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RESEARCH ARTICLE

Formulation and Characterization of Floating Microspheres of Glipizide

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ABSTRACT

The aim of the current research work is to formulate and characterized the floating microspheres of glipizide. Glipizide administered in a dose of 5-20 mg in once or twice daily. Glipizide rapidly and completely absorbed from GIT but when administered in single unit dosage form it produced the gastric irritation, it creates the need to develop multiparticulate dosage form. The microsphere was prepared using Emulsion solvent evaporation method. The drug to polymer ratio used to prepare the different formulations was 1:2. The polymer content was a mixture of Eudragit S100 and Ethyl Cellulose 22cps in varying concentration. The floating microspheres were then subjected to FTIR, DSC, SEM, particle size, size distribution, % yield, drug content, entrapment efficiency, In-vitro dissolution, release kinetics and stability studies. FTIR confirms that there was no chemical interaction between Glipizide and polymer. The DSC obtained for there was no interaction between the Glipizide and the polymer in the solid state. The % Practical Yield was found to be 68.33-78.66. The particle size of floating microsphere was found to be 483.63 to 511.56. The drug entrapment efficiency of floating microspheres was found to be 81.96 to 39.34. The Glipizide floating microspheres with smooth surface was observed upon SEM. %Buoyancy of microspheres was in the range 60.78% to 76.19% after 12 hrs. *In-vitro* drug release was found in the range of 92.21% to 72.66% over the 12 hrs. Kinetics and mechanism of drug release from F1 formulation was evaluated on the basis of Higuchi equation, Zero order, First order, Hixoncrowell equation and Peppas model. Correlation coefficient (r^2) and slope value for each equation in the range of (r^2=0.71-0.998 and n=0.51-39.00) was calculated. After performing the dissolution of F1 batch after 3 months the percentage drug release was found to be 90.73%.

KEYWORDS

Microspheres, Glipizide, Bioavailability, Gastric irritation

INTRODUCTION

Diabetes is a condition where the amount of glucose in your blood is too high because the body cannot use it properly. This is because your pancreas doesn't produce any insulin, or not enough insulin, to help glucose enter your body's cells or the insulin that is produced does not work properly (known as insulin resistance). As of 2013, 382 million people have diabetes worldwide. Type 2 makes up about 90% of the cases. This is equal to 8.3% of the adult population with equal rates in both women and men.[3]

Glipizide is an oral rapid and short-acting anti-diabetic drug from the sulfonylurea class. It is classified as a second generation sulfonylurea, which means that it undergoes enterohepatic

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circulation. Second-generation sulfonylureas are both more potent and have shorter half-lives than the first-generation sulfonylureas. Glipizide administered in a dose of 5-20 mg in once or twice daily. Glipizide rapidly and completely absorbed from GIT but when administered in single unit dosage form it produced the gastric irritation, it creates the need to develop multiparticulate dosage form. The conventional dosage form having the drawbacks like poor patient compliance, dose dumping, and fluctuation in drug release profile, produce gastric irritation, low bioavailability and low stability. To overcome these all problems needs of multiparticulate formulation.

One novel multiparticulate formulation is floating microspheres. Floating systems was first described by Davis in 1968. Floating drug delivery system is an effective technology to prolong the gastric residence time in order to improve the bioavailability of the drug. FDDS are low-density systems that have sufficient buoyancy to float over the gastric contents and remain in the stomach for a prolonged period.[1]

In floating types the bulk density of microspheres is less than the gastric fluid, so remains buoyant in stomach without affecting gastric emptying rate. The drug is released slowly at the desired rate, if the system is floating on gastric content and increases gastric residence and increases fluctuation in plasma concentration. Moreover it also reduces chances of striking and dose dumping. One another way it produces prolonged therapeutic effect and therefore reduces dosing frequencies.[2]

MATERIAL & METHODS

Material

Glipizide gift sample received from USV Pharma, Mumbai, Eudragit S100 LR, Ethyl Cellulose 18-22cps, Poly vinyl Alcohol Hot, Calcium Chloride, Dichloro Methane, Methanol, Conc. Hydrochloric acid procured from Research-Lab Fine Chem Industries, Mumbai. All other chemicals and reagents used were LR grade.

