dx.doi.org/10.14227/DT180311P72

Gastroretentive Orlistat Microspheres: Formulation, Characterization, and In Vitro Evaluation

email: sbsateesh@gmail.com

S. B. Sateesha[1,*], B. Prakash Rao[1], A. J. Rajamma[2], and L. V. G. Nargund[3]

[1]*Department of Pharmaceutics, Acharya and BM Reddy College of Pharmacy, Bangalore-560090, India*

[2]*Department of Pharmacognosy, KLE University College of Pharmacy, Bangalore-560010, India*

[3]*Department of Pharmachemistry, Nargund College of Pharmacy, Bangalore-560085, India*

ABSTRACT

The present study involves the preparation and evaluation of floating microspheres of orlistat for improving drug bioavailability by prolongation of gastric residence time. The microspheres were prepared by a solvent diffusion–evaporation technique using inert polymers (Eudragit RL100, cellulose acetate, and ethyl cellulose). The effect of three formulation variables (i.e., drug/polymer ratio [D/P], polymer amount, and stirring speed) on floatability, encapsulation efficiency, percentage fines, and release mechanism were studied. The results of Fourier transform infrared spectroscopy show no interaction between the drug and the polymers. In vitro dissolution studies were performed in 0.1 N HCl (pH 1.2) for 12 h, and samples were analyzed by HPLC using a UV–vis detector at 205 nm. The grading curve was constructed for the selected formulations, and D_{30}, D_{60}, and D_{90} values were determined to characterize the size distribution. A comparison of r^2 values for Higuchi, Korsmeyer–Peppas, and zero-order kinetic models for different batches of microspheres shows Fickian and non-Fickian diffusion kinetics. The orlistat microspheres prepared with cellulose acetate (D/P 1:2) at the stirring speed of 900 rpm show maximum floatability and optimum encapsulation efficiency, and exhibited a prolonged release for almost 12 h with a Fickian diffusion release mechanism.

INTRODUCTION

Dyslipidemia is a disorder of lipoprotein metabolism. Dyslipidemia is the elevation of plasma cholesterol, triglycerides, or both. It can also be manifested by the elevation of low-density lipoprotein (LDL) cholesterol and the decrease of high-density lipoprotein (HDL) cholesterol in the blood (*1*).

Dyslipidemia is a primary risk factor *that contributes to the development of atherosclerosis* in the general population and in diabetic patients. Most people with high serum cholesterol also have elevated LDL because much of the serum cholesterol is transported in LDL. The concept therefore has emerged that LDL is the predominant atherogenic lipoprotein. The remarkable finding that LDL-lowering therapy reduces the risk for subsequent coronary events even in patients with advanced atherosclerotic disease discloses a role for LDL in late stages of atherogenesis (*2*).

Orlistat, [(1S)-1-[(2S, 3S)-3-hexyl-4-oxo-oxetan -2-y1] methyl] dodecyl] (2S)-2-formamido-4-methyl–pentonate (Figure 1), also known as tetrahydrolipstatin, is designed to treat obesity. It reduces the LDL concentration in the blood by inhibiting gastric and pancreatic lipases (the enzymes that break down triglycerides in the intestine). The primary effect of orlistat is local lipase inhibition within the GI tract after an oral dose. When lipase activity is blocked, triglycerides from the diet are not hydrolyzed into absorbable free fatty acids and are excreted undigested instead, thereby reducing caloric intake (*3*, *4*). A single dose of orlistat will prevent approximately 30% of dietary fat from being absorbed, which indicates its effectiveness in controlling dyslipidemia. It also exhibits antiproliferative and antitumor properties in prostate and breast tissues (*5*).

In a study conducted in an obese population over four years, the incidence of type-2 diabetes was reduced with orlistat (6.2%) when compared with placebo (*6*, *7*). Hence, orlistat is an important drug in prophylactic management of obesity and for the management of type-2 diabetes. Orlistat has a short half-life (<2 h) and requires administration multiple times a day. The absorption window is restricted to the upper part of the gastrointestinal tract, which may lead to variability and nonuniform absorption, and makes the bioavailability unpredictable (*8*). The shorter residence time of the conventional dosage form in the stomach and the variable gastrointestinal transit time may affect the efficacy of this drug. Hence, a beneficial delivery system would be one that can control and prolong the gastric emptying time and deliver drugs in higher concentration to the absorption site. One such approach is a multiparticulate spherical dosage form having a density less than that of gastric fluids (*9*). A formulation that can deliver the drug for a prolonged time would be ideal. Recently, Jain et al. (*10*) evaluated a floating delivery system of orlistat using calcium silicate. Such dosage forms are better because they reduce the

*Corresponding author.

