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pH Independent Immediate Release Formulation of Glipizide Using Air Jet Milled Ternary Complex:*In-vitro* Characterization and Molecular Modelling Studies

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ABSTRACT

The objective of this study was to develop pH independent immediate release (IR) tablet formulation of Glipizide (GPZ) incorporating β-cyclodextrin (β-CD) and a ternary agent produced by high energy air jet milling complexation technique. GPZ (pKa of 5.9) is poorly water soluble (3.9µgm/ml) exhibiting pH dependent solubility (1.1µgm/ml at pH 2.0 and 26.6 µgm/ml at pH 6.8) owing to which it demonstrates dissolution rate limited absorption and bioavailability. Several complexation techniques involving formation of binary and ternary complexes were evaluated for achieving pH independent release of GPZ. Ternary complex involving GPZ:β-CD: Arginine (1:2:1) prepared using high energy air jet milling was found to be most promising in terms of significant enhancement in solubility, further attaining pH independent dissolution. Molecular modelling (MM) was carried out in order to understand the GPZ:β-CD orientation and GPZ group interactions with the cyclodextrin cavities. Molecular modelling suggested interaction of cyclohexyl and methylpyrazinecarboxamido groups of GPZ with the cyclodextrin cavities and favorability of head to tell (HT) orientation due to its minimum interaction energy. XRD and DSC study showed partial amorphization of GPZ. Scanning electron microscopy (SEM) studies revealed formation of new solid phase of ternary complex indicating partial amorphization. Tablets prepared using optimized ternary complex of GPZ showed immediate and pH independent drug release as compared to marketed tablet formulation and plain drug.

Keywords: Glipizide, Immediate release, β-Cyclodextrin, Ternary complex, pH independent

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INTRODUCTION

In recent drug discovery and development, the number of drug candidates exhibiting low solubility has increased tremendously. Around 70% of the emerging drug candidates possess poor aqueous solubility[1]. At present approximately 40% of the marketed immediate release (IR) oral drugs are categorized as practically insoluble (<100 µg/mL) limiting their overall therapeutic potential[2]. The aqueous solubility of a drug is a critical determinant of its dissolution rate. Enhancement of the dissolution rate of the drugs is thought to be a key factor for improving the bioavailability of BCS class II drugs which generally display dissolution-limited absorption[3].

Existing Pharmaceutical technology provides many approaches to enhance the dissolution rate of poorly soluble drugs such as crystal modification, particle size reduction, self-emulsification, pH modification, amorphization and cyclodextrin complexation which are considered to be effective for improving the dissolution behavior of BCS class II drugs[4].

Cyclodextrins and their derivatives increase the apparent solubility of poorly water-soluble drugs by forming inclusion complexes. Solubility enhancement ratio by complexation has been reported to be 1.1–46-fold compared with that of control formulations of crystalline and lyophilized drugs[5]. Cyclodextrin with ternary components which modify pH in solid dosage forms are considered to be an augmented approach for ionizable drugs to further improve their solubility and dissolution rate. The pH induced by the ternary component significantly influences the saturation solubility of an ionizable drug by dissociation, and can alter the micro environmental pH[6,7]. Arginine and Tromethamine were evaluated as the ternary agents in the complex.

GPZ which is a poorly aqueous soluble oral hypoglycemic agent belonging to BCS class II and is one of the most commonly prescribed drug for the treatment of patients with type II diabetes mellitus[8]. Its low aqueous solubility and poor dissolution in upper gastric fluid limits its therapeutic application by delaying rate of absorption and finally the onset of action. Its dissolution is considered to be a rate determining step in its absorption from the gastrointestinal tract.

Particle size reduction with ternary cyclodextrin complexation approach was used to increase dissolution rate of GPZ. Hence the primary objective of this research work was to developed cyclodextrin based ternary system by employing high energy air jet milling which is highly efficient and can be industrial scalable technique for complexation. Air jet milling technique enabled effective inclusion complexation, with micronization followed by partial amorphization

of ternary complex.

Molecular modeling study gives an insight about the groups of GPZ interacting with β-CD and their actual Head to Head (HH), Tell to Tell (TT) or Head to Tell (HT) orientation with GPZ . Their potential and interaction energies with optimum distance of interactive groups to cyclodextrin cavities were calculated by Schrödinger maestro software and most preferred orientation for complexation was studied.

MATERIALS AND METHODS

Materials

Glipizide (GPZ) was provided by USV Limited, India. B-CD was procured from Signet Chemical Corporation, India. Arginine was purchased from SD Fine Chemicals, tromethamine procured from Merck India. Other excipients like lactose DCL21, microcrystalline cellulose (PH101), Ac-Di-Sol (SD-711), colloidal silicon dioxide (Aerosil 200) and magnesium stearate were supplied by Signet Chemical Corporation, India. Solvents used were of HPLC grade; other chemicals used were of analytical grade.

