Preparation and evaluation of phospholipid complex to improve solubility of curcumin

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Abstract
Water solubility, drug permeability, and metabolism are some of the elements that affect a medication's oral bioavailability. In medication development, poor solubility is a problem that frequently calls for particle size reduction methods like nanosization, which improves bioavailability. Important model terms and the significance of model reduction are revealed by statistical analysis. Additionally, thermal examination revealed a melting peak at 204.3°C and confirmed the successful synthesis of Cur-phospholipid complex with an average particle size of 170.2 nm, demonstrating curcumin's excipient compatibility. In comparison to traditional solutions, the improved formulation showed delayed drug release and a drug encapsulation effectiveness of 93.98%. Higher concentrations of the formulations reduced cell survival, according to studies on cell viability.

Keywords: Curcumin, phospholipid complex, pluronic, micelles

Introduction

A drug's oral bioavailability is significantly affected by factors such as water solubility, dissolving rate, drug permeability, and both first pass and pre-systemic metabolism. Solubility is crucial for achieving the necessary bloodstream concentration to elicit pharmacological effects. Recent advances in the pharmaceutical industry include technologies like computer modeling, high throughput screening, and combinatorial chemistry to create drugs with better affinity and selectivity; however, these often lead to low bioavailability [1-5]. The biopharmaceutical classification system identifies 50% of new drugs as class II, indicating limited water solubility. According to the U.S. Pharmacopoeia, a drug is considered water-soluble if it can dissolve in 250 mL of aqueous solutions with a pH between 1 and 7.5. Enhanced methods for improving solubility and bioavailability include complexation, particle size reduction, and crystal engineering [6-9]. Poor water solubility is a persistent challenge in drug discovery, with estimates suggesting that up to 70% of new compounds and 40% of marketed drugs face this issue. Strategies during pharmaceutical development must focus on improving drug solubility, as it is critical for drug release and absorption, particularly for oral medications. Effective oral bioavailability requires the drug to pass through cell membranes in dissolved form. Solutions are necessary for conducting tests essential for assessing toxicity and pharmacokinetic properties, as poor solubility restricts a drug's biological applicability. Finally, achieving effective drug delivery necessitates a successful sequence of disintegration, dissolution, and absorption in oral solid dosage forms, further complicated by the rates of disintegration and membrane penetration [10-14].

Because of the drug's low bioavailability, a high dose is recommended to reach the systemic circulation and maintain the minimal therapeutic level. Drug waste, toxicity, and adverse consequences result from this [15]. The reduction in particle size may cause a drug that is not particularly soluble to become super saturated insoluble. Because intrinsic solubility seldom varies (Fick's first law, Noyes-Whitey equation), the most obvious effect of particle size reduction is an increase in the dissolving rate through an increase in the particle surface area [16]. Experiments also revealed that the thickness of the diffusion layer decreased with decreasing particle size, which caused solvated molecules to migrate into bulk solution and dissolve more quickly [27].

Raising the surface area by increasing the dissolving velocity is part of the very basic classical micronization process. Particle size is reduced to 2 to 5 µm using micronization [18]. Micronization was insufficient to maximize bioavailability because many recently developed novel medicines had very weak solubilities. The particles started the nanosization process, the next stage of size reduction following micronization, when their size was reduced to less than 1 µm and their size distribution fell between 200 and 500 nm [19]. The bioavailability and solubility rates of nanosized particles were significantly higher than those of micronized particles because of the substantial increase in surface area. Aim of this study was to prepare phospholipid complex of curcumin to improve solubility.

Material And methods

Material

Pluronic F127 was supplied by Himedia Laboratories in Thane, Maharashtra, India, whereas curcumin was provided by SRL Pharma. In Mumbai, India, Merk Specialties Pvt. Ltd. sold us DMSO and other solvents. The investigation also made use of analytical-grade solvents. Milli-Q water served as the solvent.