Method

Microsphere containing Glipizide was prepared using Emulsion solvent evaporation method. The drug to polymer ratio used to prepare the different formulations was 1:2. The polymer content was a mixture of Eudragit S100 and Ethyl Cellulose 22cps in varying concentration. The drug polymer mixture was dissolved in a mixture of Dichloromethane and Methanol (1:1$^{v/v}$). The mixture was dropped in to 0.4%$^{w/v}$ Poly vinyl alcohol solution (400ml) containing Calcium Chloride by 22 gauge needle. The solution was stirred with a propeller-type agitator and magnetic stirrer at 40^{oC} for 1 h at 300 rpm. The formed floating microspheres were filtered by whattmann filter paper washed with water and dried at 40^{oC} overnight. [4, 5, 6, 7, 8, 9, 10]

Evaluation

%Practical Yield[4]

The percentage yield of different formulations was determined by weighing the hollow microspheres after drying. The percentage yield was calculated as follows.

$$\% \text{ Practical yield} = \frac{\text{Total weight of dried Microspheres}}{\text{Total weight of Drug + Polymer}} \times 100$$

Particle Size Determination[11, 12]

The particle size of microspheres was determined by optical microscopy method, approximately 100 microspheres were counted for particle size using a calibrated optical microscope. The microspheres were uniformly spread on a slide. The measurement was done under 450x (10x eye piece and 45x objective) and100 particles were calculated.

Bulk Density[6]

Apparent bulk density (ϱb) was determined by pouring the mass in to a graduated cylinder. The bulk volume (Vb) density was calculated in g/cm^3 by using following formula:

$$\varrho b = M/Vb$$

Tapped Density[6]

The measuring cylinder containing known amount of blend was tapped for a fixed time. The minimum tapped volume (Vt) occupied in

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Table: 1 Batch design

Batches	Drug : Polymer (1:2)			PVA Solution (% w/v)	$CaCl_2$ (%w/v)
	Drug (mg)	Eudragit S100 (mg)	Ethyl Cellulose 22cps (mg)		
F1	300	500	100	0.4	5
F2	300	400	200	0.4	5
F3	300	300	300	0.4	5
F4	300	200	400	0.4	5
F5	300	100	500	0.4	5
F6	300	500	100	0.4	4
F7	300	500	100	0.4	8
F8	300	500	100	0.4	12

the cylinder and weight of the (M) mass was measured. The tapped density was calculated in g/ cm^3 by using following formula:

$$\varrho t = M/Vt$$

Angle of Repose[6]

The angle of repose of the microspheres, which measures resistance to particle flow, was determined by the fixed funnel method and calculated by using following formula:

$$\Theta = \tan^{-1} h/r$$

Hausner's Ratio[6]

Tapped density and bulk density were measured and the Hausner ratio was calculated using following formula

$$\text{Hausner ratio} = \varrho t/\varrho b$$

Carr's Index[6]

The bulk density and tapped density was measured and compressibility index was calculated using following formula:

$$\text{Carr's index} = TD\text{-}BD/TD \times 100$$

%Drug Entrapment Efficiency[12, 13]

The various formulations of the microspheres were subjected for drug content. The microspheres containing approx 25mg drug from all batches were accurately weighed and crushed. The powdered of microspheres were dissolved with 10ml Methanol in 100ml volumetric flask and makeup the volume with 0.1 N HCl. This resulting solution is then filtered through whattmann filter paper No. 44. After filtration, from this solution 10 ml was taken out and diluted up to 100 ml with 0.1 N HCl. Again from this solution 2 ml was taken out and diluted up to 10 m1 with 0.1 N HCl and the absorbance was measured at 276 nm against 0.1 N HCl as a blank.

%Drug Loading Efficiency[13]

The drug loading efficiency of microspheres were calculated by using following formula:

$$\%\text{Drug loading} = \frac{\text{Actual Drug content}}{\text{Total weight of microspheres}} \times 100$$

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Scanning Electron Microscopy[4]

From the formulated batches of microspheres, the batch which showed an appropriate results including percentage release were examined for surface morphology and shape using scanning electron microscope JEOL, JSM-670F Japan (Diya Labs, Mumbai). Sample was fixed on carbon tape and fine gold sputtering was applied in a high vacuum evaporator. The acceleration voltage was set at 3.0KV during scanning. Microphotographs were taken on different magnification and higher magnification was used for surface morphology.

