Box-Behnken Experimental Design in Development of Glimepiride Floating Matrix Tablets

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Abstract: Floating matrix tablets of Glimepiride were developed to enhance its bioavailability by prolonging the gastric residence time in which Glimepiride was chosen as a model drug because of its has incomplete absorption due to its low gastric residence time. Floating matrix tablets were prepared using melt granulation technique. Bees wax was used as a hydrophobic meltable material. Hydroxypropylmethyl cellulose (Hypromellose K4MCR), sodium bicarbonate (sodium bicarb.) and ethyl cellulose (EC) were used as matrixing agent, gas generating agent and floating enhancer, respectively. Tablets were evaluated for physical characteristics such as weight, thickness, hardness, % friability and drug content. Tablets also were subjected to *in vitro* evaluation as buoyancy test (floating lag time), floating duration and drug release profile for 24 hours. A Box – Behnken design was applied to investigate the combined effect of 3 formulation variables including amount of hypromellose (X1), sodium bicarbonate (X2) as well as ethyl cellulose (X3). Fifteen batches were prepared and evaluated. Floating lag time, Flag (Y1), percent of drug released in 5 hours (Y2) and percent of drug released in 12 hours (Y3) were taken as responses. Obtained results of multiple regression analysis indicated that, high level of hypromellose (50 mg), high level of sodium bicarbonate (20 mg) and intermediate level of ethyl cellulose (15 mg) should be used to manufacture the tablet formulations with the desired *in vitro* floating time and dissolution. In addition; Formulations developed using Box – Behnken design, were fitted to various kinetic models for drug release. Formulation F7 was selected as a promising formulation.
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Key words: Floating Matrix tablet, Box – Behnken design, Glimepiride, Melt granulation technique.

1. Introduction

Glimepiride is a FDA approved sulphonyl urea oral antidiabetic drug, which has rapid and complete absorption after oral administration **(Rubina *et al*., 2011)**. Diabetes affects the gastric emptying rate, thus incomplete absorption of the drug is often accompanied by lesser bioavailability **(Shweta *et al*., 2005)**. Enhancement of the gastric retention would enable extending the absorption phase of the drug **(Singh and Kim, 2000, Chawla, Bansal, 2003)**. Oral administration of gastroretentive dosage form of glimepiride would attain the retained dose in the stomach due to floating **(Deshpande *et al*., 1997, Menon *et al*., 1994)** and causes the drug to be released in sustained manner, so that the drug could be released continuously to its absorption site in the upper gastro intestinal tract. This mode of administration would be best achieving the hypoglycemic effect of the drug.

Based on this idea, an attempt was made to formulate floating matrix tablets of glimepiride using different ratios of hypromellose. The prepared tablets were evaluated physically for their weight, thickness, hardness and % friability. All the tablets were evaluated for *in vitro* floating capacity, *in vitro* drug release profile and release kinetics. Optimization was carried out using Statgraph Box – Behnken technique to choose the most promising formula.

2. Materials and methods:

Materials:

Glimepiride was kindly supplied by Egyptian Group for drug trading Co. (EGD), Cairo, Egypt. Bees wax was purchased from Sigma – Aldrich, USA. Hydroxypropylmethyl Cellulose (Hypromellose K4MCR, 4000 cps) and ethyl cellulose were gift samples from SEPPIC, France. Sodium bicarbonate was purchased from Loba Chemie, Mumbai, India. Other chemicals were purchased from ADWEC, Egypt and they were of analytical grade and were used as received.

Methods:

Preparation of glimepiride floating tablets:

Bees wax was melted in a large plate then the required quantity of glimepiride was added to the molten mass. Previously prepared mixture of HPMC K4MCR and/or EC and sodium bicarbonate was added to the molten glimepiride – bees wax mixture and stirred well to mix. The mass was then removed from the hot plate and subjected to scraping until it attained room temperature. The coherent mass was passed through a 60-mesh sieve and the resulting granules were resifted on a 100-mesh sieve to remove the fines.

