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Original Article

# QUICK/SLOW BIPHASIC RELEASE OF A POORLY WATER SOLUBLE ANTIDIABETIC DRUG FROM BI-LAYER TABLETS

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**ABSTRACT**

**Objective:** The objective of the present work is to develop a bi-layer tablet consisting of an Immediate Pulse Release [IPR] layer and a sustained release [SR] layer that can produce a distinct biphasic release having two different drug release rates. The IPR layer is intended to release a fraction of the dose rapidly at a faster rate and the SR layer is meant for slow release of the remaining dose at a slower rate for a desired period of time.

**Methods:** The quantitative determination was carried out by UV spectrophotometer. Solid dispersion was prepared by melt method. IPR layer was prepared by direct compression method, SR layer was prepared by wet granulation method. *In-vitro* drug release study from tablets was carried out in USP II tablet dissolution rate test apparatus. FTIR, DSC, XRD studies were performed.

**Results:** 89% of the incorporated drug was released within 30 min in acid solution of pH 1.2 from the IPR tablet prepared with the highest amount of gelucire due to solid state transformation of the drug. The SR layer (SR8) comprising of SAL, CG and CMC produced prolonged drug release (70% in 10 h). The optimized IPR3 layer and SR layers were compressed to form bi-layer tablets from which 23-37% drug was released immediately in 30 min, and the remaining drug was released slowly for 7 to 10 h depending upon the compositions of the tablets.

**Conclusion:** This study revealed that bi-phasic release of GPZ consisting of an initial quick release and subsequent slow release could be achieved by formulating bi-layer tablets using the existing tablet technology, and such formulation may be able to control hyperglycaemia effectively for a longer period of time

**Keywords:** IPR, SR, Glipizide, Solid Dispersion, Normoglycaemia Bilayer Tablets, Biphasic release.

## INTRODUCTION

During the last several years' development of modified release drug delivery system (DDS) remains on the thrust areas of research in pharmaceutical product development. Oral SRDDS have been developed to provide release of a drug at a constant rate for better compliance and therapy to the patients who need long term and continuous therapy. Oral site specific DDS, also known as slow/quick DDS, have been developed to minimize the drug release in upper gastrointestinal tract and to provide rapid release in the colon for local therapeutic effect [1]. However, for the treatment of many diseases, an ideal dosage regimen require an immediate attainment of therapeutic drug concentration at the site of action, and then to maintain the level for the desired period of time/treatment. This type of dosage forms is necessary to arrest the symptoms and to achieve maximum relief immediately, and then to provide sustained drug release for avoiding repeated administration. SR dosage forms generally lead to delayed attainment of therapeutic plasma drug concentration and are unable to provide a rapid disposition of the dose immediately after administration [2]. Such drug delivery systems can be modified by applying barrier layers to one or both sides of the tablets to restrict the surface area exposed to a fluid [3, 4]. However, many of the multilayer systems provide a constant release rate of drugs from tablets instead of a biphasic release [5]. Thus, with a view to achieve an immediate relief of symptoms associated with a disease and then to maintain the relieved condition for a longer period of time, the concept of biphasic DDS have emerged. This delivery system is capable of releasing a drug at two different rates, i.e. in a quick/slow manner depending on the need of the patients.

Diabetes mellitus is a chronic disease which is characterized by high blood glucose level due to either insufficient secretion of insulin from the pancreas or ineffective utilization of glucose by the body [6, 7]. Sustained hyperglycaemia may lead to several complications and organ failure [8]. The rationality in diabetes treatment is to achieve normoglycaemia/euglycaemia quickly, and then to maintain the normoglycaemic condition over a desired period of time [9]. For such therapy, a DDS should be designed in such a way that a precalculated dose of the drug is released rapidly to produce normoglycaemia, and the remaining dose is released slowly sufficient to maintain the normoglycaemia over a desired period of time.

Formulation of tablets using the existing and well established tablet technology appears to be very suitable for achieving biphasic drug release. In recent years, various researchers have reported the development of bi-layer tablets of either single drug [5, 10] or a combination of drugs within the same therapeutic category [11, 12] for quick/slow type of biphasic release of drugs. Biphasic release of drugs was also achieved through formulation of gastro retentive floating multiple tablets [13], musoadhesive matrix tablets [14]. Most of the reports deal with the compression of an IPR layer over a SR layer to form a bilayer tablet. The principle involved in achieving quick release is rapid disintegration of IPR layer using super disintegrants. However, for a poorly water soluble drug, IPR is dissolution rate limited. Solid dispersion is a technology which induces solid state transformation/molecular dispersion of drugs enhancing the rate of dissolution considerably.

In the present investigation, GPZ, an antidiabetic drug, has been selected as a model drug for developing bi-layer tablets. The drug is characterized by a short biological half life of 3.4±0.7 h [15] requiring administration in a dose of 2.5 to 10 mg in 2-3 divided doses [16]. Moreover, it falls under class II category of Biopharmaceutical Classification System exhibiting poor aqueous solubility and thus making absorption as dissolution rate limited.

The objective of the present work is to develop a bi-layer tablet consisting of an IPR layer and a SR layer that can produce a distinct biphasic release having two different drug release rates. The IPR layer is intended to release a fraction of the dose rapidly at a faster rate and the SR layer is meant for slow release of the remaining dose at a slower rate for a desired period of time. To achieve instant release of a fraction of the dose, the solubility and dissolution rate of GPZ was increased by solid dispersion technique, using gelucire as a

water soluble carrier. Gelucire has been reported to be non toxic and is suitable for use in tabletting, capsule filling [17] and have high solubilizing property [18, 19]. The resulting solid dispersion was converted into IPR layer of the bi-layer tablet. The release rate retarding SR layer was formulated using sodium alginate (SAL) with or without carboxymethyl cellulose (CMC) and CG. Initially the IPR and SR layers were optimized separately, and finally the Bi-layer tablet was evaluated for distinct biphasic release.

