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Development and Validation of the UV-Spectrophotometric Method for Determination of Tetrahydrocurcumin

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Abstract

The main objective is to develop and validate the UV-spectrophotometric method for the estimation of Tetrahydrocurcumin as per ICH guidelines.

Materials and Methods: A simple, rapid, accurate, and economical UV-spectrophotometric method has been developed for the estimation of tetrahydrocurcumin.

Results: Theλ_{max} of tetrahydrocurcumin was found to be 280nm. The drug follows linearity in the concentration range 2-12 µg mL^{-1} with a correlation coefficient value of 0.9975. The accuracy of the method was checked by recovery experiment performed at three different levels, i.e., 50%,100% and 120%. The % recovery was found to be in the ranged between 97.81-101.39 with < 1 %RSD, thereby indicating the low variability and a strong agreement between experimental and true values. Ruggedness of the proposed method was studied with the help of two analysts**.**

Conclusion: The above method was a rapid tool for routine analysis of tetrahydrocurcumin

Keywords: Quantitative determination, Tetrahydrocurcurcumin, UV, validation

Introduction

Tetrahydrocurcumin, a significant polyphenolic metabolite of curcumin, was discovered in dried Curcuma longa rhizomes (C21h2406; mol. wt. 372.42Da). THC is an extremely potent, colourless antioxidant [1]. When compared to curcumin, THC has superior antioxidative activity, a longer half-life in the plasma, and greater stability in 0.1 M phosphate buffer at various pH levels [2]. Insoluble in water and more polar than curcumin, THC is soluble in acetone, alcohol, and glacial acetic acid [3]. The antibacterial,

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antitumor, antioxidant, and anti-inflammatory properties of THC have been demonstrated to be superior to those of curcumin, as well as its antiviral, anti-diabetic, anti-metastatic, anti-cancer, anti-hypertensive, neuroprotective, hepatoprotective, wound-healing, and anti-aging properties [4].

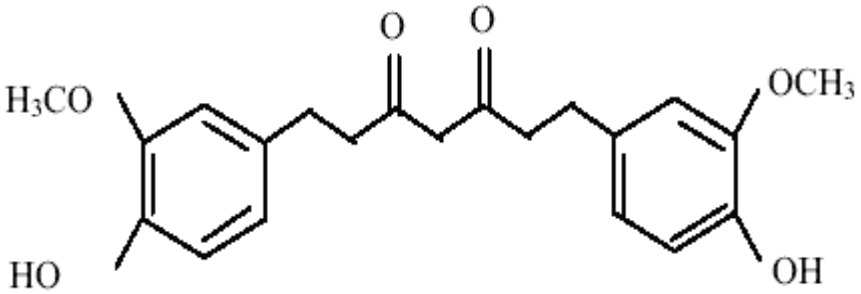
THC is frequently used in numerous skincare procedures. Tyrosinase is inhibited by THC, which makes it a bleaching agent. It lessens melanin synthesis. It is superior to other whitening agents like kojic acid, arbutin, and vitamin C in terms of effectiveness. It has a broad spectrum of anti-inflammatory and antibacterial properties. Skin inflammation and UVB-induced skin damage can both be treated with THC [5]. By easing pain and inflammation more quickly, IT has a favourable impact on the treatment of minor burns, skin irritation, and acne scars. It has been established that this makes it a safe and effective ingredient in high-end cosmetics that treat a variety of skin conditions[6]. This is due to its anti-inflammatory and high potential antioxidant activity.Despite having many positive attributes, THC's bioavailability is limited by its low aqueous solubility (0.0056 mg/ml) and ineffective skin absorption.(log P value of 2.98). Because of this, THC has a poor oral and systemic bioavailability and is typically only found in serum and urine as a glucuronide. THC is most likely to be eliminated through bile, where it is found as a conjugate of glucuronide. As a result, we require a different and more appropriate route of administration to deliver THC to the body in therapeutic doses while boosting its bioavailability and shielding it from metabolism [7].

In this regard, several accepted manuscripts on the development of nanotechnology-based drug delivery systems have been published. These include vaginal nano microbicides, self-emulsifying floating delivery systems, THC-based porous scaffolds, and electron spun nanofibres. Tetrahydrocurcumin, a metabolite of curcumin, is thought to be helpful for treating local inflammation [8].

