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Research Article

FORMULATION AND *IN VITRO* EVALUATION OF DAPAGLIFLOZIN AND SAXAGLIPTIN BILAYERED TABLETS

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Keywords:
Dapagliflozin, Saxagliptin, Bilayered tablets, Karaya gum, HPMC K15M.

ABSTRACT

Dapagliflozin (DG) is a sodium glucose cotransporter-2 (SGLT-2) inhibitor and Saxagliptin (SG) is a dipeptidyl peptidase-4 (DPP-4) inhibitor. The aim of the present work is to formulate a bilayered tablet (BT) of DG as immediate release (IR) layer and SG as sustained release (SR) layer by direct compression method for the effective treatment of type 2 diabetes mellitus. Type and concentration of superdisintegrant among [sodium starch glycolate (SSG)/Lycoat RS720/ Ludiflash] was optimized to enhance the dissolution rate (DR) of DG from the IR layer of BT. Type and concentration of SR polymer among (Carbapol 940/ Karaya gum/ HPMC K15M) was optimized to extend the release of SG up to 12 h with zero order release profile from the SR layer of BT. It was concluded that the optimization of the ratio of SG: SR polymer (HPMC K15M), had significant effect on extending the release profiles of SG. The ratio of SG: HPMC K15M at 1:18 respectively forms a better matrix for the extending the release of SG up to 12 h from the SR layer of BT. The optimized formulation; BT9 [IR9 (6% w/w Ludiflash as superdisintegrant and SR9 (with 60% HPMC K15M as SR polymer)] releases 100% of DG from the IR layer with in 45 min and extends the release of SG up to 12 h with a better zero order release profile (r^2=0.994). It passes the accelerated stability studies as per ICH guidelines. A combination of these two classes [SGLT-2 inhibitors (DG) and DPP-4 inhibitors (SG)] of glucose-lowering agents and formulating them as a BT is more effective in the treatment and maintenance of type 2 diabetes mellitus.

INTRODUCTION

The main objective of combination therapy is to encourage the utilization of lower doses of drugs to treat patients and also to minimize dose dependent side effect and adverse reactions.[1-2] Bilayered tablets can be a primary option to avoid chemical incompatibilities between different drugs by physical separation, and to enable the development of different drug release profiles [immediate release (IR) with extended release (ER)]. Applications of bilayered tablets are mainly used in the combination therapy; to deliver the loading dose and sustained dose of the same or different drugs and are used to deliver the two different drugs having different release profiles [3]. Type 2 diabetes mellitus is a progressive disease with multiple underlying patho-physiologic defects. Monotherapy alone cannot maintain glycemic control and leads to treatment failure. Ideally, a combination of glucose-lowering agents should have complementary mechanisms of action that address multiple patho-physiologic pathways, can be used at all stages of the disease, and be generally well tolerated with no increased risk of hypoglycemia, cardiovascular events, or weight gain. Two classes of glucose-lowering agents that meet these criteria are Dapagliflozin (DG); a sodium glucose cotransporter-2 (SGLT-2) inhibitors and Saxagliptin (SG); a dipeptidyl peptidase-4 (DPP-4) inhibitors. The aim of this present work is to formulate a bilayered tablet of Dapagliflozin (as IR layer) and Saxagliptin (as SR layer) by direct compression method using various extended release polymers such as HPMC K15M, Karaya gum and

Carbapol 940, for the effective treatment of type 2 diabetes mellitus.

MATERIALS AND METHODS

Materials

Dapagliflozin (DG) and Saxagliptin (SG) were obtained as gift samples from M/s Concord Drugs Ltd., Hyderabad, India. Sodium starch glycolate (SSG), Lycoat RS 720, Ludiflash, Carbapol 940, Karaya gum, HPMC K15M, PVP K30, Microcrystalline cellulose (MCC), Magnesium stearate and Talc were purchased from S.D. Fine-Chem Ltd., Chennai, India. All the excipients used in study are of analytical grade.

Drug-excipient compatibility studies by FT-IR

FT-IR spectra of pure drug(s) and drug: polymer (1:1) physical mixtures were recorded out, in the region of 400-4000 cm^{-1} at spectral resolution of 2 cm^{-1}, by the potassium bromide pellet method using (Shimadzu-1800, Japan) [4]. The interpretation results of FT-IR spectra were shown in Fig. 1 and Table 1.

Calibration curve of Dapagliflozin and Saxagliptin in 0.1N HCl and Saxagliptin in pH 6.8 Phosphate buffer

100 mg of pure drug was dissolved in 100 mL of 0.1N HCl/ pH 6.8 Phosphate buffer (stock solution-I; 1000 µg/mL) and then placed in an sonicator for 10 min, from this 10 mL of solution was taken and the volume was adjusted to 100 mL with 0.1N HCl/ pH 6.8 Phosphate buffer (stock solution-II; 100 µg/mL). The stock solution-II; was suitably diluted with 0.1N HCl/ pH 6.8 Phosphate buffer to obtain the series of working dilutions: 5, 10, 15, 20, 25 and 30 µg/mL of drug solution. The median concentration was scanned for λ_{max} and at the respective λ_{max} working dilutions were analyzed by using a double beam UV-Vis spectrophotometer (PG instruments T60, UK). The standard calibration curve was plotted by taking concentration on X-axis and absorbance on Y-axis was shown in Fig. 2.