## MATERIALS AND METHODS

Glipizide (GPZ), Indian Pharmacopoeia, was a generous gift from Stadmed Pvt Ltd, Kolkata India. Gelucire 44/14 and Microcrystalline Cellulose (MCC) were donated respectively by Gattefosse, Saint-Priest Cedex-France, and Dr Reddy's Lab, Hyderabad, India. Lactose Monohydrate and Calcium Gluconate (CG) from Himedia, Mumbai, India, SAL, SD Fine chem. Pvt Ltd Mumbai, India, CMC, Loba Chemie, Mumbai, India, Magnesium Stearate and all other ingredients were obtained commercially and used as received. Double distilled water was used throughout the experiment.

### Preparation of solid dispersion

Solid dispersion of GPZ and gelucire was prepared by fusion method using three different ratios of GPZ/Gelucire (weight ratios of 1:1, 1:3, 1:5). Accurately weighed amount of gelucire was taken in an aluminium pan and melted on a water bath at 60 °C with constant stirring. Required amount of GPZ was added into the melted carrier with stirring to achieve a homogeneous mass which was then allowed to cool to room temperature. Lactose was added to the mass during cooling. The resulting solid dispersion was sieved through # 85 mesh BS screen and stored in desiccator until use.

### Solubility study

Excess drug was added in 10 ml double distilled water contained in 50 ml stopper conical flask and was shaken for 48 h on a mechanical shaker at 37 °C. The mixture was filtered through whatman filter paper no 1, and aliquot, following suitable dilution, was analyzed at 276 nm using a spectrophotometer (Multiscan Go, Thermoscientific, USA). The solubility of the drug was determined from calibration curve. Similarly, the solubility of GPZ was determined in acid solution (0.1 (N) HCl, pH 1.2) and phosphate buffer (PB) solution (pH 6.8). All experiments were done in triplicate.

### Phase solubility study

For phase solubility study, required amount of gelucire (0**,** 2, 4, 6, 8, 10 and 12 %w/v) was added in 10 ml acid solution (pH 1.2) in a stopper 50 ml conical flask. Excess powdered drug was added in each of the conical flasks and shaken for 48 h on a mechanical shaker at 37 °C. The mixture was filtered, and an aliquot, following suitable dilution, was analyzed at 276 nm spectrophotometrically to determine the solubilities. The experiment was repeated in a similar fashion in PB solution (pH 6.8). The solubility of GPZ in presence of different amount of gelucire has been shown in [table 1].

**Table 1: Phase solubility and $G_0tr$ of glipizide at different concentration of gelucire 44/14**

| Conc of Gelucire 44/14 (%w/v) | Solubility of GPZ and its associated gibbs free energy in acid buffer ph 1.2 | | Solubility of GPZ and its associated Gibbs free energy in phosphate buffer ph 6.8 | |
|---|---|---|---|---|
| | Solubility of GPZ (µg/ml)±SD(n=3) | $-g_0tr$ (J/K/mol) | Solubility of GPZ (µg/ml)±SD(n=3) | $-g_0tr$ (J/K/mol) |
| 0 | 1.383333±0.11 | 0 | 5.035±0.017 | 0 |
| 2 | 4.05±0.16 | -2770.47 | 35.02333±1.83 | -5002.16 |
| 4 | 8.1±1.63 | -4558.14 | 45.32333±1.46 | -5667.06 |
| 6 | 13.53333±0.96 | -5881.94 | 51.61333±1.52 | -6002.23 |
| 8 | 18.21333±0.28 | -6647.92 | 57.09333±1.85 | -6262.48 |
| 10 | 22.16667±0.23 | -7154.54 | 62.53333±0.69 | -6497.2 |
| 12 | 21.52333±2.17 | -7078.58 | 58.94333±0.30 | -6344.72 |
| 14 | 18.76667±0.43 | -6725.1 | 37.59±1.02 | -5184.56 |
| 16 | 17.10333±0.24 | -6485.75 | 26.85±1.61 | -4316.78 |

**Table 2: Compositions of IPR tablets**

| Formulation Code | GPZ(mg) | Gelucire 44/14 (mg) | GPZ: Gelucire | Lactose (mg) | Avicel PH 102(mg) | Mag St(mg) | Total(mg) |
|---|---|---|---|---|---|---|---|
| Control | 1.5 | -- | -- | 53.55 | 124.95 | 2 | 182 |
| IPR1 | 1.5 | 1.5 | 1:1 | 53.1 | 123.9 | 2 | 182 |
| IPR2 | 1.5 | 4.5 | 1:3 | 52.2 | 121.8 | 2 | 182 |
| IPR3 | 1.5 | 7.5 | 1:5 | 51.3 | 119.7 | 2 | 182 |

**Table 3: Compositions of SR tablets**

| Formulation Code | GPZ(mg) | SAL (mg) | HVCMC(mg) | CG(mg) | Polymer: CG | Mag St | Total (mg) |
|---|---|---|---|---|---|---|---|
| SR1 | 3.5 | 100 | 0 | 100 | 1:1 | 2 | 205.5 |
| SR2 | 3.5 | 100 | 0 | 130 | 1:1.3 | 2 | 235.5 |
| SR3 | 3.5 | 100 | 0 | 150 | 1:1.5 | 2 | 255.5 |
| SR4 | 3.5 | 100 | 0 | 180 | 1:1.8 | 2 | 285.5 |
| SR5 | 3.5 | 86.6 | 0 | 129.9 | 1:1.5 | 2 | 222 |
| SR6 | 3.5 | 60.62 | 25.98 | 129.9 | 1:1.5 | 2 | 222 |
| SR7 | 3.5 | 43.3 | 43.3 | 129.9 | 1:1.5 | 2 | 222 |
| SR8 | 3.5 | 25.98 | 60.62 | 129.9 | 1:1.5 | 2 | 222 |

### Preparation of IPR tablet

IPR tablets, the compositions of which are shown in [table 2], were prepared by direct compression method. Solid dispersion (equivalent to 1.5 mg of GPZ), MCC and Mg-Stearate were blended manually for 10 min and compressed into tablets using 8.7 mm flat face punch in a tablet machine (RIMEK, Karnavati Engineering Ltd., Gujarat, India). The crushing strength and weight of the tablets were kept constant respectively at 2 kg and 182 mg. Control tablets without containing solid dispersion were prepared in a similar way.