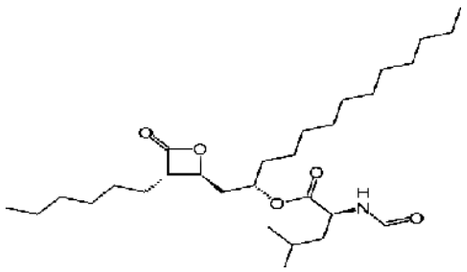

Figure 1. Chemical structure of orlistat.

inter-subject absorption variability and lower the probability of dose dumping.

In the present work, we have evaluated the suitability of different polymers (i.e., Eudragit RL100, cellulose acetate, and ethyl cellulose) for the development of floating microspheres of orlistat. Three independent formulation parameters (i.e., drug/polymer ratio (D/P), polymer type, and stirring speed) were studied. Formulations were optimized based on the encapsulation efficiency, percentage yield, particle size distribution, and physical appearance of the microspheres.

MATERIALS AND METHODS

Materials

Orlistat was generously supplied as a gift sample by Biocon, Bangalore, India. Eudragit RL100 was received as a gift sample from Zydus–Cadila Healthcare, Ltd., Mumbai, India. Ethyl cellulose, cellulose acetate, and polyvinyl alcohol (PVA) were purchased from Sigma-Aldrich Company, Mumbai, India. All other chemicals were of analytical reagent grade.

Methods

Compatibility Studies

Fourier transform infrared spectroscopy (FTIR) was used to quantify the interaction between the drug and carrier used in formulation. Spectra were recorded for pure drug and 1:1 physical mixtures of drug and polymer on a Shimadzu 8400S FTIR spectrophotometer (Kyoto, Japan). Samples were prepared as KBr discs at a pressure of 150–200 kg/cm^2 and scanned over 400–4000 cm^{-1} with a resolution of 4 cm^{-1}.

Preparation of HPLC Calibration Curve

The standard calibration curve of orlistat was developed using solutions of pure orlistat equivalent to 40, 80, 120, and 160 µg/mL in methanol, mobile phase (90:10 acetonitrile/phosphoric acid), and 0.1 N HCl (pH 1.2). A weighed quantity of orlistat (100 mg) was dissolved initially in 100 mL of methanol to give a 1000 µg/mL solution (SS-I) and further diluted with respective media. The absorbances were recorded by HPLC (Waters 2489, Milford, MA, USA) method (*11*) using 90:10 (v/v) acetonitrile/phosphoric acid as mobile phase delivered at 1 mL/min by a Waters 515 pump. Twenty microliters was injected onto an Agilent Eclipse XDB-C_{18} column (150 × 4.6 mm) at room temperature. The column eluent was monitored at 205 nm using a UV–vis detector.

Preparation of Floating Microspheres

Microspheres were prepared in 30-g batch sizes by the solvent diffusion–evaporation method (*12*, *13*). The drug and polymer were dissolved at room temperature in one of several solvent systems (Table 1), which was then poured into 200 mL of a 0.1 M acidic solution of PVA and maintained at 25–30 °C. The solution was subsequently stirred at different speeds (Table 1) for 2 h to allow

Table 1. Orlistat Microsphere Formulations

Formulation Code	Polymer	Drug/polymer ratio (D/P)	Stirring Speed (rpm)	Organic Solvent System (1:1)	PVA concentration (M)
Fca1	cellulose acetate	1:1	600	ethyl acetate/acetone	0.1
Fca2	cellulose acetate	1:2	600	ethyl acetate/acetone	0.1
Fca3	cellulose acetate	1:3	600	ethyl acetate/acetone	0.1
Fca4	cellulose acetate	1:2	900	ethyl acetate/acetone	0.1
Fca5	cellulose acetate	1:2	1200	ethyl acetate/acetone	0.1
Feu1	Eudragit RL100	1:2	600	acetone/ethanol	0.1
Feu2	Eudragit RL100	1:2	900	acetone/ethanol	0.1
Feu3	Eudragit RL100	1:2	1200	acetone/ethanol	0.1
Fec1	ethyl cellulose	1:2	600	ethyl acetate/ethanol	0.1
Fec2	ethyl cellulose	1:2	900	ethyl acetate/ethanol	0.1
Fec3	ethyl cellulose	1:2	1200	ethyl acetate/ethanol	0.1

complete evaporation of the volatile solvent. The formed microspheres were collected by filtration using a nylon cloth and washed repeatedly with distilled water. They were dried in vacuum and subsequently stored in an amber container over fused calcium chloride. All experiments were performed under subdued light conditions to prevent photodegradation of drug.