Methods

Phase solubility studies

Phase solubility study was carried out based on Higuchi and Connors method[9]. In brief, an excess amount of GPZ in purified water, simulated gastric fluid pH 1.2 and pH 6.8 pH buffer containing various concentrations of β-CD (0-12 mM) was shaken for 24 hrs at 37^0C on a water bath shaker rotating at 100 rpm (Orbital Shaker Incubator, Remi, India). The equilibrated aliquots were filtered through 0.22 μm PVDF filter (Millipore, India) followed by dilution and analyzed spectrophotometrically at 276 nm (Jasco V-530 UV/Vis Spectrophotometer). The stability constant K_s for 1:2 GPZ-βCD was calculated using following equation 1:

$$Ks(1:2) = \frac{\text{Slope}}{\text{So}\,(1-\text{Slope}\,)} \qquad (1)$$

Where, S_0 is the solubility of the GPZ in the absence of β-CD.

Preparation of solid complexes

The solid complexes of GPZ and β-CD (binary system) with or without ternary agent were prepared by the following techniques. The stoichiometric ratio of binary system was kept 1:2, whereas that of ternary systems was kept as 1:2:1.

The physical mixture

Binary mixtures were prepared by gently mixing drug: β-CD. These mixtures were passed through an (60#) sieve prior to use.

Kneading

GPZ and β-CD in stoichiometric ratio of 1:2 were triturated using small volume of ethanol and water (1:1) to obtain a thick paste, which was kneaded for 30 minutes and then dried at 40^0C in an oven. The dried mass was then passed through 40 mesh sieve[10].

Freeze drying

GPZ and β-CD was accurately weighed and dissolved in distilled water (10 mL). The whole solution was stirred on magnetic stirrer for 30 min. The resulting solution was then freeze dried. The dried powder was passed through 80 mesh sieve and stored in a desiccator until further evaluation.

Ball Milling

The ball milling was performed in a high-energy planetary mill (Kalweka HD-410AC, India) at room temperature. Stainless steel milling jars (500 mL) containing an appropriate varying sizes of balls (15, 25, 50 mm) of the same material were employed[11]. Twenty gram each of GPZ: βCD (1:2), GPZ: β-CD:Arginine (1:2:1) and GPZ: βCD: Tromethamine (1:2:1) were milled individually at 50 rpm for 6 hours with a ball : sample weight ratio of 20:1.

Air jet milling

In case of Air jet milling pre-mixing of powders was performed by geometric mixing followed by mixing in a double cone blender (Kalweka HD-410AC, India) operated at 25 rpm for 15 min.[12]. Three different premixed powders viz. GPZ : βCD (1:2), GPZ : βCD : Arginine (1:2:1), and GPZ:βCD:Tromethamine (1:2:1), (100 gm. each) were processed in a high energy air jet mill unit (Rucha Pharma, India). Solid feeding rate (SFR) of 5 g/min, feeding pressure (FP) of 20 psi and grinding pressure (GP) in range of 70-80 psi were used. Particle size of binary and ternary air jet milled complex was determined using laser diffraction (LD) method (Mastersizer equipped with Hydro 2000MU, Malvern Instruments, UK). Water with a refractive index (RI) of 1.33 was used as measuring medium.

Molecular modelling

Conformational analysis of GPZ and βCD was performed using Lig Pre module within Schrödinger maestro software keeping rest of the constraints default. Semiempirical calculations were carried out on the host-guest complexes (GPZ:β-CD) and energy minimization was performed within macro model of Schrödinger suit by keeping molecular mechanics force field OPLS 2005 with default constraints. After minimizing host and ligand energies (β-CD and GPZ) their minimum energy conformers were selected for complexation conformational analysis. To locate the centroid of β-CD for complexation, the glycosidic oxygen atoms of β-CD were placed

onto the *XY* plane, and the centres of all atoms were defined. As the ligand (GPZ) is a slightly bend molecule it was divided into two equal parts to fix its centroid. In the complex formation, the ligand enters through the secondary hydroxyl groups of β-CD into the cavity, either head or tail where the initial distance between the centroids of the ligand and that of the CDs is kept at 10 Å[13]. The distance was decreased by 2.0 Å (i.e.10.0, 8.0, 6.0, 4.0, 2.0, 0.0) whereby the ligand came close to the secondary hydroxyl groups of β-CD with decreasing distance. All complexes were optimized with respect to their potential energy. The lowest potential energy complex was selected for further complexation conformational analysis. At another end of ligand second β-CD with its centroid located by previously mentioned procedure was brought closer to the centroid of ligand complexed with β-CD having minimum energy. The distance was similarly decreased by 2.0 Å (i.e.10.0, 8.0, 6.0, 4.0, 2.0, 0.0) as in the former case. Three possible orientations of both the β-CD units in the complex, head-to-head (HH), head-to-tail (HT), and tail-to-tail (TT) were studied and their lowest interaction energies were calculated by using following equation 2,

$$\Delta E_{int} = E_{com(1:2)} - E_{\beta CD} - E_{ligand} \qquad (2)$$

Where,

ΔE_{int} is the total interaction energy of the complex, $E_{com(/1:2)}$ is where E com(1:2) is the complex energy in 1:2 stoichiometry, $E_{\beta\text{-CD}}$ is the β-CD energy, and E_{ligand} is the ligand energy.