**Preparation of SR tablet**

SR tablets, the compositions of which are shown in [table 3], were prepared by wet granulation method. GPZ, SAL, CMC and CG were blended, moistened with required amount of water, and granulated using #18 mesh BS screens. The granules were dried at 60 °C for sufficient time till the moisture content in the granules reduced to about 2% w/w. The dried granules were passed through #22 mesh BS screen, mixed with Mg-Stearate and compressed into tablet using 8.7 mm flat face punch. The crushing strength of the tablets was kept constant at 4 kg.

**Preparation of Bi-Layer tablets**

For the preparation of bi-layer tablet, initially the granules of SR layer were compressed in the tablet machine using 8.7 mm flat face punch. Upon this layer, the granules of the IPR layer were placed manually and further compressed. Bi-layer tablets, the compositions of which are shown in [table 4] were designated as BL1 to BL4**.** Total amount (5 mg) of the drug in bi-layer tablet was divided into two fractions (1.5 mg in IPR layer and 3.5 mg in SR layer) following the report published elsewhere [20]. The crushing strength of the tablets was kept constant at 4 kg.

**Table 4: Compositions of bi-layer tablets**

| Formulation code | IPR code | SR code |
|---|---|---|
| BL1 | IPR3 | SR5 |
| BL2 | IPR3 | SR6 |
| BL3 | IPR3 | SR7 |
| BL4 | IPR3 | SR8 |

**Evaluation of IPR, SR and BL tablets**

**Weight variation test**

Twenty tablets were weighed individually in an electronic pan balance (XB 600 M-C, Precisa, Switzerland). The weight of each tablet was compared with the average weight of the tablets.

**Crushing strength of tablets**

Crushing strength of the tablets was determined using Monsanto type Tablet Hardness Tester (Campbell Electronics, Mumbai, India) and average value of 10 determinations was reported.

**Thickness of tablets**

The thickness of each tablet was measured with a Digimatic Caliper (CD-6"CS, Mitutoyo Corporation, Japan) and average of 10 determinations was calculated.

**Friability test**

Ten tablets were weighed and placed in the plastic drum of a Friabilator (EF2, Electro Lab, Mumbai, India). After 100 revolutions, the tablets were dedusted with a soft brass and reweighed. The percentage of weight loss was calculated.

**Disintegration test of IPR tablets**

*In-vitro* disintegration study of IPR tablets was carried out in USP II tablet disintegration test apparatus (Electrolab tablet disintegration tester (USP) ED-2L) at a frequency of 28-32 cycles per minute. One tablet was placed in 1000 ml acid solution of pH 1.2 maintained at 37±2 °C. The time (min) required for complete disintegration (when no residue of the tablet remains on the screen of the basket or, if any residue remains, it consists of fragments of disintegrated parts of tablets) of the tablets were noted.

**Drug content of the IPR and SR tablets**

One IPR tablet was weighted and powdered in glass mortar. The powder was quantitatively transferred into a 250 ml volumetric flask with 150 ml phosphate buffer solution of pH 6.8 was added. The stoppered flask was shaken for 24 h in a mechanical shaker and volume was made up to the mark with the buffer solution. The mixture was filtered and an aliquot following suitable dilution was analyzed spectrophotometrically at 276 nm for GPZ content.

The potency of the tablet was determined in the usual manner using a calibration curve constructed in buffer solution (pH 6.8). Average drug content of 10 tablets was considered as potency of the tablets. The reliability of the above method was judged by conducting recovery analysis for three consecutive days at three levels of spiked drug solution in the presence or absence of the excipients. The recovery averaged 98.51±1.14%. The drug content of SR tablets was determined in the similar way.

***In-vitro* drug dissolution study**

*In-vitro* drug release study from IPR tablets was carried out in USP II tablet dissolution rate test apparatus (model TDP-06P, Electro Lab, Mumbai, India) at a paddle speed of 75 rpm. One tablet was placed in 1000 ml acid solution (pH 1.2) at 37±0.5 °C. At predetermined time intervals, aliquots were withdrawn and replenished immediately with the same volume of fresh medium maintained at 37 °C. The aliquots following suitable dilution were analyzed spectrophotometrically at 276 nm to calculate the amount of drug released.

*In-vitro* drug release study from SR tablets was performed following the method described for delayed release tablets in USP30-NF25. Initial drug release study was carried out in 750 ml of acid solution for 2 h. Thereafter, the pH of the medium was adjusted within 5 min to 6.8 by adding 250 ml of 0.2 (M) tribasic sodium phosphate solutions and the dissolution study was continued. The amount of drug released was determined using the calibration curves drawn respectively in acid solution and phosphate buffer solution. Drug release from BL tablets was determined in a similar way.

**Drug-excipient and polymer compatibility study**

The compatibility of GPZ with the excipients and polymers used in the preparation of IPR and SR tablets was studied through FTIR, DSC, XRD studies.

**Fourier transforms infrared (FTIR) analysis**

FTIR spectra of GPZ, physical mixture, and powdered IPR tablet were recorded in a FTIR spectrophotometer (Perkin Elmer, RX-1, UK). The samples were mixed with KBr and converted into pellets at 6 ton pressure with a hydraulic press. The spectra were taken in the wave number region of 4000-400/cm. The FTIR spectra of physical mixture and powdered SR tablets were determined in a similar way.