Granules from both 60- and 100- mesh sieves were collected (about 5 gm) and mixed with 2% w/w talc and 1% w/w magnesium stearate **(Patel *et al*., 2007)**. This lubricated homogenous blend was compressed into tablets using 8 mm flat-face standard concave punch and die tablet compression machine, single punch tablet press, Erweka, type EK:0 Erweka

Apparatabeous, Frankfurt, Germany. Compression force was adjusted to obtain tablets with hardness in range of 5 to 6 kg/cm^2. Tablet formulation and evaluation results of preliminary trials' batches (P1 to P7) are shown in table (1).

Table (1): Floating matrix formulation and Evaluation of preliminary trials.

Formulation ingredient	P1	P2	P3	P4	P5	P6	P7
Glimepiride(mg)	4	4	4	4	4	4	4
Beeswax(mg)	15	15	15	15	15	25	30
HPMC K4MCR(mg)	70	60	50	40	25	40	35
Sodium bicarbonate(mg)	10	10	10	10	10	10	10
Ethyl cellulose(mg)	0	10	20	30	45	20	20
Floating lag time(sec)	320	300	260	253	250	240	340
Floating time without rupture of tablets(min)	<190	<190	<190	<190	<190	>730	>730

HPMC K4MCR indicates hypromellose K4MCR. All batches contain 1% magnesium stearate and 2% talc, thus total weight of each floating matrix equals 102 mg.

Box-Behnken experimental design:

A Box-Behnken statistical design with 3 factors, 3 levels and 15 runs was selected for the optimization study (**Box and Behnken 1960, Nirav *et al*., 2010**).The experimental design consists of a set of points lying at the midpoint of each edge and the replicated center point of multidimensional cube. Independent variables and intervals selected to perform the mixture design are listed in table (2). Table (3) summarizes the composition of glimepiride floating tablet formulations and their responses according to Box-Behnken design.

Table (2): Variables and intervals selected to perform Box-Behnken design.

Independent variable	Level		
	Low	Medium	High
Hypromellose (X1)	30	40	50
Sodium bicarbonate (X2)	10	15	20
Ethyl cellulose (X3)	5	15	25
Transformed value	-1	0	1

The weight of independent variable X1, X2 and X3 is measured in mg.

Table (3): Composition of glimepiride tablet formulations and their responses according to Box-Behnken design.

	Independent variable			Response		
Formula	HPMC (X1)	Sod bicarb(X2)	EC(X3)	F lag (Y1)	Release 5(Y2)	Release 12(Y3)
F1	30	10	15	164.0	42.96	70.38
F2	30	15	5	175.0	43.23	59.98
F3	40	15	15	181.0	47.69	62.34
F4	30	15	25	156.0	52.79	68.64
F5	40	10	25	167.0	49.62	63.29
F6	40	10	5	190.0	46.72	70.32
F7	50	20	15	210.0	40.32	80.63
F8	40	15	15	181.0	47.69	62.34
F9	50	10	15	226.0	43.72	74.32
F10	50	15	25	214.0	42.79	83.29
F11	40	20	25	160.0	40.29	72.39
F12	30	20	15	176.0	47.62	84.52
F13	40	20	5	176.0	41.29	78.92
F14	50	15	5	214.0	47.69	83.34
F15	40	15	15	181.0	47.69	62.34

The amount of glimepiride was fixed at 4mg, beeswax was fixed at 15 mg and total weight of the floating matrix tablet was fixed at 125 mg by completing the weight by anhydrous lactose as filler. The weight of independent variable X1, X2 and X3 is measured in mg.

Physical characterization:

The fabricated tablets were characterized for weight variation (n=20) using Electric balance, (Mettler J100 Switzerland), hardness (n=6) using Tablet Hardness Tester, Type PTB 301, Germany, thickness using Tablet Thickness Apparatus, Planimeter, India and % friability (n=20) using Tablet Friability Test apparatus USP Standards, DA-6D, Bombay-400-069, India.