CHARACTERIZATION OF THE COMPLEX

In-vitro dissolution studies

In-vitro dissolution studies of plane GPZ, physical mixtures of βCD and GPZ, and other binary complexes prepared by different methods were carried out in stimulated gastric fluid (900 ml) at pH 1.2 maintained at 37 ± 0.2^{0}C and stirred at 50 rpm. Samples were withdrawn at 5, 10, 15, 20, 30 and 45 min interval, filtered through whatman filter paper, and analysed spectrophotometrically at 276 nm[14]. The methods of complexation which exhibited significant enhancement in dissolution were further used for formation of complexes involving ternary agents. Arginine and Tromethamine were employed as ternary agents. The ternary complexes obtained by ball milling and air jet milling were compared on dissolution basis as described earlier for selection of the most appropriate method for complex formation.

UV spectroscopy

Solution state complex formation was characterized by UV spectrophotometer[15]. For UV characterization drug concentration was kept constant (2 X 10^{-6} M) and βCD concentration was increased from 2, 4, 6, 8, 10 mM and scan between 200 and 400 nm was performed.

Differential scanning calorimetry

Thermal characteristics of pure GPZ and its complexes were analyzed under dry nitrogen purge (20 mL/min) at a heating rate of 10^0C/min using a Differential Scanning Calorimeter (Perkin Elmer, Pyris-6 DSC, USA).

Powder X-ray diffractometry

Powder X-ray diffraction patterns of pure GPZ and its solid complexes were recorded using Phillips P Analytical X'Pert PRO powder X-ray diffractometer using Ni-filtered, Cu K_α radiation, a voltage of 40 KV and a current of 30 mA. The scanning rate employed was 1^0/min and samples were analyzed between 2θ angles of over 10–40^0.

Fourier transforms infrared (FTIR) spectroscopic analysis

FTIR spectra of plane GPZ and its solid complexes were recorded on a FTIR-5300 Spectrophotometer (Jasco, Japan) employing KBr disk method using Hydrostatic press to prepared compact disc of samples. The scanning range was 4,000–400 cm^{-1}.

^{1}H-NMR spectroscopy

The ^{1}H-NMR spectra of GPZ, β-CD and the solid ternary complex with Arginine prepared by air jet milled were recorded using Ultrashield 700 Plus Bruker (500 MHz) Fourier Transform Nuclear Magnetic Resonance (FTNMR) instrument at 298 K. The spectra were recorded in Deuterated Water (D_2O) solvent systems.

Scanning electron microscopy

Morphological characteristics of complexes were studied using Scanning Electron Microscopy (SEM). The purpose of morphological study was to evaluate the actual size and surface topology of milled complex. The samples were examined in a Jeol Scanning Electron Microscope (JSM-6380 LA) at an acceleration voltage of 10 kV.

Tableting of ternary complex

Solid ternary complexes of GPZ obtained by ball milling and air jet milling were compressed into tablets. MCC PH101, anhydrous lactose and magnesium stearate used as a diluents. PVP K-30 was used as binder with IPA as solvent. Tablet was compressed by using 7.5mm concave punches with weight and hardness was set at 200 mg and 5 kg/cm^2 respectively.

Different pH different media (DPDM) dissolution studies

Dissolution of the air jet milled solid ternary complex incorporated into tablets and Marketed tablet was carried out using USP type II apparatus (Electrolab, India) in different media (900 ml); 0.1N HCl pH 1.2, acetate buffer pH 4.5, phosphate buffer pH 6.8 and water. Dissolution was carried out at 37 ± 0.2^0C and stirred at 50 rpm. Samples were withdrawn at 5, 10, 15, 20, 30

and 45 min interval, filtered through Whatman filter paper, and analyzed spectrophotometrically at 276 nm

