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Preparation and *in vitro* evaluation itraconazole SD using hot melt and solvent evaporation method

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ABSTRACT:

Infections of the fingernails and toenails caused by fungi are frequently treated with itraconazole (ITR). ITR has a poor aqueous solubility and may exhibit restricted absorption upon dissolution. Polyethele glycol (PEG 5000), gelucire (GLR 44/14), and β-cyclodextrin (βCD) were the polymers used to prepare the solid dispersion (SD) of ITR by physical mixing (PM), hot melt and solvent evaporation method, which were 1:1, 1:3, and 1:5 respectively. The physical characteristics, solubility, *in vitro* dissolution tests, and Fourier Transform Infrared (FT-IR) Spectroscopy of the SD were all studies. All formulations were determined to have a high and evenly distributed ITR content. The USP paddle (type II) dissolving apparatus was used for the *in vitro* dissolution investigations. The prepared SD exhibited a significantly higher ITR dissolving rate than the pure ITR. While comparing the aqueous solubility (0.9745 ± 0.029), drug content (94.55 ± 2.89), and dissolution rate (96.17 ± 3.89 %) of GLR 44/14 (1:5) by fusion technique to other dispersions prepared by physical mixing (PM) and fusion method using PEG 5000 and βCD (1:3 and 1:5), the latter revealed a slower rate of dissolution and the out of the three carriers, the ITR dissolved more readily in GLR 44/14 based SDs. It was concluded that the SD may improve the ITR's dissolution and that SDs based on PEG6000 were more successful in doing so.

Keywords: Drug release, Itraconazole, Drug potency, Drug carriers, Gelucire 44/14, β-cyclodextrain, Polymer, Solid dispersion

INTRODUCTION

Improving the oral bioavailability of poorly soluble drugs continues to be one of the most difficult elements of itraconazole (ITR) development. The drugs solubility behaviour, in addition to its permeability, is a key factor in determining its oral bioavailability. There have always been some ITR whose solubility has made it difficult to prepare an oral formulation that works well [1]. One of the most common and significant issues facing formulation scientists in the pharmaceutical business today is the formulation of poorly soluble chemicals for oral delivery.

ITR has several formulation challenges primarily due to its poor water solubility and variable bioavailability. Key formulation issues include the poorly soluble in water, which complicates its formulation into oral and injectable forms. Poor solubility can lead to inconsistent absorption and therapeutic levels in the body [2]. The bioavailability of oral ITR can be highly variable that the capsule form, for example, requires an acidic environment for optimal absorption, which can be problematic for patients taking acid-reducing drugs.

ITR has a complex pharmacokinetic profile with a long half-life, necessitating careful dosing to avoid toxicity. Its absorption can be affected by food intake, and there is significant variability among patients. ITR is a strong inhibitor of the cytochrome P450 3A4 (CYP3A4)

enzyme, leading to numerous potential drug interactions [3]. This requires careful management and monitoring when ITZ is co-administered with other drugs.

However, improving the solubility through formulation techniques is the most alluring alternative for speeding up the release rate. There are practical limitations to the final dosage form production, solubilization, and particle size reduction methods that are frequently employed to speed up the dissolve rate and, in turn, the oral absorption and bioavailability of low water-soluble ITRs [4]. A workable approach that allows for the removal of many of the restrictions associated with improving the bioavailability of poorly water-soluble ITRs. This process, subsequently named "solid dispersion (SD)," required melting the physical mixes of the ITRs to prepare a eutectic combination with water-soluble carriers [5].

MATERIALS AND METHODS

Itraconazole (ITR) was a gift sample obtained from Lifecare, Chandigarh, India. Poly ethelene glycol (PEG 5000), Sodium hydroxide (NaOH), Potassium di□hydrogen phosphate was purchased from Sigma - Aldrich, Bangalore, India. Beta-cyclodextrin (βCD) and gelucire 44/14 ware of gift samples from BASF Corporation, Mumbai, India; All the chemicals and regents used were of analytical grade.

Preparation of physical mixture (PM)

The desired drug/carrier ratio (as indicated in table 1) was prepared by slightly grinding drug ITR and carriers (PEG 5000, GLR 44/14, and βCD) in a mortar for two minutes to prepare the drug and carrier PM. Subsequently, the powder was run through sieve number 80. The final product was kept in a desiccator so that additional analysis could be performed [6].

Preparation of SD

Melt Method

PEG 5000 and GLR 44/14, the precisely weighed amounts of carriers, were melted in a water bath to prepare SDs, and the ITR was then distributed in the molten solution. The process of fusion was employed to prepare the SDs. A appropriate amount of ITR was placed in a china dish, and the necessary amounts of carriers (PEG 4000 and PEG 6000) were added to generate the necessary drug to carrier ratio for the formulations indicated in Table 1. After that, the mixture was heated to a predetermined temperature while being constantly stirred to melt the ITR and carrier. After cooling in an ice bath, the melted mixture was poured to porcelain tile to solidify [6]. The made SDs were put in a desiccator after being ground up and sieved (80#).