Percentage Yield and Encapsulation Efficiency of Microspheres

The percentage yield of the formulation was calculated using the following equation.

% yield = (practical yield / theoretical yield) × 100

The quantitative determination of drug in microspheres was performed using the HPLC method described earlier. Microspheres were crushed thoroughly by trituration and suspended in a minimal amount of methanol to dissolve the drug. The solutions were sonicated and filtered using 47-mm, 0.45-µm pore size membrane filters (Millipore). Clear filtrate was suitably diluted with a filtered and degassed mixture of mobile phase (90:10 acetonitrile/phosphoric acid). The percentage encapsulation efficiency was determined to evaluate the effect of three independent factors.

Encapsulation Efficiency (%) =

$$\frac{\text{amount of encapsulated drug}}{\text{amount of added drug}} \times 100$$

Particle Size Analysis

The *USP* procedure (Method I) for particle size analysis was followed (*14*). A set of seven standard sieves in the range of 0.15–1.18 mm and 25-g samples of dried microspheres were used. The percent retained was plotted versus sieve opening size to determine formulation homogeneity. The size distribution was also evaluated with the help of a grading curve (i.e., log sieve size vs percent fines) (*15*, *16*). From the grading curve, D_{30}, D_{60}, and D_{90} values, corresponding to 30%, 60%, and 90% fines, respectively, were determined.

Flow Properties

Powder properties of all preparations were measured using a tap density tester (Electrolab ETD-1020) at increments of 250, 500, and 750 taps with 250 drops/min. Bulk density, tapped density, Carr compressibility index, and Hausner ratio were determined. The angle of repose was also measured to provide a measure of the flow properties and compressibility of the microspheres.

Floatability Study

An in vitro floatability study (*17*) was conducted by placing 1 g of microspheres over the surface of 900 mL of a 0.1 N HCl (pH 1.2) solution containing 0.02% Tween 80 as a dispersing medium in a USP dissolution Apparatus 2 (paddle) (TDT-08T, Electrolab). The medium was agitated with a paddle rotating at a speed of 75 rpm for 12 h. The floated microspheres were collected, dried, and weighed at intervals of 2 h, and the floatability percentage was calculated using the following formula (*18*)

Floatability (%) = $(W_f / W_f + W_s) \times 100$

where W_f and W_s are the weights of the floating and settled microparticles, respectively. All determinations were made in triplicate.

In Vitro Release Study

The formulations Fca2, Fca4, Fca5, Feu2, and Fec2 were selected for release-rate studies based on the optimization. The studies were conducted using amber colored jars by USP Apparatus 2 method for 12 h. An accurate weight of 100 mg of pure drug or microspheres equivalent to 100 mg of drug was placed in 900 mL of a 0.1 N HCl (pH 1.2) solution containing 0.02% Tween 80 (to maintain perfect sink condition) maintained at 37 ± 0.5°C and stirred at 75 rpm (*19*). A 10-mL sample was withdrawn from the dissolution medium at 1-h intervals up to 12 h. The dissolution medium was replenished at each sampling with an equal volume of prewarmed fresh dissolution medium. Samples were stored at 2–8 °C until analysis. The amount of orlistat released was analyzed by HPLC using a UV–vis detector at 205 nm. All experiments were performed in triplicate.

Release Kinetics

Data obtained from in vitro dissolution studies were evaluated using different mathematical models to describe the kinetics of the drug release from microspheres. The kinetics of orlistat release was evaluated using Higuchi, Korsmeyer–Peppas, and zero-order models to check the phenomena controlling drug release from the microspheres. The goodness of fit was evaluated using the correlation coefficient values (r^2).

The Higuchi model was developed based on Fick's law, and it describes the fraction of drug released from a matrix as proportional to the square root of time (*20*, *21*)

$$Q_t = K_H \sqrt{t}$$

where K_H is the Higuchi rate constant and Q_t is the amount of drug released at time *t*. If a plot of square root of time versus cumulative amount of drug released yields a straight line with a slope that is greater than or equal to one, then the particular dosage form is considered to follow Higuchi kinetics of drug release.