**Differential scanning calorimetry (DSC) study**

DSC thermograms of GPZ, physical mixture, and powdered IPR tablet were obtained using Perkin Elmer (Pyris Diamond TG/DTA, Singapore) differential scanning calorimeter which was calibrated against indium. Weighed amount of the sample was kept in a hermetically sealed aluminum pan and heated at a rate of 10°C/min over a temperature range 30-300°C under constant nitrogen flow of 150 ml/min. Same procedure was followed for the physical mixture and powdered SR tablets.

**X-Ray powdered diffraction (XRD) study**

The qualitative XRD studies of GPZ, physical mixture, and powdered IPR tablet were performed using an X-ray diffractometer (ULTIMA-III, Rigaku, Japan). The samples were scanned from 5 ° to 50 ° diffraction angle (2θ) range under the following measurement conditions: Source, Ni-filtered Cu-Kα (λ = 1.54) radiation; voltage, 40 kV; current, 40 mA; scan speed, 1 °/min. The X-ray diffraction

patterns of the physical mixture and powdered SR tablet were determined in a similar way.

**Data treatment**

**Diffusion coefficient of drug**

Diffusion coefficient [$D_c$, $cm^2$/sec] of GPZ from the tablets were determined using [Eq (1)] which was used to calculate the diffusion coefficient ($D_c$) of drug from spherical matrices [21]

$$D_c=\pi (r\Theta/6M_\infty)^2 \text{.....[1]}$$

Where r is the equivalent spherical radius of tablet, Θ is the slope of linear portion of $M_t/M_\infty$ versus $t^{1/2}$ plot, $M_t$ is the amount of drug released at time t (sec), and $M_\infty$ is the total amount of drug loaded. The equivalent spherical diameter [cm] of the tablets was calculated from the relationship:

$$d= (6hr_c)^{1/3} \text{.....[2]}$$

Where d, $r_c$, and h represent equivalent spherical diameter, radius, and height (cm) of the tablets respectively.

**Drug release mechanism**

To study the kinetics and mechanism of GPZ release from various tablets, the *In-vitro* drug release data were fitted in the following equations [table 5]:

**Zero order model** [22]

$$M_t=M_0+K_0t \text{.....[3]}$$

Where $M_t$ is the amount of drug released in time t, $M_0$ is the initial amount of drug, and $K_0$ is the zero order release constant.

**First order model** [23]

$$\log M=\log M_0-K_1 (t/2.303) \text{.....[4]}$$

Where M is the amount of drug released in time t, $M_0$ is the initial amount of drug, and $K_1$ is the first order release constant.

**Korsmeyer-Peppas model**

$$Mt/M\infty = Kt^n \text{.....[5]}$$

Where $Mt/M\infty$ is the fraction of drug released at time t, K is the release rate constant and n denotes the diffusional exponent indicative of transport mechanism. In case of tablet, n=0.45 indicates Fickian diffusion, 0.45<n>0.89 indicates non-Fickian/anamolous transport, n=0.89 is for case II transport, and n>0.89 indicates super case II transport [24, 25]. Anamolous refers to a combination of both erosion and diffusion controlled release and case II refers to erosion of polymeric chain.

**Mean dissolution time (MDT)**

MDT in min was calculated from [Eq 6] [26] using the values of n and k obtained from [Eq. 5].

$$MDT= (n/n+1).\ k^{(-1/n)} \text{.....[6]}$$

**RESULTS**

Solid dispersions of GPZ and gelucire prepared in weight ratio of 1:1, 1:3, and 1:5 were semi solid in nature and were converted into free flowing powder by adding lactose as adsorbent.

The equilibrium solubility of GPZ in water, acid solution (pH 1.2), and PB solution (pH 6.8) were found respectively 4.90, 1.48 and 5.04 µg/ml. The results of phase solubility study are shown in [table 1] which indicated a linear increase in solubility with increase in concentration of gelucire in both acid solution and PB solution.

It was further noted that the maximum solubility of the drug was attained at 10% w/v concentration of gelucire and beyond which the solubility decreased. At 10% w/v concentration of gelucire, the solubility of GPZ was enhanced by 15.27 and 12.35 times in acid solution and PB solution respectively compared to the solubility in water at 37 °C. The Gibb's free energy was calculated from the [Eq. 7]

$$G\,^\circ_{tr}=-2.303RT \text{ Log } (S_0/S_s) \text{.....[7]}$$

Where, $S_0/S_s$ is the ratio of the molar solubility of GPZ after and before treatment with carrier. The value of gas constant (R) is 8.314 J/K/mol and T is temperature in Kelvin (K). It was found that there was an increase in the negative values of Gibbs free energy with the increase in the concentration of the carrier up to 10% w/v, beyond this concentration the negative values of Gibb's free energy decreased.

**Table 5: Derived release parameters and release kinetics from IPR, SR, BL tablets**

| Formulation code | Zero order $R^2$ | First order $R^2$ | Korsmeyer-Peppas | |
|---|---|---|---|---|
| | | | n | $R^2$ |
| Control | 0.980 | 0.988 | -- | -- |
| IPR1 | 0.945 | 0.982 | -- | -- |
| IPR2 | 0.879 | 0.968 | -- | -- |
| IPR3 | 0.783 | 0.940 | -- | -- |
| SR1 | 0.997 | 0.980 | 0.811 | 0.996 |
| SR2 | 0.997 | 0.865 | 1.006 | 0.996 |
| SR3 | 0.998 | 0.831 | 0.987 | 0.991 |
| SR4 | 0.996 | 0.868 | 0.980 | 0.996 |
| SR5 | 0.997 | 0.942 | 0.821 | 0.979 |
| SR6 | 0.995 | 0.886 | 0.951 | 0.943 |
| SR7 | 0.993 | 0.856 | 1.14 | 0.959 |
| SR8 | 0.987 | 0.842 | 0.986 | 0.981 |
| BL1 | 0.995 | 0.752 | 0.236 | 0.94 |
| BL2 | 0.993 | 0.782 | 0.262 | 0.972 |
| BL3 | 0.999 | 0.854 | 0.356 | 0.988 |
| BL4 | 0.994 | 0.869 | 0.379 | 0.986 |

IPR, SR and BL tablets complied with specified limits of USP with respect to weight variation, drug content, and friability. The variations in the thickness of the tablets were within±5% and the crushing strength of IPR tablets was kept constant at 2 kg. Control tablets which were prepared without gelucire disintegrated within 2 min. However, the disintegration time of IPR tablets was found to be protracted from 3-4 min to 9-10 min with increase in the amount of gelucire.