Assay of tablets:

Twenty tablets from each batch were weighed and powdered. Powder equivalent to 4 mg of glimepiride was accurately weighed and transferred into a 100 ml volumetric flask and shake with 100 ml of methanol for 10 minutes. Then 10 ml of methanol solution was diluted up to 100 ml with 0.1 N HCl with 0.5% w/v sodium sulphate and sonicated for 5 minutes to get a concentration about 4 µg/ml. A portion of the sample was filtered through 0.45 µ membrane filter and analyzd by UV spectrophotometer, Jenway 6105UV/Vis, England at 230 nm, which was previously determined by UV scanning (**Rubina *et al*., 2011**).

Buoyancy test (floating lag time):

The prepared floating matrix tablets of glimepiride were subjected to the buoyancy test (n=6) as described by **Rosa *et al* (1994)**. The tablets were placed in a 100 ml glass beaker containing 0.1 N HCl (simulated gastric fluid, SGF) and the time required for the tablet to rise to the surface and float was determined as floating lag time (Flag).

***In vitro* dissolution studies:**

The drug release profile of fabricated glimepiride floating matrix tablets (n=6) were determined using USP apparatus I (Dissolution Apparatus USP Standards, Scientific, DA-6D, Bombay, 400-069, India). The dissolution medium was 900 ml of Simulated Gastric Fluid (SGF), 0.1 N HCl with 0.5% w/v sodium lauryl sulphate at 37 ± 0.5 °C with 50 rpm. Samples were withdrawn at regular intervals over 24 hours, filtered through 0.45 µ membrane filter. Filtered samples were analyzed spectrophotometrically at λ_{max} 230 nm. The amount released was calculated from the regression line of the standard curve developed in the same medium (**Rubina *et al*., 2011**).

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Drug release kinetics:

Formulae that showed reasonable delay in drug release were subjected to kinetic analysis by fitting the release data to different kinetic models to explain the release kinetics of glimepiride from various floating tablets. These kinetic data were estimated using different kinetic orders. Zero order as cumulative amount versus time (Equation 1), first order as log cumulative amount of drug remaining versus time (Equation 2) and Higuchi's model as cumulative percentage of drug released versus square root of time (Equation 3)**(Higuchi,1963)**.

$C = K_o t$ ---------- (1)

$Log C = Log C_o – Kt/2.303$ ---------- (2)

$Q = kt^{1/2}$ ---------- (3)

Linear regression analysis was performed on those data. A linear relationship indicates a constant or near zero order release for cylindrical homogenous matrix systems. The release data was further treated by the equation of **Ritger and Peppas (1987)** (Equation 4) . This equation was treated logarithmically to determine the value of (n).The value of (n) determines the type of drug release as revealed in table (4).

$M_t / M_\infty = kt^n$ ---------- (4)

Where M_t/M_∞ is the fractional solute release, it is the release line. K is the kinetic constant characteristic of the drug polymer system and n is an exponent that characterizes the mechanism of the drug release

Table (4) Transport mechanisms from a polymer tablet under sink conditions.

n^a	Transport Mechanism
0.5	Fickian diffusion (Higuchi release)
0.5 < n < 1.0	Non – Fickian (anomalous)
1.0	Time – independent linear transport (zero order release)
n > 1.0	Super case II transport

3. Results and Discussion:

Preliminary trials:

Bees wax was selected as a hydrophobic meltable material to impart sufficient integrity to the tablets. Hypromellose K4MCR was selected as a matrixing agent, considering its widespread applicability and excellent gelling activity in sustained release formulations. Sodium bicarbonate generates CO_2 gas in the presence of hydrochloric acid present in dissolution medium. The gas generated is trapped and protected within the gel (formed by hydration of HPMC K4 MCR), thus decreasing the density of the tablet. As the density of the tablet falls below 1(density of water), the tablet becomes buoyant. EC was used as floating enhancer. It also works as a dissolution retardant, being insoluble in gastric pH. Five preliminary batches (P1-P5) were prepared using the same amounts of sodium bicarbonate and beeswax but different amounts of

hypromellose K4MCR and EC. The amount of hypromellose K4MCR was decreased, while the amount of EC was increased from batch P1 to P5. **(Patel *et al*., 2007)**. From the evaluation results (Table 1), it was observed that as the amount of EC was increased from 0 to 45 mg, the Flag decreased and this effect was significant on reducing Flag to 30 mg of EC. Hence, it was decided to optimize the amount of EC between 5 and 25 mg. As the amount of hypromellose K4MCR was increased from 25 to 70 mg, the Flag increased, indicating that a high amount of hypromellose K4MCR is undesirable to achieve low Flag. Below 25 mg, hypromellose K4MCR might not give sufficient strength to the matrix to prolong drug release up to 12 hours. Hence, it was decided to optimize hypromellose K4MCR between 30 and 50 mg.