Compatibility Study by DSC and FTIR

A reliable technique for doing compatibility studies of materials, particularly in the creation of pharmaceutical

formulations, is the combination of Fourier-transform infrared (FTIR) spectroscopy with differential scanning calorimetry (DSC). While FTIR offers details on molecular interactions and chemical structure, DSC sheds light on a material's thermal characteristics.
The interaction between the components required to produce the Cur-phospholipid complex was examined by obtaining Cur, Phospholipon® 90H, and their PM IR spectra. The FTIR spectra of the Cur-phospholipid complex were also recorded using an FTIR spectrophotometer (FTIR-8300).

Preparation of Cur-phospholipid complex
Two milliliters of ethanol were used to dissolve 100 milligrams of weighted Ccm. About 100–300 mg of Phospholipon® 90H were used with this solution. 1,4-dioxane was added to further dissolve the mixture. After that, the entire mixture was refluxed in a water bath at 60 to 70°C for four to six hours. The reaction mixture was heated in a water bath and then condensed. A small amount of n-hexane (around 3–4 mL) was added to this saturated mixture. For the night, the resultant mixture was refrigerated. The next day, the mixture was allowed to precipitate using distilled water. The next day, the mixture was allowed to precipitate using distilled water. The supernatant was decanted and dried the next day at room temperature. For analysis, an additional batch of Cur-phospholipid complex with a yield of about 90% was stored in an airtight glass vial at room temperature.

Physicochemical characterization of Cur-PhP
Particle Size and Zeta Potential
To characterize nanocarriers and ensure their effectiveness, particle size and zeta potential are two of the most critical properties to measure. In summary, deionized water was used to disperse and transfer 5 mg of dried Cur-phospholipid complex in cuvetes. This was placed within the instrument chamber of the Malvern Zetasizer, and measurements were taken at 25 °C. A Malvern Zetasizer nanosizing device was used to test the zeta potential of the Cur-phospholipid complex between -200 and +200 mV.
Determination of drug content
Drug content, also known as drug loading content (DLC), is a critical quality attribute for nanocarriers that indicates the amount of a drug loaded into the formulation. It is typically expressed as a percentage of the total weight of the nanocarrier system or as the mass of the drug per unit mass of the nanocarrier.
Entrapment efficiency
Entrapment efficiency is calculated by measuring the amount of drug that was not loaded ("free drug") and comparing it to the total amount of drug added during formulation. As previously reported in literature, centrifugation was utilized to separate the free medication from the Cur-phospholipid complex in order to evaluate the degree of entrapment. After being diluted, the supernatant was extracted and used for analysis using a micropipette. The samples were analyzed at 427 nm. The following formula was used to determine the entrapment efficiency.

$$\% EE = \frac{(Total\ Drug - Unentrapped\ Drug)}{Total\ Drug} \times 100 \qquad \text{Eq 1}$$

In vitro drug release from nanocarriers
The Franz diffusion cell's donor compartment was filled with the aqueous suspension of the Cur-phospholipid complex. Next, the receptor compartment was filled with pH 7.4 phosphate buffer saline. The diffusion cell's donor and receptor compartments were separated by the dialysis membrane. The Cur-phospholipid complex aqueous solution, which included about 10 mg of medication in an equivalent volume of Cur-phospholipid complex powder (~20 mg), was poured into the donor compartment. An equivalent volume of pH 7.4 PBS was added to the medium after samples were taken from the side arm at prearranged intervals. A UV spectrophotometer was used to measure the acquired samples at 427 nm.
Scanning electron microscopy (SEM)
Scanning electron microscopy (SEM), which creates high-resolution, three-dimensional images of the external shape of nanocarriers, is a basic technique for characterizing them. Important characteristics of nanocarriers, including size, shape, surface roughness, and aggregation, are directly and thoroughly revealed by this visual study.

Result And discussion
Drug's excipient compatibility
The excipient compatibility of the drug was confirmed through physical mixing and thermal analysis of curcumin. Differential Scanning Calorimetry (DSC) showed a significant melting peak for pure curcumin at approximately 204.3°C, and a glass transition for PF 127 was observed between 50.8 and 54.5 °C (see Figure 1). Fourier Transform Infrared (FTIR) spectroscopy identified characteristic peaks for functional groups in curcumin at specific wave numbers, including hydroxyl, carboxyl, alkyl, and ester groups, with notable assignments for C–H and carbonyl stretching vibrations (see Figure 2). These findings suggest potential chemical changes or novel molecular interactions.