There have been numerous reports on various nano-vesicular systems for enhanced topical/transdermal delivery of bioactive molecules. One type of nanovesicle that can be used to deliver medications for the treatment of skin conditions is the ethosome. Ethosomes are non-invasive, soft, malleable nanocarriers [9]. The lipid bilayer's organisation is disturbed by the presence of a high concentration of ethanol in the vesicles, which makes it easier for them to enter deeper skin layers [10].

They possess special qualities like biodegradability, biocompatibility, the capacity to entrap hydrophilic and hydrophobic drugs, drug protection from degradation, lowered side effects, enhanced skin penetration, and drug retention [11]. Accordingly, the objective of this study was to develop the UV-spectrophotometric method for the estimation of tetrehydrocurcumin as per ICH guidelines.

Structure



Material and Method

Table 1. List of Instruments

S. No.	Equipment	Manufacture
1.	Electronic Balance	IIAXIS, Model-AGN-204PO
2.	Digital pH meter	LAB INDIA
3.	UV-Vis spectrophotometer	LAB INDIA Analytical UV-3200, India
4.	IR- spectrophotometer	Bruker Alpha II
5.	Magnetic stirrer	REMI-2MLH
6.	Freezer	Godrej Edge, India
7.	Bath Sonication	Remi
8.	Ultracentrifuge	REMI Electro Technik LTD, INDIA

Table 2. List of chemicals

S. No.	Materials	Source
1.	Tetrahydrocurcumin	Sami Labs Limited, Bangalore, India
2.	Ethanol	Fisher Scientific, Mumbai, India
3.	Phospholipid® 90G	Lipoid Gumbo, Germany
4.	Potassium dihydrogen phosphate	Himedia laboratories, India
5.	Sodium hydroxide	Himedia laboratories, India

Method Development

In order to develop a UV spectrophotometric method for the estimation of THC, preliminary studies were carried out to screen a suitable solvent while taking into account the solubility of THC and the formulation of the nano-vesicles.

Preparation of stock and standard solution

A stock solution of THC (1 mg/mL) was prepared by dissolving the pure drug in HPLC grade methanol. Standard solutions were prepared by diluting the stock solution was the methanol to yield concentrations of 2, 4, 6, 8, 10 and 12 µg/ml respectively[12].

Selection of wave length of maximum absorption (λ_{max})

A standard solution containing 10 µg/mL of THC was scanned between the wavelength regions of 400 - 200 nm against methanol as blank.

Method Validation:

The developed UV-Spectrophotometric method was validated as per International Conference on Harmonization (ICH) guidelines Q2 (R1) in terms of system suitability, linearity, precision, accuracy, ruggedness and robustness.

System suitability

The system suitability was tested by measuring the absorbance of the standard solution of THC and determining the % relative standard deviation (%RSD) of the peak area[13].

Specificity and selectivity

The developed method was evaluated for specificity and selectivity by comparing the UV spectra of methanol and standard solution of THC[14].

Linearity

The calibration curve was obtained by analyzing the six different concentrations of THC standard solutions ranging from 2-12 µg/ml in triplicates. A graph between the concentrations versus peak area gives slope, intercept and coefficient of correlation (r^2)[15].

Sensitivity

The limit of detection (LOD) and limit of quantification (LOQ) was calculated from the standard calibration curve using the following equations[16].

$$LOD = \frac{3.3 \times SD}{\alpha}$$

$$LOQ = \frac{10 \times SD}{\alpha}$$

Where SD - Standard deviation of the response; α – Slope of the calibration curve

Precision

The precision of the developed method was established by performing both intra-day and inter-day variation studies. Intra-day precision was carried out by analyzing the three different concentrations (2, 6, and 12 µg/mL) three times in a day (morning, afternoon and evening) whereas inter-day precision was performed by three different concentrations on three different days. The peak area was measured and % RSD was calculated[17].

Accuracy

The accuracy of the developed method was assessed by determining the recovery value. It was carried out by adding known concentrations of THC standard solutions at three different concentration levels (50, 100 and 150 %) to the fixed concentration of pre-analyzed THC solution. The percentage recovery was calculated by comparing the theoretical amount and recovered amount[18].

