaction between the drug and excipients. From the data obtained from FTIR spectra, it was inferred that the preferred functional group frequencies in pure Gymnemic acid drug were reproducible in Gymnemic acid with excipients. It was confirmed that the drugs and excipients used in the formulations were found to be compatible with each other[10].

The following melting points were observed as endothermic peak readings in DSC thermogram; Naringin at 171.05°C; Naringin with excipients thermogram shows reproducibility in thermogram peak at 170.43 (Drug peak). From the data, it was inferred that on performing the DSC studies for formulation, which ensures that the drug was effectively compatible with excipients. This thermal behavior confirms that both drugs exist in an amorphous form or molecularly dispersed in nature and also the excipients are highly compatible to the drug i.e., the drug property will not be affected by the excipients used in the formulation[1,2,5,8].

CONCLUSION:

Nanophytosomes are lipid-based nano-carriers that can improve the solubility, stability, and bioavailability of bromelain, potentially enhancing its therapeutic effects. Gymnema sylvestre extracts, containing gymnemic acids, can lead to a decrease in body weight and fat accumulation, particularly in animal models. The Pre-formulation Parameters of Nano-phytosomes of selected phytoconstituents having anti-obesity activity was done.

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