RESULTS AND DISCUSSION

Phase solubility studies

Phase solubility study is pre-requisite for the optimization of the inclusion complex as it defines the affinity between β-CD and drug molecule in water[9]. The phase solubility curve of GPZ in the presence of varying concentrations (mM) of β-CD (Figure 1). The curve in water showed a linear increase in solubility of GPZ with an increase in concentrations of β-CD. However the curve in pH 1.2 showed little negative deviation indicating the poor solubility of complex in acidic pH relative to water. Increasing the concentration of βCD increased the solubility of GPZ in pH 1.2 as well as in aqueous medium. The stability constant (Ks) for the βCD-GPZ complex at 37^{0}C, assuming a 1:2 stoichiometry, calculated from the slope of phase solubility was 159.95 M^{-1} in pH 1.2 and 567.43 M^{-1} in water and 773.011 M-1 in 6.8 phosphate buffer which indicated a stable complex formation

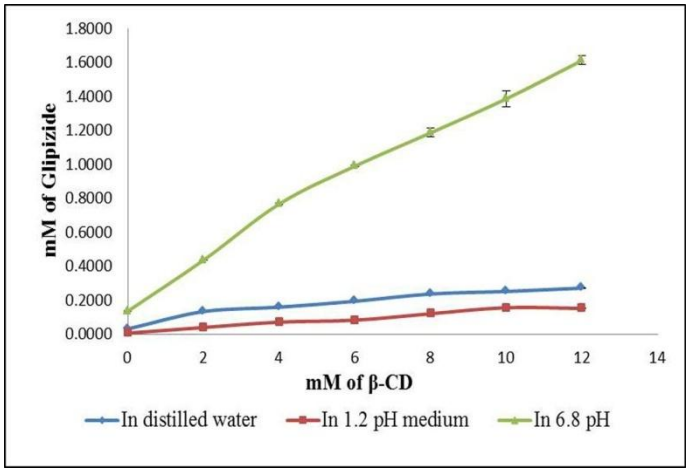

Figure 1: Phase solubility of GPZ in presence of β-CD.

Characterization of solid complexes

By UV spectrophotometry

UV characterization showed that as the concentration of β-CD increased the absorbance λmax of GPZ at 276nm is shifted by1 nm to higher values confirming the possible interaction of aromatic portion of GPZ in solution state with β-CD.

Differential scanning calorimetry

Thermal behaviour of GPZ and its solid complexes along with its individual components are shown in Figure.2. GPZ showed a sharp endotherm at 218.18^{0}C with ΔH= 86.879 J/g whereas β-CD showed a broad endotherm from 65 to 100 ^{0}C, which attributed to the loss of moisture. Being

highly crystalline, arginine also showed a sharp melting peak at 218.18^{0}C. Thermograms of the solid complex prepared by physical mixture showed an exothermic peak at 214.74^{0}C and in air jet milled complex showed at 204 ^{0}C with ΔH=20.3238 J/g which could be correspondingly attributed to the melting of arginine and GPZ. Slightly Lower shift in exothermic peak of GPZ is attributed to the presence of ternary component and micronized having partial amophorous nature, as the melting point of a material is known to shift to the lower scale in the presence of other substances[16].

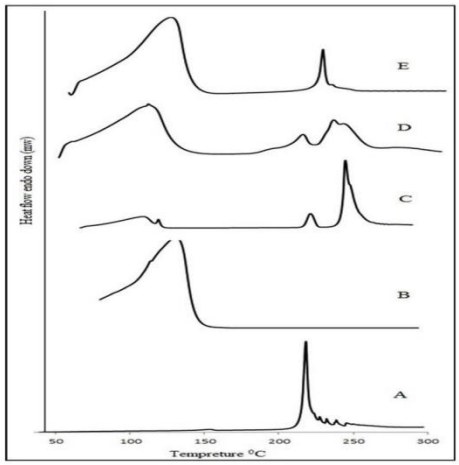

Figure 2: DSC curves of (A)GPZ, (B)β-CD, (C)Arginine, (D) Ternary physical mixture of GPZ: β-CD : Arginine (1:2:1) and (E) GPZ: β-CD: Arginine (1:2:1) air jet milled omplex

Powder X-ray diffractometry (PXRD)

The PXRD patterns of plane GPZ and solid complexes prepared by air jet milled technique along with its individual components (Figure 3).

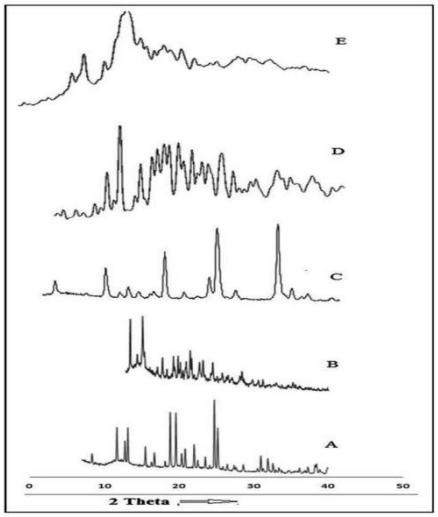

Figure 3: XRD spectra of (A) GPZ, (B) β-CD, (C)Arginine, (D) Ternary physical mixture of GPZ: β-CD : Arginine (1:2:1), (E) GPZ:β-CD: Arginine (1:2:1) air jet milled complex

The diffractograms of GPZ and arginine exhibited a series of intense peaks, which are indicative of their crystalline character. The PXRD of β-CD showed broad intense peaks, indicating presence of crystalline state. An X-ray diffraction pattern of physical mixture was constituted by some of the characteristic peaks of GPZ indicating an absence of stoichiometric complexation. Whereas ternary air jet milled complex showed significant reduction in peak intensity (partial amorphization) thus confirming the ternary complex formation.