Formulations P1 to P5 were subjected to *in vitro* dissolution study. All matrix floating tablets ruptured within 3 hours with more than 80% drug release. This result might be due to poor strength of tablets or to insufficient binding provided by beeswax, which failed to keep the matrix intact. Formulations P6 and P7 were prepared using 25 and 30 mg of beeswax, respectively, and were found to remain intact for more than 12 hours under stirring at 75 rpm in the dissolution studies. Formulation P 7 exhibited floating lag time of 340 seconds. These results might be due to poor penetration of simulated gastric fluid in a tablet core due to the presence of a high amount of beeswax. Hence, it was decided to keep the beeswax at 15 mg. It was decided to optimize sodium bicarbonate between 10 and 20 mg to decrease Flag to less than 3 minutes. Box-Behnken design was used in the present study to optimize the formulation for acceptance criteria (i.e Flag less than 3.5 minutes, release after 5 hours is 52% and release after 12 hours is 90%).

Box-Behnken design:

The amounts of matrixing agent (hypromellose K4MCR, X1), gas-generating agent (sodium bicarbonate, X2) and floating enhancer (EC, X3) were selected as independent variables in Box-Behnken design. The floating lag time (Flag, Y1), release % after 5 hours (Release 5, Y2) and release % after 12 hours (Release 12, Y3) were taken as responses. Based on the experimental design, the factor combinations yielded different mean responses **(Palamakula *et al*., 2004)**. Table (3) summarizes the experimental runs, their factor combinations and the levels of experimental units in the study as well as Flag, % release after 5 hours and % release after 12 hours. In order to determine the levels of factors which yielded optimal Flag and % release, mathematical relationships were generated between the dependent and independent variables. Using Box-Behnken design, the model was fitted to the data. Regression analysis of the data was carried out in SAS (Statistical Analysis System) by a special cubic model **(Kincl *et al*., 2005) (Panchagnula *et al*., 2007) (Abdel-Fattah *et al*., 2007) (Dalia *et al*., 2008).**

Standard physical tests:

Quality control tests of the prepared floating matrices of glimepiride were summarized in table (5).

Dissolution studies:

Figures 1 and 2 illustrate the release rate profile of formulation batches, F1 to F7 and F8 to F15, respectively. It is obvious that most of drug (about 60%) released after about 5 hours and thus release 5 was considered as one of the responses (Y2). Also, about 80% of drug released after 12 hours and thus, release 12 was considered as one of the responses (Y3).

Table (5): Quality control tests of glimepiride' floating matrices.

Formula	Mean weight (mg)	Thickness (mm)	Drug content (%)	Hardness (kg)	Friability (%)	Disintegrating time(min)	Flag (sec)
F1	124.9	5.42	97.3	5.22	0.099	swelling	165
F2	124.8	5.33	98.6	5.39	0.097	swelling	175
F3	125.3	5.31	99.7	5.83	0.102	swelling	181
F4	125.3	5.52	99.4	5.49	0.1004	swelling	157
F5	124.7	5.42	98.9	5.52	0.096	swelling	167
F6	125.4	5.63	99.02	5.87	0.098	swelling	190
F7	124.9	5.57	98.62	5.93	0.102	swelling	210
F8	125.3	5.31	99.7	5.83	0.102	swelling	181
F9	125.3	5.72	99.37	5.37	0.100	swelling	227
F10	124.9	5.62	99.42	5.97	0.100	swelling	214
F11	125.1	5.53	100.32	5.39	0.102	swelling	161
F12	125.4	5.97	102.61	5.79	0.103	swelling	176
F13	125.2	5.39	99.86	5.79	0.100	swelling	177
F14	125.3	5.49	98.97	5.88	0.998	swelling	215
F15	125.3	5.31	99.7	5.83	0.102	swelling	181