Ruggedness and Robustness

Ruggedness of the method was determined by measuring the absorbance of the standard solution (6µg/ml) by two different analysts and two different instruments[19]. The robustness was assessed by measuring the absorbance at two different wave lengths (278 and 282 nm). The % RSD of the absorbance values was calculated[20].

Applicability of the Developed Method

Preparation of THC loaded nanoethosomes

The THC loaded ethosomal formulations (THC-ETH) were prepared using the classical cold method introduced by Touitou et al., 2000 with slight modifications. Briefly, phospholipon® 90G (600 mg) and THC (40 mg) were dissolved in ethanol (8 ml) in an airtight container under magnetic stirring at 700 rpm (Remi 2MLH, REMI ELEKTROTECHNIK LTD., India) at 30 °C ± 1 °C. The phosphate buffer saline (PBS, pH 6.4, 12 ml) pre-heated to 30 °C ± 1 °C was added slowly to the ethanolic solution at a constant rate (200

µl/min) with continuous mixing. The temperature was kept at 30 °C during the experiment. The stirring was continued for an additional 10 min, and the resultant ethosomal suspension was maintained at 4 °C overnight for swelling. The ethosomal suspension was bath sonicated using an ultrasonic sonicator (Riviera Glass Private Limited, Mumbai, India) for 5 min at 4 °C to get smaller vesicles. The sonicated ethosomal vesicles were stored at about 4 °C until further characterization[21].

Vesicle Morphology

An optical microscope (Radical scientific Equipments Pvt. Ltd, RXL-4T, India) was used to examine the formation of vesicular structure and shape of the prepared vesicles prior to sonication. A drop of diluted vesicular suspension was kept on a clear microscope slide and spread uniformly by placing the cover slip and examined under an optical microscope at a magnification of 20 X[22].

Determination of entrapment efficiency

The amount of THC entrapped inside the ethosomal system was determined by the centrifugation technique . An aliquot of the vesicular suspension was placed in the centrifuge tubes and centrifuged at 9,000 rpm for 1 h using a Remi R-12 C plus centrifuge (REMI ELEKTROTECHNIK LTD., India). Following centrifugation, the free THC present in the supernatant was separated from the pellet, appropriately diluted and quantified for THC content by using UV-spectrophotometer . The percentage entrapment efficiency (% EE) was determined as follows: EE % = (T−S)/T x 100, where T is the total amount of THC in the ethosomal dispersion; S is the amount of THC present in supernatant only, and T-S is the amount of THC present inside the nanovesicles[23].

Results and Discussions

Method development

Method development involves selection of suitable solvent which can solubilize the drug and provide sufficient sensitivity. Several solvents such as water, ethanol, methanol, dimethyl sulfoxide (DMSO) and dimethylformamide (DMF) were tried. THC and THC-ETH were very soluble in methanol hence methanol was selected as common solvent for development of UV-spectrophotometric method for estimation of THC in bulk drug and nanoformulation.

Selection of λ_{max}

The UV spectrum of THC in the wave length range of 200-400 nm is shown in Figure 3. The wavelength of maximum absorption (**λ_{max}**) for THC was found to be 280 nm. Hence 280 nm was selected for the analysis of THC.

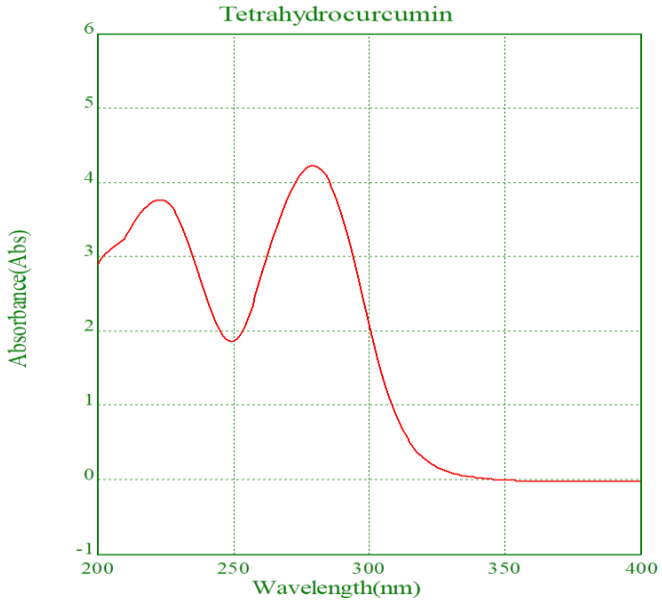


Figure 1: UV spectrum of THC in methanol

Method validation

The primary purpose of analytical method validation is to ensure that the methods are suitable for their intended use.

