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Formulation Development and In-Vivo, In- Vitro Evaluation of Sitagliptin Phosphate Floating Microspheres

[1]Aftab Alam*,[2]Dr. S. Rajasekaran

[1]Faculty Of Pharmacy, Department of Pharmaceutics, Bhagwant University Ajmer

[2]Department of Pharmacology, Karuna College of Pharmacy, Palakkad, Kerala

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KEYWORDS
Floating microspheres, Sitagliptin Phosphate, In-vitro release, Bioavailability

ABSTRACT:

The purpose of the present investigation was the development and characterization of gastro-retentive floating drug delivery system for anti-diabetic drug Sitagliptin Phosphate that would retain the drug in stomach and continuously release the drug in controlled manner up to a predetermined time leading to improved bioavailability. Different formulations of Sitagliptin Phosphate were prepared as the floating microspheres using Hydroxypropyl methylcellulose (HPMC) and Eudragit RS100 polymers by emulsion solvent evaporation technique. The dried floating microspheres were evaluated for drug content, particle size analysis, incorporation efficiency, floating behavior and in-vitro drug release studies. The developed gastro retentive floating drug delivery system of Sitagliptin Phosphate showed excellent physicochemical properties and controlled drug release pattern, thereby improving the bioavailability of the drug and also manage the complicacy of the diabetes in a better manner.

1. Introduction

Gastro retentive delivery systems are designed to be retained in the stomach for a prolonged time and release their active ingredients and thereby enable sustained and prolonged input of the drug to the upper part of the gastrointestinal (GI) tract. Gastro retentive delivery system can be classified as follows.

- Bio-adhesive Drug Delivery System
- Expandable Drug Delivery System
- Floating Drug Delivery System and
- High-density systems

Among these systems, FDDS have been most commonly used. Floating drug delivery systems is one of the important approaches to achieve gastric retention to obtain sufficient drug bioavailability.

The Floating Drug Delivery System design appears to be one of the most effective and rational approaches for the controlled oral drug delivery. This FDDS appears to have a distinct advantage in delivering the drugs that are absorbed mainly in the upper part of the GI tract and drugs having stability and solubility problem in the lower part of intestine. This area of research therefore should be aimed at developing the dosage forms to increase the pharmacokinetic profile of the drugs. There are various natural polymers and gums, which need to be explored to find their use in the designing of floating dosage forms.The Floating systems were first described by Davis in (1968). The floating delivery systems can be retained in the stomach and assists in improving the oral sustained delivery of drugs that have an absorption window in a particular region of the gastrointestinal tract. These systems help in continuously releasing the drug before it reaches the absorption window, thus ensuring optimal bioavailability.

1.1 Effervescent Floating Drug Delivery System

These buoyant delivery systems utilize matrices prepared with swellable polymers such as polysaccharides, e.g., chitosan, and effervescent components, e.g., sodium bicarbonate and citric or tartaric acid [47, 48]. or matrices containing chambers of liquid that gasify at body temperature. The matrices are fabricated so that upon arrival in the stomach, carbon dioxide is liberated by the acidity of the gastric contents and is entrapped in the gellified hydrocolloid. This produces an upward motion of the dosage form and

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maintains its buoyancy. A decrease in specific gravity causes the dosage form to float on the chyme. The carbon dioxide generating components may be intimately mixed within the matrix, in which case a single-layered tablet is produced a bilayered tablet may be compressed which contains the gas generating mechanism in one hydrocolloid containing layer and the drug in the other layer formulated for a SR effect. This concept has also been exploited for floating capsule systems. The floating capsules by filling with a mixture of sodium alginate and sodium bicarbonate. The systems were shown to float during in gastric fluid as a result of the generation of CO_2 that was trapped in the hydrating gel network on expo- sure to an acidic environment.

Recently a multiple-unit type of floating microparticles, which generates carbon dioxide gas, has been developed.[48] The system consisted of controlled-release microparticles surrounded by double layers. The inner layer was an effervescent layer containing both sodium bicarbonate and tartaric acid. The outer layer was a swellable membrane layer containing mainly polyvinyl acetate and purified shellac. Moreover, the effervescent layer was divided into two sublayers to avoid direct contact between sodium bicarbonate and tartaric acid. Sodium bicarbonate was contained in the inner sublayer and tartaric acid was in the outer layer (figure1.1). When the system was immersed in a buffer solution at 37˚C, it sank at once in the solution and formed swollen microparticles, like balloons, with a density much lower than 1 g/ ml. The reaction was due to carbon dioxide generated by neutralization in the inner effervescent layers with the diffusion of water through the outer swellable membrane layers.

1.2 Non-Effervescent Floating Drug Delivery System

Non-effervescent floating dosage forms use a gel forming or swellable cellulose type of hydrocolloids, polysaccharides, and matrix-forming polymers like polycarbonate, polyacrylate, polymethacrylate, and polystyrene. The formulation method includes a simple approach of thoroughly mixing the drug and the gel-forming hydrocolloid. After oral administration this dosage form swells in contact with gastric fluids and attains a bulk density of < 1.

One of the approaches to the formulation of such floating dosage forms involves intimate mixing of drug with a gel-forming hydrocolloid, which swells in contact with gastric fluid after oral administration and maintains a relative integrity of shape and a bulk density of less than unity within the outer gelatinous barrier [38, 39].

When such dosage forms come in contact with an aqueous medium, the hydrocolloid starts to hydrate by first forming a gel at the surface of the dosage form. The resultant gel structure then controls the rate of diffusion of solvent into and drug out of the dosage form. As the exterior surface of the dosage form goes into solution, the gel layer is maintained by the immediate adjacent hydrocolloid layer becoming hydrated. As a result, the drug dissolves in and diffuses out with the diffusing solvent, creating a 'receding boundary' within the gel structure [38, 39].

A multilayered, flexible, sheet-like medicament device that was buoyant in the gastric juice of the stomach and had SR characteristics. The device consisted of at least one dry, self supporting carrier film made up of a water-insoluble polymer matrix having a drug dispersed and a barrier film overlaying the carrier film. This system worked as floating system.