***In-vitro* Buoyancy Test**[14, 15]

In-vitro buoyancy studies were carried out for each formulation using 300mg of drug loaded floating microspheres were spread over the surface of USP Type II (paddle) dissolution apparatus filed with 900ml of 0.1 N HCl containing 0.02% of Tween 80. The medium was maintained at 37°C and agitated with a paddle rotating at 100 rpm for 12 hrs. At the end of this period, the layer of buoyant particles on the surface of the medium was collected and the sinking particulates were separated by filtration. Both particle types were dried overnight at 40°C. Dried weights were measured and buoyancy was determined by the weight ratio of the floating particles to the sum of floating and sinking particles.

$$\% \text{ Buoyancy} = \frac{\text{Dry wieght of floated microsphere}}{\text{Dry weight of floated microspheres + Setteled microspheres}} \times 100$$

In-vitro Drug Release Study[15, 16, 17]

The *In-vitro* drug release were performed using paddle type dissolution apparatus. In this method, a weighed quantity of the microsphere which is equal to dose is placed in muslin cloth and tie to the paddle. The dissolution study performed using 900ml 0.1 N HCl (pH 1.2) for 12 hr at 37±0.5^{0C} stirred 50rpm. 1ml sample was pipet out per hour and maintain sink condition. Then analyzed all the sample on UV-Spectrophotometer at 276nm.

Drug Release Kinetic[4, 18, 19]

Several kinetic models have been proposed to describe the release characteristics of a drug from microspheres. The dissolution profile of all the formulations was fitted to Higuchi, Zero order, First order, Hixoncrowell and Korsemeyer-Peppas to ascertain the kinetic modeling of drug release.

The value of 'n' gives an indication of the release mechanism. When n = 1, the release rate is independent of time (typical zero order release / case II transport); n = 0.5 for Fickian release (diffusion/ case I transport); and when 0.5 < n < 1, anomalous (non-Fickian or coupled diffusion/ relaxation) are implicated. Lastly, when n > 1.0 super case II transport is apparent. 'n' is the slope value of log Mt/M∞ versus log time curve.

The results obtained from *in vitro* release studies were plotted in four kinetics models of data treatment as follows.

- ✓ Cumulative percentage drug release Vs. √T (Higuchi's classical diffusion equation)
- ✓ Cumulative percentage drug release Vs. Time (zero order rate kinetics)
- ✓ Log cumulative percentage drug retained Vs. Time (first order rate kinetics)
- ✓ $W_0^{1/3}$-$W_t^{1/3}$ Vs. Time (Hixoncrowell equation)
- ✓ Log of cumulative percentage drug release Vs. log Time(Peppas exponential equation)

Stability Studies[12, 20, 21]

By placing the microspheres in screw capped glass container and stored them at 40^{oC} and 75 Rh. It was carried out of a 90 days and the drug content and drug release of the microsphere was analyzed.

RESULTS AND DISCUSSION

FT-IR

FT-IR obtained for pure Glipizide, Glipizide-Eudragit S100, Glipizide-Ethyl Cellulose and Glipizide-Eudragit S100 & Ethyl Cellulose there was no chemical interaction between

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Table: 2 FT-IR spectrum ranges of formulations

Sr. no.	Transition	Ranges (cm^{-1})	Drug R1	R2	R3	R4	R5
1	N-H str	3000-3700	3251.13	3251.13	3250.16	3435.34	3250.16
2	C-H str	2700-3300	294.44	2943.13	2943.47	2929.00	2943.47
3	C=O	1650-1700	1688.73	1688.73	1688.73	1688.73	1688.73
4	-CONH	1600-1750	1649.19	1651.12	1649.19	1641.48	1649.19
5	C-H bend (Cyclohexane)	1345-1450	1375.29	1308.75	1308.75	1383.97	1387.83
6	S=O str	1149-1180	1159.26	1159.26	1159.26	1159.26	1159.26
7	C-H bend (Benzene)	650-900	686.68	686.68	686.68	668.36	6868.68

(**R1**-PureDrug, **R2**-Drug+ES100**, R3**-Drug+EC**, R4**-ES100+EC & **R5**-Drug+ES100+EC)

Glipizide and polymer and it can be concluded that the characteristics bands of Glipizide were not affected after successful loading.