Preparation of immediate release layer

DG, superdisintegrants (SSG/Lycoat RS 720/ Ludiflash), PVPK30 and MCC were weighed and co-sifted through sieve No. # 40 (ASTM), blended in a poly bag for 10 min and lubricated with sieve No. # 60 (ASTM) passed magnesium stearate and talc by mixing in the same poly bag, for additional 2-3 min; which is used as upper IR layer [5].

Preparation of sustained release layer

SG, SR polymer (Carbapol 940/ Karaya gum/ HPMC K15M), PVP K30 and MCC were weighed were co-sifted through sieve No. # 40 (ASTM), blended in a poly bag for 10 min and lubricated with sieve No. # 60 (ASTM) passed magnesium stearate and talc by mixing in the same poly bag, for additional 2-3 min; which is used as lower SR layer. Composition of Dapagliflozin (IR) layer and Saxagliptin (SR) layer of bilayered tablets is given in Table 2.

Pre-compression studies

Directly compressible tablet blends of DG-IR layer and SG-SR layer were evaluated for [angle of repose (θ), bulk density (BD), tapped density (TD), Carr's Index (CI) & Hausner's Ratio (HR)]. Limits for powder flow characteristics as per USP are mentioned in Table 3. The consolidated results of pre-compression studies of IR and SR layers were tabulated in Table 4 [6,7].

Compression of bilayered tablets

Bilayered tablets were prepared by direct compression method, initially the SR layer was compressed and later the upper punch was lifted and the blend of IR layer was poured into the die and compressed using (Rimek mini press I, India.) fitted with a 5 mm standard flat circular punches with an average weight of 300 mg and average hardness of 6.5 Kg/cm^2.

Post-compression studies of bilayered tablets

Average weight of tablets

20 tablets (n=20) were randomly selected from each batch and their weight was determined by an electronic balance (Sartorius, Germany).

Thickness

6 tablets (n=6) were randomly selected from each batch and their thickness was measured using a vernier calipers (Mitutoyo Corporation, Japan.),

Hardness

6 tablets (n=6) were randomly selected from each batch and their hardness was measured using a Monsanto hardness tester (Secor, India).

Friability

The friability of the 20 tablets (n=1) from each batch was tested by a friabilator (Roche Friabilator, Germany) at a speed of 25 RPM for 4 min. The tablets were then de-dusted, re-weighed, and percentage weight loss was calculated by the equation below,

$$\% \text{ Friability} = \frac{(\text{Initial Wt.} - \text{Wt.after friability})}{\text{Initial Wt.}} \times 100 \quad \text{Eq. No. 1}$$

Assay

6 tablets from each batch (n=6), were randomly selected from each batch IR and SR layers were separated by scrapping and crushed in a mortar with pestle separately; the quantity of blends equivalent to 100mg of drugs (DG/SG) was suspended in 100 mL of 0.1N HCl in a volumetric

flask and sonicated for 2 min. The dispersion was filtered through 0.45µm membrane filter, suitably diluted with 0.1N HCl and analyzed by a double beam UV-Vis spectrophotometer (Shimadzu-1800, Japan) by measuring absorbance at obtained λ_{max} of the drug (DG/SG). The consolidated results of post compression studies of bilayered tablets are tabulated in Table 5.

In vitro dissolution studies

In vitro dissolution studies of IR layer

For to optimize the composition of IR layer, 6 tablets (n=6) with only compressed IR layer, were randomly selected from each batch and undergone dissolution in the USP-II (paddle) dissolution apparatus (Lab India DS 8000, India), each flask was filled with 900 mL of 0.1N HCl; speed of paddle was maintained at 50 rpm, the temperature was kept constant at 37°C ± 0.5°C. At time points 0, 5, 10,15,20,25,30, 35, 40 & 45 min, 5 mL of dissolution media was withdrawn, filtered through 0.45µm membrane filter, suitably diluted and analyzed at respective λ_{max} of DG using a double beam UV-Vis spectrophotometer (Shimadzu-1800, Japan). Each sample withdrawn was replaced with an equal amount of fresh 0.1 N HCl, to keep the volume constant. *In vitro* dissolution profiles of DG-IR tablets were shown in Fig. 3.

***In* vitro dissolution studies of bilayered tablets**

Bilayered tablets containing optimized IR layer and varying SR layers *in* vitro dissolution studies were carried out by randomly selecting 6 tablets (n=6) from each batch using USP dissolution apparatus type II/ paddle (Lab India DS 8000, India) in 900 mL of 0.1N HCl for first 2 h and in 900 mL of pH 6.8 Phosphate buffer up to 12 h. Speed of paddle was maintained at 50 RPM, the temperature was kept constant at 37°C ± 0.5°C. Samples were collected at time points 0, 30 (both DG & SG), 60, 120, 180, 240, 300, 360, 420, 480, 540, 600, 660, 720 min, 5 mL of dissolution media was withdrawn, filtered through 0.45µm membrane filter, suitably diluted and analyzed at respective λ_{max} of DG & SG at 30 min and for SG at other time points using a double beam UV-Vis spectrophotometer (Shimadzu-1800, Japan). Each sample withdrawn was replaced with an equal amount of fresh 0.1N HCl, to keep the volume constant. *In vitro* dissolution profiles of DG/SG bilayered tablets were shown in Fig. 4.