Fourier transforms infrared (FT-IR) spectroscopic analysis

The chemical interaction between the drug and the carrier often leads to peculiar changes in the infrared (IR) profile of complexes[17]. An IR spectrum confers the information regarding hydrogen bonding that likely to occur between guest and host. The principal characteristic peaks corresponded to the structural features of GPZ are found due to N–H stretching of sulphinido group at 3325 cm^{-1}, N–H stretching of amido at 3,250, cylohexyl CH_2 stretching at 2,943.3 cm^{-1} and aromatic –CH stretching at 2854.3.The FT-IR spectrum of the β-CD is characterized by intense bands at 3,300–3,500 cm^{-1} due to O–H stretching vibrations. The vibration of the CH=CH and CH_2-CH_2 groups appears in the range of 2,800–3,000 cm^{1}. In IR study solid complex of physical mixture and ternary air jet milled complex showed combination of the peaks. The overlays of IR spectra are presented in (Figure 4).

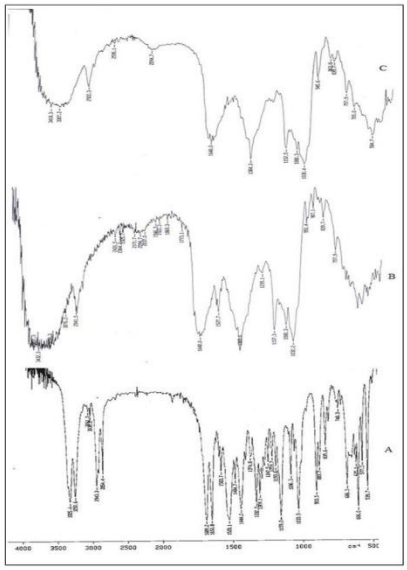

Figure 4: Infrared spectra of (A) GPZ, (B) Ternary physical mixture of GPZ: β-CD: Arginine (1:2:1) and (C) GPZ:β-CD: Arginine (1:2:1) air jet milled complex

The principal peaks of the GPZ were largely affected. GPZ crystals showed a characteristic absorption band at 3325.4, 3250.6 cm^{-1} of N-H stretching and 2943.3, 2854.4 $cm^{-1}$of C-H and CH_2 –CH_2 stretching of aromatic and cyclohexyl group. The FT-IR spectra of ternary solid

complex were compared with the ternary physical mixtures, ternary air jet milled and GPZ. Where stretching of GPZ disappeared along with reduced intensity of the other band. Changes in the characteristic bands of GPZ confirm the existence of the complexes a new compound with different spectroscopic bands. Thus from the IR studies it can be concluded that aromatic and cyclohexyl ring, with functional group of GPZ interact with O–H group of β-CD through Hydrogen bonding.

Nuclear magnetic resonance (NMR) spectroscopy

The chemical shift changes in the ^{1}H-NMR spectra have been used to monitor the complex formation process. If a guest is incorporated into the cyclodextrin cavity, the hydrogen atoms located in the interior of the cyclodextrin cavity (H-3 and H-5) will be considerably shielded by the guest molecule causing a significant up field shift whereas the protons on the exterior surface of the torus (H-1, H-2, H-4 and H-6) will either be unaffected or experience a marginal shift[18]. The different protons of GPZ have been labelled as shown in (Figure 5). The ^{1}H NMR spectrum of β-CD in D_2O consists of six protons the H-3 δ 3.913, a unresolved broad peak consisting of H-5 and H-6 at δ 3.822 and δ 3.808, H-4 triplet appears at δ 3.540 (Table 1).

Table 1: ^{1}H-NMR spectra of Free GPZ and ternary solid air jet milled complex showing shifting of δ values (chemical shift) in free state and in complex state.