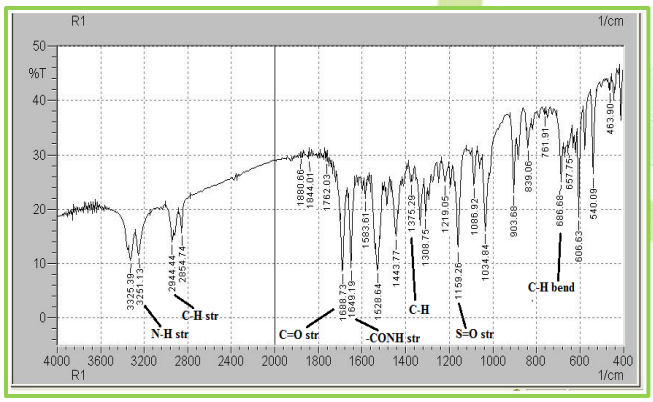

Figure: 1 FT-IR Spectra of Pure Glipizide

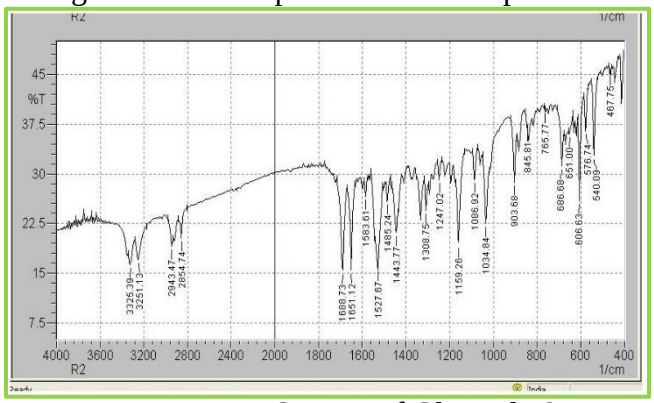

Figure: 2 FT-IR Spectra of Glipizide & Eudragit S100

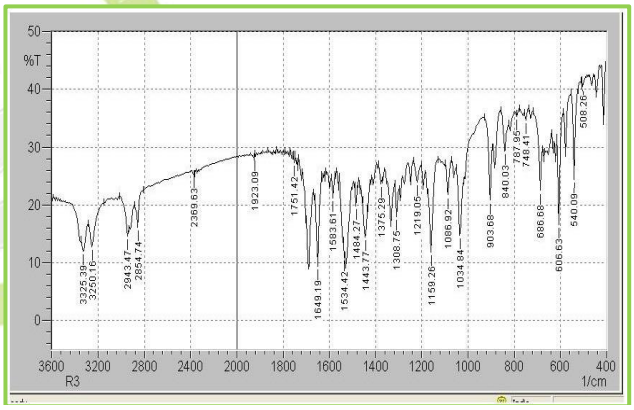

Figure: 3 FT-IR Spectra of Glipizide & Ethyl Cellulose

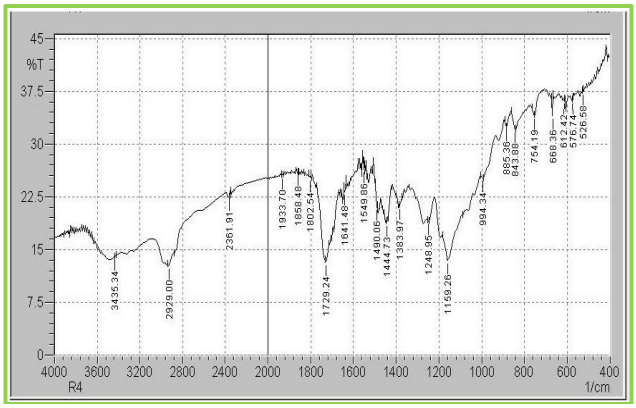

Figure: 4 FT-IR Spectra of Eudragit S100 & Ethyl Cellulose

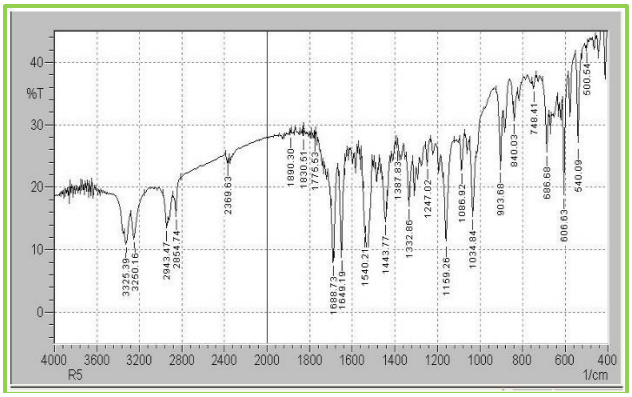

Figure: 5 FT-IR Spectra of Glipizide, Eudragit S100 & Ethyl Cellulose

DSC

The DSC obtained for there was no interaction between the Glipizide and the polymer in the solid state. The melting point range of Glipizide is between 200-205°C, thus indicating there is no change of Glipizide in pure state, physical mixture of drug and polymer.

%Practical Yield

The %Practical Yield was found to be 78.66, 75.21, 71.39, 70.50, 68.33, 78.22 77.80 and 78.55 of F1, F2, F3, F4, F5, F6, F7 and F8 respectively as shown in table 3.

Particle Size Determination

The particle size of floating microsphere was found to be 483.63 to 511.56 it has been observe that as increasing the concentration of Ethyl cellulose increasing the size of microspheres due to high viscosity as shown in table 3.

%Drug Entrapment Efficiency

The drug entrapment efficiency of floating microspheres was found to be 81.96 to 39.34 as increasing the concentration of Eudragit S100 and Calcium Chloride to aqueous phase as shown in table 3.

