Formulation and characterization of gastro retentive hollow microspheres filled with Nateglinide for diabetes care

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Abstract

The study aimed to develop microspheres containing Nateglinide, which could be used in the gastrointestinal to enhance the drug's retention and bioavailability. Various polymers, such as carbopol 934, methylcellulose, and eudragit, were used in the solvent diffusion emulsion process to create the microspheres. The micrometric properties, in vitro lightness, percent yield, capturing efficiency, and *in vitro* tests were evaluated, along with FT-IR and SEM. The results showed that the size of the microspheres was directly proportional to the skimming time, and the in vitro drug release tests revealed significant nateglinide release for over eight hours, with ethylcellulose formulations exhibiting better results across all formulations. The release behavior was also customized within Peppas and zero-order equations. These microspheres with gastric coating have higher yields and could be used effectively to monitor blood glucose levels for nateglinide administration.

Keywords: Diabetes, Nateglinide, Eudragit, Microsphere, Solvent diffusion emulsion technique, etc.

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1. Introduction

Diabetes mellitus is when the body has trouble regulating blood glucose levels due to insulin secretion or resistance issues. This condition is widespread and poses a significant global health challenge[1]. To manage diabetes, it is essential to maintain healthy blood glucose levels to prevent complications such as cardiovascular diseases, neuropathy, nephropathy, and

retinopathy. Oral drugs are a convenient and non-invasive way to treat diabetes, but they often have poor bioavailability and short half-lives. This means they are only sometimes effective in regulating blood glucose levels [2-3]. Researchers have created gastroretentive drug delivery systems (GRDDS) to increase the therapeutic effectiveness of antidiabetic medications. These systems are intended to maintain medications in the stomach for longer, enhancing absorption and bioavailability and delivering more consistent drug administration[4]. One type of GRDDS is gastroretentive hollow microspheres filled with the antidiabetic drug nateglinide. *Nateglinide* is a meglitinide analog that stimulates insulin secretion from pancreatic beta cells, reducing postprandial blood glucose levels. However, its short half-life and fast absorption from the gastrointestinal tract make it challenging to administer[5-6]. Hollow microspheres are a promising solution to this problem because they can carry a high drug payload and stay in the stomach longer, prolonging drug release and improving therapeutic efficacy[6-7]. Our study aims to formulate and characterize gastroretentive hollow microspheres loaded with Nateglinide for diabetes care. We will use biocompatible polymers to create microspheres of various shapes and sizes. Then, we will evaluate the drug-loaded microspheres' properties, including morphology, size distribution, drug content, and drug release kinetics[8]. This research has the potential to improve the treatment of diabetes and help patients maintain healthy blood glucose levels.

2. Materials

Nateglinide as a gift from Hetero Drugs Ltd in Hyderabad. Additionally, Vijaya Chemicals in Hyderabad procured Eudragit RS 100, Ethylcellulose, Carbopol 934, Methanol Dichloromethane, and other excipients.

2.1 Methods

2.1.1 Preparation and examination of microspheres with Nateglinide

Microspheres containing Nateglinide were created through a *Solvent diffusion emulsion technique* utilizing Eudragit, Carbopol 934, and ethylcellulose as polymers. The process involved the following steps: Nateglinide, its active pharmaceutical ingredient, was dissolved in 5 cc of distilled water. Various ratios of drug-to-polymer (1:1, 1:2, and 1:3) were created by dissolving the appropriate quantities. Using a Remi Lab Magnetic stirrer for around 10 minutes at 500 rpm, the drug-polymer solutions were mixed and emulsified into a stable water-in-oil (w/o) emulsion. The stable without emulsion was progressively added to a 200 ml aqueous solution of 1% polyvinyl alcohol (PVA) while constantly stirring at 1000 rpm using a mechanical stirrer fitted with a room temperature three-bladed propeller (Remi engines,

India). This procedure was continued for two hours to allow for the full breakdown of the solvent. The microspheres were separated by filtering and repeatedly rinsed with sterile water to remove any remaining PVA. The produced hollow microspheres underwent a 24-hour drying process. The dried microspheres were vacuum dried at 25°C for 2 hours in a vacuum chamber to eliminate residual solvents, ensuring the complete removal of any remaining solvents[9-11].

2.1.2 Evaluation of hollow microspheres

Particle size analysis

Release and floating properties depend on particle size analysis. We measured hollow microspheres using 14, 16, 18, 22, 30, and pan sieves. The sieves were in ascending sequence. Each filter retained a percentage weight of hollow microspheres. Then, for all formulations, the mean particle size was determined by dividing the formula weight by the percentage of hollow microsphere weight maintained[12].