Sr. No.	Group involved in Complexation and ^{1}H of Glipizide	δ Free GPZ	δ Complex	δComplex - δ Free GPZ
1	Cyclohexyl ^{1}H (Hg, Hh, Hi, Hj, Hk)	-CH_2= 1.74,1.49 1.21,1.11 1.49,1.46	Very small change in δ values upfield	Not found significant change as β-CD peaks merge. -0.038
	Hf	Hf = 3.54	3.502	
2	Methyl 2- pyrazine ^{1}H	(Ha) -CH_3 = 2.33	2.513	+ 0.183
		(Hb) -CH = 8.70	8.457	-0.2429,
Chemical shifts (ppm) for the protons of β-CD in the free state and in the inclusion complex				
Sr. No.	^{1}H of of β-CD	δ Free β-CD	δ Complex	δComplex - δ Free β-CD
1	H-3	3.913	3.811	-0.102
	H-4	3.534	3.515	-0.0189
	H-5	3.822	3.792	-0.030
	H-6	3.808	3.773	-0.034

It is known that the H-3 and H-5 protons are located in the interior of the β-CD cavity and it is therefore the inclusion of GPZ with β-CD will specifically affect the chemical shifts of these two protons. The complexation of GPZ with β-CD experiences upfield shifts however, it is not the same for all protons. The ^{1}H NMR spectrum of β-CD complex shows H-3 signal of pure β-CD is shifted and most likely merged with that of the unresolved peak of H-5 and H-6 protons (19-20).

H-3 signal Proton showed upfield δ -0.003 ppm. Since H-3 is located in the interior of the cavity hence H-5 signal shows upfield shifts because both are located inside the cavity. The upfield shifts observed for H-3 and H-5 protons confirmed the inclusion inside the cavity.

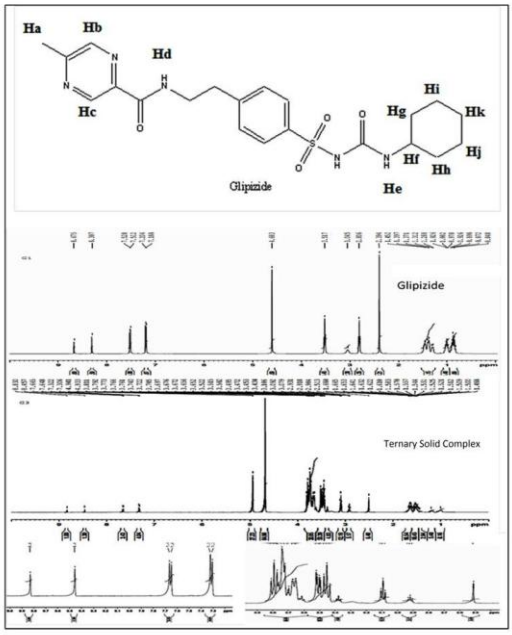

Figure 5: ^{1}H NMR spectra of Free GPZ and ternary solid complex

Molecular modeling

The complexation is solely due to the existence of noncovalent interactions; and when a multitude of noncovalent interactions simultaneously operate, they mutually influence each other (13). Molecular modelling confer understanding of group involve with βCD cavities in 1:2 stoichiometric ratio of (β-CD: GPZ) complex where the cyclohexyl and methylpyrazinecarboxamido group interact with β-CD cavities with formation of hydrogen bonds with primary hydroxyl group of β-CD. Different orientation of β-CD were studied with their optimum distance that gives lowest interactive energy (IE) from each of the centroid of β-CD and GPZ Fig.6. β-CD which are oriented in the HH manner and distance between centroids of the cyclodextrin to centroid of GPZ is around 5Å having IE found to be 938.52 kcal/mole, whereas for HT and TT orientations IE found to be 139.18, 150.35 Kcal/mole having 2Å and 6Å distance respectively. In other, HH and TT orientations IEs are more when compared with HT orientations (Figure 7A., B.,C). In the HT orientation, the Cyclohexyl and methylpyrazine ring buried in the β-CD cavity with an exposed to the primary hydroxyl groups of β-CDs with lowest IEs (Figure 6 and Figure7). Other orientations of β-CDs such as HT and TT are not highly favourable for complexation by considering their IE of β-CD complexes and the H-bond

interaction which prefers the HT orientation.

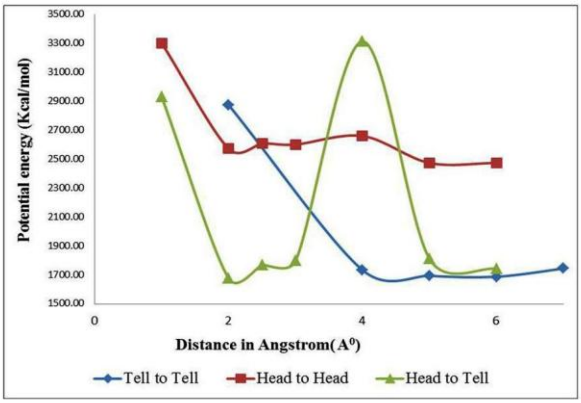

Figure 6: Interaction energy (Kcal/mole) versus distance (Å) of 1:2 stoichiometry complexes of β-CD and GPZ obtained at different orientation

