INTRODUCTION

Sulfasalazine is a sulfa drug. It is a derivative of mesalazine. It is formed by combination of sulfapyridine with salicylate by an azo bond. It can be abbreviated by SSZ. Sulfasalazine and its metabolite 5-aminosalicylic acid (5-ASA) are poorly absorbed from gut so its main mode of action is believed to be inside the intestine. It exhibits slow GI absorption rate and inter individual variation of its bioavailability. Thus solubility enhancement and dissolution enhancement of sulfasalazine from its dosage form is an important issue for its in vivo bioavailability and therapeutic efficacy. It was planned to improve the solubility of sulfasalazine by using polymer like Gelucire50/13. Different ratio were employed as 1:1, 1:3, 1:5 and Solid dispersion were prepared by Solvent evaporation method, Physical mixture and Kneading method. Preformulation study was done before going to formulation in that melting point, solubility and compatibility study was done. The prepared solid dispersion also evaluated for percentage yield, percent drug content. Solubility study was done in water. Solid dispersion also characterized by FTIR, DSC, PXRD. In-vitro dissolution study was done in pH6.8 phosphate buffer using USP dissolution test apparatus type II.

KEYWORDS: Solid dispersion, Sulfasalazine, Gelucire 50/13, BCS Class 2 drug.

Fig 1: Structure of sulfasalazine.
Mechanism of solubilization
Step-I Holes opens in the solvent

Step-II Molecules of the solid breaks away from the bulk.

Step-III The freed solid molecule is integrated into the hole.

Fig no 2: Mechanism of Solubilisation

MATERIALS AND METHODS
Sulfasalazine was provided by Glenmark Pharmaceutical Ltd, nashik, India and gelucire 50/13 from Glenmark Pharmaceutical Ltd., Sinnar, India as a gift sample. All other chemicals and reagents were used of analytical grade.

Phase Solubility Study
Solubility measurements were performed according to Higuchi and Connors method. An excess amount of the drug was added to 10 ml volumetric flask containing 1%, 3%, 5%, 7%, 9% aqueous solution of gelucire50/13. The samples were shaken for 48 hours at room temperature 25±1°C on an orbital shaker incubator. After 48 hours of shaking to achieve equilibrium, 5 ml of aliquots were withdrawn after 1 hour and filtered immediately using membrane filter (0.45 μ). The filtered samples were diluted suitably and assayed for absorbance at 359 nm using UV/Visible spectrophotometer (Shimadzu 1800). Solubility studies of physical mixtures, solid dispersion and Kneading method were also performed in same manner. Three determinations were carried out for each sample to calculate the solubility of sulfasalazine.

Preparation of Solid dispersion
The solid dispersion was prepared by using Gelucire50/13 as a hydrophilic carrier. Different drug and polymer ratios were employed as 1:1, 1:3, 1:5 and prepared by Solvent evaporation method, Physical mixture method and Kneading method.

a) Solvent evaporation method
Solid dispersions were prepared using a SE method. Drug & polymers (Gelucire 50/13) were dissolved in methanol in different ratio & the solutions were made homogeneous by continuous stirring and solvent was evaporated by subjecting the solution with constant stirring at 70 to 80°C till complete evaporation of solvent. The obtained SD’s were dried and subsequently pulverized by triturating in pestle mortar and screened through 60 mesh sieve and stored in desiccator till further evaluation.

b) Physical Mixture method
Solid dispersions were prepared using a PM method. Drug & polymers (Gelucre50/13) were weighed and transferred to mortar and mix it using pestle. The Physical mixture were sieve through 60 mesh and stored in desiccator till further evaluation.

c) Kneading Method
A mixture of accurately weighed drug and carrier is wetted with solvent and kneaded thoroughly for some time in a glass mortar. The paste formed is dried and sieved and stored in desiccator till further evaluation.

Characterization of Prepared solid dispersion
1. Drug Content
About 10 mg drug equivalent of SD was weighed accurately and transferred to 100 ml volumetric flask. From this stock solution (100 μg/ml), 1 ml was withdrawn and further diluted up to 10 ml with PBS pH(6.8). This solution was used for the assay for drug content by UV spectrophotometer at 359 nm. Concentration of drug in stock solution was calculated by using calibration curve and from which percent drug content was calculated.

\[
\text{% Drug Content} = \frac{W_a}{W_t} \times 100 \quad \text{(2)}
\]

Where,
Wa = Actual drug content,
Wt = Theoretical drug content.

2. Fourier Transform-Infra Red Spectroscopy
FT-IR spectra of plain SSZ and solid dispersions with carrier by using Fourier transform infrared spectrophotometer. Solid dispersion were then scanned using FT-IR spectra of mixture were compared with that of plain drug for change or shift in any principle peak of spectra of plain drug.

