

Formulation development and optimization: Encapsulated system of atenolol and Glyburide

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Abstract: Purpose: Objective of this study is to develop; tablet- in- a capsule system, to deliver Atenolol 25mg and Glyburide 5mg in the hard gelatin capsule. In order to improve patient compliance and reduce problems associated with complex therapeutic regimen Atenolol (cardio-selective beta-blocker) and Glyburide (anti-diabetic; sulfonyleurea) are commonly, prescribed to the diabetic hypertensive patient. Method: In present work six different formulations of Atenolol (AF1-AF6) and Glyburide (GF1-GF6) were prepared by direct compression method using Avicel, Lactose DC, Crospovidone and Magnesium Stearate in different proportions and encapsulated in hard gelatin shells. Post compression parameters i.e. weight variation, diameter variation, thickness variation, hardness variation, % friability, disintegration, % drug release were determined at different pH 1.2, 4.5 and 6.8, and subjected to dissolution profile comparison through similarity factor (f_2). Results: Stability studies were performed and shelf lives were calculated by R-Gui Stab R console 2.15.2 and determined to be 15 and 27 months for Atenolol and Glyburide respectively. The percentage drug contents of Atenolol and Glyburide were estimated spectrophotometrically at 286nm and 314.7nm respectively. Formulations CF1-CF6 (encapsulated) were subjected to weight variation, disintegration and dissolution tests and subjected to model dependant analysis for dissolution studies. The simultaneous quantitation of Atenolol and Glyburide for content assay was done by HPLC method of analysis. Conclusion: formulation CF6 is showing highest coefficient of correlation values for all models applied. So we can conclude that the proposed system can improve patient compliance by increasing the ease of administration of two drugs together.

Keywords: Tablet-in-a-capsule, atenolol, glyburide, direct compression, simultaneous estimation.

INTRODUCTION

To prevent the complications associated with the diabetes, it is important to control blood glucose levels and blood pressure, effective control of blood glucose levels and blood pressure can reduce the risk of diabetic retinopathy, nephropathy, strokes and death by one third (Leslie, 1999). In this comorbid conditions the patient is bound to take a number of medicines for long period of time but this reduce the patient compliance, results in more sever complications.

Multiple drug therapy causes non-compliance, which is a major obstacle to the effective delivery of health care. Estimates from the World Health Organization indicate only 50% of patients followed number of drug treatment recommendations. This may affect patient health, and affect the wider society when it causes complications from chronic diseases, formation of resistant infections, or untreated psychiatric illness (Sabat e, 2003, Sweileh *et al.*, 2004).

Several efforts have been made to improve the patient compliance by formulating once-a-day dosage form. Encapsulated tablet system can simplify the therapy and reduce or eliminate the chances of improper administration (Rao *et al.*, Rao *et al.*, 2011).

This formulation offers number of advantages like

- Making simple and potent formulation of low therapeutic dosage regimen
- Ease of product formulation by manufacturing the dosage form with direct compression method
- Enhancement of patient compliance, which requires life time therapy
- Reduced inter and intra subject variation
- Minimized local gastric irritation
- Improved safety and efficacy, etc (Gujral *et al.* 2013).

It is a multifunctional and multiple unit system for oral use and can be developed by filling versatile tablets in a hard gelatin capsule, for example: Immediate release mini tablets, Rapid-release Mini-Tablets (RMTs), Sustained release Mini-Tablets (SMTs), Pulsatile Mini-Tablets (PMTs), and Delayed-onset Sustained release Mini-Tablets (DSMTs) (Li and Zhu, 2004, Rao *et al.*, Rao *et al.*, 2011, Tehseen *et al.*, 2013). In this technology we can reduce the size of the tablet such that it could be enclosed in a capsule, and then deployed within one single dosage form (Pozzi *et al.*, 1994, Rao *et al.*, 2011, Sabat e, 2003) (fig. 1).

Atenolol is the cardio selective beta-blocker and extremely used in hypertension, angina pectoris, arrhythmia and myocardial infarction. And Glyburide is the anti diabetic drug belong to the class sulfonyleurea, commonly prescribed to type II diabetic patients (Leslie,

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1999, Sweileh *et al.*, 2004) this combination is supposed to be the best combination for diabetic-hypertensive patients (Colhoun *et al.*, 1998, Dey *et al.*, 2008, Moser and Franklin, 2007).