Under some experimental situations, when the release mechanism deviates from Fick's equation and follows an anomalous behavior (non-Fickian release), a more generic equation that can be used is the Korsmeyer–Peppas model. It describes drug release from the polymeric system in which release deviates from Fickian diffusion (*22*), as expressed in the equation

$$M_t/M_\infty = K t^n$$

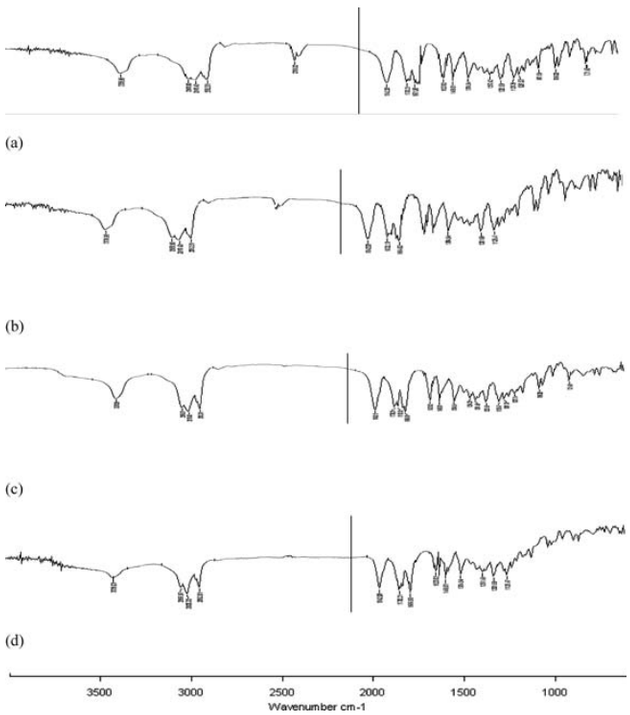

Figure 2. FTIR spectra of (a) orlistat; (b) physical mixture of orlistat and ethyl cellulose; (c) physical mixture of orlistat and cellulose acetate; and (d) physical mixture of orlistat and Eudragit RL100.

where M_t/M_∞ corresponds to the amount of drug released at time *t* and after an infinite time, *K* is a constant comprising the structural and geometric characteristics of the microsphere, and the release exponent *n* is a parameter that depends on the release mechanism. Peppas used this *n* value to characterize different release mechanisms. If *n* is 0.5 or less, the release mechanism follows Fickian diffusion, and higher values ($0.5 < n < 1$) for mass transfer follow a non-Fickian model (anomalous transport). The drug release follows zero-order and case-II transport if *n* equals 1. When *n* is greater than 1, the mechanism of drug release is regarded as super case-II transport. This model is used to analyze the release of pharmaceutical polymeric dosage forms when the release mechanism is not well known or when more than one type of release phenomenon was involved. The *n* value could be obtained from the slope of a plot of log M_t/M_∞ versus log time.

Zero-order kinetics describes the system in which drug release rate is independent of concentration (*23*)

$$Q_t = Q_o + K_o t$$

where Q_t corresponds to amount of drug dissolved at time *t*, Q_o is the initial amount of drug in the solution, which is often zero, and K_o is the zero-order release rate constant.

Stability Studies

The optimized formulations were placed in screw-cap, amber glass containers and stored at ambient humidity and different temperatures such as 25 ± 2°C, 30 ± 2°C and 40 ± 2°C for a period of 3 months. The samples were analyzed for physical appearance and for drug content at regular intervals of 30 days.

RESULTS AND DISCUSSION

Compatibility Studies

Drug–excipient interactions play a vital role with respect to biological performance and formulation stability. FTIR spectroscopy was used to study the physical and chemical interactions between drug and excipients. The characteristic absorption peaks obtained for drug alone and in the presence of polymers (1:1) are depicted in Figure 2. From the spectra, it is clear that the main drug peaks and the frequencies of peaks observed were within the standard range (Table 2). This indicates that the drug was compatible with the formulation components.

Standard Calibration Curve of Orlistat

The regression coefficient (r^2) values found for orlistat calibration curve developed in methanol, mobile phase, and 0.1 N HCl solutions are 0.9936, 0.9991, and 0.9991, respectively. The standard calibration curves are linear over the concentration range of 40–160 µg/mL and follow Beer's law with high r^2 values in all media. The standard curve developed in mobile phase was used to estimate drug formulation concentrations.