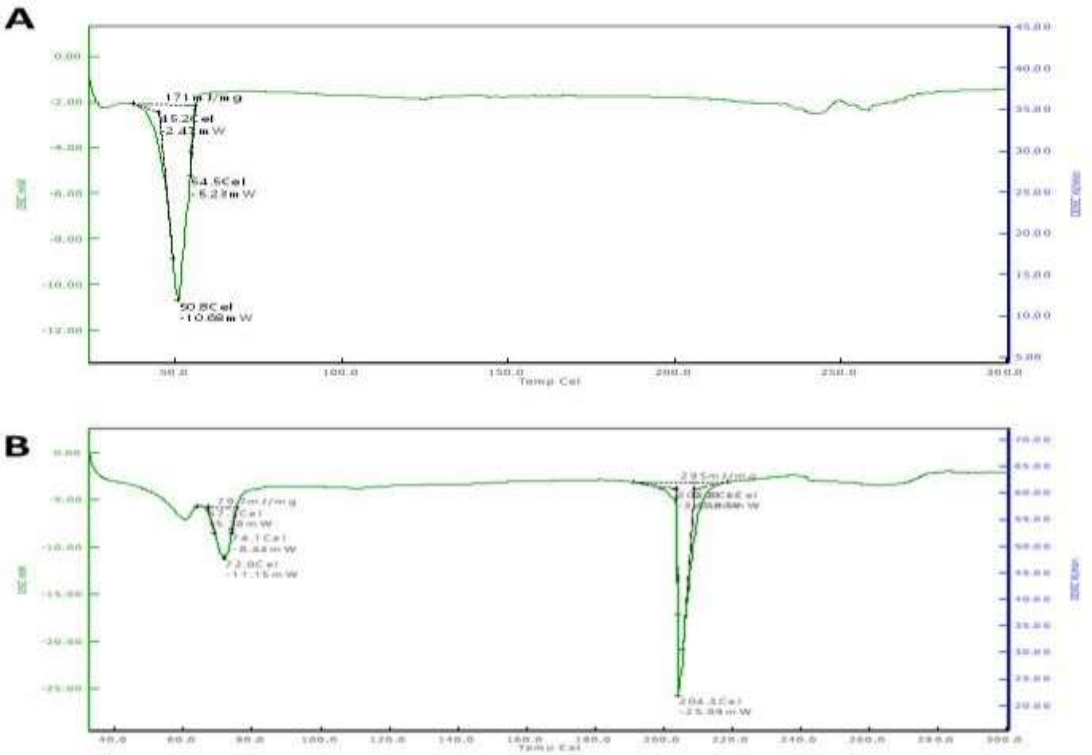

Figure 1: DSC of curcumin and their mixture with other excipients

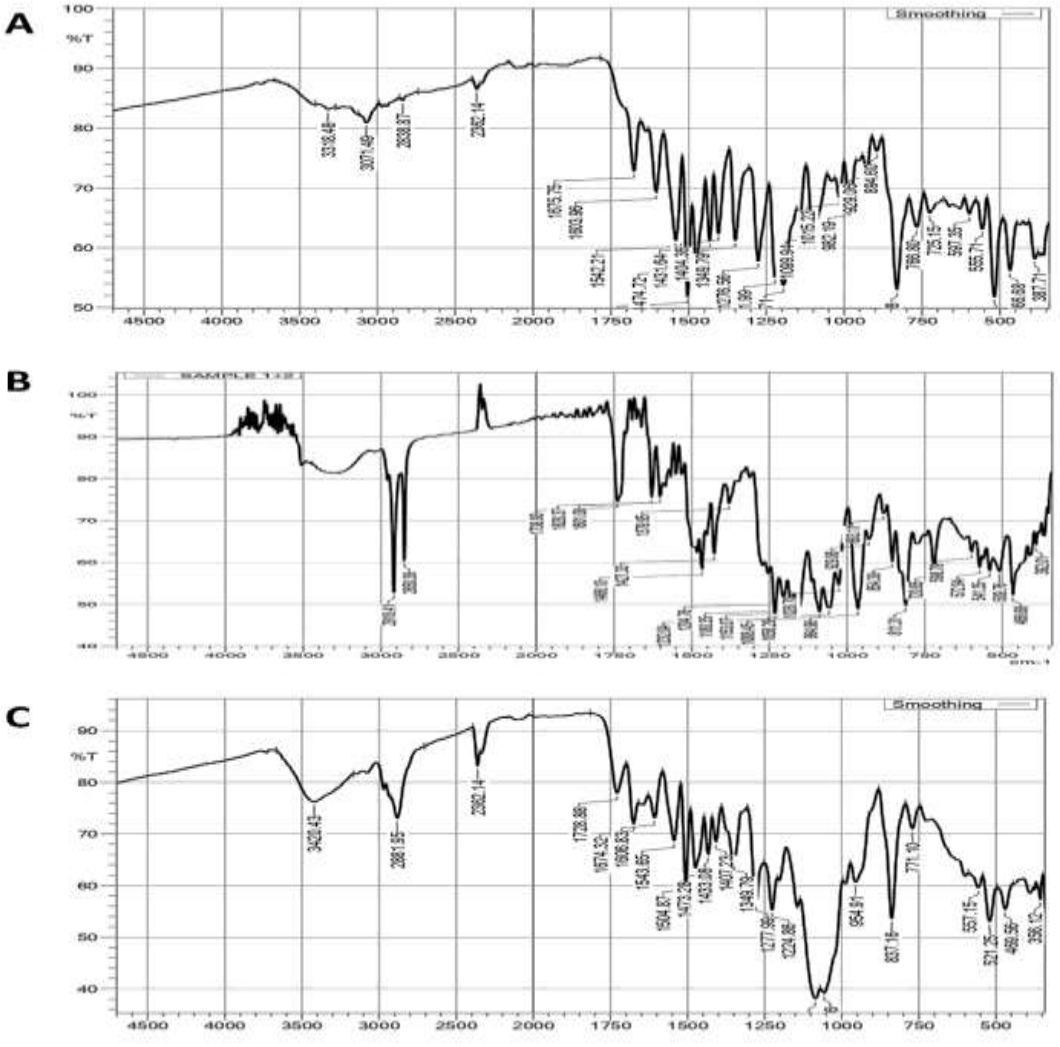

Figure 2: FTIR spectra of curcumin A), PF127 B) and their mixture C)

Particle Size

A crucial stage in the development of Cur-phospholipid complex is the measurement of their particle size utilizing a DLS-based nanoparticle sizing device. Drug transport across the semipermeable membrane is influenced by particle size. Previous research has shown that as the particle size falls between 200 and 400 nm, the drug's bioavailability increases [21, 22]. The average particle size of the curcumin phospholipid complex was 170.2 nm (see Figure 3A). The Cur-phospholipid complex were found to have zeta potentials of approximately -15.10 mV (see Figure 3B).