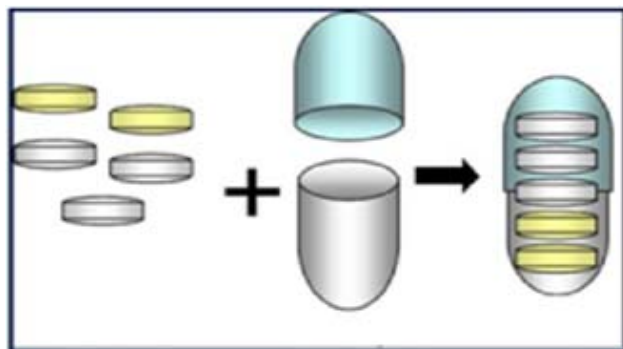


Fig. 1: Tablet-in-a capsule dosage form

The aim of this study is to design a cost effective tablets of Atenolol and Glyburide and to encapsulate in order to improve the patient compliance and increase the ease of administration. This approach will be helpful in simplifying the therapeutic regimen. In current study directly compressible and encapsulated tablets were subjected to pharmaceutical quality assessment tests and compared with the marketed brands (Colhoun *et al.*, 1998, Manmohan *et al.*, 2012, Martinello *et al.*, 2006).

MATERIAL AND METHODS

Material

Atenolol and Glyburide were gifted by *Searle Pakistan Limited (SPL)*, Avicel PH101 (*FMC Corporation, USA*), Lactose-DC (*Riedel-de Haën*), Cross-povidone (*ISP Technologies, Inc. Wayne, NJ*), Magnesium stearate (*Dow Chemicals, USA*), Potassium dihydrogen phosphate (*Riedel-de Haën*), Sodium hydroxide (*Merck, KGaA, and darmstadt, Germany*), Methanol HPLC grade (*RDH, Sigma-Aldrich GmbH, Seelze, Germany*), Ortho-phosphoric acid (*Merck, KGaA, darmstadt, Germany*) and all other chemicals used were of analytical grade and purchased from commercial sources.

Equipments

Weighing balance (*Mettler Toledo B204-S, Switzerland*), Vernier caliper, Hardness tester (*Osk-Fujiwara Seiki. Co. Ltd*), Roche friabilator (*Roche friabilator GmbH D-63150 type TA 200*), USP Dissolution apparatus I and II (*Erweka, Heusenstamm Germany*), USP Disintegration apparatus (*Erweka. D-63150Heusenstamm Germany*), UV-VIS Spectrophotometer (*UV- 1800 Shimadzu Corp.*) and Single punch compression machine (*KORSCH Erweka, Frankfurt Germany*).

The liquid chromatographic system Shimadzu model LC-10ATVP pump and a Shimadzu model SPD-2AT VP. A chromatographic system was integrated via Shimadzu

model CBM- 102 Communication Bus Module. Analysis was conducted on a Purospher STAR RP, C18 (Sum, 250×4.6 mm) column.

Method

Formulation development

The trial formulations of Atenolol and Glyburide were prepared in the ratios mentioned in table 1-2. The blends were subjected to the evaluation of pre-compression parameters.