1.3 Various attempts have been done to retain the dosage form in the stomach as a way of increasing retention time.

1.3.1 High-density systems [33, 35]

High-density systems having density of ~3 g/cm^3 are retained in the rugae of the stomach. The only major drawbacks with such systems is that it is technically difficult to manufacture them with a large amount of drug (>50%) and to achieve the required density of 2.4–2.8 g/cm^3.

1.3.2 Swelling systems

Swelling systems are capable of swelling to a size that prevents their passage through the pylorus. As a result, the dosage form is retained in the stomach for a longer period of time.Upon coming in contact with gastric fluid the polymer imbibes water and swells.[4, 5]

1.3.3 Bio/mucoadhesive systems [26, 27]

Bio/mucoadhesive systems bind to the gastric epithelial cell surface or mucin and extend the GRT by increasing the intimacy and duration of contact between the dosage

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form and the biological membrane. The epithelial adhesive properties of mucin have been applied in the development of gastroretentive drug delivery systems. The use of mucoadhesive microspheres consisting of a drug and Carbopol 934P (polyacrylic acid, polymerized in benzene and highly cross-linked with allyl sucrose), dispersed within a waxy matrix of polyglycerol esters of fatty acids has been reported.

1.3.4 Size-increasing drug delivery systems

Another approach to retaining a pharmaceutical dosage form in the stomach is by increasing its size above the diameter of the pylorus. However, owing to significant inter individual variations; the cut-off size cannot be determined exactly. Roughly, the dosage forms should be larger than 13 mm, but even bigger units have been found to be emptied from the stomach. In order to facilitate swallowing, it is highly desirable to design dosage forms with an initially small size that once in the stomach significantly increase in size. The expanded state should be achieved rapidly in order to prevent premature emptying through the pylorus. Conversely, the systems should also guarantee their clearance from the stomach after predetermined time intervals to avoid accumulation upon multiple administrations.

1.4 Factors affecting the efficacy of microspheres

Density -GRT is a function of dosage form buoyancy that is dependent on the density; density also plays an important role in the determining the location of the delivery system in the stomach. If density of the delivery system is higher than the gastric contents, then it sinks to the bottom of the stomach while low density drug delivery systems float on the surface.

Single or multiple unit formulation – multiple unit formulations show a more predictable release profile and insignificant impairing of performance due to failure of units, allow co-administration of units with different release profiles or containing incompatible substances and permit a larger margin of safety against dosage form failure compared with single unit dosage forms.

Fed or unfed state – under fasting conditions, the GI motility is characterized by periods of strong motor activity or the migrating myoelectric complex (MMC) that occurs every 1.5 to 2 hours. The MMC sweeps undigested material from the stomach and, if the timing of administration of the formulation coincides with that of the MMC, the GRT of the unit can be expected to be very short. However, in the fed state, MMC is delayed and GRT is considerably longer.[36,37]

Nature of meal – feeding of indigestible polymers or fatty acid salts can change the motility pattern of the stomach to a fed state, thus decreasing the gastric emptying rate and prolonging drug release.

Caloric content – GRT can be increased by four to 10 hours with a meal that is high in proteins and fats.

Frequency of feed – the GRT can increase by over 400 minutes when successive meals are given compared with a single meal due to the low frequency of MMC.

1.4.1 Practical approaches to design FDDS The concept of floating drug delivery system was described in the literature as early as 1968, when David disclosed a method of overcoming the difficulty experienced by some person of gagging or choking while swallowing medicinal pills. The author suggested that such difficulty could be overcome by providing pills having a density less than 1 g/ml so that pill will float on the surface of water.On the other hand Rouge and coworkers showed that multiple unit dosage from decreases the inter subject variability in absorption and minimizes probabilities of dose dumping by uniform distribution within the gastric content and provides longer duration of action . In the designing of FDDS, following rationale should be sought:

A. Rational in the stomach as per the clinical demand or need

B. Convenience for patient

C. Ability to load substantial amount of drug with different physiochemical properties and release them in a controlled manner

D. Complex matrix integrity of control formulation in the stomach, inexpensive optimization between floatation time and release rate, lag time must be less.

1.4.2 The advantages of floating microspheres

1. Improves patient compliance by decreasing dosing frequency.

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2. Bioavailability enhances despite first pass effect because fluctuations in plasma drug concentration is avoided, a desirable plasma drug concentration is maintained by continuous drug release.
3. Better therapeutic effect of short half-life drugs can be achieved.
4. Gastric retention time is increased because of buoyancy.
5. Drug releases in controlled manner for prolonged period.
6. Site-specific drug delivery to stomach can be achieved.
7. Superior to single unit floating dosage forms like tablets since microspheres release drug uniformly and there is no risk of dose dumping.

1.4.3 In Vivo Studies

Diabetes mellitus is a chronic disease characterized by high blood glucose level due to absolute or relative deficiency of circulating insulin levels.

Diabetes affects about 5% of the global population and management of diabetes without any side effects is still a challenge to the medical system. Diabetes mellitus is a disease that affects more than 100 million people and may attain about five times more subjects in the next 10 years. Its control involves exercise, diet and chemotherapy. Type 2 diabetes is also referred to as non-insulin dependent diabetes mellitus (NIDDM), or adult onset diabetes mellitus (AODM). In type 2 diabetes, patients can still produce insulin, but do so relatively inadequately for their body's needs. In many cases this actually means the pancreas produces larger than normal quantities of insulin.

A major feature of type 2 diabetes is a lack of sensitivity to insulin by the cells of the body. In addition to the problems with an increase in insulin resistance, the release of insulin by the pancreas may also be defective and sub optimal. In fact, there is a known steady decline in beta cell production of insulin in type 2 diabetes that contributes to worsening glucose control.

The alloxan and the product of its reduction, dialuric acid, establish a redox cycle with the formation of superoxide radicals. These radical undergo dismutation to hydrogen peroxide.