%Drug Loading Efficiency

The drug loading efficiencies of microspheres were in the range of 13.11 – 27.32% w/w as shown in following table 3.

Surface Morphology (SEM)

The surface morphology of the Glipizide floating microspheres was studied by SEM. SEM photographs of F1 formulation was shown in fig. no. 6. The Glipizide floating microspheres with smooth surface was observed.

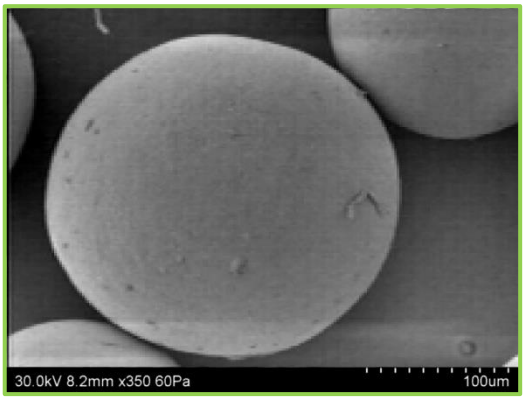

Figure: 6 SEM of F1

%Buoyancy

The microspheres floated for prolonged time over the surface of the dissolution medium without any apparent gelation. As increasing the concentration of Ethyl Cellulose 22cps increases the buoyancy time. Percentage buoyancy of the microspheres was in the range 60.78% to 76.19% after 12 hrs. The results obtain are given in table 3.

%Drug Release

The in vitro performance of Glipizide floating microspheres showed sustained release of Glipizide. The results of the *In-vitro* dissolution studies shows as Ethyl Cellulose 22cps concentration increases the drug release from the floating microsphere decreases. *In-vitro* drug release was found in the range of 92.21% to 72.66% over the 12 hrs. The results are shown in table 4.

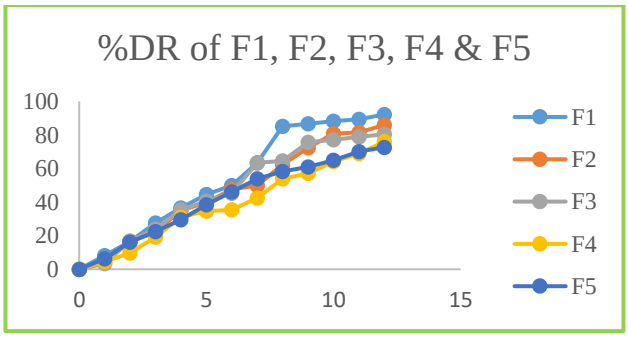

Figure: 7 %DR of F1, F2, F3, F4 & F5

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Table: 3 Results of %Practical Yield, Size, %DEE, %DLE & %Buoyancy