Solvent evaporation method

After the ITR was dissolved in the ethanol (90%), the carrier (βCD) was distributed and agitated using a magnetic stirrer (REMI-2MLH) at 38 ± 0.5 °C. Transferring the same solution to a petri dish and allowing it to evaporate at 50 ± 0.5 °C is necessary to extract the solvent with a hot plate. The final product was dried for twenty-four hours at 40 °C (Yadav et al. 2009).

Drug uniformity

Using a SD of 100 mg equivalent of ITR in pH 5.8 phosphate buffer as a solvent, the drug content uniformity was evaluated. At 256 nm, the estimation was carried out in a UV/visible spectrophotometer [7].

The following formula was used to get the yield percentage in relation to the initial amount.

$$\% \text{ Yield } = \frac{\text{Mass of the SD}}{\text{Total mass of the drug and carrier}} \text{ X } 100$$

Evaluation of SDs

Aqueous solubility study

Conical flasks holding 10 ml of distilled water were filled with the excess amount of the formulations (PMs and SDs), and they were shaken on a rotary shaker for 48 hours at 37°C. The flasks were then taken out and filtered. After the proper dilution with distilled water,

suitable aliquots were removed from the filtered solution and examined for drug content, comparing with pure drug solubility [8].

Fourier Transform Infrared (FT-IR) Spectroscopy

To evaluate a potential interaction (difference in structure) between the drug and carriers, FT-IR analysis was conducted. Using an FT-IR spectrophotometer (Perkin Elmer FTIR spectrometer, spectrum 1000 Germany), the IR spectrum of the solid samples was examined in the solid powder using the KBr disc method in the wavenumber range of 4000–400 cm^{-1} at a scan speed of 1 cms^{-1}.

***In vitro* release studies**

A precise weighted quantity of the material was extracted for the dissolution investigations. At predetermined intervals, aliquots of the material were taken out and their drug release was assessed by measuring the absorbance at 256 nm in a dissolving media of double distilled water. The identical amount of new medium (double distilled water) was used to replace the volume that was removed at each time interval [8].

Stability study

The ITR-SD complex physicochemical stability was examined for three months at 45 ± 0.5 oC and 60 ± 5% RH in a stability compartment (Wadegati TM Labe Quip (P) Ltd., Model No. HTC-3003, Andheri (E), Mumbai, India). At one-month intervals, ITR-SD investigated dissolution, drug content, and physical changes [8].

Statistical Analysis

Every preparation for an SDs formulation and every analysis was done twice. Analysis of variance (ANOVA: single factor) was used to determine the significance of the effect of formulation variables release parameters (t50% and t80%) with the help of Origin Pro 2023. When $p < 0.05$, the difference was deemed significant.

RESULTS AND DISCUSSION

Saturation solubility

The ITR has an inherent solubility of 0.0001±0.0003 mg/mL in double distilled water at ambient temperature, it is nearly insoluble in water (Table 1). The carrier GLR 44/14 comprising PM and SD had the highest saturation solubility among PMs and SDs (1:5). This could be because different carriers have distinct properties that affect how well they hydrate, dissolve, and maybe compound with different drugs [9].

Table 1. Aqueous solubility of ITR-PM and SD complexes

Aqueous solubility (mg/mL)				
Pure CMN at 37 °C			**0.001 ± 0.0003**	
D:C ratio*	**PM**	**1:1**	**1:3**	**1:5**
CMN-PM complex				
GLR 44/14	0.236 ± 0.032	0.7644 ± 0.087	0.7987 ± 0.033	0.8712 ± 0.061
GLR 44/14	0.2855 ± 0.071	0.8298 ± 0.088	0.8920 ± 0.012	0.9745 ± 0.029
β-CD	0.1927 ± 0.049	0.2988 ± 0.051	0.3582 ± 0.047	0.3976 ± 0.011

FT-IR outcome

The –NH group of ITR and the –OH group of GLR 44/14 may form intermolecular hydrogen bonds as a result of these notable alterations. Significant alterations were seen in the comparison spectra of PM-GLR and SD-GLR with ITR and carrier. Nonetheless, minor variations in the stretching vibration caused by the -CH_2 groups of GLR were seen at two distinct wave numbers, 2880.1 and 2869.8 cm^{-1}, respectively, indicating a potential variation in the extent of drug-carrier interaction in PM and SD. The ternary amide peak of PEG 5000

(at 1639.4 cm^{-1}) or the peak of the –NH group of ITR (at 3320.7 cm^{-1}) may show a drop in intensity, suggesting that intermolecular hydrogen bonding between the drug and carrier occurs in both PMs and SDs [10]. The quantity of chemicals may also be the cause of a drop in band intensity. Thus, the absence of interaction between two chemicals by FTIR might be concluded with some scepticism.