2. Materials & Methods

2.1 Method selection

2.1.1 Single emulsion solvent evaporation method

In this method the drug – sitagliptin phosphate and polymer- Eudragit RS100 and ethyl cellulose in 1:2 ratios were dispersed in 50ml ethanol. This mixture was stirred on a magnetic stirrer till it appeared as a clear solution. Magnesium stearate (10%wt/wt) was dispersed in the drug and polymer solution which acted as a droplet stabilizer. Drug-polymer mixture was added to 250ml of light liquid paraffin with continuous mechanical stirring at 1000rpm and solidifying agent added i.e.30ml n-hexane. The drug polymer mixture was added with continuous stirring with the help of mechanical stirrer. Solidifying agent was added drop by drop while stirring for rigidization of microspheres. Stirring was continued for two hour, until ethanol evaporated completely. The microspheres formed were collected by filtration in vacuum, washed 4-5 times with 50 ml petroleum ether each and dried at 30±2°C for 24 hours.

Table 2.1 Characterization of method

Methods	**Shape**	**% drug entrapment (wt/wt)**
Double emulsion method	Irregular particle	46
Multiple emulsion method	Porous particle	60
Solvent evaporation method	Regular and smooth surface of microspheres	80

A blunt end cannula fitted with plastic syringe was used to administer the optimized formulation (floating microspheres of sitagliptin phosphate), and standard drug (sitagliptin phosphate). The treatments were administered orally. Animals of all groups were treated with an oral D-glucose load of 2gm/kg by means of cannula. Group III, IV were treated orally with floating

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microspheres of sitagliptin phosphate at doses level of 100mg and standard drug (sitagliptin phosphate) 200mg/kg b.w. solution. Blood samples were withdrawn from the tail of each rat using sharp sterile blade under light ether anesthesia after 0min, 1hrs, 2hrs, 4hrs and 8 hours.

2.2 Optimization of drug and polymer ratio different formulations

Various batches F1, F2, F3, F4, F5, F6, F7, F8, F9 with different drug: polymer ratio were prepared by single emulsion solvent evaporation method. Keeping stirring speed and polymer ratio as independent variables by using factorial design $(3)^2$, to find out the dependent variable. The droplet stabilizer magnesium stearate (150mg) and solidifying agent, floating microspheres were prepared by single solvent evaporation method.

Table 2.2 Actual and code value of independent variables

Code value	Actual value		Drug concentration (mg)
	Polymer concentration	Stirring Speed	
	(mg) X1	X2 (rpm)	
-1	200	800	100 in all Batches
0	400	1000	
1	600	1500	

Table 2.3 Different formulation selected with drug polymer concentration with stirring speed

Batch Code	Drug (mg)	Polymer Ratio(X1)	Stirring speed (rpm) (X2)
F1	100	-1	0
F2	100	0	0
F3	100	-1	-1
F4	100	1	-1
F5	100	0	1
F6	100	1	1
F7	100	-1	-1
F8	100	0	0
F9	100	1	1

Table 2.4 Different formulation using with drug and polymer ratio concentration find out the particle size, drug loading, drug entrapment efficiency, yield and floating ability of floating microspheres of sitagliptin phosphate

Batch (Drug Polymer Ratio)	Mean ± S. D., n=20	Mean ± S. D., n=3				
	Average Diameter (µm)	Floating Duration (Hrs)	Floating Ability (%)	Yield (%wt/wt)	DEE (%wt/wt)	Drug Loading (%wt/wt)
F1	206.3(±2.497)	9	82.673(±0.577)	78.27(±0.015)	58.27 (±0.020)	25.32(±0.010)
F2	223.7(±2.946)	11	84.83 (±0.763)	79.02 (±0.009)	62.36 (±0.025)	38.88 (±0.015)
F3	259.8(±4.315)	12	86.01(±0.215)	81.05 (±0.040)	74.22 (±0.015)	50.24 (±0.010)
F4	259.1(±4.818)	12	88.60 (±0.360)	81.18 (±0.042)	80.51 (±0.015)	69.60(±0.020)
F5	255.1(±2.726)	12	95.47 (±0.416)	89.56 (±0.035)	85.38 (±0.015)	83.61(±0.010)
F6	293.5(±2.838)	10	80.78 (±0.503)	80.99 (±0.100)	75.32 (±0.010)	80.38(±0.010)
F7	314.7(±7.040)	10	75.72	79.23 (±	53.32	76.42(±0.00

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			(±0.249)	0.035)	(±0.015)	5)
F8	354.5(±5.949)	9	69.47 (±0.551)	77.82 (± 0.020)	42.73(±0.02 6)	64.58(±0.01 7)
F9	375.1(±8.252)	8	67.90 (±0.360)	74.55 (± 0.026)	41.04 (±0.051)	53.97(±0.00 5)

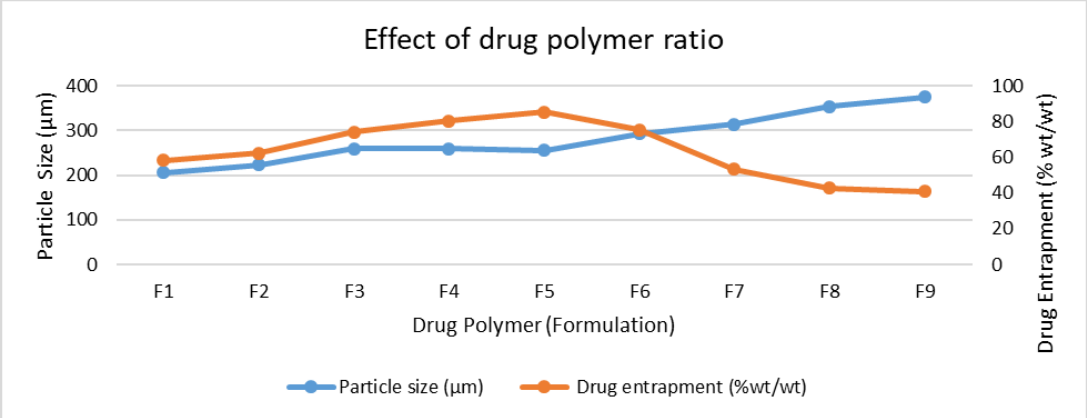

Figure 2.1 Effect of Drug-polymer ratio on average particle size and percent drug entrapment

2.3. Optimization of stirring speed

Stirring speed plays an important role in controlling the particle size and drug entrapment of floating microspheres. Floating microspheres were prepared by the method as described in 3.1.1. with optimized ratio of drug and polymer with different stirring speed. keeping the quantity of droplet stabilize and hardening agent constant, utilizing four different speeds i.e. 800, 1000 and 1500 rpm.