Batch	%Practical Yield	Size (µm)	%DEE	%DLE	%Buoyancy after 12 Hrs
F1	78.66	483.63	68.85	22.95	60.78
F2	75.21	488.04	59.01	19.67	67.30
F3	71.39	496.86	55.73	18.58	71.18
F4	70.50	505.68	49.18	16.39	73.77
F5	68.33	511.56	39.34	13.11	76.19
F6	78.22	479.22	62.29	20.77	61.22
F7	77.80	482.16	75.41	25.14	59.61
F8	78.55	480.69	81.96	27.32	62.96

Table: 4 %DR of Batch F1-F8

Time (Hrs)	F1	F2	F3	F4	F5	F6	F7	F8
1.	8.11	4.79	3.31	4.42	6.27	7.37	5.53	7.74
2.	16.59	16.59	16.96	9.59	16.22	18.81	8.48	16.59
3.	27.66	22.13	23.97	19.18	22.50	22.13	15.49	20.28
4.	36.51	35.04	35.40	30.98	29.50	34.67	26.92	23.60
5.	44.63	39.83	40.57	34.67	38.36	40.20	37.25	40.94
6.	49.79	47.58	45.36	35.40	46.10	51.63	40.57	53.85
7.	63.44	49.79	63.44	42.41	53.85	63.07	58.27	62.33
8.	85.20	62.33	64.54	53.85	58.27	64.54	67.86	66.76
9.	86.68	72.29	75.61	57.17	60.86	73.77	70.08	73.03
10.	88.15	80.77	77.09	64.18	64.91	81.14	81.51	84.83
11.	89.26	81.51	78.93	68.97	70.08	88.52	85.94	89.26
12.	92.21	85.94	80.40	75.98	72.66	91.10	92.58	91.84

(Note: All values are n=3**)**

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Table: 5 Release kinetic of F1

Formulation code	Higuchi	Zero Order	First Order	Hixon crowell	Korsemeyer-Peppas	
	r^2	r^2	r^2	r^2	r^2	N
F1	0.96	0.95	0.84	0.71	0.98	1.02

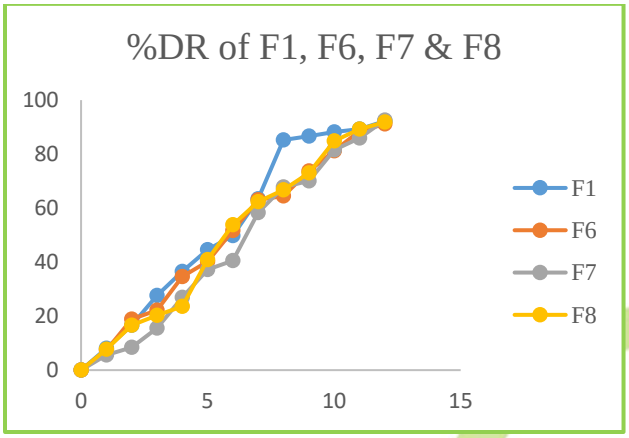

Figure: 8 %DR of F1, F6, F7 & F8

Drug Release Kinetic

Drug release pattern was evaluated in 0.1 N HCl of F1 formulation. Kinetics and mechanism of drug release from F1 formulation was evaluated on the basis of Higuchi equation, Zero order, First order, Hixoncrowell equation and Peppas model. Correlation coefficient (r^2) and slope value for each equation in the range of (r^2=0.71-0.998 and n=0.51-39.00) was calculated.

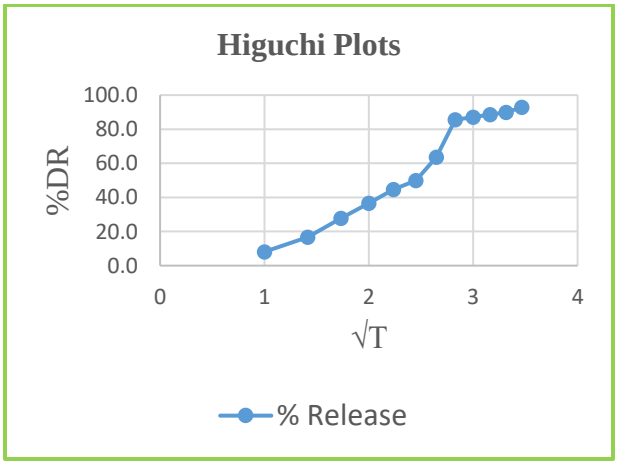

Figure: 9 Higuchi Plots

The diffusion exponent 'n' values of Korsemeyer-Peppas model was found to be in the range of 1.02 for the Glipizide floating microspheres prepared with Eudragit S100 and EC22cps indicating Super Case II transport of drug through Glipizide floating microspheres.