In vitro dissolution kinetics analysis of bilayered tablets

The *in vitro* drug release data of all batches were fitted into Zero order, First order, Higuchi and Korsemeyer-Peppas models to ascertain the drug release kinetics [8]. The drug release from the hydrophilic matrix whether depends on drug's concentration or not was explained by zero and first order models. Higuchi model describes whether the drug release is predominantly by diffusion or not. The Korsemeyer- Peppas model further explains the mechanism of diffusion by plotting the first 60% of drug release. The respective models were defined by the equations below.

Zero order: Qt=Q0+K0t Eq. No. 2

First order: Log Q = Log Q0 – K1t /2.303 Eq. No. 3

Higuchi model: Qt = KH t1/2 Eq. No. 4

Korsemeyer-peppas model: Mt/Mα = K t n Eq.No. 5

Where Q_t is the amount of drug dissolved at time, *t*; Q_0 is the initial amount of drug in the solution at time *t=0*, *Q* is the amount of drug remaining at time, *t*; M_t/M_α is the fraction of drug released at time, *t* and n is diffusion exponent. K_0, K_1, K_H and *K* refer to the rate constants of respective kinetic models. Drug release mechanisms based on n-values, for cylindrical shape, as per Korsmeyer-Peppas model, were tabulated in Table 6. The consolidated drug release kinetic data of SR layer of DG/SG bilayered tablets were tabulated in Table 7.

Accelerated stability studies on optimized formulation

Optimized formulation (BT9 or IR9/SR9), of 20 tablets in 10 CC HDPE pack up to 3 months were carried according to International Conference on Harmonization (ICH) guidelines by placing in a humidity chamber (NSW-175, Narang Scientific work, India) maintained at 45°C ± 2°C and 75% ± 5% RH [9]. At the end of every month up to 3 months, the samples were withdrawn and evaluated for post compression studies. The consolidated results of accelerated stability studies were tabulated in Table 8. Comparative *in vitro* dissolution profiles of initial and accelerated stability samples were shown in Fig. 5. The chemical stability of drug in the 3M-accelerated stability sample of optimized formulation (BT9); which will influence the *in vitro* and *in vivo* dissolution characteristics was investigated using FT-IR studies. The FT-IR spectra were recorded out in the region of 400-4000 cm^{-1} at spectral resolution of 2 cm^{-1}, by the potassium bromide pellet method using (Shimadzu-1800, Japan) and the comparative FT-IR spectra of optimized BT9-Initial and 45°C/75RH-3M were shown in Fig. 6.

Table 1: Interpretation of Dapagliflozin and Saxagliptin (pure drugs) FT-IR spectra

Dapagliflozin			Saxagliptin		
Function group	**Standard Frequency (cm^{-1})**	**Observed Frequency (cm^{-1})**	**Function group**	**Standard frequency (cm^{-1})**	**Observed Frequency (cm^{-1})**
C=O Carbonyl Stretching	1680-1760	1244	N-H stretching	3000-3700	3433.12
-OH Stretching	3200-340	3390.12	C-H stretching	2850-2960	2919.18
C=C Aromatic stretching	1620-1680	1611.92	-OH stretching	3200-3400	3301
			C-N stretching	1600-170	1619.77
			C-O stretching	900-1300	1033.8

Table 2: Composition of Dapagliflozin (IR) layer and Saxagliptin (SR) layer of bilayered tablets

Dapagliflozin (IR) tablets									
Ingredients	**IR1**	**IR2**	**IR3**	**IR4**	**IR5**	**IR6**	**IR7**	**IR8**	**IR9**
Dapagliflozin	10	10	10	10	10	10	10	10	10
SSG	3	6	9	-	-	-	-	-	-
Lycoat RS 720	-	-	-	3	6	9	-	-	-
Ludiflash	-	-	-	-	-	-	3	6	9
PVP K30	20	20	20	20	20	20	20	20	20
MCC	107	104	101	107	104	101	107	104	101
Mg. stearate	6	6	6	6	6	6	6	6	6
Talc	4	4	4	4	4	4	4	4	4
Total	**150**	**150**	**150**	**150**	**150**	**150**	**150**	**150**	**150**