Release profiles of GPZ from control and various IPR tablets are shown in [fig 1]. The control tablets which was prepared without gelucire released 16.53±1.62% and 39.26±2.06% of the loaded drug respectively in 30 min and 2 h in acid solution of pH 1.2. The release of drug from IPR tablets was higher than that obtained from the control tablets. Moreover, increase in the amount of gelucire in the IPR tablets increased the drug release considerably. IPR 3 tablet

which was prepared with the highest amount of gelucire, released 89.27±1.95% and 100.09±0.7% of GPZ respectively in 30 min and 2 h.

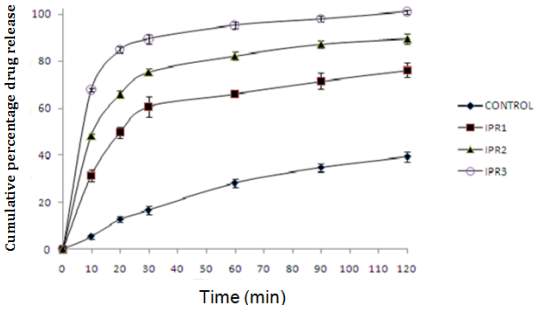


**Fig. 1: GPZ release from IPR tablets in acidic solution of pH 1.2 for 2 h. Filled rhombus control tablet, filled square IPR1, filled triangle IPR2, unfilled circle IPR 3. Average±SD,n=3**

*In-vitro* drug release profiles from SR tablets (SR1-SR4) are shown in **[**fig. 2]. SR layer was prepared with various ratios of SAL and CG. Increase in amount of CG, which was used as a crosslinking agent, decreased the drug release from tablets in the following order SR1>SR2>SR3. Further increase in the amount of CG increased the drug release as in SR4 tablets. MDT and diffusion coefficient [table 6] also followed the same pattern. Increase in the amount of CG increased the values of MDT and decreased the values of diffusion coefficient. However, in case of SR4, prepared with the highest amount of CG, the values of MDT decreased and that of diffusion coefficient increased, as shown in table 6. SR tablets, designated as SR6, SR7 and SR8, were prepared by substituting SAL with increasing amount of CMC, and the drug release profiles of the tablets are shown in [fig 3]. The release of GPZ was found to decrease with the increase in the amount of CMC.

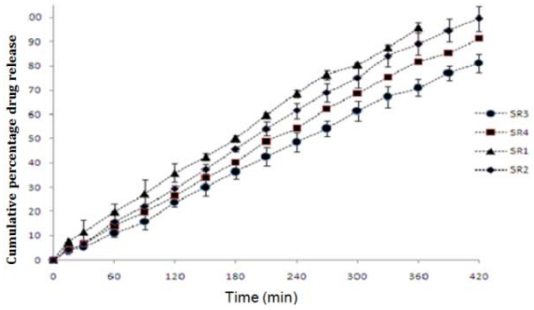


**Fig. 2: GPZ release from SR tablets in acidic solution of pH 1.2 for 2 h followed by phosphate buffer solution (pH 6.8). Filled triangle SR1, filled rhombus SR2, filled circle SR3, filled square SR4. Average±SD, n=3**

**Table 6: MDT and diffusion coefficient of drug calculated from the release profile of various SR tablets**

| Formulation code | MDT (min) (mean±SD, n=3) | Diffusion coefficient ($cm^2/s$) (mean±SD, n=3) |
|---|---|---|
| SR1 | 18.82±0.35 | 0.019±0.05 |
| SR2 | 28.24±2.07 | 0.012±0.03 |
| SR3 | 38.32±0.57 | 0.008±0.03 |
| SR4 | 33.13±0.55 | 0.011±0.05 |

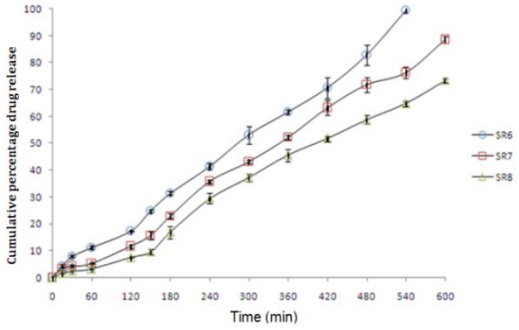


**Fig. 3: GPZ release from SR tablets in acidic solution of pH 1.2 for 2 h followed by phosphate buffer solution (pH 6.8). Unfilled circle SR6, unfilled square SR7, unfilled triangle SR8. Average±SD, n=3**

BL tablets designated as BL1 to BL4, were prepared with optimum formulations of IPR and SR layer. BL tablets immediately released a fraction of the loaded drug ranging from 23-37% within first 30 min of dissolution study in acid solution of pH 1.2. Thereafter, it slowly released the remaining amount of drug. While drug release from BL1 tablet extended up to 7 hr, BL4 released 70% of the drug in 10 hr [fig 4].