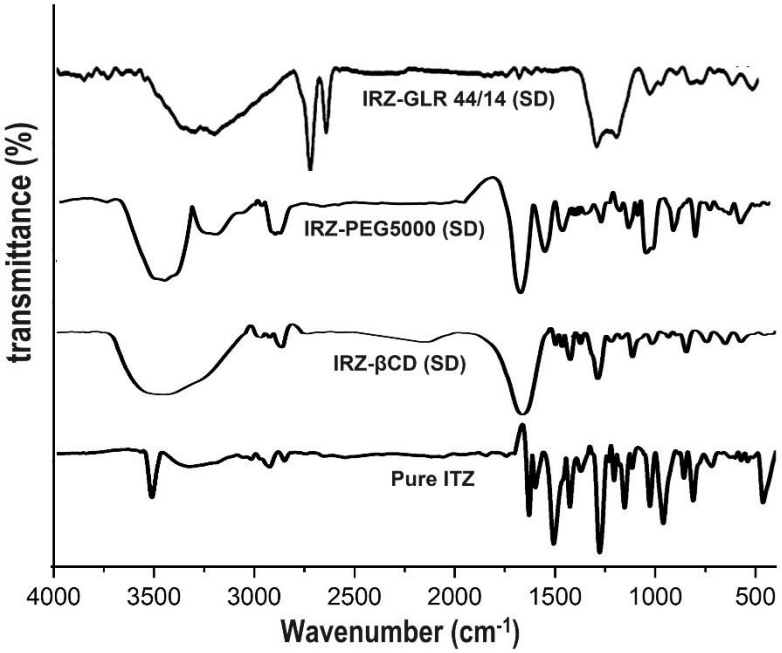

Figure 1. FT-IR spectrum of pure ITR, SDs with the carriers of βCD, PEG 5000 and GLR 44/14.

Drug content analysis

The percentage of drugs discovered ranges from 94.55 ± 2.89 to 98.10 ± 5.142 % from 1: to 1:5. Low standard deviations of the results and a high drug content were present in all of the PMs and SDs. The drug shown to be evenly distributed throughout the powder composition with all the carriers. As a result, it seems that the procedure utilised in this work to prepare SDs can be repeated.

The percentage yield of the all the carriers except βCD shown as reduced yield due to the sticky nature with the preparing vessels. In conclusion, drug content uniformity and percentage yield shown significant for all the preparation as per the result revealed by Mohamed et al (2021) [11].

Table 2. Various carriers ITR-SD of drug content and % yield

Drug content (%)			
D:C ratio*	**1:1**	**1:3**	**1:5**
GLR	98.10 ± 5.142	97.43 ± 6.213	94.55 ± 2.89
PEG 5000	98.16 ± 4.411	97.89 ± 4.441	97.11 ± 4.457
β-CD	98.33 ± 5.235	98.21 ± 3.543	97.16 ± 4.541
Total yield (%)			
GLR	87.34± 7.317	88.40 ± 7.478	90.99 ± 6.985
PEG 5000	89.34 ± 6.181	90.10 ± 5.479	91.90 ± 5.412
β-CD	90.91 ± 4.878	92.17 ± 3.450	96.58 ± 3.681

Every PM and SD exhibits a high drug content (> 95%) and ITR dissolved more readily in the PMs than in pure ITR. The ITR solubility was greatly enhanced by the formulations of ITR in SDs. When compared to PEG 5000 and βCD, the SDs of ITR using the same amount of PEG 5000 as a carrier were better at dissolving ITR. As the quantity of carrier increased from 1:1 to 1:5, the ITR dissolution from the SDs of PEG 5000 increased.

***In vitro* dissolution study**

To choose the best carriers, the SD of ITR was prepared with a variety of carriers, including GLR, PEG 5000, and βCD. The fact that these carriers underwent no chemical changes in the process of preparing SD was considered encouraging. In pH 5.8 phosphate buffer, the SD of ITR with carriers PEG 5000, GLR 44/14, and βCD demonstrated a significant increase in the dissolution rate. The dissolution of ITR increased as the fraction of carriers, and the T50% and T80% values were lowest when the GLR-SDs (i.e., 1:1–1:5) were considered [12]. The ratio of ITR:GLR 44/14 demonstrated a maximum of 1:5, surpassing all other formulations. This indicates that an enhanced concentration of matrix generated with PEG 5000 at a ratio of 1:5 boosted the dissolution rate (Figure 2). These findings show that the dissolution of SDs is improved when the concentration of carriers increases. This could be because the carrier increases the drug's wet-ability, the drug particle size is reduced during the preparation of the SD, the drug crystals undergo polymorphic transformation, and there are chemical interactions between the drug and carrier. Overall the ITR:carriers of all the PM and SD increases as the amount of carriers increases.