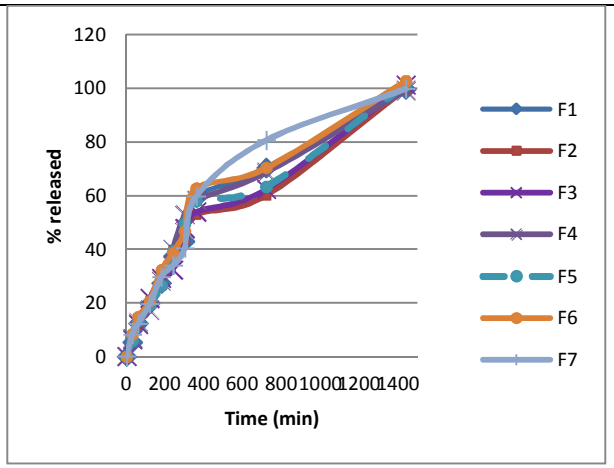


Figure (1): Dissolution profile of glimepiride floating matrix formulations (F1-F7)

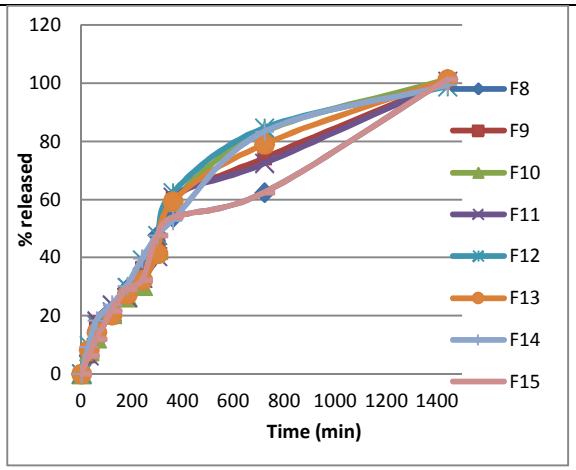


Figure (2): Dissolution profile of glimepiride floating matrix formulations (F8-F15)

Table (6) reveals the dissolution kinetic parameters of glimepiride from floating matrices which are the release exponent (n), correlation coefficient (R^2) and kinetic constant (K). On applying data in Table (4), it was obvious that all the investigated formulae followed Non-Fickian diffusion model of drug release and also indicate a coupling of the diffusion and erosion mechanism so called anomalous diffusion which may indicate that the drug release profile was controlled by more than one process **(Carmelo et al.,2006)**.

Table (6): Kinetic parameters of dissolution from glimepiride' floating matrices.

Formula	Parameters		
	Release exponent(n)	Correlation coefficient (R^2)	Kinetic constant (K)
F1	0.759	0.982	0.129
F2	0.721	0.984	0.157
F3	0.717	0.985	0.163
F4	0.723	0.978	0.164
F5	0.729	0.979	0.155
F6	0.664	0.986	0.241
F7	0.696	0.989	0.192
F8	0.717	0.985	0.163
F9	0.691	0.977	0.200
F10	0.707	0.988	0.179
F11	0.679	0.973	0.212
F12	0.637	0.986	0.292
F13	0.677	0.990	0.213
F14	0.621	0.991	0.319
F15	0.717	0.985	0.163

Simple Lattice design:

The amount of matrixing agent (HPMC K4MCR, X1), gas-generating agent (sodium bicarbonate, X2) and floating enhancer (EC, X3) were selected as independent variables in a simplex lattice design. The floating lag time (Flag), release % after 5 hours (release 5) and release % after 12 hours (release 12) were taken as responses. A statistical model incorporating 7 consecutive terms was used to evaluate the responses.

$$Y = bo + b1x1 + b2x2 + b3x3 + b12x1x2 + b23x2x3 + b13x1x3 + b123x1x2x3 \quad (5)$$

The regression equations of the fitted models relating the responses Flag, release 5 and release 12 to the transformed factor are shown in the following equations 4, 5 and 6, respectively.