The FTIR spectra of GPZ, physical mixture, and powdered IPR tablets are shown in [fig 5]. The peaks at 3251 & 3325/cm for NH stretching, 3060/cm for aromatic CH, 2854 to 2941/cm for CH stretching, 1649/cm for NH group, 1525 & 1687/cm for C=O group, 1163 for S=O and 1442 & 686 for CH bending respectively appeared in the spectrum of the drug. The peaks were also found in the spectra of physical mixture and IPR tablets almost at the same wave numbers. The FTIR spectra of the physical mixture and the powdered SR tablets also exhibited the characteristics peaks of the drug almost at the same wave numbers.

The DSC thermograms of GPZ, physical mixtures and powdered IPR tablets are shown in **[**fig 6]. The thermogram of GPZ showed a sharp melting endotherm at 216 $^{o}$C. The endotherm was, however, absent in the thermograms of physical mixture and IPR tablet. On the other hand, melting endotherm at 216$^{o}$C was also observed in the thermograms of the physical mixture and powderd SR tablets.

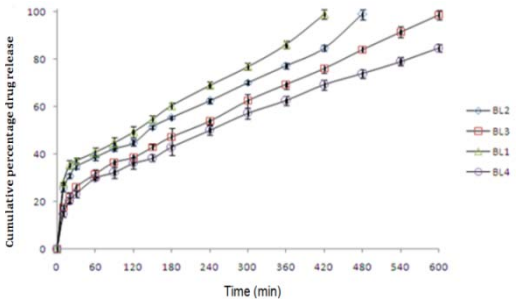


**Fig. 4: GPZ release from BL tablets in acidic solution of pH 1.2 for 2 h followed by phosphate buffer solution (pH 6.8). Unfilled triangle BL1, unfilled rhombus BL2, unfilled square BL 3, unfilled circle BL4. Average±SD, n=3**

The XRD of GPZ, physical mixtures and powdered IPR tablets are shown in [fig 7]. The diffractogram of GPZ shows sharp peaks at 7.49, 11.24, 15.67, 17.10, 18.67 and 21.86 ° 2 theta values. The peaks were, however, absent in the diffractograms of physical mixture and IPR tablet. The diffractograms of physical mixture of the drug and excipients used for SR tablet as well as for SR tablet showed the characteristics peaks at the respective 2 theta values.

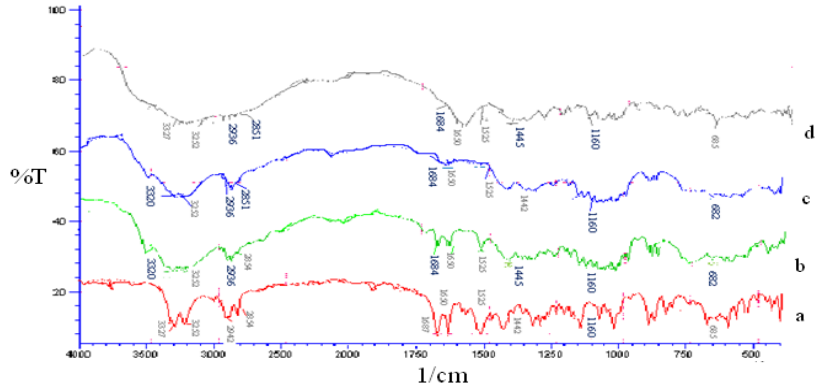


**Fig. 5: FTIR trace of a) GPZ, b) Solid dispersion of GPZ/Physical mixture of IPR tablets, c) powdered IPR tablets d) powdered SR tablets**

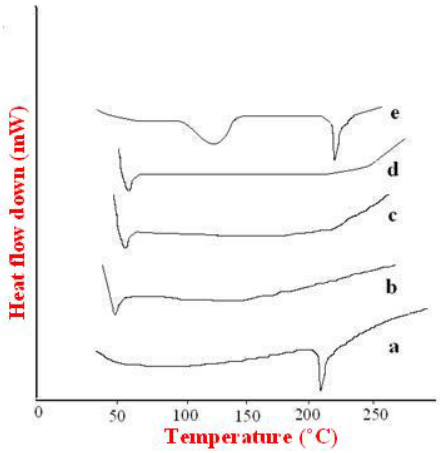


**Fig. 6: DSC thermograms of a) GPZ, b) Gelucire 44/14, c) Physical mixture of IPR tablets, d) powdered IPR tablets, e) powdered SR tablets**

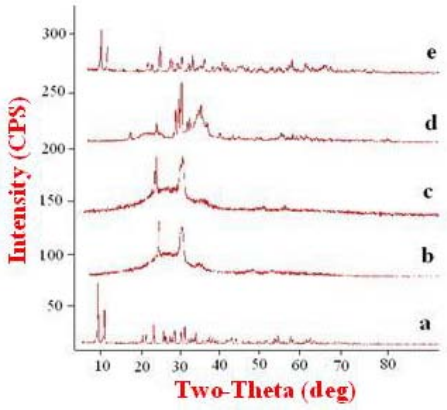


**Fig. 7: XRD pattern of a) GPZ, b) Gelucire 44/14, c) Physical mixture of IPR tablets, d) powdered IPR tablets, e) powdered SR tablets**