Table 2.5 Optimization of stirring speed

Batch (RPM)	**Mean ± S. D., n=20**	**Mean ± S. D., n=3**				
	Average Diameter (µm)	**Floating Duration (Hrs)**	**Floating Ability (%)**	**Yield (%wt/wt)**	**DEE (%wt/wt)**	**Drug Loading (%wt/wt)**
F1	199.0(±1.730)	09	81.87(±0.412)	77.27(±0.028)	59.30 (±0.022)	38.32(±0.110)
F2	208.3(±2.477)	08	85.47 (±0.406)	82.56 (± 0.035)	63.32 (±0.027)	42.88 (±0.215)
F3	240.5(±2.790)	11	80.56(±0.331)	75.27(±0.020)	67.51(±0.020)	60.24 (±0.210)
F4	242.5(±1.890)	11	78.23(±0.530)	73.27(±0.040)	79.38 (±0.015)	79.60(±0.220)
F5	256.1(±2.626)	12	96.47 (±0.406)	90.56 (± 0.035)	84.38 (±0.015)	84.61(±0.010)
F6	203.5(±2.738)	10	80.78 (±0.513)	80.91 (± 0.200)	78.32 (±0.110)	81.38(±0.210)
F7	334.7(±6.140)	10	75.72 (±0.239)	78.13 (± 0.135)	59.32 (±0.115)	76.42(±0.105)
F8	304.5(±4.949)	9	71.57 (±0.541)	76.82 (± 0.220)	52.73(±0.116)	74.58(±0.117)
F9	325.1(±7.252)	8	69.90 (±0.260)	76.55 (± 0.126)	49.04 (±0.351)	63.97(±0.105)

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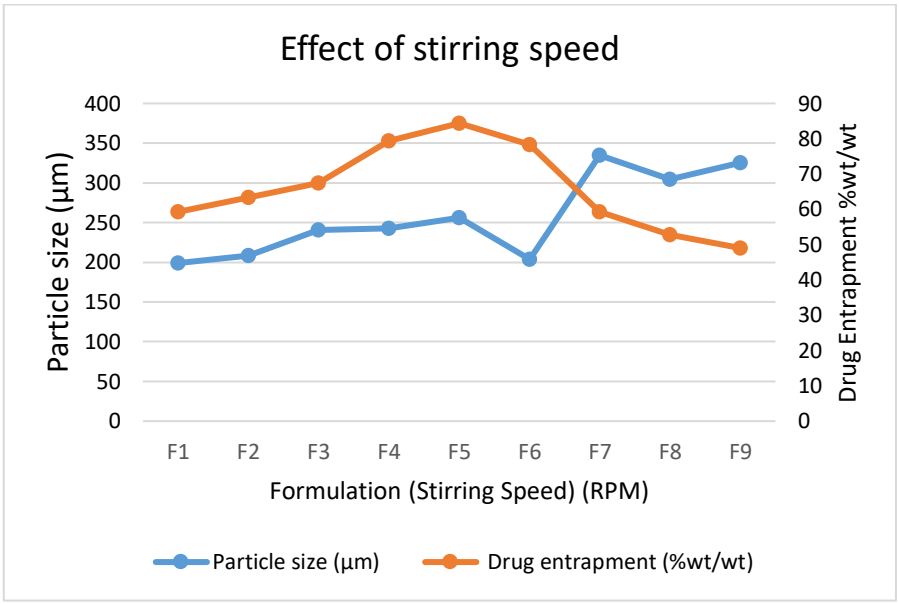

Figure 2.2 Effect of stirring speed on average particle size and percent drug entrapment

2.4 Floating microspheres characterization

2.4.1 Scanning electron microscopy

A microsphere is observed under an electron microscope. They are mounted directly onto the SEM sample stub using sided tape and coated with gold film under reduced pressure.

2.4.2 Particle size

Particle size of microspheres is measured by optical light microscopy. Data of size discussed earlier (Table no. 3.4-3.4)

2.4.3 Drug loading, Drug Entrapment Efficiency

A quantity of microspheres containing equivalent to 50mg of the drug was taken for evaluation. The amount of drug entrapped was estimated by crushing the microspheres and extracting with aliquots 100ml of 0.1 N HCl repeatedly. The extract was transferred to a 100 ml volumetric flask and the volume was made up using 0.1N HCl. The solution was filtered and the absorbance was measured after suitable dilution spectrophotometrically at 267nm against appropriate blank. The amount of drug entrapped in the microspheres was calculated by the following formula:

Drug Entrapment Efficiency = (Amount of drug actually present / Theoretical drug load expected) X 100

Drug loading = (Amount of drug actually present/ total weight of microspheres) X 100

2.4.4 Percent yield

The total amount of microspheres obtained was weighed and the percentage yield calculated taking into consideration the weight of the drug and polymer.

$$\text{Percent yield} = \frac{\text{practical yield X100}}{\text{theoretical yield}}$$

2.4.5 In-Vitro evaluation of floating ability

An in vitro floating study was carried out using simulated gastric fluid USP containing 1% Tween 80 as a dispersing medium. Microspheres were spread of over the surface of 900 ml of dispersion medium at 37±0.5°C and agitated by a paddle rotating at 100rpm.