Dapagliflozin (IR) and Saxagliptin (SR) bilayered tablets									
Ingredients	**BT1**	**BT2**	**BT3**	**BT4**	**BT5**	**BT6**	**BT7**	**BT8**	**BT9**
IR9	**150**	**150**	**150**	**150**	**150**	**150**	**150**	**150**	**150**
Saxagliptin	5	5	5	5	5	5	5	5	5
Carbapol 940	30	60	90	-	-	-	-	-	-
Karaya gum	-	-	-	30	60	90	-	-	-
HPMC K15M	-	-	-	-	-	-	30	60	90
PVP K30	20	20	20	20	20	20	20	20	20
MCC	q.s.	q.s.	q.s.	q.s.	q.s.	q.s.	q.s.	q.s.	q.s.
Talc	4	4	4	4	4	4	4	4	4
Mg.stearate	6	6	6	6	6	6	6	6	6
Total	**300**	**300**	**300**	**300**	**300**	**300**	**300**	**300**	**300**

Table 3: Limits for powder flow characteristics (as per USP)

Flow Character	AR (°)	CI (%)	HR( )
Excellent	25–30	≤ 10	1.00-1.11
Good	31–35	11-15	1.12-1.18
Fair	36–40	16-20	1.19-1.25
Passable	41–45	21-25	1.26-1.34
Poor	46–55	26-31	1.35-1.45
Very Poor	56–65	32-37	1.46-1.59
Very, very Poor	>66	> 38	> 1.60

Table 4: Results of pre-compression studies of Dapagliflozin (IR) layer and Saxagliptin (SR) layer

Dapagliflozin (IR) layer						Saxagliptin (SR) layer					
F. Code	AR	BD (gm/cm²)	TD (gm/cm²)	CI (%)	HR	F. Code	AR	BD (gm/cm²)	TD (gm/cm²)	CI (%)	HR
IR1	23.54±0.38	0.384±0.012	0.424±0.026	10.81	1.12	SR1	33.56±1.08	0.58±0.34	0.68±0.30	14.48	1.18
IR2	23.98±0.25	0.388±0.024	0.438±0.018	11.11	1.12	SR2	33.38±1.01	0.62±0.25	0.83±0.28	12.51	1.16
IR3	24.32±0.24	0.365±0.034	0.424±0.022	13.15	1.15	SR3	28.63±0.86	0.63±0.45	0.88±0.19	15.20	1.19
IR4	22.42±0.29	0.388±0.017	0.438±0.029	13.51	1.15	SR4	28.40±0.89	0.68±0.28	0.86±0.54	09.48	1.12
IR5	23.95±0.22	0.389±0.012	0.424±0.028	10.81	1.12	SR5	32.15±0.94	0.65±0.51	0.85±0.24	13.18	1.18
IR6	24.43±0.23	0.368±0.012	0.424±0.019	13.15	1.15	SR6	33.53±0.89	0.63±0.12	0.86±0.18	14.68	1.19
IR7	22.34±0.28	0.318±0.032	0.418±0.037	11.01	1.16	SR7	31.26±0.99	0.62±0.22	0.69±0.53	10.28	1.13
IR8	23.41±0.26	0.353±0.026	0.448±0.019	13.21	1.13	SR8	33.46±0.98	0.68±0.46	0.88±0.82	09.22	1.12
IR9	24.05±0.12	0.369±0.014	0.414±0.032	10.41	1.11	SR9	32.60±0.64	0.61±0.16	0.68±0.24	13.06	1.18

Table 5: Results of post-compression parameters of Dapagliflozin (IR) and Saxagliptin (SR) bilayered tablets

F. Code	Avg. wt. (mg)	Thickness (mm)	Hardness (Kg/cm²)	Friability (%)	Assay (%)	
					DG	SG
BT1	300±1.09	4.42±0.3	6.31±0.40	0.18	98.18±0.90	95.24±0.49
BT2	299±0.94	4.41±0.5	6.52±0.36	0.14	96.42±0.40	98.41±0.62
BT3	301±0.59	4.38±0.6	6.11±0.16	0.18	95.90±0.90	98.96±1.06
BT4	300±1.01	4.43±0.4	6.25±0.22	0.32	98.88±0.10	96.90±0.54
BT5	300±1.36	4.44±0.4	6.18±0.18	0.12	96.22±1.15	96.19±0.54
BT6	301±1.58	4.41±0.5	6.14±0.04	0.23	95.44±0.80	98.68±0.16
BT7	300±0.49	4.42±0.8	6.31±0.01	0.15	95.09±2.15	98.88±0.95
BT8	300±1.46	4.40±0.9	6.50±0.62	0.13	98.18±0.90	99.58±1.49
BT9	300±0.95	4.39±0.9	6.42±0.14	0.18	99.14±1.45	96.38±1.21

Table 6: Diffusion exponent and drug release mechanisms for cylindrical shape (Korsemeyer-Peppas model)

Diffusion exponent (n)	Drug release Mechanism
0.45	Fickian diffusion
0.45 < n <0.89	Anomalous (Non- Fickian) diffusion
0.89	Case II transport
n > 0.89	Super Case II transport