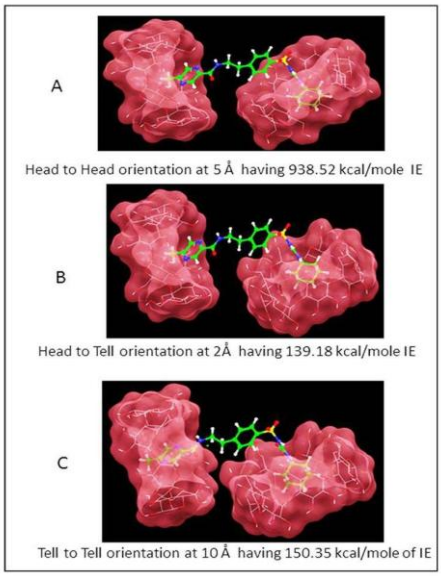

Figure 7: Stable Orientation of GPZ:BCD in 1:2 stoichiometric ratio at varying distance from centroid of GPZ and their respective interaction energy kcal/mole (A)Head to Head, (B) Head to Tell and (C) Tell to Tell

Scanning Electron microscopy

When pure GPZ was micronized in the high energy air jet milled, micronized particles are found to adhere to each other due to strong inter-particle force and form irregular shape agglomerates Figure. 8. In addition to β-CD and arginine reduces the cohesion force among particles; as a result tendency to form agglomerates decreased substantially. SEM images clearly showed the micronized particle with change in surface topology gives idea of change in crystallinity (partially amorphized) of drug because of reduction in particle size ranging from 2-4 µm having uneven crystal shape.

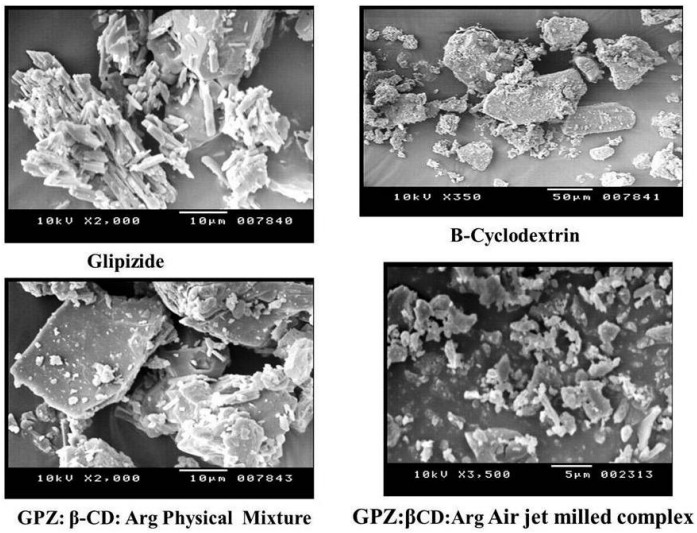

Figure 8: SEM images of plane GPZ, β-CD, ternary physical mixture and air jet milled Complex

In-vitro dissolution studies of complexes

The tablet dissolution profiles of plane GPZ, physical mixture and the GPZ:β-CD (1:2) binary complexes prepared by various methods (Figure 9). According to these results, the binary inclusion complexes prepared by air jet milling and ball milling technique confer release up to 60% and 55% respectively as compare to plane drug which exhibited only 4-5 % drug release in 1.2 pH medium. This may be attributed to better inclusion complexation with its hydrophobic cyclohexyl and methylpyrazinecarboxamido group which are mainly responsible for its low aqueous solubility. In addition reduction in particle size also leads partial amorphization of complex and favors enhanced dissolution. To enhance further dissolution of GPZ binary complexes prepared by ball milling and air jet milling technique were subjected for ternary complexation with ternary components like arginine and Tromethamine (Figure10) Ternary complex GPZ:β-CD:Arg (1:1:1) prepared by Air jet milling technique found to achieve 100 % dissolution with respect to all pH medium than ternary complex prepared by ball milling technique which showed dissolution around 80 % (Figure10). This may attributed to addition of arginine which is better alkalizer than tromethamine and sufficiently maintain high pH in proximity of drug owing to its highly basic guanidinium residue of arginine having pKa 12.48. Guanidinium group which is positively charged in neutral, acidic and even most basic environments, and thus imparts basic chemical properties to arginine. Because of the conjugation between the double bond and the nitrogen lone pairs, the positive charge is delocalized, enabling the formation of multiple H-bonds and ionic bonds with sulfamoyl group of GPZ. Thus

ionization of sulfamoyl group takes place; microenvirmenatl alkaline pH results and therefore improvement in solubility. Further we anticipate that formation of hydrogen bond between drug and arginine further facilitate its solubility. Ternary agent plays crucial role as it adsorb in micronized form on the surface of GPZ to generate micoenvirmental pH in dissolution medium [21,9]. In addition, it significantly influence the saturation solubility of an GPZ by dissociation results improved dissolution rate and confer pH independent release[8]. Optimized tablet formulation with arginine as ternary agent prepared by air jet milled exhibited pH independent dissolution and compare with marketed formulation (Figure 11 and Figure 12).