System suitability

The system suitability test was done to guarantee the performance of the UV-spectrophotometer during the analysis. The system suitability test results are summarized in Table 3. The % RSD of the absorbance values was within 1%, indicating the reproducibility and repeatability of the UV-spectrophotometer (% RSD < 1).

Table 3. System suitability test results (n = 6)

Runs	Concentration (µg/ml)	Absorbance
1	6	0.2758
2	6	0.276
3	6	0.2763
4	6	0.2767
5	6	0.2764
6	6	0.2762
Average		0.2762
STDEV		0.0003
%RSD		0.11

n = number of replicates

Specificity and selectivity

A specificity test was performed to ensure that none of the components interfered with the drug's quantification. The UV absorption spectrum of methanol and THC standard solution was depicted in the Figure 4. The zero absorbance value at 280 nm in the UV spectrum of solvent indicates the specificity and selectivity of the developed method.

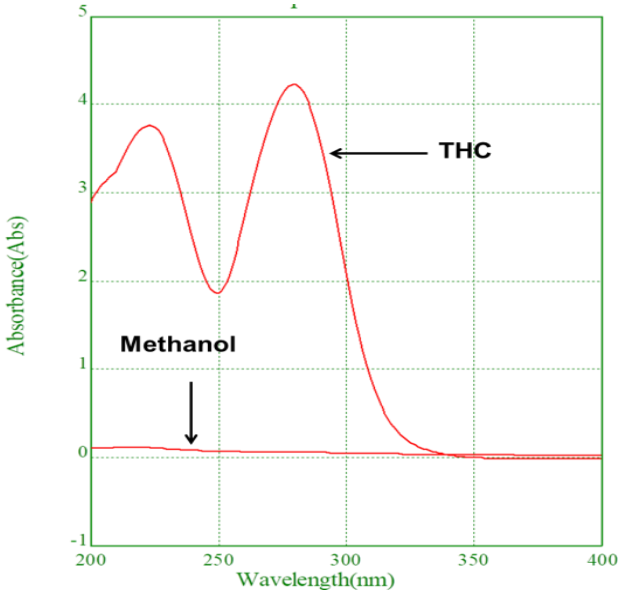


Figure 2. UV spectra of methanol and tetrahydrocurcumin

Linearity

Linearity is the ability of the analytical method to obtain test results that are directly proportional to the concentration of the analyte in the sample. The absorbance values of the standard THC solutions were plotted against their respective concentrations and subjected to the least square regression analysis to calculate the calibration equations and correlation coefficients (Figure 5). The calibration curves for THC was linear over the tested concentration range of 2-12 µg mL^{-1}. The regression equations for THC was y = 0.0577X + 0.0082. The correlation coefficient's values for THC were greater than 0.9975 suggesting excellent correlation and good linearity for the developed method.

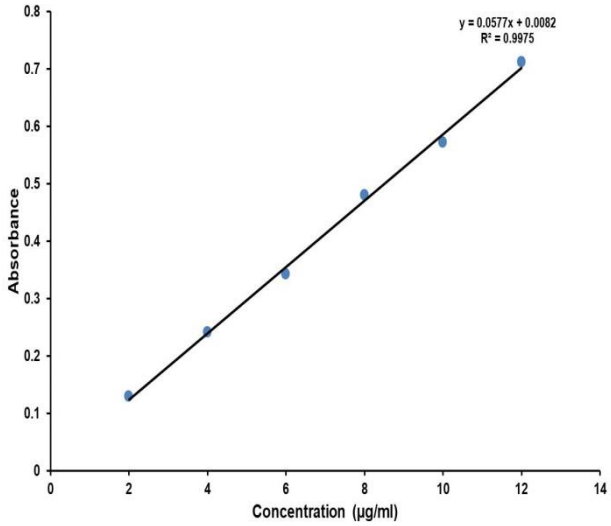


Figure 3. Calibration curve of THC in methanol

Sensitivity

Limit of detection (LOD) and limit of quantification (LOQ) were calculated to confirm the sensitivity of the method. The LOD and LOQ values for THC were 0.68 and 2.07 µg/ml respectively. The low values of LOD and LOQ indicate that developed method was sufficiently sensitive to be used for determination of entrapment efficiency of THC in nanovesicular system.