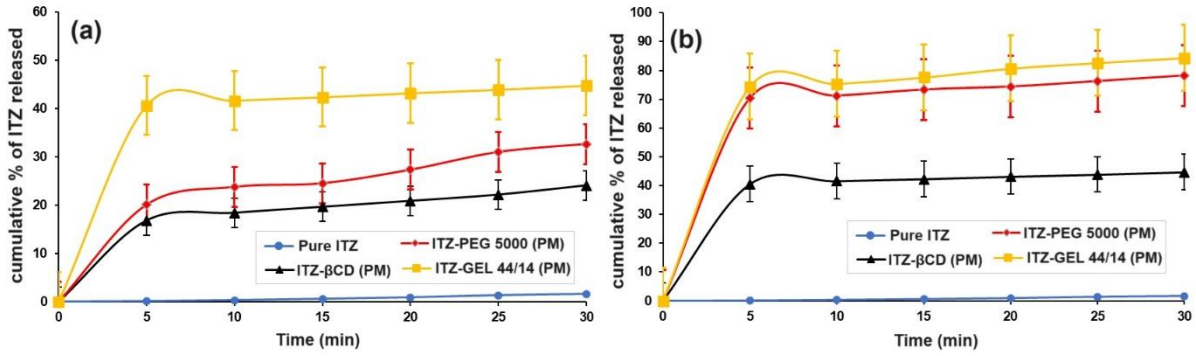

Figure 2. Cumulative percentage of ITR released form pure ITR, GLR, PEG 5000, and βCD (a) PM and (b) SD

The end result indicates that SDs have the potential to promote ITR dissolution, and that SDs based on GLR 44/14 are more successful in doing so. The many techniques used to prepare SD was given in the introduction. The dissolution of a drug that is poorly or sparingly soluble depends on its bioavailability and the method utilised for particle size reduction [13].

The drug concentration was best estimated using the spectrophotometric approach, which was also best suited for studying the dissolution of different SDs of ITR. It was used all the way through the inquiry. The formulation of ITR's SD using different carriers, such as GLR, PEG 5000, and BCD, was evaluated to see which carriers would work best. These carriers were thought to be promising because they did not experience any chemical changes while the SD was being prepared. The dissolution apparatus outlined in the USP Paddle II was utilised to prepare SDs of ITR in pH 5.8 phosphate buffer. It was determined that the dissolving studies findings were satisfactory. This shown that there was a significant increase in the rate of ITR dissolution in pH 5.8 phosphate buffer when the SD of ITR with carriers PEG 5000, GLR 44/14, and BCD was used [14]. The ratio of ITR: GLR 44/14 demonstrated a maximum of 1:5, surpassing all other formulations. This indicates that a larger concentration of matrix generated with a ratio of 1:5 enhanced the dissolution rate. The development of SD of ITR with carriers,

which leads to size reduction, is responsible for the enhancement in the *in vitro* characteristics. Apart from the crystalline substance's size reduction, the drug's rapid dissolution rate could be attributed to its exceptional wettability and dispersibility in a SD system made using water soluble carriers. A comparative analysis of the in-vitro dissolution profiles of various ITR ratios and carriers was conducted.

Stability studies

Stability tests were conducted for three months at room temperature on three formulations (SD-GLR, SD-PEG 5000, and βCD) that demonstrated encouraging outcomes. The physical characteristics and pharmacological content of SDs were unchanged after three months. This suggests that the ITR remained stable in SDs even following three months of short-term storage. Initially, solubility tests were carried out to examine solubility of ITR in various solvents and buffers [15]. The PM and SD of ITR with various carriers (GLR, PEG 5000, and βCD) were prepared using a solvent method as part of the formulation studies. Overall, the stability tests conducted for three months at room temperature on a few chosen formulations revealed no alterations in the drug content or physical characteristics.

CONCLUSION

In this study, itraconazole physical mixture and solid dispersions were prepared and evaluated using Gelucire 44/14, PEG 5000, and β-cyclodextrin as carriers. The Fourier Transform Infrared Spectroscopy (FTIR) analysis confirmed the compatibility and absence of significant chemical interactions between itraconazole and the carriers. Aqueous solubility studies demonstrated that Gelucire 44/14 significantly enhanced the solubility of itraconazole compared to PEG 5000 and β-cyclodextrin. Furthermore, dissolution studies indicated that itraconazole solid dispersions with Gelucire 44/14 exhibited superior dissolution profiles, leading to improved drug release rates. Therefore, among the three carriers studied, Gelucire 44/14 emerged as the most effective carrier for enhancing the solubility and dissolution of itraconazole, making it the best choice for formulation development.

Acknowledgment

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