FTIR and Scanning Electron Microscopy (SEM)

To analyze the properties of Nateglinide, we exposed it to FTIR and obtained an infrared spectrum within a range of 400-4000 cm^{-1}. We then examined the formulation that displayed an acceptable balance between buoyancy and percentage release. We examined the surface morphology and shape of the hollow microspheres using the Hitachi, Japan, scanning electron microscope. We fixed the sample on carbon tape in a high vacuum evaporator to prepare for the examination and applied fine sputtering gold. We set the acceleration voltage at 20kV during scanning and took various magnifications with microphotographs[13]. For surface morphology, we used higher magnifications.

Drug Entrapment

Hollow microspheres containing Nateglinide that have been weighed and crushed. It took 12 hours to store powdered microspheres in 10 cc of ethanol. Filtering solution [14] was Whatman filter paper No. 44. A UV spectrophotometer (Shimadzu 1700) measured absorbance at 216 nm after diluting with a fresh solvent to calculate the percentage of drug entrapment.

$$\%\mathrm{DE} = \frac{\text{Calculated drug conten}}{\text{Calculated drug content Theoretical drug conten}} t \times 100$$

In vitro drug release study

To measure the drug release rate from the microspheres, we employed a six-station dissolving test device (LABINDIA, DS8000) of the paddle type. At 37 0.5 °C and 100 rpm, we suspended

the microspheres in 0.1 N HCl (1.2 pH), and their total weight was equivalent to 16 mg of the medication. We kept everything in the sink for the duration of the trial. Spectrophotometric analysis at 216 nm was performed on 1 ml samples taken at 30-minute intervals filtered through a 5-µm membrane[15-17].

Kinetic modeling of drug release

Several mathematical models were used for the nateglinide-loaded microspheres better to understand the kinetics and mechanism of drug release. The results of the in vitro drug release study of microspheres were fit by a variety of kinetic equations, including Higuchi's model (cumulative percentage of drug release v/s square root of time), Korsemeyer Peppas model (log cumulative percentage of drug release v/s log time), and Hixson Crowell model (cube root of percentage drug remaining v/s time). Plots of the data on releases were made. Regression analysis of these plots yielded linear segments from which R2 and slope values were derived[18-19].

2.1.3 Stability Study

Stability tests were performed to ensure the hollow microspheres had the proper density and release percentage[20-22]. The microspheres were stored for 90 days in borosilicate screw-capped glass containers at ambient temperature (27 ± 2 °C) and oven temperature (42 ± 2 °C) to test the stability of the formulation.

3. Results and Discussion

3.1 Particle size analysis

The sieving procedure was used to determine particle size. Their size greatly influences microspheres' buoyancy and drug release. The medication was released most quickly from hollow microspheres with a size of 500 nm, with a mean particle size of 756 nm. Table 1 shows that particle size increased along with polymer concentration.

Table 1 Formulations of Nateglinide hollow microspheres and particle size

F. no	**Polymer**	**Drug and polymer ratio**	**Mean particle size* (nm)**
F1	Eudragit	1:1	758
F2	Eudragit	1:2	762
F3	Eudragit	1:3	788
F4	Ethylcellulose	1:1	764
F5	Ethylcellulose	1:2	781
F6	Ethylcellulose	1:3	804
F7	Carbopol 934	1:1	756
F8	Carbopol 934	1:2	767
F9	Carbopol 934	1:3	799

3.2 Scanning Electronic Microscopy (SEM)

SEM was used to examine hollow microspheres to identify their form and surface characteristics, as shown in Fig 1. At 40x magnification, the improved F6 formulation's surface morphology was studied, and the microsphere's tiny hollow cavity, responsible for the floating feature, was visible inside the microsphere. The particle size (F6 formulation) was measured to be 804 mm. They imagined the hollow microspheres to be perfectly round. (Fig 1)