% floating microspheres = (Weight of floating microspheres / initial weight of floating microspheres) X 100

2.4.6 In-Vitro drug release study

The in vitro drug release studies were carried

out by paddle method. A quantity of microspheres equivalent to 100 mg of the drug was used. The 900ml of 0.1N HCl was used as dissolution fluid. The paddle was rotated at a speed of 100 rpm and the whole system was thermally controlled at 37±1ºC. Five ml of the

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aliquots were withdraw at predetermined time intervals and filtered through whatman filter paper. The samples were suitable diluted with 0.1N HCl and the solutions were analyzed for the drug content spectrophotometrically at 267nm against reagent blank. The dissolution medium was replaced with same volume of 0.1N HCl to maintain sink condition. From this percentage drug release is calculated and plotted against function of time study the pattern of drug release

Table 2.5 Cumulative % release of sitagliptin phosphate from floating microspheres, in 0.1N HCl.

Time(h)	Cumulative % drug release								
	F1	F2	F3	F4	F5	F6	F7	F8	F9
01	21.45	20.79	22.76	21.45	24.33	21.58	11.74	8.46	7.28
02	23.94	23.15	27.33	26.43	31.15	24.46	18.3	18.82	11.47
03	33.12	29.58	34.43	35.75	38.63	32.33	27.87	22.5	21.31
04	41.91	33.12	39.68	40.99	44.14	37.19	34.3	27.48	26.04
05	44.67	36.27	45.59	47.55	50.00	44.54	38.5	31.94	30.63
06	46.77	44.01	55.56	55.95	59.74	47.55	42.31	37.84	35.35
07	51.87	47.55	64.74	66.97	70.5	51.1	46.37	42.83	40.86
08	59.23	57.39	68.2	75.63	77.68	57.26	49.39	45.72	45.06
09	66.31	67.5	71.96	81.4	85.66	62.12	53.59	47.82	47.29
10	70.91	72.48	73.96	83.5	87.29	68.81	57	53.06	51.1
11	73.79	74.71	74.45	84.42	88.34	72.74	60.54	56.34	53.59
12	74.84	75.37	76.68	85.21	90.04	73.79	63.69	60.41	58.31

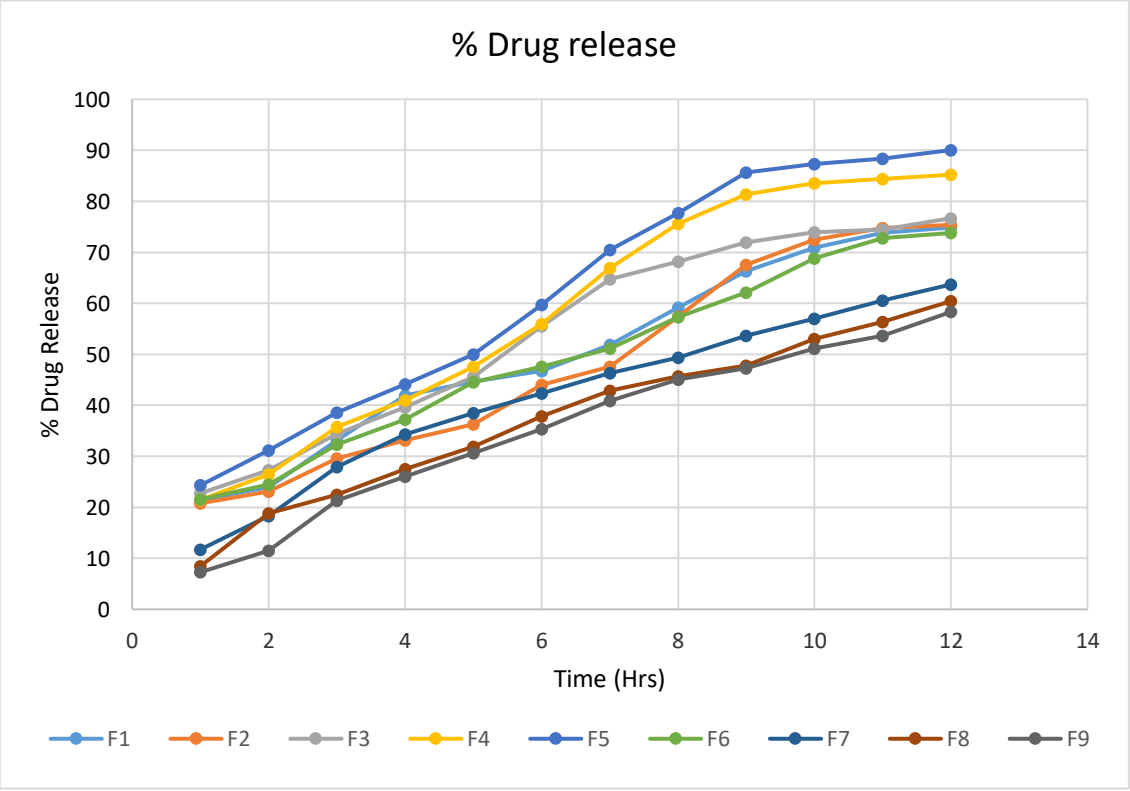

Figure 2.3 In-vitro release profile of floating microspheres of sitagliptin phosphate in 0.1N Hcl.

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2.4.7 Release kinetics

The release kinetics were studied by various kinetic models such as first order, zero order, Higuchi plot and modified Hixon Crowell model. All formulated batches followed the Higuchi model, which supports the release behavior of the optimized batch also.