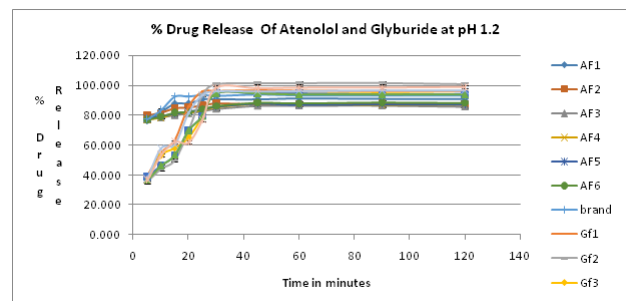


Fig. 2: % Drug Release of Atenolol and Glyburide at pH 1.2

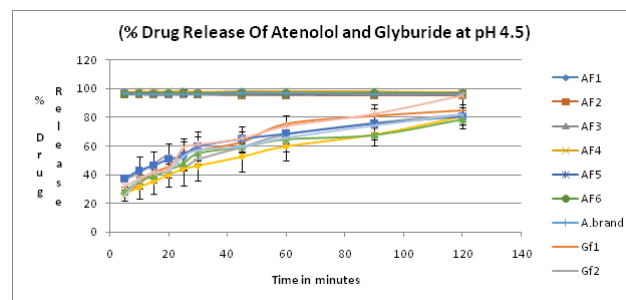


Fig. 3: % Drug Release of Atenolol and Glyburide at pH 4.5

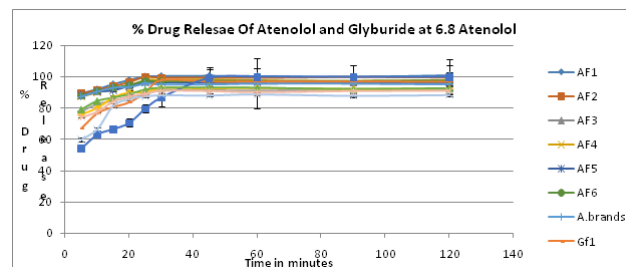


Fig. 4: % Dug Release of Atenolol and Glyburide at pH6.8

Evaluation of pre-compression parameters

Properties of powder blends were evaluated as follows:

Angle of repose

Angle of repose of all six formulations of Atenolol and Glyburide were determined by funnel method, by measuring the height of heap form by the blend to its base

Bulk density

Bulk Densities of all powder blends were determined, by pouring the pre determined quantity of powder in 100ml

graduated cylinder, bulk volumes were measured and bulk densities were calculated. Where,
Bulk density=mass/bulk volume (1)

Tapped density

Tapped densities were found by tapping the graduated cylinder 100 times (till no further contraction in volume observed). Then tapped density was calculated as follows (Jaimini *et al.*, 2007):

Tapped density=mass/tapped volume (2)

Carr's index

Compressibility index can be used to predict the flow property and is based on density measurements. Carr's index can be calculated as. Carr's index (%)=(tapped density-poured density)/ tapped density × 100 (3)

Hausner's ratio

Flow ability of powder can be assessed by Hausner's ratio. Hausner's ratio=tapped density /poured density (4) (Patel *et al.*, 2011)

Blending rate constant

In order to provide uniformity of drug distribution, determining appropriate time for mixing is prerequisite. Blending time was determined by mixing the powder blend in poly bag for different time periods that is 3,6,9 and 12 minutes, followed by tablet compression. Content assay of 10 tablets were determined and % RSD was calculated. For formulations AF1-AF6 and GF1-GF6, blending time was found to be 9 and 3 minutes respectively (Carstensen, 1974).

Preparation of tablets

Six different formulations (F1-F6) of Atenolol 25mg and Glyburide 5mg were formed by direct compression method, all ingredients given in table 1 and 2 were weighed accurately and sieve from 20 mesh sieve and properly mixed by tumbling action in poly bag. Magnesium Stearate (2-5%) was added and mixed for five minutes, and finally subjected to compression on a single punch tablet machine (Korsch Erweka, Germany).

Evaluation of post compression parameters

Tablets of Atenolol and Glyburide prepared by direct compression method were subjected to different pharmacopeial and non pharmacopeial tests including weight variation (Mettler Toledo B204-S, Switzerland), thickness and diameter test variation tests, (Vernier caliper), hardness tests was determined by OSK Fujiwara seiki co ltd and friability was evaluated using Roche friabilator (Roche friabilator (GmbH D-63150 type TA 200). % friability was calculated as % Friability=(initial weight-final weight)/ initial weight × 100 (5)

Disintegration

Disintegration test of all trial batches AF1-AF6 and GF1-GF6 were carried out by using USP disintegration

apparatus (Erweka, Heuesnstanm, Germany) in 900ml purified water at 37±2°C.

Assay of atenolol tablets and Glyburide tablets

Sample of 20 tablets was crushed and solution of 75ug/ml for Atenolol and 67.5ug/ml for Glyburide formed in methanol and content determined by double beam UV-Visible spectrophotometer (Shimadzu UV-Visible spectrophotometer 1800, England) at 286.08nm and 314.7nm (22.Viahnu P, 2010, Belal *et al.*, 2011, El-Massik *et al.*, 1996).

Dissolution profile comparison

The dissolution profile of AF1-AF6 and GF1-GF6 were determined by using USP apparatus II at 50 rpm and 75 rpm respectively, in 900 ml of pH 1.2, 4.5 and 6.8 and compared with respective reference brands. The 10ml sample were drawn and filtered at 5, 10, 15, 20, 25, 30, 45, 60, 90 and 120 minutes and replaced with the fresh medium. The percentage drug release was determined by using (Shimadzu UV -Visible spectrophotometer 1800) at 286nm and 314nm for Atenolol and Glyburide respectively.