## DISCUSSION

The objective of the present study was to develop bilayer tablets of GPZ that can produce euglycaemia rapidly through an IPR of a fraction of the dose, and then maintain the euglycaemic level for extended period of time through slow release of the remaining fraction of the dose. As GPZ exhibits poor aqueous solubility, the enhancement of solubility became the first criteria to achieve IPR of the drug. Solid dispersion is one of the widely used and established methods for enhancing aqueous solubility of poorly water soluble drugs [27]. Various water soluble carriers such as polyethylene glycols [28], polyvinyl pyrrolidone [29],-β-Cyclodextrin [30] have been widely used to develop solid dispersions of poorly water soluble drugs like diazepam [31], valdecoxib [28], aceclofenac [32]. Gelucire 44/14 (where suffixes 44 and 14 represent its melting point and HLB value respectively) has been selected in the present study to prepare solid dispersions of GPZ**.** Gelucire, having low melting point, increases aqueous solubility of many poorly water soluble drugs considerably [33, 18]. Phase solubility study indicated that the solubility of GPZ in both acid solution (pH 1.2) and PB solution (pH6.8) increased with increase in the concentration of gelucire, reached a limiting value at 10% w/v of gelucire, and then decreased [table 1]. The above phenomenon can be explained by thermodynamic principles. The solubilization process is thermodynamically favored only if the overall Gibb's free energy of the solution is decreased. Associated Gibb's free energy was calculated from the equation 7. Table 1 shows that the negative values of Gibb's free energy increased with the increase in the concentration of gelucire, reached a maximum value, and then decreased. The increase in the negative values of Gibbs free energy with the increase in the concentration of the carrier indicates the spontaneity of the solubilization process and that the reaction becomes more favorable up to concentration of 10 %w/v. beyond that concentration, the negative values of Gibb's free energy decreased, indicating the saturation of solubilization.

IPR tablets, the compositions of which are shown in table 2, were prepared by direct compression method using solid dispersion of GPZ with gelucire at three different levels. The wet and sticky solid dispersions were converted into free flowing powder for ease of compression by adding lactose. Lactose is a common water soluble diluent [34], acts as a surface adsorbent [35,36], and enhances the dissolution rate of hydrophobic drug by increasing the surface area of the solid dispersion [35]. The IPR tablets complied with the pharmacopoeial limits of USP with respect to the weight variation, content uniformity, friability and thickness. The disintegration time of IPR tablets were higher than that of control tablets prepared without gelucire. While the disintegration time of control tablets was 1-2 min, the same increased from 3-4 min to 9-10 min with increase in the concentration of gelucire although the hardness of all the tablets were kept constant at 2 kg. Increase in disintegration time of the IPR tablets with increase in gelucire concentration might be related to the soft and waxy nature of the carrier. Such carriers often plasticize, melt or soften during compression and fill the pores of the tablets leading to increase in the disintegration time [37]. The release of GPZ from IPR tablets was higher than that of control tablets which were prepared without gelucire. Moreover, the drug release increased with increase in concentration of gelucire. While 39.26±2.05% drug was released from the control tablets in acid solution in 2 h, 100.90±0.70% drug was released from IPR3 tablets containing highest amount of gelucire in the same time interval (fig 1). The faster release of GPZ from IPR tablet was attributed to the enhancement of solubility of the drug by solid dispersion which might have induced solid state transformation of the drug. GPZ did not interact with any of the excipients as the characteristic peaks

(3251 & 3325/cm for NH stretching, 3060/cm for aromatic CH, 2854 to 2941/cm for CH stretching, 1649/cm for NH group, 1525 & 1687/cm for C=O group, 1163 for S=O and 1442 & 686 for CH bending respectively) of GPZ [38,39], in the physical mixture and IPR tablets appeared at almost the same wave numbers [fig 5]. DSC thermograms of GPZ produced a sharp melting endotherm at 216 °C corresponding to the melting point of the drug [15]. However the melting endotherm was not evident in either the physical mixture or the IPR tablets [fig 6]. Moreover the characteristics peaks of GPZ at 2theta values of 7.49, 11.24, 15.67, 17.10, 18.67 and 21.86were not evident in the XRD traces of the physical mixture and IPR tablets [fig 7]. The results indicate that although the drug was compatible with excipients of IPR tablets, the crystalline drug was converted into amorphous state. Solid state transformation of drugs during solid dispersion process is well documented [40].

SR tablets of GPZ were prepared by wet granulation method using different ratios of SAL and CG. The compositions of the various tablets are shown in [table 3]. All the tablets complied with the pharmacopoeial limits with respect to the weight variation, content uniformity, friability and thickness. GPZ was compatible with all the excipients used in preparation of SR tablets. FTIR study revealed that the characteristics peaks of the drugs were present in the spectra of both physical mixture and powdered SR tablets [fig 5]. The melting endotherm of GPZ at 216 °C was evident in the DSC thermograms of the physical mixture and SR tablets [fig 6]. In addition XRD diffractograms demonstrated the presence of the peaks of the drugs almost at the same 2theta values as found with the pure drug [fig 7]. However the intensity of the peaks was found to be reduced, which may be due to dilution effect of the drug in the formulation [39]. The studies confirmed that GPZ was compatible and did not undergo any polymorphic change in the SR tablets.

The drug release from SR tablets was studied under the dynamic pH shift condition where the release study was conducted in acid solution of pH 1.2 for the first 2 h and thereafter the pH of the medium was increased to pH 6.8 and the results are shown in [fig 2]. The release of GPZ was faster from the tablet SR1 prepared with highest ratio of SAL/CG. Decrease in the ratio of SAL/CG, as in tablets SR2 and SR3, decreased the drug release, and after a certain ratio as in SR4, the drug release increased. Variation in drug release due to addition of CG in SR layer was further ascertained from the values of MDT which was calculated from [Eq. (6)]. One way analysis of variance (ANOVA) shows that MDT increased significantly [$p<0.05$] with increase in the amount of CG, and after a certain amount of CG, MDT decreased [table 6]. SAL, a biopolymer obtained from marine brown algae, is composed of varying proportion of D-mannuronic acid (M) and L-glucoronic acid (G) residues which are arranged in MM or GG blocks interspersed with MG block [41]. It has a unique property of forming calcium alginate gel through ionotropic gelation with $Ca^{2+}$ions in simple and mild conditions [42-44]. SR tablets consisting of SAL and CG as a source of $Ca^{2+}$ions were prepared by wet granulation method. $Ca^{2+}$ions which were generated from CG in contact with water during the wet massing step as well as dissolution study reacted with the acid residues of SAL and crosslinked the polymer chain. Crosslinking reduced the mobility of the polymer chain and resulted in the formation of a true gel layer around the tablet surface in contact with water [45]. Increase in the amount of $Ca^{2+}$ions increased the crosslinking density producing higher gel strength and reducing the macromolecular mesh size [46, 47]. This resulted in a decrease in diffusion and hence the release of GPZ through the gel layer. The diffusion coefficient of GPZ through the gel layer formed around the SR layer tablet was determined using [Eq. (1)], which was used to calculate the diffusion coefficient of drug from spherical matrices [21, 48]. Table 6 shows that the values of diffusion coefficient of the drug decreased as the amount of CG in the SR tablets was increased, and beyond a certain amount, the diffusion coefficient increased. The drug release profiles also followed the same trend. Increase in drug release at higher level of CG might be due to the presence of excess un reacted CG in the tablet, which acted as a channeling agent [49]. Among the various SR tablets, SR3 produced prolonged release of GPZ extending up to 8 h**.** But it was not practically feasible to compress both SR3 and IPR3 tablet together in 8.7 mm die punch for the preparation of bi-layer tablets. So to accommodate both the layers in the die punch, the weight of SR3 tablet was reduced to 222 mg while keeping the ratio of SAL/CG same. The GPZ release profile of the resulting SR tablet (SR5) was compared with that of SR3 using similarity factor $f_1$ and dissimilarity factor $f_2$. The values of $f_1$ and $f_2$ were calculated respectively from the Eq. 8 and Eq 9 [50, 51]