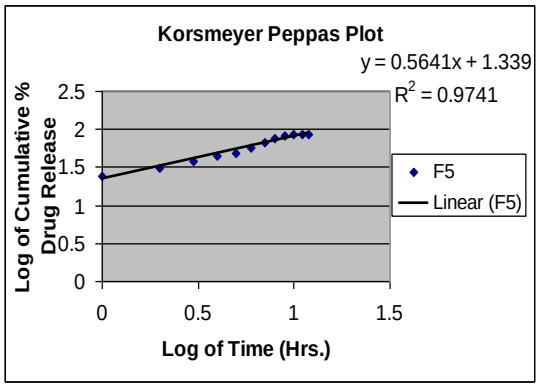

Figure 2.5 Korsmeyer-Peppas plot of the optimized batch

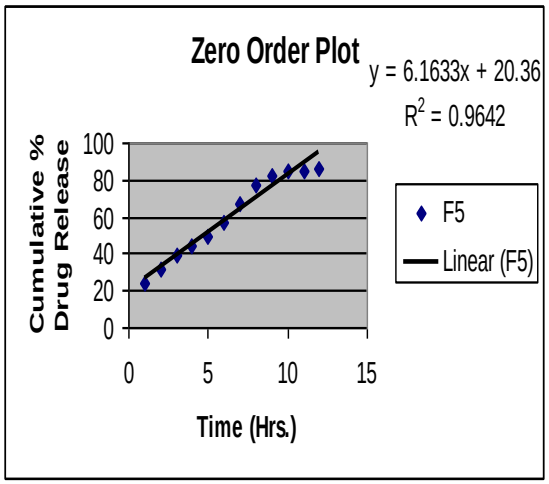

Figure 2.6 Zero order plot of the optimized batch

2.2.8 Statistical analysis

The data obtained from release rate determination studies of Sitagliptin phosphate floating microspheres were analyzed statistically with one- way ANOVA and Newman-Keuls multiple comparison test by using Graphpad Prism. Formulation F5 significantly differ from other formulations ($P<0.05$). So that at the polymer ratio of F5 drug release was more compared of other formulations.

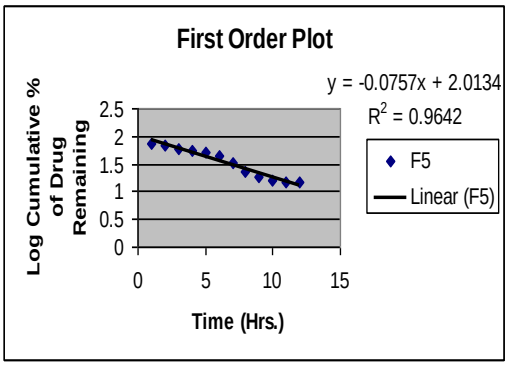

Figure 2.7 First order plot of the optimized batch

3. Material and methods

3.1. Experimental animals and their housing

The in vivo activity was required for controlled released action of floating microspheres over on conventional dosage form. The in vivo activity has been approved by Institutional Ethical Committee, Male Wister rats weighing 150-230g were obtained from the Animal House, Faculty of Pharmacy, MET Faculty of Pharmacy Ram Ganga Vihar Moradabad Uttar Pradesh.

The animals were housed in well ventilated animal house under natural photoperiodic condition in large polypropylene cages and were fed standard pallet diet and water ad libitum.

3.1.2 Experimental Induction of Diabetes in Rats

Diabetes was induced in adult male Wister rats by single intraperitonial injection of alloxan monohydrate [100mg/kg body weight], dissolved in normal saline [0.9% w/v] for three consecutive days. Diabetes was confirmed on 4th day after alloxan monohydrate dose administration by determining blood glucose concentration using glucometer (Accu- Check). Only animals with blood glucose level ≥220mg/dL were used for study.

3.2 Experimental design and procedure

The animals used for experiment were adult male Albino rats (Wistar strain150- 250g). Total of 24 rats were taken, 6 rats in each group, (table 5.1) and were fasted 16 hours before the day of experiment with free access to water. Diabetes was induced in all rats by intraperitonial injection of alloxan monohydrate. The dose of alloxan monohydrate was 125mg/kg body

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weight. [1, 3] The vehicle used for preparation of alloxan monohydrate was normal saline.

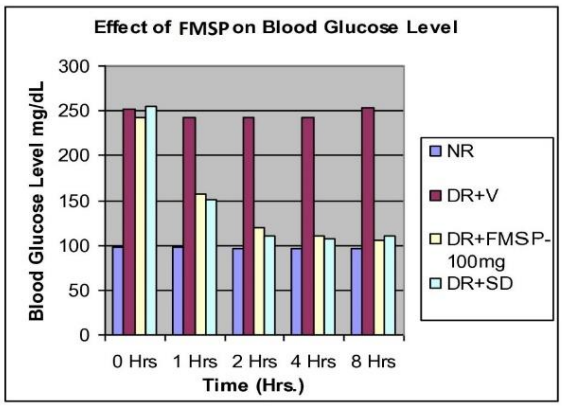

Figure 3.1 Effect of FMSP on blood glucose level

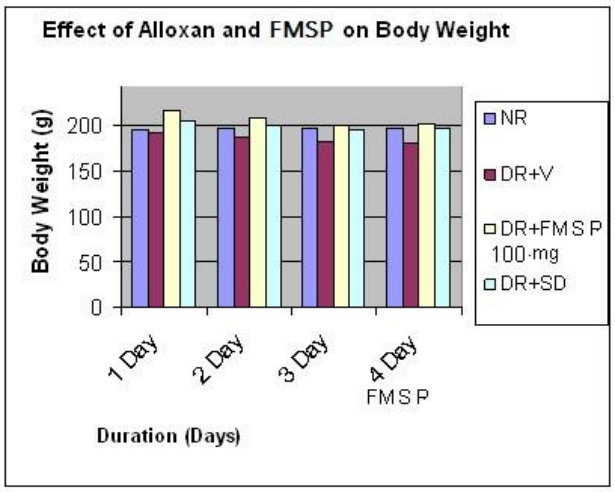

Figure 3.2 Effect of alloxan and FMSP on body weight

4. Results and Discussion

4.1 Selection of the method of preparation

Solvent evaporation technique was selected as a method of choice in the present research due to its advantages described as follows. The versatility and flexibility of methods allows for the use of different polymers and solvents. Solvent evaporation technique permits higher polymer concentration per batch production improving the microparticles yield batch. It can be used for entrapment of hydrophilic drug. The fast evaporation rate of the solvent permits a reduction in the processing time; moreover the evaporation rate may be used to control the microspheres size as compared with other methods.

In double emulsion method the formed microspheres were observed to have a irregular shape and low drug entrapment. This may be attributed to instability of primary emulsion, as droplets tend to agglomerate when second phase is added.

4.2 Optimization of processing parameters

The various processing parameters involved in the method were optimized including quantity of drug: polymer concentration, stirring speed.