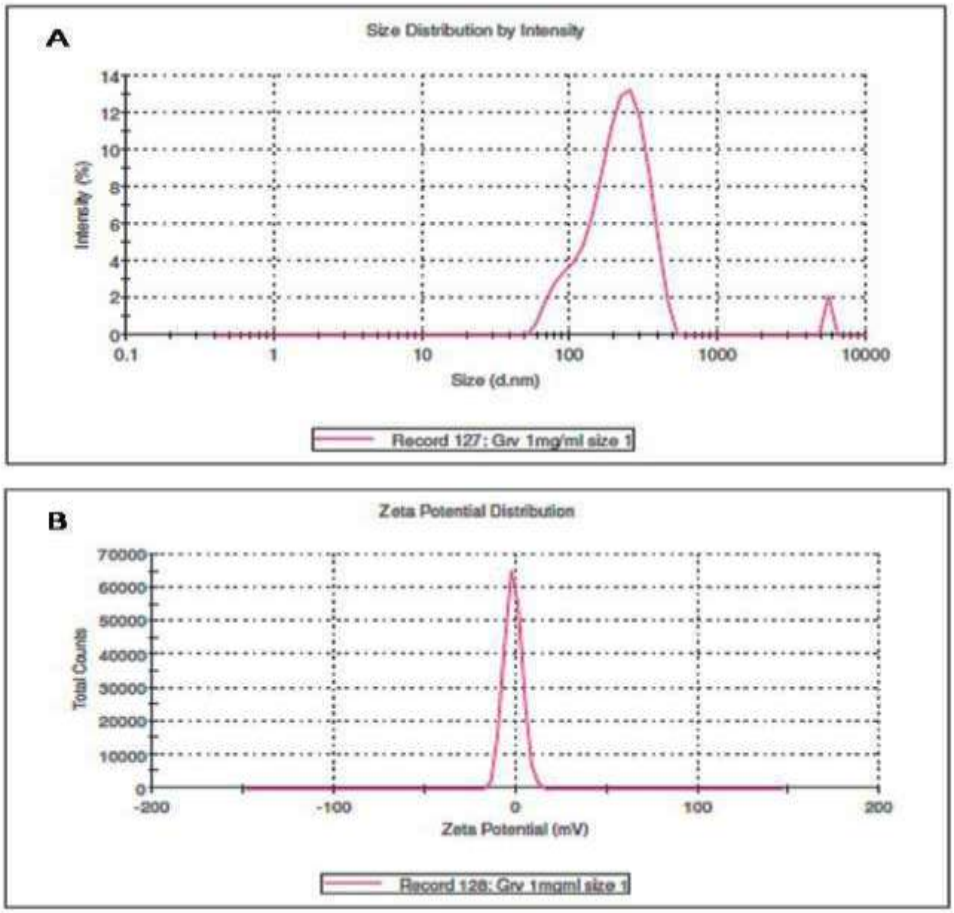

Figure 3: Particle size and zeta potential of Cur-phospholipid complex

Entrapment efficiency

It was discovered that the improved nanocarrier formulation's %EE was 93.98 ± 1%.

***In vitro* drug release study**

Using the dialysis method, the drug release pattern of Cur from the standard curcumin solution was assessed during a 24-hour period. The optimized batch of Cur-phospholipid complex underwent a similar process (see Figure 6). The improved batch of Cur-Ph Cur-phospholipid complex P and the traditional curcumin solution are compared (see Figure 6). The improved batch demonstrated a 98.11±0.22% release after a full day. Thus, compared to traditional curcumin solution, Cur-phospholipid complex exhibit a higher percentage of drug release over a longer duration.

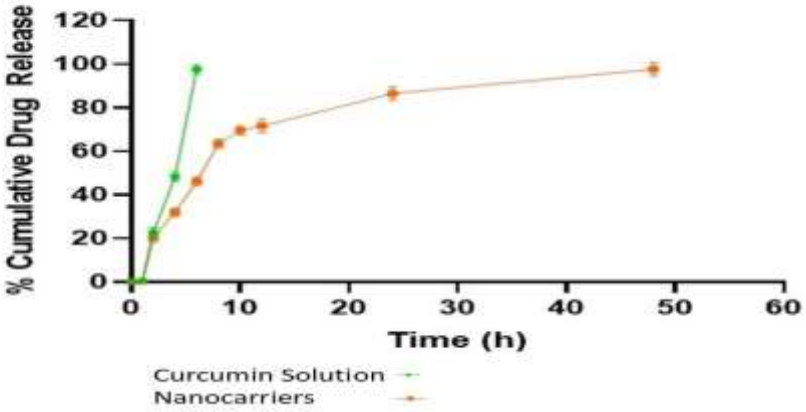

Figure 4: *In vitro* drug release from Cur-phospholipid complex

SEM study

Figure 5 displays scanning electron microscopy pictures of the Cur-phospholipid complex in cluster form with tiny nanosize spherical shapes. This demonstrates that Cur-phospholipid complex formation was successful.