Effect of Variables on Microsphere Characteristics

It is difficult to assess the effect of variables individually or in combination. However, the effects of three variables,

Table 2. Frequency of Peaks Observed in FTIR Spectra of Orlistat Pure Drug and Physical Mixtures with Polymers

Functional Group	Standard Range (cm^{-1})	Orlistat (cm^{-1})	Orlistat and cellulose acetate (cm^{-1})	Orlistat and ethyl cellulose (cm^{-1})	Orlistat and Eudragit RL100 (cm^{-1})
C=O stretching	1700–1725	1708	1710.7	1709.6	1710.5
C–H stretching in CH_2	2850–2960	2920	2920	2923.1	2921.5
N–H stretching	3500–3300	3336.6	3338.6	3337.8	3337
C–H deforming	875–895	877.7	877.5	878.2	877
C=C aromatic stretching	1450–1600	1521.7	1458.1	1462	1466

Table 3. Particle Size Distribution of Selected Formulations

	Particle Size Distribution (mm)				
Parameter	**Fca2**	**Fca4**	**Fca5**	**Feu2**	**Fec2**
D_{30}	0.38	0.26	0.16	0.16	0.15
D_{60}	0.74	0.45	0.25	0.49	0.20
D_{90}	1.10	0.80	0.74	1.10	0.58

namely (1) drug–polymer ratio (D/P), (2) polymer type, and (3) stirring speed on the orlistat encapsulation efficiency, production yield, and particle size were studied. Micro- spheres were also characterized for powder properties, floatability, and release kinetics.

Powder Properties

Microspheres were prepared by gradually increasing polymer concentration in combination with varying stirring speed to assess the effect of these variables on production yield and encapsulation efficiency. The largest yield was observed for Feu3 (95.64 ± 1.3%) and the least was for Fec1 (62.32 ± 2.1). The formulations prepared with Eudragit RL100 (Feu1, Feu2, Feu3) gave a better yield (>82%) than those prepared with other polymers. The production yield of microspheres prepared from cellulose acetate was satisfactory (>80% except at lower concentrations of cellulose acetate) from a formulation perspective, but the yield obtained with ethyl cellulose was relatively poor (<73%). The encapsulation efficiency of drug increased with increasing polymer concentration. The reason may be that at a higher polymer solution viscosity

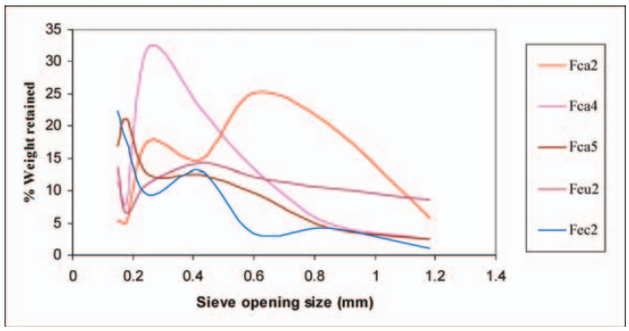

Figure 3. Particle size distributions of selected orlistat floating-microsphere formulations.

(at the highest polymer concentration), the diffusion of the drug into the external phase is expected to decrease, which would result in higher encapsulation efficiency.

Lower polymer concentration with increasing stirring speed results in smooth and smaller particles. However, the encapsulation efficiency was low. A combination of lower stirring speed and higher polymer concentration results in large irregular particles. The percent fines significantly increased with increasing stirring speed, which agrees with the values calculated for D_{30}, D_{60}, and D_{90} (Table 3) for different polymers (1:2) at varying stirring speeds. Hence, the stirring speed of 900 rpm was chosen as the optimum speed, and all formulation blends were prepared at the polymer concentration of 1:2.

The floating ability of the microspheres also increases at higher concentrations of polymer, as shown in Table 4. All formulations floated over the surface of the dissolution medium for over 10 h without any apparent gelation.