Precision

The precision of analytical method indicates the closeness of agreement between a series of measurements obtained from multiple samplings of the same sample under the similar analytical conditions. The results of intra-day and inter-day precision expressed in terms of %RSD for the determination of THC are presented in Table 4. The % RSD values for intra-day precision ranged from 0.39 – 0.59 % and those for inter-day precision were ranged between 0.10-0.87 % respectively. % RSD values in both cases were lower than 2% indicating the high precision of the developed method.

Table 4. Intraday and interday precision of THC

Analyte	Content (µg/mL)	Intraday (n = 6)		Inter day (n = 3)					
				Day 1		Day 2		Day 3	
		Found (µg/mL)	% RSD	Found (µg/mL)	% RSD	Found (µg/mL)	% RSD	Found (µg/mL)	% RSD
THC	2	1.95	0.59	2.05	0.87	2.07	0.97	1.99	0.21
	6	6.32	0.42	6.00	0.10	6.09	0.35	5.84	0.80
	12	12.12	0.39	11.89	0.67	12.11	0.39	11.99	0.13

n = number of replicates; RSD = relative standard deviation

Accuracy (recovery)

Recovery studies are carried out to measure how close the experimental value is to the true value or an accepted reference value. Recovery studies were performed by spiking pre-analysed THC samples with known concentrations of THC at three different levels (high,
middle and low) and the mean percentage recoveries along with their % RSD are summarized in Table 5. The mean percentage recoveries obtained for THC were ranged between 97.81-101.39 with < 1 %RSD, thereby indicating the low variability and a strong agreement between experimental and true values.

Table 5. Recovery studies of THC (n = 3).

Compound	Contents (µg)	Quantity added (µg)	Theoretical amount (µg)	Recovered amount (µg)	Recovery (%)	% RSD
THC	4.77	1.41	6.18	6.04	97.81	0.41
	4.77	4.79	9.56	9.69	101.39	0.27
	4.77	5.19	9.99	10.03	100.77	1.37

n = number of replicates; RSD = relative standard deviation

Ruggedness and robustness

Ruggedness and robustness is the measure of method's capacity to remain unaffected by the change in the analyst, instrument and in the spectrophotometric condition and to provide an indication of its reliability during the normal usage. The results of robustness data are represented in Table 6. The % RSD values calculated for THC was found to be less than 2% which indicates the ruggedness and robustness of the developed method.

Table 6. Results of ruggedness and robustness study (n = 6)

Parameter	Absorbance						
	Ruggedness				Robustness		
	Analyst I	Analyst II	Instrument I	Instrument II	278 nm	280 nm	282 nm
6 µg/ml	0.3425	0.3420	0.3425	0.3418	0.3420	0.3425	0.3421
6 µg/ml	0.3429	0.3421	0.3429	0.3415	0.3425	0.3429	0.3452
6 µg/ml	0.3426	0.3423	0.3426	0.3421	0.3427	0.3426	0.3422
Average	0.3426	0.3421	0.3426	0.3418	0.3424	0.3426	0.3431
SD	0.0002	0.0001	0.0002	0.0003	0.0003	0.0002	0.0017
% RSD	0.06	0.044	0.060	0.087	0.10	0.06	0.51

Applicability of the Developed Method

Preparation of THC loaded nanoethosomes

The THC loaded nanoethosomes were successfully prepared by using phospholipon® 90G and ethanol using thin film hydration technique.

Vesicle shape and morphology

The preliminary characterization of THC loaded ethosomes (prior to sonication) was carried out using an optical microscope. The optical microscopic images of THC loaded nanovesicles are shown in Figure 6, which confirms the formation of vesicles having a spherical shape and different lamellarity. Further, the images also showed the existence of a large population of vesicles with no aggregation and/or fusion among the vesicle.