Similarity factor (f2)

For dissolution profile comparison of test and reference brands, model-independent approach was described by many scientists (Maggio *et al.*, 2008) FDA has approved following equation to determine similarity factor; Where as

$$f_2 = 50 \times \log \left\{ \left[1 + \frac{1}{N} \sum_{i=1}^N (R_i - T_i)^2 \right]^{-0.5} \right\} \times 100 \quad (6)$$

R_i and T_i are % drug release of reference and test formulations at each time points, N is the total number of samples. Formulations AF1-AF6 were compared with the market available reference brand of Atenolol where as GF1-GF6 were compared with that of Glyburide. Formulations are considered to be similar if f₂ comes greater than 50%.

Encapsulation of tablet

After evaluation of post compression parameters, tablets of both drugs were manually incorporate into the hard gelatin capsule of size "00". The encapsulated systems were further subjected to quality assessment, and following parameters were evaluated.

Weight variation

The USP weight variation test is run by weighing 20 capsule individually (Mettler Toledo B204-S, Switzerland), average weight was calculated and compared the individual capsule weight to the average weight, the capsule meet the USP test, i.e. not more than two capsules, shall fall outside the percentage limit (±7.5%) (London, 2004).

Table 1: Atenolol

S. No.	Ingredients	F1	F2	F3	F4	F5	F6
1	Atenolol	25.000mg	25.000mg	25.000mg	25.000mg	25.000mg	25.000mg
2	Avicel	68.875mg	-	-	-	-	-
3	Lactose DC	-	68.875mg	67.000mg	65.000mg	70.000mg	69.00mg
4	Cros povidone	3.500mg	3.500mg	4.000mg	5.000mg	2.5mg	3.375mg
5	Mg Stearate	2.625mg	2.625mg	2.625mg	2.625mg	2.625mg	2.625mg

Table 2: Glyburide

S. No.	Ingredients	F1	F2	F3	F4	F5	F6
1	Glyburide	5.000mg	5.000mg	5.000mg	5.000mg	5.000mg	5.000mg
2	Avicel	88.875mg	-	-	-	-	-
3	Lactose DC	-	89.000mg	88.000mg	87.000mg	90.000mg	91.000mg
4	Cros povidone	3.500mg	3.375mg	4.375mg	5.000mg	2.375mg	2.000mg
5	Mg Stearate	2.625mg	2.625mg	2.625mg	2.625mg	2.625mg	2.625mg

Table 3: Similarity factor f_2

Formulations	Glyburide			Atenolol		
	pH 1.2	pH 4.5	pH 6.8	pH 1.2	pH 4.5	pH 6.8
	f_2 (%)					
1	47.518 (DS)	33.227 (DS)	26.208 (DS)	65.187 (S)	93.453 (S)	59.725 (S)
2	54.918 (S)	27.601 (DS)	23.953 (DS)	60.930 (S)	91.544 (S)	73.053 (S)
3	78.448 (S)	27.697 (DS)	23.817 (DS)	53.172 (S)	90.893 (S)	99.183 (S)
4	65.680(S)	29.169 (DS)	33.076 (DS)	56.795 (S)	96.556 (S)	89.404 (S)
5	63.727 (S)	29.209 (DS)	23.173 (DS)	57.491 (S)	94.177 (S)	87.102 (S)
6	53.523 (S)	33.724 (DS)	27.917 (DS)	56.764 (S)	97.935 (S)	94.384 (S)

(S)= similar and (DS)= dissimilar

Assay:

Table 4: Quality Parameters of encapsulated system

Formulation	Weight variation (mg)±SD (n=20)	Disintegration (min)	Assay (Atenolol)	Assay (Glyburide)
CF1	300.7±1.03	6.5	95.677±0.336	99.341±3.630
CF2	300.9±1.37	5.9	91.106±3.710	93.817±3.973
CF3	301.6±2.739	5.1	95.635±6.730	98.823±5.443
CF4	301.6±2.623	5.5	96.759±6.979	98.382±1.252
CF5	302.2±3.12	4.9	98.120±4.869	89.995±1.379
CF6	301.8±2.66	5.2	96.443±7.844	97.961±1.315
	Limit: ±7.5% (278.34-323,46mg)	NMT 30 min		