$$\textbf{Flag} = 263.857 - 8.0*X1 + 6.55*X2 - 1.35*EC + 0.1475*X1^2 - 0.14*X1*X2 + 0.0475*X1*X3 - 0.07*X1^2 + 0.035*X2*X3\ 0.06*X3^2 \quad (6)$$

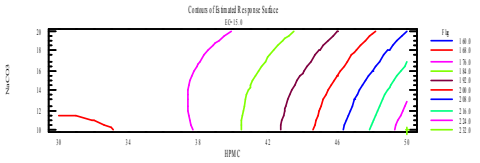
$$\textbf{Release 5} = -35.895 + 1.75175*X1 + 5.275*X2 + 1.8565*X3 - 0.00945*X12 - 0.0403*X1*X2 - 0.03615*X1*X3 - 0.1236*X2^2 \quad (7)$$

$$\textbf{Release 12} = 197.904 - 5.69238*X1 - 5.04175*X2 - 0.014375*X3 + X1^2 - 0.03915*X1*X2 - 0.021775*X1*X3 + X2^2 + 0.0025*X2*X3 + 0.0262*X3^2 \quad (8)$$

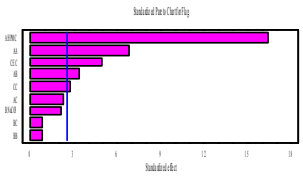
Three-dimensional (3D) response surface plots:

Three-dimensional (3D) plots for the measured responses were formed, based on the model polynomial functions to assess the change of the response surface. Also the relationship between the dependent and independent variables can be further understood by these plots **(Baumgartner et al., 2000)**. Since the model has more than two factors, one factor was constant for each diagram. Response surface plots with contour below for responses Y1, Y2 and Y3 are further presented in figures (3 - 5). Figures (3-a,b,c) show response surface plots (3D) with contour below of the effect of HPMC (X1), $NaHCO_3$ (X2) and EC (X3) on the response (Y1), F lag. While, figures (4-a,b,c) show response surface plots (3D) with contour below of the effect of X1, X2 and X3 on the response (Y2), release

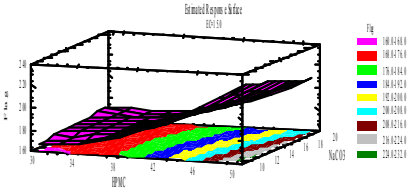
5. Finally, figures (5-a,b,c) show the effect of X1, X2 and X3 on the response (Y3), release 12. Figure (3-b, 4b and 5b), standardized pareto chart, depicts the main effect of independent variables, X1, X2 and X3 on Y1 (F lag), Y2 (release 5) and Y3 (release 12), respectively. The length of each bar in the graph indicates the effect of these factors and the level of their effects on responses. The length of the bar extending behind the reference line indicates the extent of corresponding factor effects on Y1, Y2 or Y3.



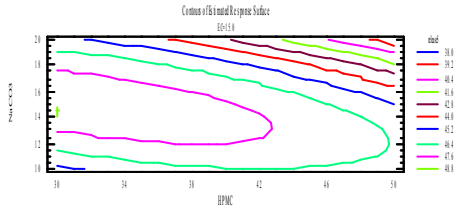
(3-a) Contour of estimated response surface



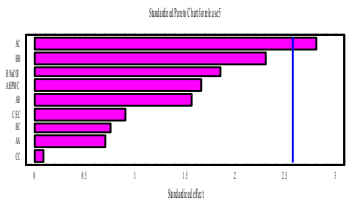
(3-b) Standard pareto chart showing the effects of independent variables HPMC(X1), $NaHCO_3$(X2) and EC(X3) and their combined effect on F lag.



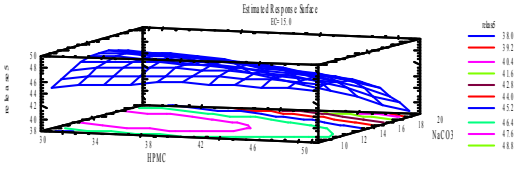
(3-c) Response surface plot (3D) with contour below showing the effect of X1, X2 and X3 on Y1 response

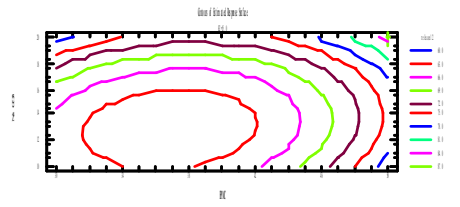


(4-a) Contour of estimated response surface.