It was observed that initially there was an increase in particle size and drug entrapment with the increasing in drug: polymer ratio. The size of microspheres increased from 199.3 µm to 256 µm and percentage drug entrapment increased from 59.30 % wt/wt to 84.38 % wt/wt on increasing drug: polymer ratio from F1 to F5 w/w. With further increase in polymer concentration the particles became irregular in shape which leads to decrease in drug entrapment. These result indicated the optimal drug: polymer ratio (F5 w/w) for required microparticles formulation (Fig.3.1,3.2).

Stirring speed was optimized in order to achieve stable formulation with average particle size, maximum drug entrapment and good floating ability. Emulsification was carried out under stress created by stirring to reduce the size of emulsion droplets. A stable microspheres formulation was achieved at 1000rpm with average particle size, maximum drug entrapment and maximum floating ability of 256µm, 84.38 % wt/wt and 96.00 % respectively (Fig3.2). A further increase in the stirring speed resulted in decreased particle size, drug entrapment and floating ability. The dispersion of drug: polymer into the droplets in oil phase depends upon the agitation speed of the system. As agitation speed was increased, beyond 1000 rpm, it resulted in high turbulence, caused frothing and adhesion of microspheres to the container wall. Thus, 1000rpm was selected as the optimized speed of stirring.

The amount of droplet stabilizer was optimized in order to obtain regular spherical particle with maximum percent drug entrapment and floating ability. The quantity of the droplet stabilizer was varied at 5, 10 and 15 % wt/wt. The mean particle size of floating microspheres was found to be increase with decreasing

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the amount of magnesium stearate. As the droplet stabilizer amount increased, the shape of microparticle became irregular. The regular shaped floating microspheres are found at drug polymer concentration 100 mg drug, 400 mg polymer at 1000 rpm speed of F5 (formulation) with 256 µm diameter, floating ability 96% floating duration 12 hrs, yield 90 % wt/wt, drug entrapment 84.88% wt/wt and drug loading 84.11% wt /wt. So that formulation (F5) has been selected optimized formulation.

5.0 Conclusion

Controlled release dosage forms where the drug is dispersed through polymer deliver the drug in the gastrointestinal tract with a low rate, leading to a more constant plasma level.

Sitagliptin phosphate was identified and characterized as per the requirements of I.P 1996 to confirm the authenticity of the drug sample. The λmax was obtained at 267nm, which is concordant with official values. The drug was also identified by IR spectroscopy. The obtained IR spectra matched with the spectra given in I.P confirms the sample to be authentic.

The mean diameters of floating microspheres prepared using various agitation speeds (i.e. 800, 1000 and 1500rpm) were 240.50, 256, and 208.30 respectively. It was found that particle size decreased with increase in speed of agitation. It was found that at 1000rpm stable microspheres formulation was achieved at 1000rpm with average particle size, maximum drug entrapment and maximum floating ability of 256 µm, 84.38 % wt/wt and 96.47 % respectively.

In case of floating microspheres of sitagliptin phosphate, the reduction in glucose levels was slower; it reached maximum reduction 8 hours after oral administration, and reductions in glucose levels were sustained over longer periods of time.

The sustain hypoglycemic effect observed over longer periods of time in the case of floating microspheres is due to the controlled release and absorption of sitagliptin phosphate over longer period of time. But in case of standard sitagliptin phosphate reduction of blood glucose level in animals was less.

The body weight of animals began decrease significantly upon induction of diabetes and regained their body weight during the course of treatment with floating microspheres of sitagliptin phosphate more in compare to standard sitagliptin phosphate. However, in the diabetic control group the body weight remained significantly low.

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References

1.‘ British Pharmacopoeia’, Controller of Her Majesty’s Stationary office, Published by Council of Europe, London Vol I, 2001, :1076.

2. Sweetman C.S. Martindale, The Complete Drug Reference, Published by Pharmaceutical Press, London. Part I, 33rd ed., 2002, :332.

3. Hardman J. G, Limbird L. E and Goodman Gilman A, Goodman & Gillman’s The Pharmacological Basis of Therapeutics, McGraw-Hill Publication 10th ed. 2001, :1705-1706.

4. Moffat C Anthony, Osselton David M and Widdop Brian. Clarke’s Analysis of Drugs and Poisons, Published by Pharmaceutical Press, UK. Vol II, 3rd ed., 2004, :1229.

5. Kumar Vinay, Abbas K. Abdul, Fausto Nelson and Mitchell N. Richard. Robbins Basic Pathology, Published by Elsevier India private Limited, New Delhi. 8th ed., 2007, :775-787.

6. Mohan Harsh. Textbook of Pathology. Published by Jaypee Brothers Medical Publisher Limited New Delhi 4th ed., 2000, :802-808.

7. Tripathi K. D. Essential of Medical Pharmacology. Published by Jaypee Brothers Medical Publishers Limited 5th ed., 2003, :248-251.

8. Benita S. Microencapsulation: methods and industrial application. Vol.73, Merecel Dekker, Inc: 1996.

9. Srivastava A.K, Ridurkar D.N, Floating microspheres of cimetidine: formulation, characterization and in vitro evaluation, ACTA Pharm. 2005; 55(3) :277-285.

10. Chowdary K.P.R, Koteshwara Rao, Ethyl cellulose microspheres of gliplizide: charaterization, in vitro

Journal of Chemical Health Risks

www.jchr.org

JCHR (2025) 15(1), 100-112 | ISSN:2251-6727

and in vivo evaluation, Indian J Pharm Sci. 2004, 66(4): 412-416.

11. Shahoo S.K, Formulation and in vitro evaluation of eudragit microspheres of stavudine ,J.Pharm. Res. 2005, 4(1): 369-375.

12. Costa P. Lobo J.M.S, Modeling and comparison of dissolution profiles, Eur J Pharm Sci. 2001; 13: 123-133.

13. S.Haznedar. B. Dortunc, Preparation and in vitro evaluation of eudragit microspheres containg acetazolamide, Int J Pharm. 2004; 131-140.

14. Baby D., Design and release kinetic pattern evaluation of indomethacin microspheres intended for oral administration, Biomaterial.9, 482-488.