Table 5a: Release kinetics of Atenolol

Models	pH 1.2						pH 4.5					
	First order		Higuchi		Hixson-Crowell		First order		Higuchi		Hixson-Crowell	
Formulations	r^2	k_1	r^2	k_H	r^2	k_{HC}	r^2	k_1	r^2	k_H	r^2	k_{HC}
1	0.791	0.408	0.966	21.715	0.866	0.037	-0.426	0.615	-0.835	25.345	-0.592	0.069
2	0.916	0.136	0.972	20.837	0.921	0.035	-0.596	0.688	-0.916	25.432	-0.753	0.069
3	0.911	0.121	0.973	19.9914	0.921	0.031	-0.193	0.784	-0.662	26.734	-0.47	0.072
4	0.932	0.134	0.985	20.949	0.937	0.034	-0.663	0.625	-0.957	25.092	-0.793	0.069
5	0.982	0.143	0.995	21.246	0.979	0.037	-0.419	0.839	-0.814	26.387	-0.649	0.071
6	0.968	0.152	0.996	21.537	0.962	0.037	-0.598	0.691	-0.933	25.965	-0.766	0.07

Table 5b: Release kinetics of Atenolol

Models	pH 6.8						pH 7.4					
	First order		Higuchi		Hixson-Crowell		First order		Higuchi		Hixson-Crowell	
Formulations	r ²	k1	r ²	k _H	r ²	K _{HC}	r ²	k1	r ²	k _H	r ²	k _{HC}
1	0.791	0.408	0.995	23.741	0.907	0.056	0.859	0.328	0.989	24.126	0.908	0.059
2	0.893	0.381	0.98	23.754	0.952	0.058	0.9	0.302	0.987	23.809	0.927	0.058
3	0.796	0.351	0.986	23.274	0.87	0.057	0.885	0.309	0.992	23.767	0.93	0.058
4	0.774	0.342	0.988	23.141	0.864	0.057	0.816	0.311	0.966	23.166	0.875	0.057
5	0.81	0.282	0.981	22.93	0.848	0.056	0.859	0.328	0.989	24.126	0.908	0.059
6	0.823	0.361	0.997	23.636	0.914	0.058	0.896	0.287	0.992	23.551	0.926	0.057

Table 5c: Release kinetics of Glyburide

Models	ph 1.2				Ph 4.5							
	First order		Higuchi		Hixson-Crowell		First order		Higuchi		Hixson-Crowell	
Formulations	r2	k1	r2	k _H	r2	k _{HC}	r2	k1	r2	k _H	r2	k _{HC}
1	0.99	0.456	0.925	27.184	0.989	0.073	0.423	0.375	0.98	13.256	0.908	0.014
2	0.996	0.221	0.971	24.063	0.985	0.058	0.814	0.285	0.859	13.394	0.9445	0.014
3	0.919	0.28	0.997	25.094	0.911	0.066	0.669	0.291	0.975	13.169	0.97	0.014
4	0.959	0.404	0.962	24.045	0.972	0.059	0.809	0.202	0.945	13.1965	0.971	0.014
5	0.941	0.296	0.9776	23.384	0.955	0.051	0.795	0.234	0.908	13.197	0.972	0.014
6	0.951	0.2	0.996	22.285	0.933	0.051	0.793	0.204	0.961	13.769	0.966	0.014

Table 5d: Release kinetics of Glyburide

Models	pH6.8						Ph 7.4					
	First order		Higuchi		Hixson-Crowell		First order		Higuchi		Hixson-Crowell	
Formulations	r ²	k1	r ²	k _H	r ²	k _{HC}	r ²	k1	r ²	k _H	r ²	k _{HC}
1	0.97	0.101	0.93	19.051	0.966	0.014	0.791	0.288	0.962	23.173	0.871	0.057
2	0.959	0.099	0.964	18.833	0.966	0.027	0.759	0.292	0.954	23.081	0.871	0.057
3	0.959	0.079	0.971	17.232	0.97	0.021	0.779	0.29	0.959	23.154	0.862	0.057
4	0.991	0.105	0.995	18.962	0.993	0.027	0.76	0.266	0.944	22.016	0.808	0.054
5	0.985	0.099	0.996	18.879	0.991	0.026	0.73	0.261	0.927	21.861	0.775	0.054
6	0.995	0.099	0.992	18.873	0.997	0.026	0.817	0.275	0.982	22.398	0.852	0.055

Disintegration test

Disintegration test was carried out by taking random sample of six capsules of all trial formulations, using USP disintegration apparatus (Erweka,, Heuesstanm, Germany) in 900ml purified water at 37±2°C (London, 2004).