Table 7: *In vitro* drug release kinetic analysis of bilayered tablets

F. Code	Zero	First	Higuchi	Krosmeyer-Peppas	
	r^2	r^2	r^2	r^2	n
BT1	0.986	0.741	0.926	0.934	0.892
BT2	0.955	0.944	0.931	0.928	0.989
BT3	0.955	0.618	0.95	0.874	0.834
BT4	0.988	0.671	0.963	0.987	0.754
BT5	0.987	0.741	0.874	0.76	0.814
BT6	0.98	0.681	0.909	0.802	0.763
BT7	0.991	0.677	0.927	0.779	0.782
BT8	0.987	0.868	0.914	0.784	0.781
BT9	0.994	0.868	0.942	0.863	0.792

Table 8: Results of post-compression parameters of accelerated stability samples of optimized Dapagliflozin (IR) and Saxagliptin (SR) bilayered tablets (BT9)

Time duration	Avg. wt. (mg)	Thickness (mm)	Hardness (kg/cm²)	% Friability (%)	Assay (%)	
					DG	SG
Initial	300±0.95	4.39±0.90	6.42±0.14	0.18	99.14±1.45	96.38±1.21
1 month	299.8±0.21	4.34±0.12	5.94±0.11	0.24	98.88±0.10	96.23±0.18
2 month	300.5±0.08	4.32±0.21	5.82±0.20	0.32	98.18±0.90	96.31±0.21
3 month	300.4±0.70	4.23±0.23	5.78±0.18	0.41	96.42±0.40	96.26±0.12

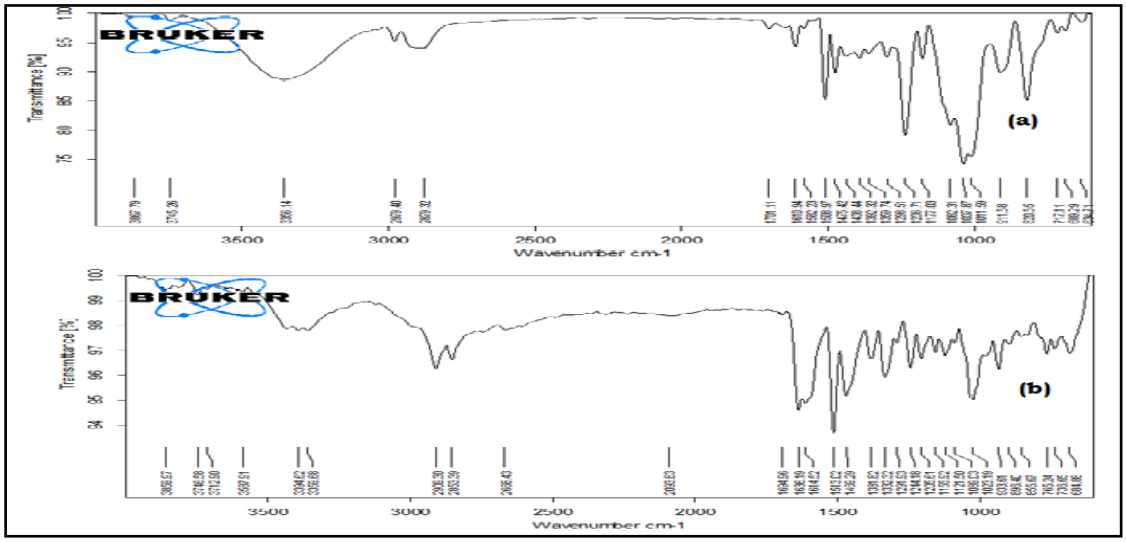

Fig. 1: FT-IR spectra of a) Dapagliflozin (pure drug); b) Saxagliptin (pure drug)

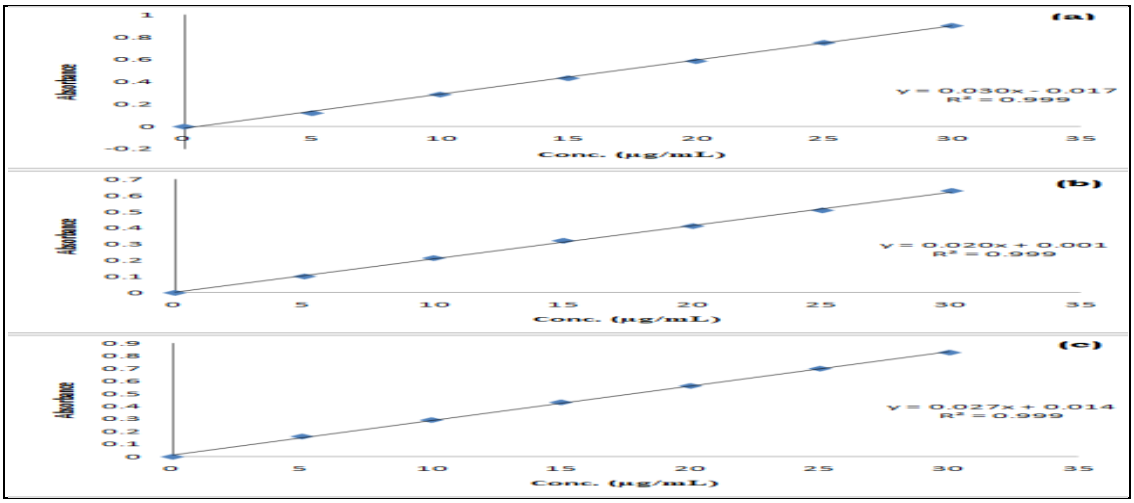

Fig. 2: Calibration curve of a) Dapagliflozin in 0.1N HCl at 222 nm; b) Saxagliptin in 0.1N HCl at 214 nm & c) Saxagliptin in pH 6.8 Phosphate buffer at 214 nm