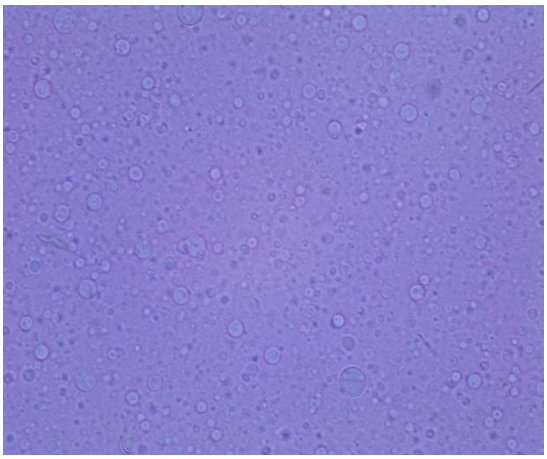


Figure 4. Optical photomicrographs of THC loaded ethosomes

Determination of entrapment efficiency

Entrapment efficiency refers to the percentage fraction of total drug incorporated inside the nanovesicular formulation. It is very essential to estimate the entrapment efficiency as it determines the drug holding capacity and ultimate delivery potential of a system. The developed UV spectrophotometric method was applied to quantify THC encapsulated within the developed nanovesicular systems. The % entrapment efficiency for THC was found to be 95.71 ± 2.14%. The higher entrapment efficiency of THC in ethosomes may be due to the co-solvent effect of ethanol. THC is a lipophilic drug which is soluble in ethanol. Hence the higher amount of ethanol would enhance the drug solubilization which would subsequently improve its entrapment within the both inner hydro-ethanolic core and lipid bilayers of the vesicle .

Conclusion

The proposed method for the determination of tetrahydracurcumin was found to be precise, selective, rapid and economical. Tetrahyracurcumin exhibited maximum absorption at 280 nm and obeyed Beer's law in the concentration range of 5 - 40 µg/ ml. The proposed method for the determination of Tetrahydracurcumin showed linear regression Y = 0.0577x + 0.0082 with correlation coefficient (R2) of 0.9975.

The % RSD for analysis of formulation was found to be within the limit. Our studies revealed a recovery percentage of 97.81-101.39 with<1% RSD which indicates that the developed method was found to be accurate. The proposed method can be used for the drug analysis in routine quality control and method proves to be more economical.

References

1. Alvarez-Ricardo Y, Meza-Morales W, Obregón-Mendoza MA, Toscano RA, Núñez-Zarur F, Germán-Acacio JM, Puentes-Díaz N, Alí-Torres J, Arenaza-Corona A, Ramírez-Apan MT, Morales-Morales D. Synthesis, characterization, theoretical studies and antioxidant and cytotoxic evaluation of a series of Tetrahydrocurcumin (THC)-benzylated derivatives. Journal of Molecular Structure. 2023 Feb 5;1273:134355.
2. Trivedi MK, Panda P, Sethi KK, Gangwar M, Mondal SC, Jana S. Solid and liquid state characterization of tetrahydrocurcumin using XRPD, FT-IR, DSC, TGA, LC-MS, GC-MS, and NMR and its biological activities. Journal of pharmaceutical analysis. 2020 Aug 1;10(4):334-45.
3. Kakkar V, Kaur IP, Kaur AP, Saini K, Singh KK. Topical delivery of tetrahydrocurcumin lipid nanoparticles effectively inhibits skin inflammation: in vitro and in vivo study. Drug development and industrial pharmacy. 2018 Oct 3;44(10):1701-12.
4. Sharma JB, Bhatt S, Saini V, Kumar M. Development and validation of uv-visible spectrophotometric method for the estimation of curcumin and tetrahydrocurcumin in simulated intestinal fluid. Research Journal of Pharmacy and Technology. 2021;14(6):2971-5.

5. Zhang ZB, Luo DD, Xie JH, Xian YF, Lai ZQ, Liu YH, Liu WH, Chen JN, Lai XP, Lin ZX, Su ZR. Curcumin's metabolites, tetrahydrocurcumin and octahydrocurcumin, possess superior anti-inflammatory effects in vivo through suppression of TAK1-NF-κB pathway. Frontiers in pharmacology. 2018 Oct 17;9:1181.

6. Panda SK, Nirvanashetty S, Missamma M, Jackson-Michel S. The enhanced bioavailability of free curcumin and bioactive-metabolite tetrahydrocurcumin from a dispersible, oleoresin-based turmeric formulation. Medicine. 2021 Jul 7;100(27).