Content assay

Assay of all encapsulated trial formulations were performed by HPLC simultaneous determination method of Atenolol and Glyburide on HPLC (Shimadzu model LC-10AT VP, SPD-2AT VP). The mobile phase consisted of methanol: water (80:20) adjusted to pH 3.4 with Ortho-phosphoric acid, the flow rate was 1ml/min and detection of 20µl sample was carried out at 235 nm using UV-Visible detector (Shimadzu Corporation, Kyoto, Japan) (Anitha et al., 2011)(Arayne et al., 2012).

Sample preparation

A sample of 10 capsules of each formulation were taken and final strength of 30ug/ml of Atenolol and 15ug/ml of Glyburide in methanol were prepared by sonication for 15 minutes, then volume was made up to 25 ml.

Percentage assay was calculated as follows:

$$\% \text{ Assay} = \frac{\text{Average peak area of sample} \times \text{conc. Of standard}}{\text{Average peak area of standard} \times \text{conc. Of sample}} \times 100 \quad (7)$$
Dissolution studies

The encapsulated trial batches CF1-CF6 were subjected to dissolution studies using USP apparatus I at 60 rpm in buffer medium pH 1.2, 4.5, 6.8 and 7.4. Sample of 10ml was drawn at same time points and replaced with fresh medium then filtered. The simultaneous determination of Atenolol and Glyburide was done spectrophotometrically, at 275nm and 229nm for Atenolol and Glyburide.

Model dependant analysis of dissolution studies

Different kinetic models were applied to dissolution data, like first order, Higuchi model and Hixson-Crowell's model. Equation

$$\text{LogQ} = \text{LogQ}_0 - kt/2.303 \quad (8)$$

Explain First order i.e. the relationship between log percentage drug remaining and time.

Higuchi model describes the relation between cumulative percentage drugs vs. square root of time equation.

$$Q = kt^{1/3} \quad (9)$$

Whereas Hixson-Crowell's model shows the relationship between cube root percentage drug remaining and time. Equation.

$$Q_0^{1/3} - Q_t^{1/3} = K_{HC} \times t \quad (10)$$

Release Kinetics of CF1-CF6 was studied through model dependant approach. Coefficient of correlation and rate constants of respective models were calculated.

Stability studies

Trial formulations CF1-CF6 were kept at room temperature, as well as under accelerated conditions (temperature 40°C, RH 75%) for stability study according to the ICH guidelines. Drug assay and dissolution profiles were studied at 3, 6, 9 and 12 months and shelf life was determined by R-GUI Stab R console 2.15.2 soft ware (Carstensen, 1974, ICH).

RESULTS

The tablets were compressed by direct compression method using the blends of different ratios of Avicel (diluent), Lactose DC (diluent), Crospovidone (super disintegrant) and Magnesium Stearate (glidant/lubricant).

Evaluation of Pre-compression parameters

Powder blends of Atenolol and Glyburide were subjected to various pre-compression evaluations such as angle of repose, compressibility index and Hausner's ratio. Results of the pre-compression parameters of all formulations were found in the range of 19.79-32.8, 9-13.17%, 1.03-1.17 and 6.699-18.77, 8.45-10.00, 1.06-1.10 for Atenolol and Glyburide respectively. In order to obtain uniformity in drug distribution blending rate constant was determined and was found within the limit (<6%), that is 12 minutes for Atenolol with % RSD of 2.54 to 5.57 and 12 minutes for Glyburide with % RSD of 1.14 to 2.99.