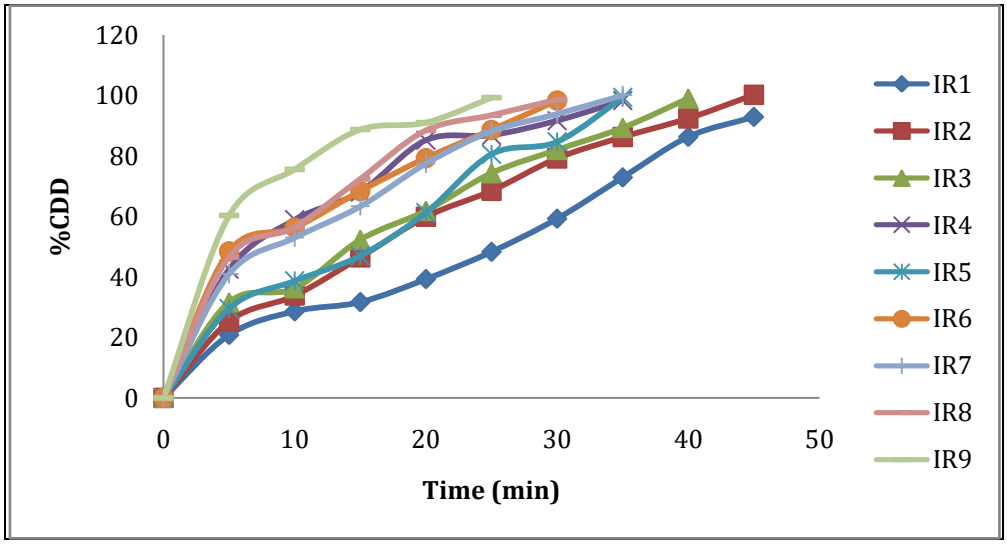

Fig. 3: Comparative *in vitro* dissolution profiles of Dapagliflozin immediate release tablets

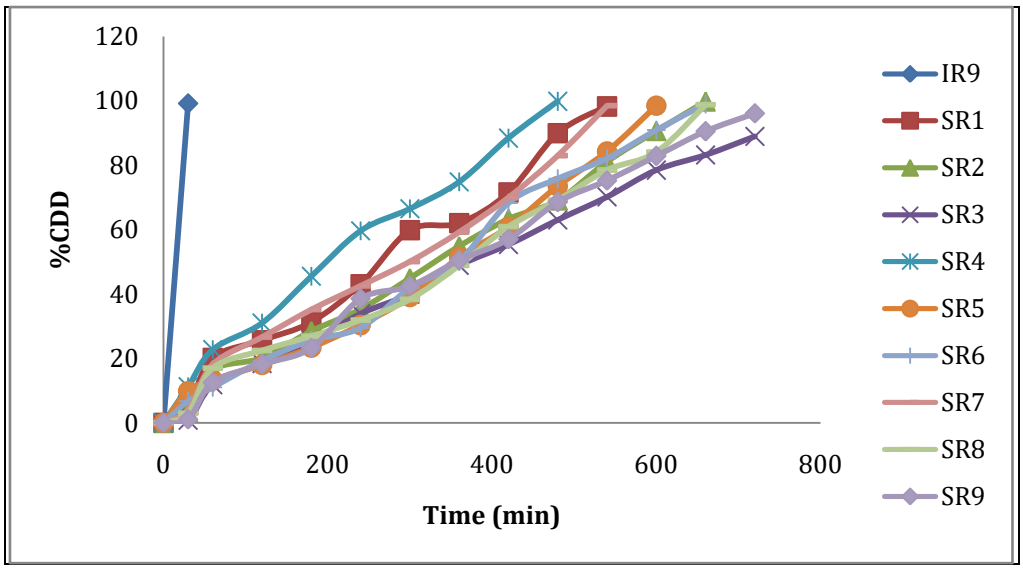

Fig. 4: Comparative *in vitro* dissolution profiles of Dapagliflozin (IR) and Saxagliptin (SR) bilayered tablets