3. Production Yield
The production yield of solid dispersion was calculated using the weight of final product after drying with respect to the initial total weight of the drug and carrier used for the preparation of solid dispersion. Percent production yield were calculated as per the formula mentioned below,

\[ \text{Py} = \frac{W_0}{W_t} \times 100 \]  

Where,
Py = Product yield,
W0 = Practical mass (solid dispersion)
Wt = Theoretical mass (carrier + drug)

4. Solubility Studies
The solubility of Sulfasalazine, Physical Mixture and Solid dispersion was determined in distilled water. The solubility of drug and solid dispersion were determined by taking 10 mg drug, equivalent quantity of solid dispersion and added them in 20 ml of distilled water, in 25 ml vials. The samples were kept at equilibrium for a period of 72 hrs. in incubator at 37± 0.5°C with occasional shaking. The supernatant collected from vials was filtered through Wattman filter paper and analyzed by UV- Visible Spectrophotometer at wavelength of 359 nm. All experiments were conducted in triplicate (n=3) and tabulated.

5. Powder X-Ray Diffraction
PXRD analysis was done by irradiating the samples with monochromatized Cu Kα radiation (1.506 Å) and analyzed between 3° and 60° (2θ) employing a Bruker AXS D8 Advance Diffractometer with Lynx Eye Detector. The step was at rate of 0.020° with step time of 32.8 sec. The diffractogram was produced by using Diffrac plus Software.

6. Differential Scanning Calorimetry
The powdered sample (3 mg) was hermetically sealed in aluminum pans and heated at a constant rate of 10 °C/min, over a temperature range of 50–300°C with nitrogen low rate of 30 ml/min. Thermograms of the samples were obtained using differential scanning calorimetry (DSC-60, Shimadzu, Japan). Thermal analysis data were recorded with Shimadzu software programs. Indium standard was used to calibrate the DSC temperature and enthalpy scale.

7. Dissolution Studies of Solid Dispersion
The in vitro dissolution study of SD was carried in USP Apparatus 2. Samples equivalent to 500 mg of SSZ was hold in muslin cloth and then added to 900 ml of phosphate buffer pH 6.8 at 37 ± 0.5° C and stirred at 50 rpm.5 ml aliquots were withdrawn at time interval of 5, 15, 30, 45, 60 min and filtered through Whatman’s (No. 41) filter paper. An equal volume of fresh dissolution medium was replaced to maintain the volume of dissolution medium. The filtered samples were analyzed spectrophotometrically at 359 nm. Cumulative percentage of labeled amount of drug released was calculated.

RESULT AND DISCUSSION
Phase solubility study
The solubility of sulfasalazine in distilled water was found to be 0.034 mg/mL. The increase in solubility was linear (r² = 0.964) with respect to the increase weight fraction of the Gelucire50/13 indicating the solvent properties of Gelucire50/13 for the drug, giving AL type solubility diagrams (Fig: 2). At 3% concentration of Gelucire50/13, the increase in solubility was around 24 fold compared with pure drug. This increase in solubility of sulfasalazine can be probably explained due to the wettability of sulfasalazine in the presence of Gelucire50/13. Recently, the increased effect of Gelucire was observed for the solubility.

![Phase Solubility Studies](image)

Fig 3: Phase Solubility Studies.

Drug Content
The selected ratios were subjected to the determination of drug content. The drug content in the selected ratios were found to be in range of 85 to 97%. Almost negligible loss of drug may have occurred probably because preparation of combinations was confined to the very small area of mortar. The results of drug content (%) are shown in table 1.
Table No 1: Percent Drug content

<table>
<thead>
<tr>
<th>Sr. No.</th>
<th>Formulation Code</th>
<th>Drug</th>
<th>Polymer</th>
<th>Ratio</th>
<th>Drug Content (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>SE 1</td>
<td>SSZ</td>
<td>Gelucire50/13</td>
<td>1:1</td>
<td>88.25±0.039</td>
</tr>
<tr>
<td>2.</td>
<td>SE 2</td>
<td>SSZ</td>
<td>Gelucire50/13</td>
<td>1:3</td>
<td>99.06±0.048</td>
</tr>
<tr>
<td>3.</td>
<td>SE 3</td>
<td>SSZ</td>
<td>Gelucire50/13</td>
<td>1:5</td>
<td>91.01±0.221</td>
</tr>
<tr>
<td>4.</td>
<td>PM 2</td>
<td>SSZ</td>
<td>Gelucire50/13</td>
<td>1:3</td>
<td>93.16±0.048</td>
</tr>
<tr>
<td>5.</td>
<td>KM 2</td>
<td>SSZ</td>
<td>Gelucire50/13</td>
<td>1:3</td>
<td>93.41±0.23</td>
</tr>
</tbody>
</table>

Production Yield
The production yield of solid dispersion prepared by solvent evaporation method was found to be 85%. Any loss in yield can be attributed to the product remaining adhered to the walls of the mortar which could not be retrieved. The production yield of PM and Kneading method given below.