Table 4. Characterization of Orlistat Floating Microspheres

Formulation Code	Yield (%)	Angle of Repose (θ)	Carr Index (%)	Hausner Ratio	Floatability after 10 h (%)	Encapsulation Efficiency (%)
Fca1	72.85 ± 4.5	26.1 ± 0.57	06.21 ± 0.011	1.065 ± 0.009	65 ± 3	43.77 ± 2.62
Fca2	86.63 ± 3.0	25.5 ± 0.75	09.09 ± 0.01	1.099 ± 0.081	72 ± 2	55.26 ± 1.81
Fca3	80.39 ± 4.1	23.6 ± 0.97	06.15 ± 0.014	1.065 ± 0.009	84 ± 2	57.48 ± 2.94
Fca4	93.41 ± 1.2	24.5 ± 0.47	11.77 ± 0.012	1.119 ± 0.017	76 ± 2	59.95 ± 1.22
Fca5	88.37 ± 2.1	23.6 ± 0.91	06.16 ± 0.009	1.076 ± 0.012	74 ± 3	51.41 ± 2.65
Feu1	89.82 ± 3.1	26.6 ± 0.97	08.96 ± 0.051	1.088 ± 0.008	73 ± 3	32.48 ± 1.18
Feu2	92.86 ± 2.3	24.8 ± 0.41	07.58 ± 0.006	1.076 ± 0.004	76 ± 2	50.14 ± 3.02
Feu3	95.64 ± 1.3	27.3 ± 0.65	11.07 ± 0.009	1.133 ± 0.108	86 ± 2	58.52 ± 1.87
Fec1	62.32 ± 2.1	26.3 ± 0.55	11.15 ± 0.009	1.138 ± 0.101	63 ± 4	36.08 ± 2.07
Fec2	69.92 ± 1.9	25.5 ± 0.75	08.95 ± 0.008	1.068 ± 0.011	68 ± 2	42.58 ± 2.07
Fec3	73.52 ± 2.0	23.2 ± 0.77	06.16 ± 0.009	1.075 ± 0.007	73 ± 2	49.47 ± 1.88

Results are the mean of 3 observations ± SD.

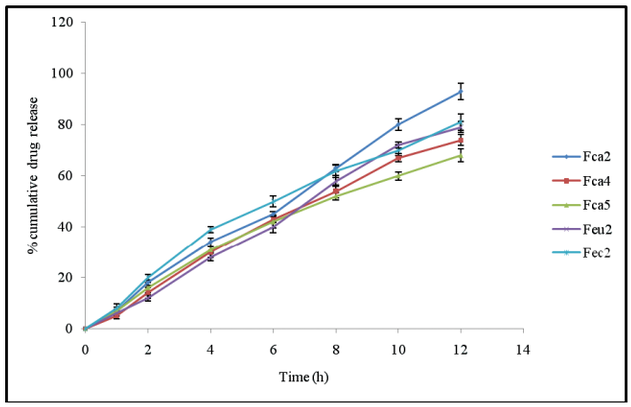

Figure 4. Release profiles of optimized orlistat floating-microsphere formulations.

The type of polymer used has a significant effect on the micromeritic properties of the formulation. Bulk flow, formulation homogeneity, surface-area-controlled processes such as dissolution, and chemical reactivity are directly affected by the size, shape, and surface morphology of the microparticles. Microspheres formulated with cellulose acetate were smaller and had a smoother surface than microspheres of Eudragit RL100 and ethyl cellulose. The homogeneity of microspheres and the percentage fines obtained with cellulose acetate-coated microspheres were satisfactory compared with others, as depicted in Figure 3. Microspheres were spherical and discrete. However, the particle size range of microspheres varied and increased with an increase in polymer concentration.

Batch flow properties were evaluated by measuring the angle of repose, the Hausner ratio, and the compressibility index; results are tabulated in Table 4. The angle of repose, the Hausner ratio, and the compressibility index are indicative of the flowability of microspheres (*24*). The better flow properties of the microspheres indicate that the microspheres produced were nonaggregated. The improved micromeritic properties of the formulated microspheres, when compared with that of the pure drug, suggest that they can be easily handled and filled into a capsule.

Kinetics of In Vitro Dissolution Studies

The release profile and kinetics of drug release are important because they correlate the in vitro and in vivo drug responses by comparing the results of pharmacokinetics and dissolution profile patterns. The in vitro release profiles of optimized orlistat microsphere formulations (Fca2, Fca4, Fca5, Feu2, and Fec2) in 0.1 N HCl (pH 1.2) for 12 h are shown in Figure 4. The cumulative release of orlistat significantly decreased with increasing polymer concentration. Smaller microspheres (formed at a lower polymer concentration with a higher stirring rate) gave rise to faster drug release due to larger surface area exposed to the dissolution medium. These results are further evidence that the optimum drug–polymer concentration should be 1:2 for better release performance. For further confirmation, results were fitted into various mathematical equations, such as Higuchi, Korsmeyer–Peppas, and zero-order release models.