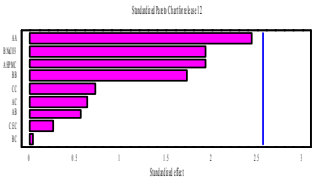


(4-b) Standard pareto chart showing the effects of independent variables HPMC(X1), $NaHCO_3$(X2) and EC(X3) and their combined effect on Release 5

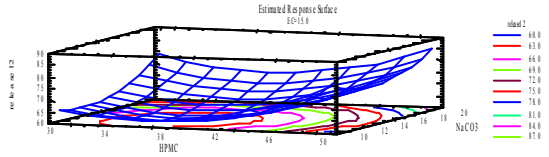




(5-a) Contour of estimated response surface.



(5-b) Standard pareto chart showing the effects of independent variables HPMC(X1), $NaHCO_3$(X2) and EC(X3) and their combined effect on Release 12.



(5-c) Response surface plot (3D) with contour below showing the effect of X1, X2 and X3 on Y3 response

Regression Analysis:

Using the software described earlier, the model was fitted to the data. Regression analysis of the data was carried out in SAS (Statistical Analysis System) by a special cubic model. From ANOVA study on the data of floating (Flag) which are given in Table (7), the statistical significance of each effect was tested by comparing the mean square against an estimate of the experimental error. In this case it was noted that 2 effects (HPMC and EC) have *p*- values less than 0.05 indicating that they are significantly different from zero at the 95% confidence level.

Table (7): ANOVA for Flag

Source	Sum of Squares	Df	Mean Square	F-Ratio	*P*-Value
A:HPMC	4656.13	1	4656.13	269.92	0.0000
B:NaHCO3	78.125	1	78.125	4.53	0.0866
C:EC	420.5	1	420.5	24.38	0.0043
AA	803.308	1	803.308	46.57	0.0010
AB	196.0	1	196.0	11.36	0.0199
AC	90.25	1	90.25	5.23	0.0709
BB	11.3077	1	11.3077	0.66	0.4549
BC	12.25	1	12.25	0.71	0.4378
CC	132.923	1	132.923	7.71	0.0391
Total error	86.25	5	17.25		
Total (corr.)	6552.93	14			

R-squared = 98.6838 percent R-squared (adjusted for d.f.) = 96.3146 percent
Standard Error of Est. = 4.15331 Mean absolute error = 1.9 Durbin-Watson statistic = 2.44348

Also, Table (8) shows the ANOVA for the data of release 5, the statistical significance of each effect was tested by comparing the mean square against an estimate of the experimental error. In this case it was noted that 0 effects have *p*- values less than 0.05 indicating that they are significantly different from zero at the 95.0% confidence level.

Table (8): ANOVA for Release 5.

Source	Sum of Squares	Df	Mean Square	F-Ratio	*P*-Value
A:HPMC	18.2408	1	18.2408	2.75	0.1581
B:NaHCO3	22.7813	1	22.7813	3.44	0.1230
C:EC	5.3792	1	5.3792	0.81	0.4091
AA	3.29732	1	3.29732	0.50	0.5122
AB	16.2409	1	16.2409	2.45	0.1784
AC	52.2729	1	52.2729	7.88	0.0377
BB	35.2545	1	35.2545	5.32	0.0693
BC	3.8025	1	3.8025	0.57	0.4830
CC	0.0531692	1	0.0531692	0.01	0.9321
Total error	33.1572	5	6.63145		
Total (corr.)	189.093	14			

R-squared = 82.4651 percent R-squared (adjusted for d.f.) = 50.9023 percent
Standard Error of Est. = 2.57516 Mean absolute error = 1.16 Durbin-Watson statistic = 2.17768Estimation

Also, ANOVA for the data of release 12 which are given in Table (9), shows the statistical significance of each effect that tested by comparing the mean square against an estimate of the experimental error. In

this case it was noted that 0 effects have *p*- values less than 0.05 indicating that they are significantly different from zero at the 95 % confidence level.