Table no 2: Determination of Production Yield.

<table>
<thead>
<tr>
<th>Sr.no</th>
<th>Formulation code</th>
<th>Ratio</th>
<th>Percentage yield</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>SE 1</td>
<td>1:1</td>
<td>73 %</td>
</tr>
<tr>
<td>2.</td>
<td>SE 2</td>
<td>1:3</td>
<td>84 %</td>
</tr>
<tr>
<td>3.</td>
<td>SE 3</td>
<td>1:5</td>
<td>86 %</td>
</tr>
<tr>
<td>5.</td>
<td>PM 2</td>
<td>1:3</td>
<td>86 %</td>
</tr>
<tr>
<td>8.</td>
<td>KM 2</td>
<td>1:3</td>
<td>85 %</td>
</tr>
</tbody>
</table>

FT-IR Study
The characteristic FTIR spectra of Sulfasalazine, Gelucire50/13, SE2, PM 2, KM 2 were shown in Fig. 3. Sulfasalazine alone showed characteristic bands belonging to SO2 NH at (1124), C=O at 1674 cm⁻¹. Gelucire 50/13 showed peaks at 2818 cm⁻¹ (-O-H stretching). The characteristic peaks of sulfasalazine and Gelucire 50/13 were observed in both solid dispersion and its corresponding physical mixture. This suggests that there is no significant interaction between the drug and carrier.

Differential Scanning Calorimetry
Drug-polymer specific interactions are thought to be of particular importance and needed to be analyzed by using Differential Scanning Calorimetry. Differential scanning calorimetry studies were carried out in order to evaluate ability of the polymer to stabilize amorphous form of drug in SD. DSC thermograph of SSZ is shown in figure 4 which shows melting endotherm at 238.85°C i.e. melting point and amorphous state of drug. DSC thermograph of SSZ:Gelucire50/13 (SE) is shown in figure 4 indicating formation of stable crystalline SD investigated by decline in melting endotherm of Gelucire50/13 from 50.41°C to 63.27°C and increase in melting endotherm of SSZ from 220.05°C to 240.0°C.
PXRD Studies
The SD was studied for prediction of crystallinity. The PXRD Pattern of SSZ is shown in Figure 5. Based on the diffractogram it can be suggested that SSZ is present in its amorphous form since it exhibits several well defined peaks at a diffractogram angle of 20. The strong peak at 20 of 21.39 was highly intense peak with 100% intensity indicating presence of amorphous SSZ.

SSZ: Gelucire50/13 (SE) Figure 5, XRD diffraction pattern revealed that the functional peak of SSZ was of low intensity and showing characteristic peak of Gelucire50/13 in the solid dispersion at 20 of 22.628 indicating presence of SSZ in crystalline state within the gelucire.

Dissolution Study
The in vitro dissolution study of SD was carried in USP Apparatus 2. Samples equivalent to 500 mg of SSZ was hold in muslin cloth and then added to 900 ml of phosphate buffer pH 6.8 at 37 ± 0.5°C and stirred at 50 rpm. 5 ml aliquots were withdrawn at time interval of 5, 15, 30, 45, 60 min and filtered through Whatman’s (No. 41) filter paper. An equal volume of fresh dissolution medium was replaced to maintain the volume of dissolution medium. The filtered samples were analyzed spectrophotometrically at 359 nm. Cumulative percentage of labeled amount of drug released was calculated. The in-vitro dissolution profiles of the drug and solid dispersion are shown in (Table 3) &Figure 6. Drug exhibited a slow dissolution, suggesting that its absorption would be dissolution rate limited, whereas solid dispersion showed a marked enhancement in dissolution rate. Thus, dissolution up to 99.22% was recorded with solid dispersion in 60 min.
CONCLUSION
The results of this study were able to conclude that the formulation of Sulfasalazine solid dispersions markedly improves the solubility and dissolution characteristics. At 3% concentration of Gelucire50/13 the increase in solubility was around 24 fold compared with pure drug. FTIR spectroscopy studies showed no well-defined interaction between Sulfasalazine and Gelucire 50/13. The DSC thermograms of SDs of carvedilol with Gelucire 50/13 showed the transformation of Sulfasalazine from a crystalline to an amorphous state, this was supported by XRD studies. Gelucire50/13 is good carrier for solubility and dissolution enhancement.

ACKNOWLEDGMENT
The authors are acknowledge to Glenmark Pharmaceutical Ltd, nashik, India for providing Sulfasalazine & gelucire50/13. We delighted to say thank you to Prof. Dr. R. B. Saudagar, Principal and also management of KCT’S R. G. Sapkal College of Pharmacy, Anjaneri, Nashik for support and providing necessary facilities to carry out the research work successfully.

REFERENCES