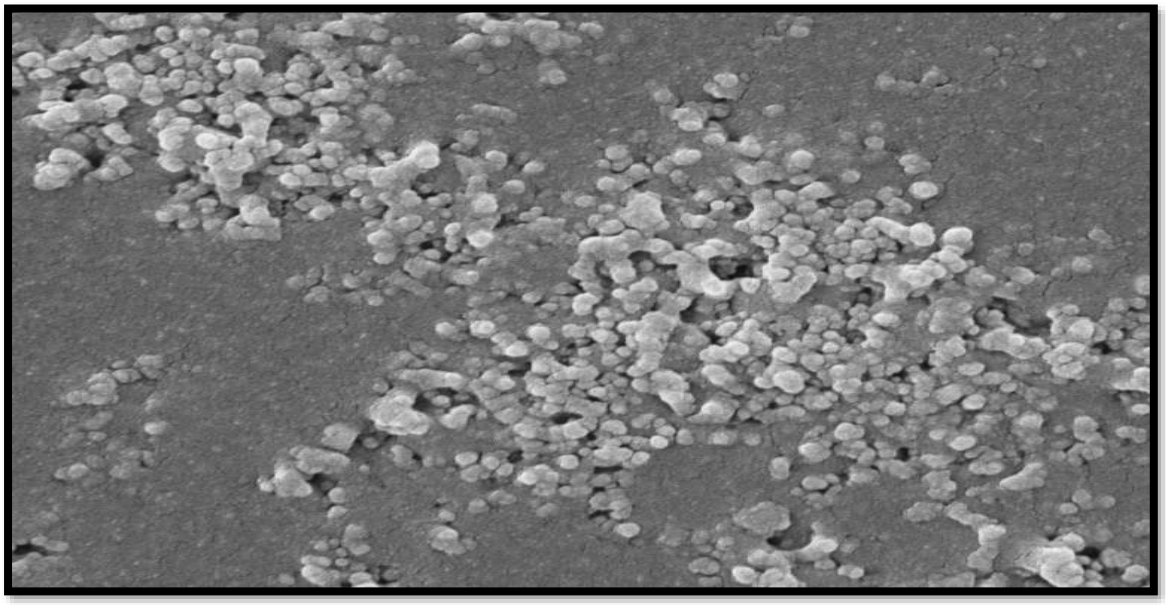

Figure 5: Scanning electron microscopic image of Cur-phospholipid complex

Conclusion

A drug's water solubility, drug permeability, and metabolism all have an impact on its oral bioavailability. Drug development is hampered by poor solubility, which calls for methods like nanosization to increase bioavailability. Thermal analysis confirmed the development of the Cur-phospholipid complex with a lower particle size and showed that curcumin is compatible with excipients. Compared to traditional solutions, the formulation demonstrated longer drug release and a higher drug encapsulation efficiency.

References

[1] Singh RP, Gangadharappa HV, Mruthunjaya K. Phospholipids: Unique carriers for drug delivery systems. J Drug Deliv Sci Technol. 2017;39:166-79.

[2] Rarokar N, Gurav S, Khedekar P. Meloxicam encapsulated nanostructured colloidal self-assembly for evaluating antitumor and anti-inflammatory efficacy in 3D printed scaffolds. J Biomed Materials Res Part A. 2021;109(8):1441-56.

[3] Saoji SD, Raut NA, Dhore PW, Borkar CD, Popielarczyk M, Dave VS. Preparation and evaluation of phospholipid-based complex of standardized centella extract (SCE) for the enhanced delivery of phytoconstituents. The AAPS J. 2016;18:102-14.

[4] Saoji SD, Dave VS, Dhore PW, Bobde YS, Mack C, Gupta D, et al. The role of phospholipid as a solubility-and permeability-enhancing excipient for the improved delivery of the bioactive phytoconstituents of Bacopa monnieri. Eur J Pharm Sci. 2017;108:23-35.

[5] Telange DR, Patil AT, Pethe AM, Fegade H, Anand S, Dave VS. Formulation and characterization of an apigenin-phospholipid phytosome (APLC) for improved solubility, in vivo bioavailability, and antioxidant potential. Eur J Pharm Sci. 2017;108:36-49.

[6] Telange DR, Nirgulkar SB, Umekar MJ, Patil AT, Pethe AM, Bali NR. Enhanced transdermal permeation and anti-inflammatory potential of phospholipids complex-loaded matrix film of umbelliferone: Formulation development, physico-chemical and functional characterization. Eur J Pharm Sci. 2019;131:23-38.