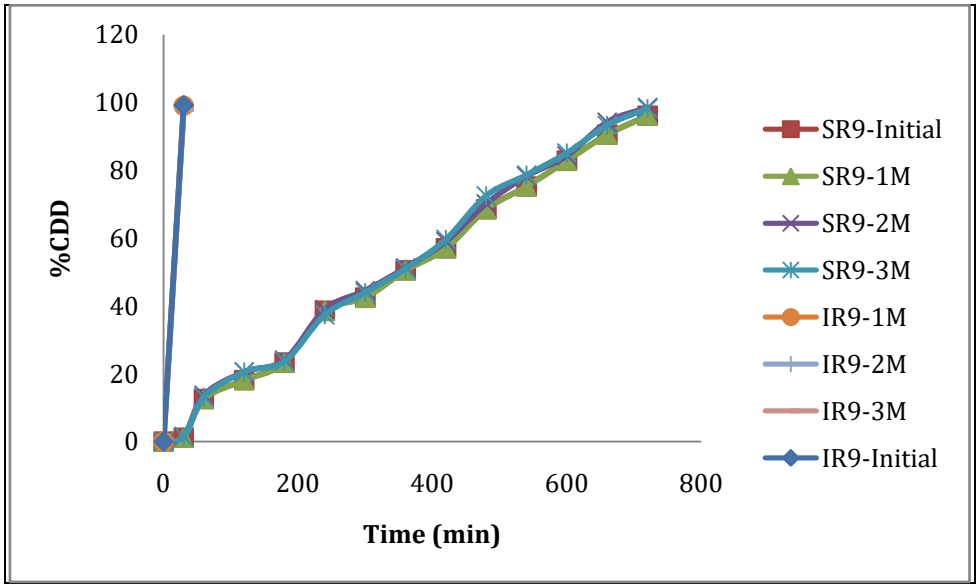

Fig. 5: Comparative *in vitro* dissolution profiles of accelerated stability studies of optimized Dapagliflozin (IR) and Saxagliptin (SR) bilayered tablets (BT9)

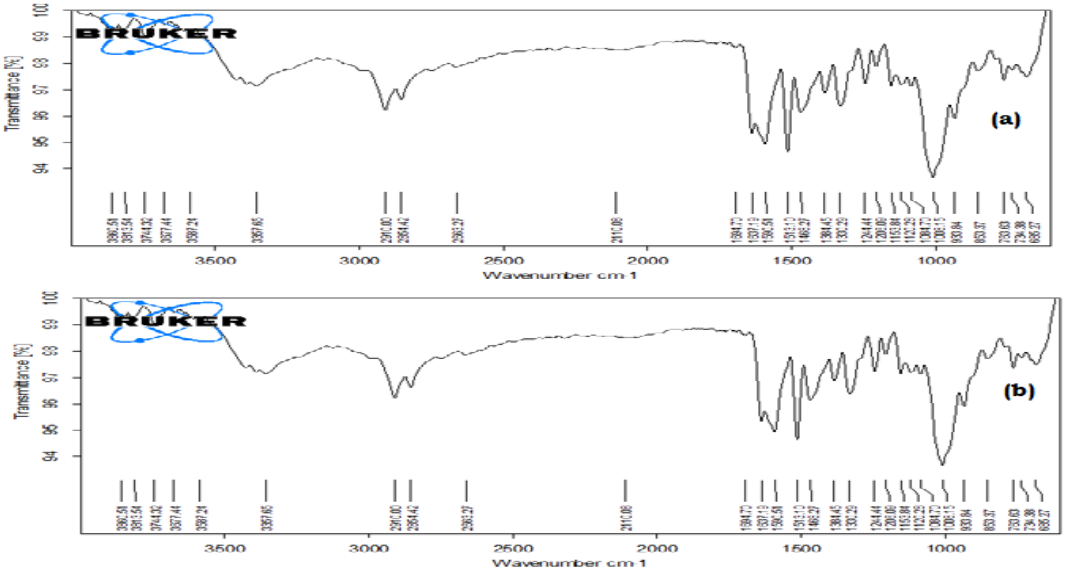

Fig. 6: Comparative FT-IR spectra of optimized formulation a) BT9-Initial b) BT9-40°C/75%RH-3M accelerated stability samples

RESULTS AND DISCUSSION

Drug-excipient compatibility studies by FT-IR

An interpretation of FT-IR spectrum of DG and SG (pure drugs) reveals that the IR bands of pure drug and drug(s) + excipients show no significant shifts or reduction in intensity of the FT-IR bands. Hence there was no incompatibility problem between the drug and excipients used in the study.

Calibration curve of Dapagliflozin and Saxagliptin

λ_{max} of DG in 0.1N HCl; SG in 0.1N HCl and SG in pH 6.8 Phosphate buffer are 222 nm, 214 nm and 214 nm respectively. The standard curves are following linearity with a regression coefficient of (r^2=0.999). They are obeying the Beer's law in the conc. range of 0-30 µg/mL. Lower standard deviation (SD) values ensured reproducibility of the method. As the excipients used in the study were not interfering and good % recovery of drug(s) indicates this spectrophotometric method was suitable for the estimation of drug(s) in dissolution studies and % assay of formulations.

Pre-compression studies

The directly compressible blends of IR layer of DG, reveals that the angle of repose was found between 22.34° ± 0.28 to 24.32° ± 0.24, Hausner's Ratio between 1.11 to 1.16 and Carr's index between 10.41 to 13.51 %. The directly compressible blends of ER layer of SG, reveals that the angle of repose was found between 28.40 ° ± 0.89 to 33.56 ° ± 1.08, Hausner's Ratio between 1.12 to 1.19 and Carr's index between 09.22 to 15.20 %. The micromeritic studies indicate a good flow and compression characteristic of all the IR and SR blends as per USP limits as mentioned in Table 3. In these IR & ER directly compressible blends MCC is used as diluent, which imparts good flow and compressibility to the blends [10].