Table (9): ANOVA for Release 12.

Source	Sum of Squares	Df	Mean Square	F-Ratio	*P*-Value
A:HPMC	181.07	1	181.07	3.73	0.1112
B:NaHCO3	181.928	1	181.928	3.75	0.1105
C:EC	3.06281	1	3.06281	0.06	0.8116
AA	289.354	1	289.354	5.97	0.0585
AB	15.3272	1	15.3272	0.32	0.5983
AC	18.966	1	18.966	0.39	0.5592
BB	145.155	1	145.155	2.99	0.1442
BC	0.0625	1	0.0625	0.00	0.9728
CC	25.3455	1	25.3455	0.52	0.5021
Total error	242.504	5	48.5008		
Total (corr.)	1057.15	14			

R-squared = 77.0606 percent R-squared (adjusted for d.f.) = 35.7697 percent
Standard Error of Est. = 6.96425 Mean absolute error = 3.26967 Durbin-Watson statistic = 2.64203

The high values of correlation coefficient for F lag (R^2 = 98.6838 %), release 5 (R^2 = 82.465%) and release 12 (R^2 =77.0606%) indicate a good fit (i.e. good agreement between the dependent and independent variables).

Prediction of the optimized glimepiride floating matrices:

After generating the model polynomial equations to relate the dependent and independent variables, the process was optimized for responses.

The promising formulation was selected on the basis of the acceptance criteria for Flag, release 5 and release 12 as mentioned earlier. Formulation F9 passed the criteria for Flag. This formulation contains HPMC K4 MCR, $NaHCO_3$ and EC with concentrations 50, 10 and 15 mg, respectively.

This indicates that, to maximize Flag (228 seconds), high level of HPMC K4MCR (50 mg), low level of $NaHCO_3$ (10 mg) and intermediate level of EC (15mg) are required.

Formulation F4 passed the criteria for release 5. This formulation contains HPMC K4 MCR, $NaHCO_3$ and EC with concentrations 30, 15 and 25 mg, respectively. This indicates that, to obtain maximum percent release after 5 hours (52%), low level of HPMC K4 MCR (30 mg), intermediate level of $NaHCO_3$ (15 mg) and high level of EC (25 mg), are required.

Formulations F7, F10, F12 and F14 passed the criteria for release 12, but F7 exhibited values which were very close to the predicted values. This formulation, (F7) contains HPMC K4 MCR, $NaHCO_3$ and EC with concentrations 50, 20 and 15 mg, respectively. This means that, to obtain maximum percent release after 12 hours, high level of HPMC K4 MCR (50 mg), high level of $NaHCO_3$ (20 mg) and intermediate level of EC (15 mg) are required.

Hence, formulation F7 was selected as a promising formulation, which was very close to all the predicted values for the three responses under experimental design after correlation between the obtained results. Table (10) reveals the observed and predicted responses and residual values for the responses, performed for the optimized glimepiride floating matrix, F7.

Table (10): observed and predicted values of the responses for the optimized glimepiride matrix (F7).

Response	Observed value
Predicted value	Residual
Flag (Y1)	210.0
208.0	2.0
Release 5 (Y2)	40.32
38.44	1.88
Release 12 (Y3)	80.63
85.03	- 4.4

Conclusion:

All the investigated glimepiride floating matrices follow non-fickian anomalous diffusion. An optimized formulation of 4 mg glimepiride floating matrix tablets was found to be gastro – retentive and controlled release over 12 hours. Design and analysis of experiments were used as good tools to obtain that experimental design methodology is a very economic way for extracting the maximum amount of complex information, a significant experiment time saving factor and moreover, it saves the materials used for analysis and personal costs as well. This means that, to obtain maximum percent release after 12 hours, high

level of HPMC K4 MCR (50 mg), high level of $NaHCO_3$ (20 mg) and intermediate level of EC (15 mg) are required. It was concluded that in near future, glimepiride floating matrices may be the drug of choice for treatment of Type II diabetes mellitus to improve the clinical efficiency.

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