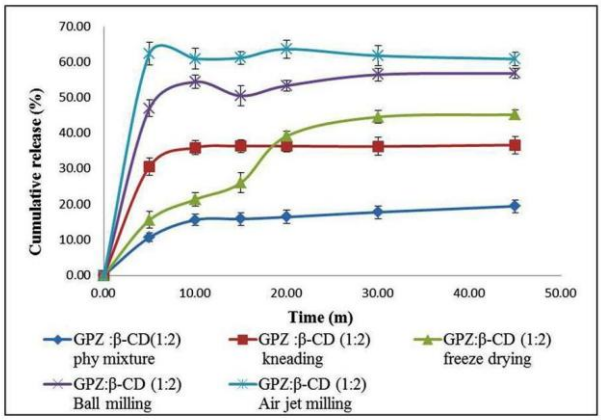

Figure 9: Dissolution of GPZ:βCD (1:2) complex by various technique in 1.2 pH medium.

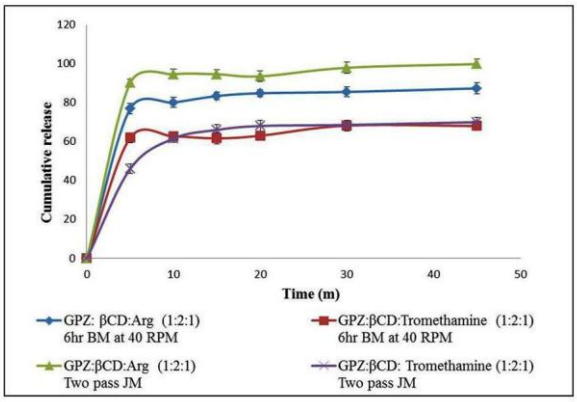

Figure 10: In- vitro Dissolution of ternary complex by Air jet mill and ball milled complex in 1.2 pH medium.

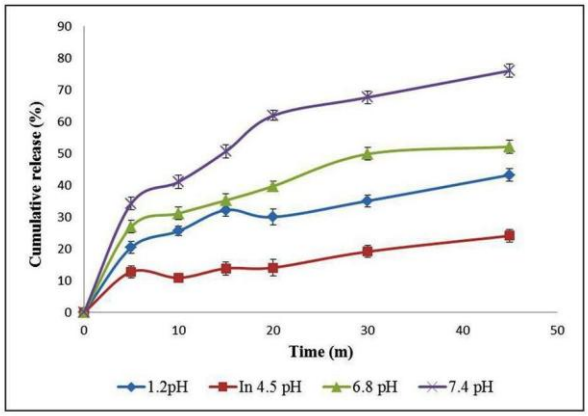

Figure11: In- vitro Dissolution of marketed tablet in diffrent pH medium.

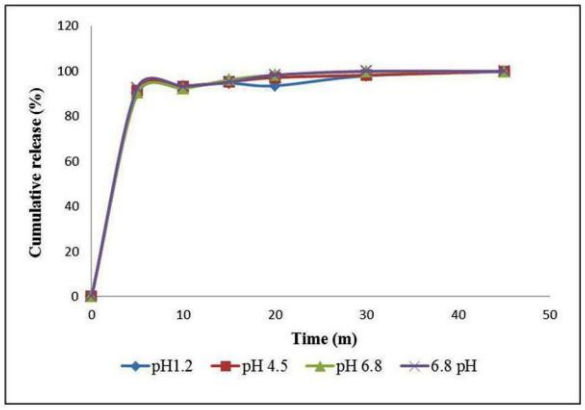

Figure12: In-Vitro Dissolution of optimized GPZ: β-CD: Arginine (1:2:1) Complex prepard by 2 pass air jet milling in different pH medium.

CONCLUSION:

The present study showed improved dissolution of GPZ using a ternary complexation technique prepared by high energy air jet milling. Microenvironmental pH generated in the ternary complex system was found to play a synergistic role not only in solubility enhancement but also

pH independent behavior of GPZ. Arginine as a ternary component appears to be significantly affecting the ternary system by combination of multiple factors and probably offers ideal interaction with GPZ, including specific hydrogen bonding and/or spatial alignment with the host. Ternary complex prepared by high energy air jet milled technique found to be simple, industrially scalable and efficient approach for solubility enhancement of the drug having poorly water and pH dependent solubility.

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