The in vitro drug release shows the highest regression coefficient values for the Higuchi model, indicating diffusion to be the predominant mechanism of drug release. The kinetic values obtained for all five formulations (Fca2, Fca4, Fca5, Feu2, and Fec2) are shown in Table 5. Among the five formulations that were introduced for the drug release study in 0.1 N HCl (pH 1.2), the release behavior of formulation Fca4 was linear and satisfactory. The regression coefficients (r^2) values of formulation Fca4 for zero-order, Higuchi, and Korsmeyer–Peppas plots are 0.9938, 0.9861, and 0.9891, respectively. The low n value of 0.36 indicates that the release approximates a Fickian diffusion mechanism. Orlistat microspheres prepared with cellulose acetate shows a desirable high-drug content, good flow properties, buoyancy, and adequate release characteristics; hence, formulations prepared by such polymers are suitable for the development of gastric retention dosage forms.

Stability Studies

The optimized formulation (Fca4), when subjected to stability studies at 25 ± 2 °C, 30 ± 2 °C, and 40 ± 2 °C,

Table 5. Values of r², k, and n for Selected Formulations

	Zero-order		Higuchi		Korsmeyer–Peppas		
Formulation	r^2	k	r^2	k	r^2	n	**Mechanism of Release**
Fca2	0.9833	0.829	0.9853	0.743	0.9898	0.44	Fickian diffusion
Fca4	0.9938	0.91	0.9861	0.794	0.9891	0.36	Fickian diffusion
Fca5	0.9973	0.856	0.9784	0.772	0.9973	0.72	Non-Fickian release
Feu2	0.9908	0.704	0.9815	0.967	0.991	0.91	Non-Fickian release
Fec2	0.9736	0.758	0.998	0.493	0.9504	0.49	Diffusion

Table 6. Stability Studies of Formulation Fca4 at Various Storage Temperatures and Ambient Humidity

Sampling interval (days)	Storage Condition		
	25 ± 2 °C	30 ± 2 °C	40 ± 2 °C
	Drug Content	Drug Content	Drug Content
01	51.55 ± 1.02	51.55 ± 1.02	51.55 ± 1.02
30	50.93 ± 0.22	48.75 ± 2.02	49.64 ± 2.22
60	50.98 ± 1.42	50.05 ± 1.02	48.05 ± 2.02
90	49.54 ± 2.22	50.75 ± 2.22	48.75 ± 2.42

Results are the mean ± SD (n = 3)

showed no significant changes in the physical and chemical properties, which confirms that the formulation (Fca4) was stable at the end of 90 days (Table 6).

CONCLUSION

The technique of preparing orlistat microspheres with cellulose acetate (1:2) by solvent evaporation–diffusion is a good and simple method to encapsulate the drug successfully. The influence of formulation variables and type of polymer on encapsulation efficiency, particle size, floatability, and extent of drug release is evident. These findings indicate that these variables can be suitably altered to achieve the desired controlled-release profile. Formulation Fca4 was stable and successfully controlled the release of orlistat in the stomach (pH 1.2) with diffusion kinetics.

ACKNOWLEDGMENTS

We are very thankful to Biocon, Bangalore, India and Zydus–Cadila Healthcare Ltd., Mumbai, India, for providing drug and polymer samples and Nargund College of Pharmacy and KLES's College of Pharmacy, Bangalore, India, for providing research facilities. We also wish to thank Prof. CRM Setty for editing this manuscript.