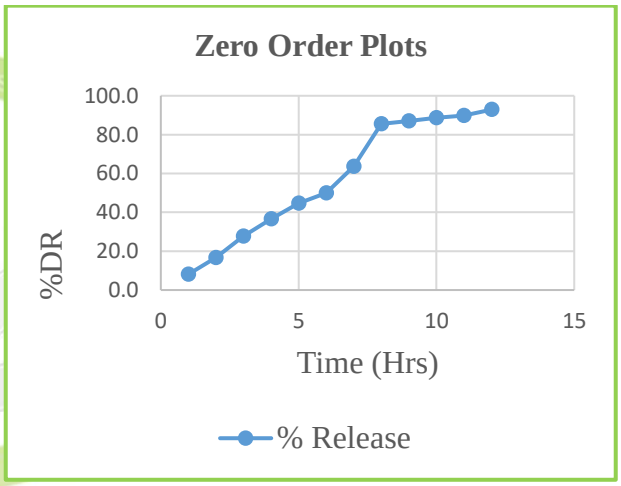

Figure: 10 Zero Order Plots

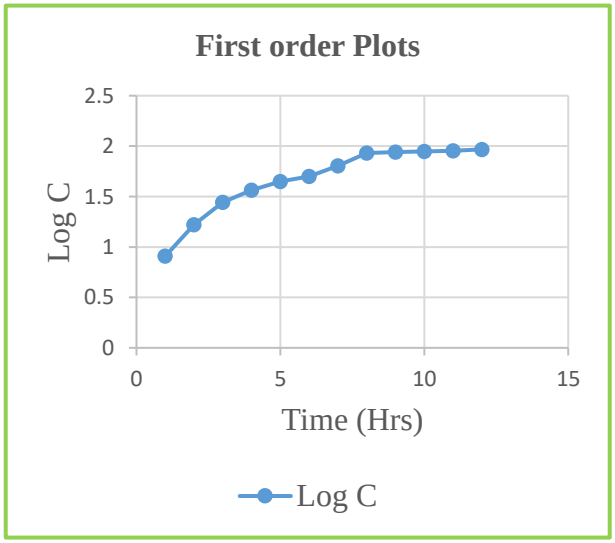

Figure: 11 First Order Plots

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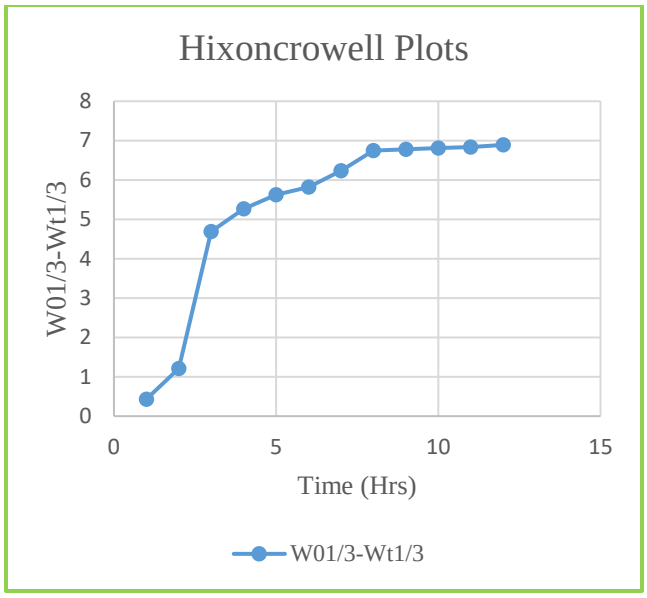