Evaluation of post-compression parameters

Various physical properties like weight variation, hardness variation, disintegration time, percentage friability and content assay influence availability of drug at site of action. In present work the tablet of Atenolol and Glyburide were compressed through direct compression method and then encapsulated to form a single unit for the simultaneous administration. The shape of the tablet kept round, the mean thickness, hardness and diameter of AF1-AF6 and GF1-GF6 were found in the range of 3.933mm, 9.915±0.0587kg and 6.25mm and 3.919±0.0022 to 3.937±0.055, 9.94±0.0587kg to 15.01±0.044, 6.25mm respectively. All the trial formulation showed weight variation within the acceptable limit due to good flow characteristics and uniform feeding of die cavity. The mean weights of trail batches of Atenolol and Glyburide were found in the range of 97.8mg±3.65 to 99.65±0.76 and 99.2mg±0.884 to 99.65±0.820, similarly % friability of both the drug formulations complies with the

pharmacoepial limits that is <1% in the range of 0.014±0.005% - 0.201±0.002% for Atenolol and 0.05±0.0005%- 0.1006±0.0001% for Glyburide .The drug content was found to range between 95.770±1.011% to 99.5±0.771 and 95.978±0.326 to 99.666±0.585% for all formulations of Atenolol and Glyburide indicating good content uniformity.

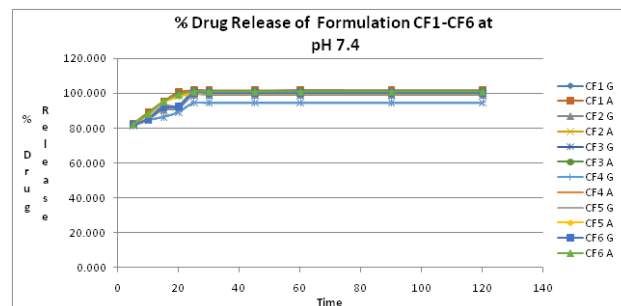


Fig. 5: % Drug Release of Encapsulation at pH 7.4

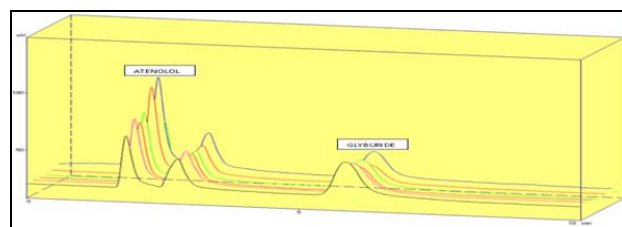


Fig. 6: Chromatogram of Simultaneous Assay

Disintegration test

To ensure the availability of the drug for dissolution and absorption disintegration were performed. (Augsburger *et al.*, 2002, Schiermeier and Schmidt, 2002) Super-disintegrants enhance the phenomenon of disintegration. In this study all the formulations of Atenolol and Glyburide showed disintegration within the USP limits that is <15 minutes. AF1 contain 3.5% of Crospovidone and 68.8% Avicel showed disintegration within 2 minutes but AF5 also disintegrated within the same time containing 2.5% Crospovidone and 70% lactose DC. Results showed that all the formulations disintegrated within 10 seconds.

Dissolution studies and profile comparison

In order to check the percentage availability of the drug, dissolution test was performed. For AF1-AF6 the percentage drug release at pH 1.2, 4.5 and 6.8 was observe within the range from 95.77±1.011% to 99.5±0.771%, where as GF1-GF6 exhibited similar drug release in the range from 93.417±1.466% to 99.66±0.585%, percentage drug release of all batches shown in figs. 2,3 and 4

The pKa of Atenolol and Glyburide are 6.9 and 5.2 respectively, showing their variable dissolution behavior at different pH. In present research the dissolution profile were determined at pH 1.2 (0.1N HCl) and phosphate

buffer pH 4.5 and 6.8 and compared with that of reference brands of Atenolol and Glyburide. Results indicated that for Glyburide % drug release was found $98.496 \pm 4.577\%$ in 30min, $95.591 \pm 8.271\%$ in 120min and $90.870 \pm 4.97\%$ in 30 min at pH 1.2, 4.5 and 6.8 respectively. Sequentially for same pH the drug release for Atenolol was $98.947 \pm 0.681\%$ in 45min, $96.789 \pm 0.735\%$ in 10min and $98.812 \pm 1.64\%$ in 25min. Similarity factor f_2 was also calculated in the same dissolution media. Data exhibit that profile of reference brands of Atenolol were formed similar with all trial formulations in all media. Whereas GF1 showed no similarity with reference at pH 1.2, but rest of the formulations showed similarity in this medium. At pH 4.5 and 6.8 GF1-GF6 showed no similarity with reference brands in table 3.

Percentage of content uniformity of Atenolol and Glyburide were performed and compared with the reference (table 4).