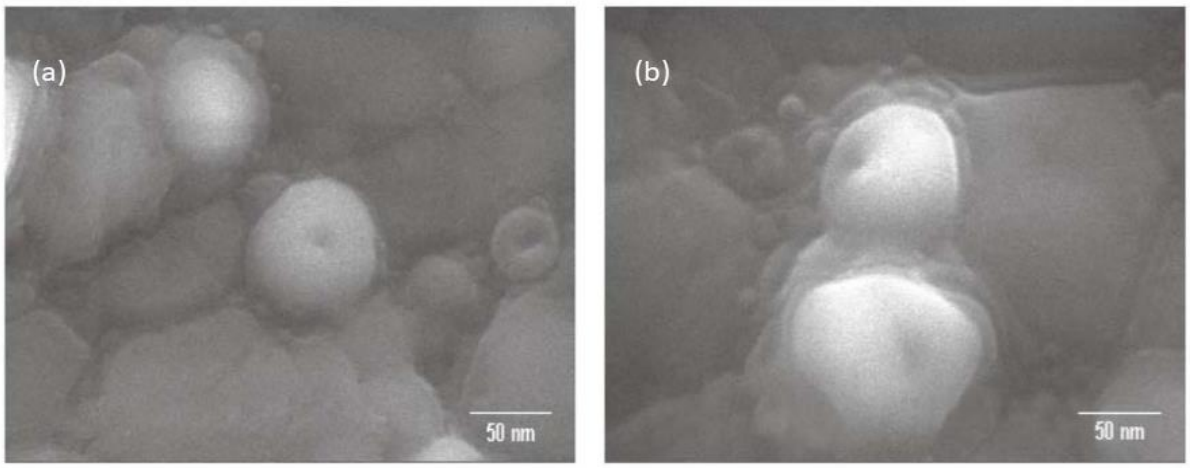

Fig 1: SEM Study of (a) Formulation F6 (b) Cross Section

3.3 FT-IR Spectrum of Nateglinide

The FTIR spectrophotometer was used to describe the FT-IR formulation of the nateglinide spectrum (Fig. 2) and F6. The formulation included all the peaks, and the physical blend revealed no chemical reactivity between the Nateglinide and the polymer. At 2972.18 (O-H stretching), 1049.31 (O-H bending), 2867.50 (C-H stretching), and 1692.11 (C=O stretching), The spectrum of Nateglinide (Fig. 2 a) has shown characteristic peaks. The peaks observed in the F6 formulation range (Fig.2 b) suggest O-H stretching at 2916.84, O-H bending at 1071.96, and C=O stretching at 1575.23. (Fig. 2) The drug's stability during the microencapsulation process was also confirmed. In the formulation and physical mixture, both peaks have emerged, suggesting no chemical reaction between Nateglinide and polymer

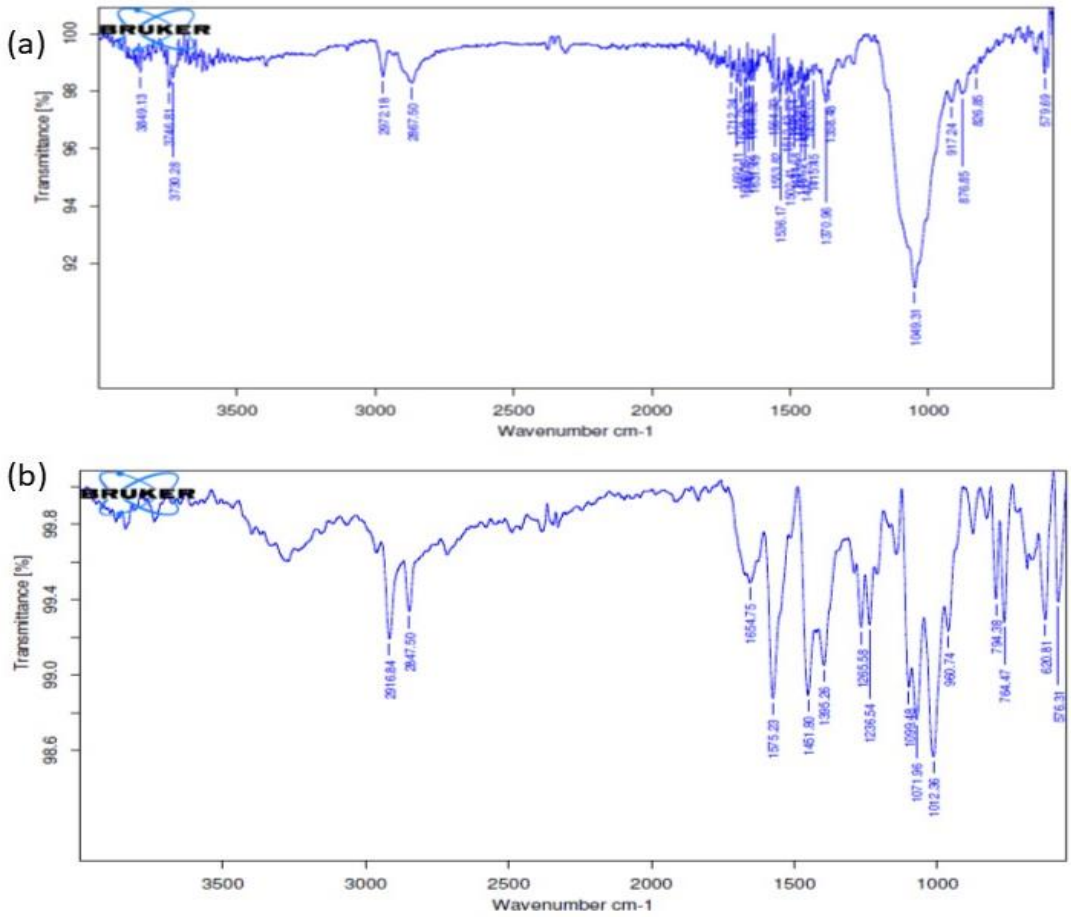

Fig 2: FTIR Spectra of a) Nateglinide b) F6 formulation

3.4 Drug Entrapment

The efficacy of various formulations for drug trapping was 68.18 percent - 84.38 percent w/w. The efficacy of drug trapping decreased marginally with increased carbopol 934 content. Due to carbopol 934's permeability properties, which might encourage the diffusion of some of the