7. Kakkar V, Kaur IP, Kaur AP, Saini K, Singh KK. Topical delivery of tetrahydrocurcumin lipid nanoparticles effectively inhibits skin inflammation: in vitro and in vivo study. Drug development and industrial pharmacy. 2018 Oct 3;44(10):1701-12.

8. Zhang ZB, Luo DD, Xie JH, Xian YF, Lai ZQ, Liu YH, Liu WH, Chen JN, Lai XP, Lin ZX, Su ZR. Curcumin's metabolites, tetrahydrocurcumin and octahydrocurcumin, possess superior anti-inflammatory effects in vivo through suppression of TAK1-NF-κB pathway. Frontiers in pharmacology. 2018 Oct 17;9:1181.

9. Godin B, Touitou E. Ethosomes: new prospects in transdermal delivery. Critical Reviews™ in Therapeutic Drug Carrier Systems. 2003;20(1).

10. Paiva-Santos AC, Silva AL, Guerra C, Peixoto D, Pereira-Silva M, Zeinali M, Mascarenhas-Melo F, Castro R, Veiga F. Ethosomes as nanocarriers for the development of skin delivery formulations. Pharmaceutical research. 2021 Jun;38(6):947-70.

11. Garg V, Singh H, Bimbrawh S, Kumar Singh S, Gulati M, Vaidya Y, Kaur P. Ethosomes and transfersomes: Principles, perspectives and practices. Current drug delivery. 2017 Aug 1;14(5):613-33.

12. Dinçer Z, Basan H, Göger NG. Quantitative determination of ambroxol in tablets by derivative UV spectrophotometric method and HPLC. Journal of pharmaceutical and biomedical analysis. 2003 Apr 1;31(5):867-72.

13. Altria KD, Rudd DR. An overview of method validation and system suitability aspects in capillary electrophoresis. Chromatographia. 1995 Sep;41:325-31.

14. Hyllbrant B, Tyrefors N, Markides KE, Långström B. On the use of liquid chromatography with radio- and ultraviolet absorbance detection coupled to mass spectrometry for improved sensitivity and selectivity in determination of specific radioactivity of radiopharmaceuticals. Journal of pharmaceutical and biomedical analysis. 1999 Jul 1;20(3):493-501.

15. Cobb Z, Shaw PN, Lloyd LL, Wrench N, Barrett DA. Evaporative light-scattering detection coupled to microcolumn liquid chromatography for the analysis of underivatized amino acids: Sensitivity, linearity of response and comparisons with UV absorbance detection. Journal of Microcolumn Separations. 2001;13(4):169-75.

16. Zhu L, Lee CS, DeVoe DL. Integrated microfluidic UV absorbance detector with attomol-level sensitivity for BSA. Lab on a Chip. 2006;6(1):115-20.

17. Eaton A. Measuring UV-absorbing organics: a standard method. Journal-American Water Works Association. 1995 Feb;87(2):86-90.

18. Gil M, Escolar D, Iza N, Montero JL. Accuracy and linearity in UV spectrophotometry with a liquid absorbance standard. Applied spectroscopy. 1986 Nov 1;40(8):1156-61

19. Mulholland M. Ruggedness testing in analytical chemistry. TrAC Trends in Analytical Chemistry. 1988 Nov 1;7(10):383-9.

20. Biter AB, Pollet J, Chen WH, Strych U, Hotez PJ, Bottazzi ME. A method to probe protein structure from UV absorbance spectra. Analytical biochemistry. 2019 Dec 15;587:113450..

21. Das SK, Chakraborty S, Roy C, Rajabalaya R, Mohaimin AW, Khanam J, Nanda A, David SR. Ethosomes as novel vesicular carrier: An overview of the principle, preparation and its applications. Current drug delivery. 2018 Jul 1;15(6):795-817.

22. Mbah CC, Builders PF, Attama AA. Nanovesicular carriers as alternative drug delivery systems: ethosomes in focus. Expert opinion on drug delivery. 2014 Jan 1;11(1):45-59.

23. Song X, Zhao Y, Hou S, Xu F, Zhao R, He J, Cai Z, Li Y, Chen Q. Dual agents loaded PLGA nanoparticles: systematic study of particle size and drug entrapment efficiency. European journal of pharmaceutics and biopharmaceutics. 2008 Jun 1;69(2):445-53.

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