Figure: 12 Hixoncrowell Plots

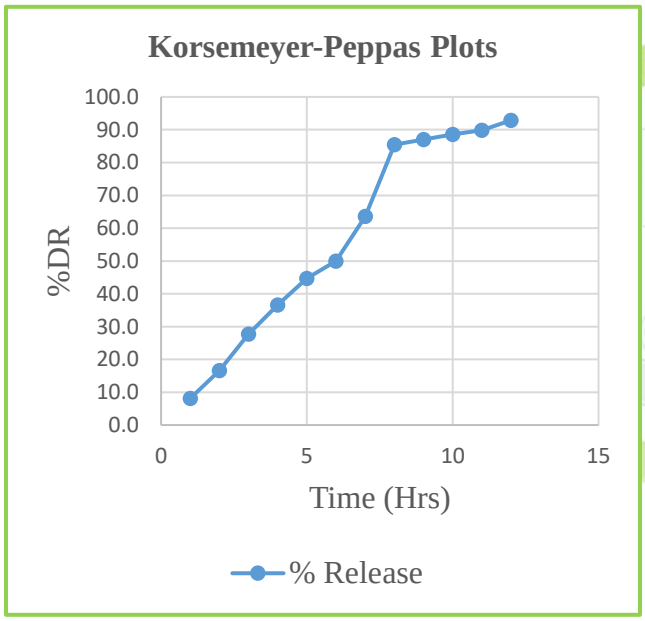

Figure: 13 Korsemeyer-Peppas Plots

Stability Studies

The stability study of F1 batch was performed at 40^{oC} and 75% Rh for 3 months. After performing the dissolution of F1 batch after 3 months the percentage drug release was found to be 90.73%. It has been observe that there is no significant difference in %drug release and %DEE after stability study as shown in table 6.

Table: 6 Results of Stability study of F1

Sr. no.	Parameters evaluated	Before stability	After stability
01	%DEE	68.85%	65.25%
02	%DR	92.21%	90.73%

In-vitro Drug Release after 3 Months

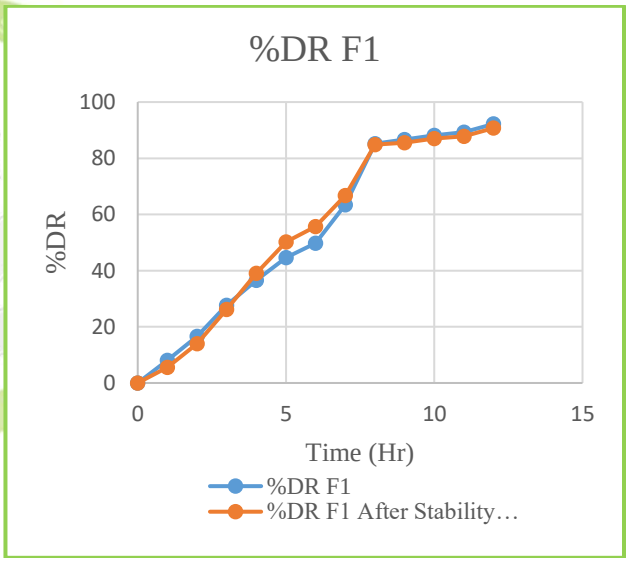

Figure: 14 Comparison of %DR of F1 before & after stability study

Table: 7 *In-vitro* Drug release of F1 after 3 months

Time (Hrs)	1	2	3	4	5	6	7	8	9	10	11	12
%DR	5.53	14.01	26.18	39.09	50.16	55.69	66.76	84.83	85.57	87.04	87.78	90.73

(Note: all the values are n=3**)**

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CONCLUSION

In the present study floating microsphere of Glipizide was prepared by emulsion solvent evaporation method by using Eudragit S100 and Ethyl cellulose as a polymer. When microspheres prepare by using Ethyl Cellulose having low viscosity does not provide proper intactness but when microspheres prepared by Ethyl cellulose having high viscosity it provide better intactness. As decreased the concentration of Eudragit S100 decrease the %Practical Yield. It has been observe that as increasing the concentration of Ethyl cellulose increasing the size of microspheres due to high viscosity. The drug entrapment efficiency of floating microspheres increase as increasing the concentration of Eudragit S100 and Calcium Chloride to aqueous phase. As increasing the concentration of Ethyl Cellulose 22cps increases the buoyancy time. Due to formation of hollow cavity.

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