REFERENCES

1. Gotto, A. M. Evolving Concepts of dyslipidemia, atherosclerosis and cardiovascular disease: the Louis F. Bishop Lecture. *J. Am. Coll. Cardiol*. **2005**, *46* (7), 1219–1224.
2. Ballinger, A.; Peikin, S. R. Orlistat: its current status as an anti-obesity drug. *Eur. J. Pharmacol*. **2002**, *440* (2–3), 109–117.
3. Tsutsumi, K. Lipoprotein Lipase and Atherosclerosis. *Curr. Vasc. Pharmacol*. **2003**, *1*, 11–17.
4. Al-Suwailem, K.; Al-Tamimi, S.; Al-Omar, M. A.; Al-Suhibani, M. S. Safety and mechanism of action of orlistat (tetrahydrolipstatin) as the first local antiobesity drug. *J. Appl. Sci. Res*. **2006**, *2* (4), 205–208.
5. Bougoulia, M.; Triantos, A.; Koliakos, G. Effect of weight loss with or without orlistat treatment on adipocytokines, inflammation, and oxidative markers in obese women. *Horm.* **2006**, *5* (4), 259–269.
6. Drew, B. S.; Dixon, A. F.; Dixon, J. B. Obesity management: Update on orlistat. *Vasc. Health Risk Manag*. **2007**, *3* (6), 817–821.
7. Anderson, J. W.; Kendall, C. W.; Jenkins, D. J. Importance of weight management in type 2 diabetes: review with meta-analysis of clinical studies. *J. Am. Coll. Nutr*. **2003**, *22* (5), 331–339.
8. Kazmi, S. A.; Khan, M.; Mashori, G. R.; Saleem, A.; Akhtar, N.; Jahangeer, A. Influence of sibutramine, orlistat and *Ispaghula* in reducing body weight and total Body fat content in obese individuals. *J. Ayub Med. Coll. Abbottabad* **2009**, *21* (2), 45–48.
9. Nath, B.; Nath, L. K.; Mazumdar, B.; Sharma, N. K.; Sarkar, M. K. Preparation and In Vitro Evaluation of Gastric Floating Microcapsules of Metformin HCl. *Ind. J. Pharm. Educ. Res*. **2009**, *43* (2), 177–186.
10. Jain, S. K.; Agrawal, G. P.; Jain, N. K. Evaluation of Porous Carrier-based Floating Orlistat Microspheres for Gastric Delivery. *AAPS PharmSciTech* **2006**, *7* (4), Article 90.
11. Souri, E.; Jalalizadeh, H.; Kebriaee-Zadeh, A.; Zadehvakili, B. HPLC Analysis of orlistat and its application to drug quality control studies. *Chem. Pharm. Bull*. **2007**, *55* (2), 251–254.
12. Gattani, Y. S.; Kawtikwar, P. S.; Sakarkar, D. M. Formulation and evaluation of Gastro retentive Multiparticulate Drug delivery system of Aceclofenac. *Int. J. ChemTech Res*. **2009**, *1* (1), 1–10.
13. Arora, S.; Ali, J.; Ahuja, A.; Khar, R. K.; Baboota, S. Floating Drug Delivery Systems: A Review. *AAPS PharmSciTech* **2005**, *6* (3), Article 47.
14. <786> Particle Size Distribution Estimation by Analytical Sieving. In *United States Pharmacopeia and National Formulary USP 31–NF 26*, Asian Edition; Tata Press Limited: Bombay, India, 2008; pp 308–311.
Li, S.; Lin, S.; Daggy, B. P.; Mirchandani, H. L.; Chien, T. W. Effect of formulation variables on the floating properties of gastric floating drug delivery system. *Drug Dev. Ind. Pharm.* **2002**, *28* (7), 783–793.
16. Singh, B. N.; Kim, K. H. Floating drug delivery systems: an approach to oral controlled drug delivery via gastric retention. *J. Controlled Release* **2000**, *63* (3), 235–59.
Timmermans, J.; Moës, A. J. Factors controlling the buoyancy and gastric retention capabilities of floating matrix capsules: new data for reconsidering the controversy. *J. Pharm. Sci*. **1994**, *83* (1), 18–24.
18. El-Kamal, A. H.; Sokar, M. S.; Gamal, S. A.; Naggar, V. F. Preparation and evaluation of ketoprofen floating oral drug delivery system. Int. J. Pharm. **2001**, *220*, 13–21.
Gohel, M. C.; Mehta, P. R.; Dave, R. K.; Bariya, N. H. A More Relevant Dissolution Method for Evaluation of a

Floating Drug Delivery System. *Dissolution Technol*. **2004**, *11* (4), 22–5.
20. Jiang, G.; Woo, B. H.; Kang, F.; Singh, J.; DeLuca, P. P. Assessment of protein release kinetics, stability and protein polymer interaction of lysozyme encapsulated poly(D,L-lactide-co-glycolide) microspheres. *J. Controlled Release* **2002**, *79* (1–3), 137–45.
21. Edlund., U.; Albertsson, A.-C. Degradable Polymer Microspheres for Controlled Drug Delivery. *Adv. Polym. Sci*. **2002**, *157*, 67–112.
22. Siepmann, J.; Göpferich, A. Mathematical modeling of bioerodible, polymeric drug delivery systems. *Adv. Drug Del. Rev*. **2001**, *48* (2–3), 229–247.
Thapa, P.; Ghimire, M.; Mullen, A. B.; Stevens, H. N. E. Controlled Release Oral Delivery System Containing Water Insoluble Drug. *Kathman. U. J. Sci. Eng. Technol*. **2005**, *1* (1), 1–10.
24. Shoaib, M. H.; Tazeen, J.; Merchant, H.; Yousuf, R. I. Evaluation of drug release kinetics from ibuprofen matrix tablets using HPMC. *Pak. J. Pharm. Sci*. **2006**, *19* (2), 119–124.