$$f_1 = \left\{ \left[ \sum_{i=1}^{P} |R-T| \right] / \left[ \sum_{i=1}^{P} R \right] \right\} \times 100 \quad \text{..... [8]}$$

$$f_2 = 50 \log \left\{ \left[ 1 + \left( \frac{1}{P} \right) \sum_{i=1}^{P} (R-T)^2 \right]^{-1/2} \times 100 \right\} \quad \text{..... [9]}$$

Where, P is the number of dissolution time points, and R and T are the mean percentage drug released at each time point t from the reference and test samples respectively. It was found that the values of $f_1$ and $f_2$ were 62.44 and 9.67 respectively. No significant difference [$p<0.05$] was obtained from students t test between $f_1$ and $f_2$ values of the two SR tablets. This indicated that the release profiles of the tablets SR3 and SR5 did not vary appreciably and hence SR5 tablet was selected for the preparation of the bi-layer tablets. In order to further prolong the drug release, SR tablet designated as SR6, SR7 and SR8 were prepared, wherein, SAL was substituted with increasing amount of high viscosity CMC. The release profiles [fig 3] revealed that increase in the amount of CMC, decreased the drug release.

Scrutiny of the release profiles of GPZ from IPR and SR tablets demonstrated that IPR 3 tablet provided fastest drug release (89% of the loaded drug in 30 min), and SR5 tablet produced the most extended drug release up to 8 h. Hence, bi-layer tablets BL1 was prepared using IPR3 and SR5. The release profiles [fig 4] revealed that 37% drug was released quickly in 30 min and thereafter, the release extended up to 7 h in a SR manner. In order to further prolong the drug release from SR layer, SAL in SR tablet was substituted with increasing amount of CMC and the tablets were designated as BL2 (SAL: CMC 70:30), BL3 (SAL: CMC 50:50), BL4 (SAL: CMC 30:70). It was noted substitution of SAL with increasing amount of CMC prolonged the drug release. While BL1 released 100% of the loaded drug in 7 h, BL4 released 70% of the loaded drug in 10 h. The release curves clearly exhibited two distinct phases of drug release. An initial fast release phase where the slope of the curve was higher and a second slow release phase where the slope of the release curve was lower [Table 7]. It was further noted that IPR in 30 min from all the BL tablets was not the same. This indicates that SR layer also contributed to the total drug release. As initial release from SR5 was more than that of SR8, the IPR from BL1 was more than that of BL4.

**Table 7: Slope of the immediate release and SR part of the bi-Layer tablets**

| Formulation code | Slope | |
|---|---|---|
| | Fast release part (time 0-10 min) | Slow release part (time 30 min-complete release) |
| BL1 | 2.783 | 0.156 |
| BL2 | 2.552 | 0.136 |
| BL3 | 1.741 | 0.124 |
| BL4 | 1.478 | 0.106 |

The data obtained from *in vitro* release profiles were fitted in various kinetic models as shown in [table 5]. It was observed that the IPR layer followed first order release kinetics with correlation coefficient of 0.940-0.988, during the initial burst release period of the first 30 min where maximum drug was released. Immediate release solid dispersion tablets showing first order release kinetics is reported in literature [52]. Drug release from SR tablets followed zero order release with super case II transport mechanisms. Bi layered tablets showed zero order release kinetics after the initial burst release, with super case II transport mechanisms. Previous literatures of bi-layered tablets with biphasic release pattern also demonstrated zero order mechanism after the initial burst release [10].

## CONCLUSION

In the present study, initially IPR and SR tablets were prepared separately to achieve respectively an IPR and slow release of GPZ, an antidiabetic drug. Solid state transformation through solid dispersion with gelucire enhanced the poor aqueous solubility of the drug and induced rapid release from IPR tablets. SR tablets produced slow release of the drug by forming a gel around the tablet surface and that was further augmented by partial replacement of SAL with CMC. The optimized IPR layer was compressed with various SR layers to develop bi-layer tablets. Resulting bi-layer tablets produced bi-phasic release of the drug at two distinct rates. A quick release amounting to 23 to 37% of the drug was obtained in 30 min, and thereafter, the release was found to prolong at a slower rate from 7 h to 10 h depending upon the compositions of the SR layers. This study revealed that bi-phasic release of GPZ consisting of an initial quick release and subsequent slow release could be achieved by formulating bi-layer tablets using the existing tablet technology, and such formulation may be able to control hyperglycaemia effectively for a longer period of time.

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## CONFLICT OF INTERESTS

The authors report no declaration of interest

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