15. Hamid A.M, Harris M.S, Once daily tablet formulation and in vitro release evaluation of cefpodoxime using hydroxypropyl methylcellulose, AAPS PharmSciTech. 2006, 7(3) :1-6.

16. Higuchi T. Mechanism of sustain action medication. Theoretical analysis of rate of release of solid drugs dispersed in solid matrices, J Pharm Sci. 1963; 52: 1145-1149.

17. Korsmeyer R.W, Peppas NA. Mechanism of solute release from porous polymer matrices, Int J Pharm ; 1983; 15:25-35.

18. Jain S.K, Awasthi A.M, Jain N.K, Agrawal G.P. Calcium silicate based microspheres of repaglinide for gastroretentive floating drug delivery: preparation and in vitro characterization, J. of Cont. Rel. 2005, 8:8.

19. Rouge N, Allemann E, Gex-Fabry M, Balant L, Cole E.T, Buri P, Comparati pharmacokinetics study of a floating multiple-unit capsule, a high-density multiple-unit capsule and an immediate-release tablet containing 25 mg atenolol, Pharm Acta Helv. 1998, 73(2): 81-87.

20. M.V. Shah, M.D. De Gennaro, H. Suryakarma, An evaluation of albumin microcapsules prepared using a multiple emulsion technique, J of Microencapsul. 4 ,1987, 223–238.

21. Singh, B.N. and Kim, K.H, J. of Cont. Rel.2000, 63 (1-2): 235-259.

22. Jain, N.K., "Controlled Novel Drug delivery", I[st] Eds., CBS publishers and distributors, New Delhi, 2002,:236-55.

23. Chien Y.W., " Controlled and Modulated Release Drug Delivery Systems", In Encyclopedia of Pharmaceutical Technology, J. Swarbrick, J.C. Boylan, New York: Dekker; 1990,:280-285.

24. Akiyama Y, Nagahara N: Novel Formulation Approaches to Oral Muco-adhesive Drug Delivery Systems. In Bioadhesive Drug Delivery Systems – Fundamentals, Novel Approaches and Development. Edited by Mathiowitz E, Chickering III DE, Lehr CM.New York: Dekker; 1999,:477-505.

25. Khosla R, Davis S.S, Effect of polycarbophil on the gastric emptying of pellets, J Pharm Pharmacol. 1987, 39:47-49.

26. Sa¨ kkinen M, Marvola J, Kanerva H, Lindevall K, Lipponen M, Kekki T, Ahonen A, Marvola M: Gamma scintigraphic evaluation of the fate of microcrystalline chitosan granules in human stomach. Eur J Pharm Biopharm 2004, 57:133-143.

27. Klausner E.A, Lavy E, Stepensky D, Friedman M, Hoffman A: Novel gastroretentive dosage forms: evaluation of gastroretentivity and its effect on riboflavin absorption in dogs. Pharm Res 2002, 19:1516-1523.

28. Lee J.H, Park T.G, Choi H.K: Development of oral drug delivery system using floating microspheres, J of Microencapsul. 1999, 16:715-729.

29. Sato Y, Kawashima Y, Takeuchi H, Yamamoto H, Physicochemical properties to determine the buoyancy of hollow microspheres (microballoons) prepared by the emulsion solvent diffusion method, Eur J Pharm Biopharm. 2003, 55:297-304

30. Streubel A, Siepmann J, Bodmeier R: Multiple unit gastroretentive drug delivery systems: a new preparation method for low density microparticles,J of Microencapsul. 2003, 20:329-347.

31. Kro¨ gel I, Bodmeier R, Development of a multifunctional matrix drug delivery system surrounded by an impermeable cylinder, J. of Cont. Rel. 1999, 61:43-50.

Journal of Chemical Health Risks

www.jchr.org

JCHR (2025) 15(1), 100-112 | ISSN:2251-6727

32. Sriamornsak P, Thirawong N, Puttipipatkhachorn S: Emulsion gel beads of calciu pectinate capable of floating on the gastric fluid: effect of some additives, hardening agent or coating on release behavior of metronidazole, Eur J Pharm Sci. 2005, 24:363-373.

33. V. Alvisi, A. Gasparetto, A. Dentale, H. Heras, A. Felletti-Spadazzi, A. D'Ambrosi, Bioavailability of a controlledrelease formulation of ursodeoxycholic acid in man, Drugs Exp. Clin. Res. 1996, 22: 29–33.

34. J.T. Fell, Targeting of drugs and delivery systems to specific sites in the gastrointestinal tract, J. Anat. 1996, 189 :517–519.

35. C.G. Wilson, N. Washington, The stomach: its role in oral drug delivery, in: M.H. Rubinstein (Ed.), Physiological Pharmaceutics: Biological Barriers to Drug Absorption, Ellis Horwood, Chichester, 1989, :47–70.

36. P.R. Sheth, J. Tossounian, The hydrodynamically balanced system (HBSE): a novel drug delivery system for oral use, Drug Dev. Ind. Pharm. 10, 1984, :313–339.

37. A.K. Hilton, P.B. Deasy, In vitro and in vivo evaluation of an oral sustained-release floating dosage form of amoxicillin trihydrate, Int. J. Pharm. 86, 1992, :79–88.

38. C. Bogentoft, Oral controlled-release dosage forms in perspective, Pharm. Int. 3 1982, :366–369.

39. P.R. Sheth, J.L. Tossounian, Sustained release pharmaceutical capsules, US Patent 4, November 21, 1978, 126, 672.

40. M.R. Franz, M.P. Oth, Sustained release, bilayer buoyant dosage form, US Patent 5, August 3, 1993,232, 704.

41. S. Desai, S. Bolton, A floating controlled-release drug delivery systems: in vitro–in vivo evaluation, Pharm. Res. 10,1993, 1321–1325.

42. S. Bolton, S. Desai, Floating sustained release therapeutic compositions, US Patent 4, 814, 179, March 21, 1989.

43. A. Dennis, P. Timmins, K. Lee, Buoyant controlled release powder formulation, US Patent 5, December 8, 1992,169, 638.