[7] Zhang Z, Lin Y, Liu F. Preparation and characterization of CdS/ZnS core-shell nanoparticles. J Disper Sci Technol. 2020;41(5):725-32.

[8] Chan JM, Zhang L, Yuet KP, Liao G, Rhee JW, Langer R, et al. PLGA–lecithin–PEG core–shell nanoparticles for controlled drug delivery. Biomaterials. 2009;30(8):1627-34.

[9] Fricker G, Kromp T, Wendel A, Blume A, Zirkel J, Rebmann H, et al. Phospholipids and lipid-based formulations in oral drug delivery. Pharm Res. 2010;27(8):1469-86.

[10] Hou Z, Li Y, Huang Y, Zhou C, Lin J, Wang Y, et al. Phytosomes loaded with mitomycin C–soybean phosphatidylcholine complex developed for drug delivery. Mol Pharm. 2013;10(1):90-101.

[11] Rajbhar K, Karodadeo GR, Kumar V, Barethiya V, Lahane A, Kale S, et al. Comparative assessment of solubility enhancement of itroconazole by solid dispersion and co-crystallization technique: Investigation of simultaneous effect of media composition on drug dissolution. Ann Pharm Fr. 2023;81(5):843-855).

[12] Preethi V, Ramesh ST, Gandhimathi R, Nidheesh PV. Optimization of batch electrocoagulation process using Box-Behnken experimental design for the treatment of crude vegetable oil refinery wastewater. J Disper Sci Technol. 2019;41(4): 592-599.

[13] Rarokar NR, Khedekar PB, Bharne AP, Umekar MJ. Development of self-assembled nanocarriers to enhance

antitumor efficacy of docetaxel trihydrate in MDA-MB-231 cell line. Int J Biol Macromol. 2019;125:1056-68.
[14] Saoji SD, Rarokar NR, Dhore PW, Dube SO, Gurav NS, Gurav SS, et al. Phospholipid based colloidal nanocarriers for enhanced solubility and therapeutic efficacy of withanolides. J Drug Deliv Sci Technol. 2022;70:103251.
[15] Rarokar N, Agrawal R, Yadav S, Khedekar P, Ravikumar C, Telange D, et al. Pteroyl-γ-l-glutamate/Pluronic® F68 modified polymeric micelles loaded with docetaxel for targeted delivery and reduced toxicity. J Mol Liq. 2023;369:120842.
[16] Rodrigues K, Nadaf S, Rarokar N, Gurav N, Jagtap P, Mali P, et al. QBD approach for the development of hesperetin loaded colloidal nanosponges for sustained delivery: In-vitro, ex-vivo, and in-vivo assessment. OpenNano. 2022;7:100045.
[17] Paruchuri VK, Nguyen AV, Miller JD. Zeta-potentials of self-assembled surface micelles of ionic surfactants adsorbed at hydrophobic graphite surfaces. Colloids Surf A: Physicochem Eng Asp. 2004;250(1-3):519-26.
[18] Zeng X, Tao W, Mei L, Huang L, Tan C, Feng SS, et al. Cholic acid-functionalized nanoparticles of star-shaped PLGA-vitamin E TPGS copolymer for docetaxel delivery to cervical cancer. Biomaterials. 2013;34(25):6058-67.
[19] Feng S, Zhang H, Xu S, Zhi C, Nakanishi H, Gao XD. Folate-conjugated, mesoporous silica functionalized boron nitride nanospheres for targeted delivery of doxorubicin. Mater Sci Eng C. 2019;96:552-60.
[20] Wang H, Li Q, Reyes S, Zhang J, Xie L, Melendez V, et al. Formulation and particle size reduction improve bioavailability of poorly water-soluble compounds with antimalarial activity. Malar Res Treat. 2013;1:769234.
[21] Singh AK, Singh A. Preparation, Characterization and In Vitro Antioxidant Potential of Boldine-phospholipid Complex. Current Drug Therapy. 2021;16(1):89-99.
[22] Pandya R, Mehta T, Gohel M. Amalgamation of solid dispersion and adsorption technique: A case study of ritonavir. J Therm Anal Calorim. 2015;120:699-709.