Post-compression studies of bilayered tablets

Reveals that the average weight of tablets was found to be 299 ± 0.94 to 301 ± 1.58 mg. The average thickness of tablets was found to be 4.38 ± 0.6 to 4.44 ± 0.4 mm. The average hardness of the tablets ranges between 6.11 ± 0.16 to 6.52 ± 0.36 Kg/cm^2, indicating satisfactory mechanical strength. The % weight loss in the friability test ranges from 0.28 to 0.48 %, which was NMT 1 % as per pharmacopoeia limits indicating a good mechanical resistance of tablets. % Assay of DG-IR layer all the batches is within 95.09 ± 2.15 to 99.14 ± 1.45 % and of SG-SR layer all the batches is within 95.24 ± 0.49 to 98.88 ± 0.95 % of the labeled content, indicating the content uniformity of drug(s) in both IR and SR layers.

In vitro dissolution studies of IR layer

For the optimization of the composition of IR layer *In vitro* dissolution studies of compressed IR layers alone of formulations IR1-IR9 were conducted in 0.1N HCl up to 45 min. Among all the formulations IR9 (6% w/w Ludiflash as superdisintegrant) shows the better dissolution efficiency at 30 min (DE30). As the concentration of superdisintegrant increases, tablet dissolution rate (DR) enhances. Among the used superdisintegrant, their order in enhancing the DR is SSG < Lycoat RS 720 < Ludiflash. Hence IR9 is selected as optimized one and in the formulation of all bilayered tablets, its composition is taken as IR layer.

In vitro dissolution studies of bilayered tablets

In vitro dissolution studies of bilayered tablets containing optimized IR layer (IR9) and varying compositions of SR layers were carried out in 0.1N HCl for first 2 h and in pH 6.8 phosphate buffer up to 12 h. The cumulative % drug dissolved values of DG is similar in all the formulations at 45 min is identical due to its similar composition. Related to the release profiles SG from SR layer, as the concentration of SR polymer increases (Carbapol 940/Karayagum/HPMCK15M) there is an increased viscosity of the gel of matrix and decrease in the effective diffusion coefficient of the drug [11]. Other factors that may contribute to differences in drug release profiles include; differences in water penetration rate, water absorption capacity, polymer swelling and drug: polymer ratio.[12] Among all factors, drug: polymer ratio is important factor affecting the rate of drug release from the matrix, which has to be optimized. Among all the formulations; BT9 (with 60% HPMC K15M as SR polymer) extends the release of SG up to 12 h with a better zero order release profile.

In vitro dissolution kinetics analysis

Among all the formulations; BT9 (with 60% HPMC K15M as SR polymer), fitted best to the zero order kinetics (as zero order, r^2 = 0.994), indicating the drug release from the matrix does not depends on drug's concentration. Drug release process is predominantly by diffusion (as Higuchi, r^2= 0.942; i.e. > 0.9); and the mechanism of diffusion is by non-Fickian diffusion [as Korsemeyer-Peppas, diffusion coefficient (n)= 0.792] (when; 0.45 < n <0.89 then non-Fickian diffusion.

Accelerated stability studies of optimized formulation

As there were no significant differences in post compression studies (weight variation, thickness, hardness, friability and *in vitro* dissolution studies) of initial and accelerated stability samples of optimized formulation BT9 in the final up to 3 months, it passes the test for stability as per ICH guide lines. Comparative FT-IR spectra of optimized BT9-Initial and 40°C/75%RH-3M, reveals there is no significant change in the functional groups peaks of the DG and SG due to interaction with polymers and other excipients in the accelerated stability studies.

CONCLUSION

In the view of above findings, optimization of type and concentration of superdisintegrants (SSG/ Lycoat RS 720/ Ludiflash) in enhancing the DR of DG from the IR layer of BT was understood. Optimization of type and conc. of SR polymers (Carbapol 940/ Karaya gum/ HPMC K15M) in extending the release of SG from the SR layer of BT was better understood. It was further concluded that the optimization of the ratio of Drug (SG): SR polymer (HPMC K15M), had significant effect on extending the release profiles of SG up to 12 h with zero order release profile. Among the three SR polymers, (SG: HPMC K15M in the ratio 1:18) respectively forms a better matrix for the extending the release of SG up to 12 h from the SR layer of BT. The optimized formulation; BT9 [IR9 (6 % w/w Ludiflash as superdisintegrant and SR9 (with 60% HPMC K15M as SR polymer)] releases 100% of DG from the IR layer and extends the release of SG up to 12 h with a better zero order release profile (r^2=0.994). A combination of these two classes [SGLT-2 inhibitors (DG) and DPP-4 inhibitors (SG)] of glucose-lowering agents and formulating them as a bilayered tablet of DG as IR layer and SG as SR layer is more effective in the treatment and maintenance of type 2 diabetes mellitus.

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