Encapsulation of tablets

In present work all trial formulations of Atenolol and Glyburide qualified the pharmaceutical quality attributes, and both the tablets of Atenolol and Glyburide were encapsulated in a hard gelatin capsule of size "00" for all the six formulations CF1-Cf6.

Quality evaluation of encapsulated system

Selection of suitable excipients in appropriate ratio makes the formulation successful. The mean weight of CF1-CF6 shown in Table 4 found in the range from $300.9 \pm 1.03\text{mg}$ to $302.2 \pm 3.12\text{mg}$ that is shown with the pharmacopeial limit that is $\pm 7.5\%$ (USP, 27, 2003). Crospovidone being super-disintegrant provided rapid disintegration to tablets. When encapsulated tablet were subjected to disintegration the mean DT of CF1-CF6 were found in the range between 4.9-6.5 minutes. Acceptable official range for capsule disintegration is NMT 30min. The drug constant of Atenolol and Glyburide after encapsulation, were also assessed and found in the range between $91.106 \pm 3.71\%$ to $98.120 \pm 4.869\%$ and $89.995 \pm 1.377\%$ to $99.341 \pm 3.630\%$ respectively showed good uniformity in all batches. The mean \pm S.D of quality attributes of encapsulated system are shown in table 4.

Dissolution study of encapsulated system

Dissolution profile of encapsulated system CF1-CF6 at pH 1.2 (0.1 N HCl), 4.5, 6.8 and 7.4 were also performed and evaluation of dissolution data first order kinetic model equation no. 8 is used extensively (Riis et al., 2007)(Gibaldi M, 1982, Gujral et al.). In present work the values of coefficient of correlation (r^2) first order for Atenolol and Glyburide in pH 1.2 (0.1 N HCl) were 0.982 and 0.996 respectively. To ascertain the drug release mechanism, drug release data at all pH were also subjected to Higuchi's diffusion plot. The value of coefficient of correlation (r^2) for Atenolol and Glyburide are given in the table 5a, b, c and d. fig. 5

Assay by simultaneous estimation

Assay of encapsulated system was performed by HPLC method and estimated simultaneously, shown in chromatogram. Simplest method for the determination of drugs was used which based on liquid-liquid or solid extraction. Peak plasma level for Atenolol and Glyburide were easily quantitated without any interference and separated at a low retention time of 10 minutes (Arayne et al., 2012) fig. 6 chromatogram. The mean \pm S.D of all Formulations are shown in table 4.

Stability studies

Stability studies of all formulations were performed according to ICH guidelines and shelf-life was calculated 15 months for Atenolol and 27 months for Glyburide by using R-GUI Stab R-console 2.15.2 software (ICH).

DISCUSSION

In order to obtain target glucose level and good blood pressure control, several drug combinations were used, and to enhance patient compliance and reduce frequency of drug administration Atenolol and Glyburide can be given in single formulation.

Powder blend with good pre compression characteristics show low weight variation and good uniformity of active pharmaceutical ingredients (Jaimini et al., 2007, Martinello et al., 2006, Pozzi et al., 1994). The results of current study indicated good flow characteristics of powder blend according to pharmacopeial specifications (Pharmacopeia, 2005).

Since penetration of dissolution medium does not guarantee the appropriate dissolution of drug, this showed there are some rate limiting steps for drug dissolution (El-Massik et al., 1996, Sathigari, 2011). In current study percentage release of all encapsulated trial batches of Atenolol and Glyburide was found in good range that showed either of the drug is not influencing the release of other and this may guarantee the better bioavailability of these drugs in the presence of each other.

The results of simultaneous estimation of assay by HPLC also confirmed the accurate delivery of drugs through encapsulated system. Similarly the stability study data also revealed that the combined formulations of Atenolol and Glyburide remained stable throughout the period of estimated shelf life.

CONCLUSION

The bi-functional encapsulated system CF1-CF6 for oral use was developed by filling of two different compressed tablets of Atenolol and Glyburide in a hard gelatin capsule. All the trial batches showed compliance with the pharmaceutical quality attributes. The release profiles

were significant at all pH 1.2 (0.1 N HCl), 4.5, 6.8 and 7.4. While formulation CF6 is showing highest coefficient of correlation values for all models applied. So we can conclude that the proposed system can improve patient compliance by increasing the ease of administration of two drugs together.

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