trapped drug to the surrounding medium during the creation of hollow microspheres, lowered the micro balloon eudragit ratio. F6 (ethyl cellulose) entrapment was found to be more (84.38 percent) for the selected formulation, which allowed us to select F6 (drug and polymer) as an optimized formulation in Table 2.

Table 2: The percentage yield and drug entrapment of various formulations

Formulation	**Drug Entrapment (% w/w)**	**Percent Yield*(%)**
F1	74.25 ±1.52	82.96±1.45
F2	71.58±0.42	81.70±1.52
F3	80.36±1.21	80.28±1.12
F4	68.18±1.41	82.56±1.14
F5	75.62±2.21	83.03±1.32
F6	84.38±1.11	86.34±1.19
F7	70.15±1.22	79.44±1.44
F8	78.20±1.24	75.10±1.26
F9	79.06±1.33	74.78±1.42

3.5 In-Vitro Drug release study

The in-vitro micro balloon drug release study was evaluated at 0.1N HCl. The formulation's Eudragit RS100 component has a low acid permeability. The release of the drug was typically minimal at 0.1 N HCl because eudragit is less soluble at acidic pH (Table 3). All (F1-F9) formulations of hollow microspheres were subjected to dissolution experiments. In the Franz diffusion cell apparatus, dissolution was conducted at 100 rpm at 10 ml of 0.1N HCl dissolution media for 8 hours. F6 showed a release rate of 97.18 percent by the end of the 8th hour of the dissolution study (Fig.3).

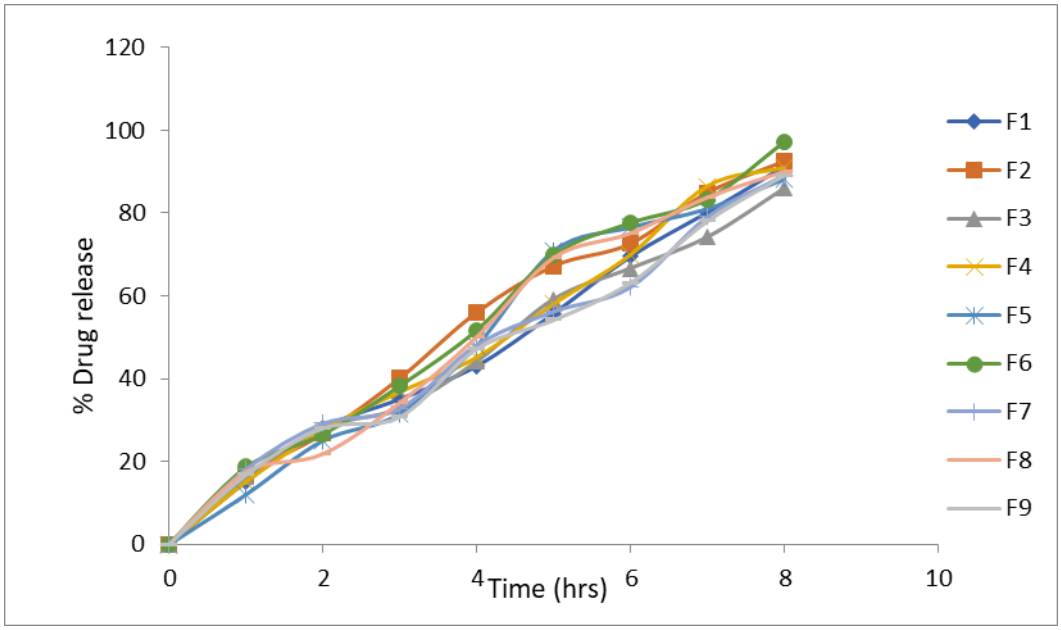

Fig 3: *In vitro* Drug release profiles of formulations

Release kinetic

The zero-request, Higuchi condition, and Pappas model analyzed the energy and part of pharmaceutical discharge from all meanings. For each scenario, Microsoft Excel agreed on the relationship coefficient (r2) and the slant and reward. For all meanings, plots of zero-request were found to be straight in the two disintegration vehicles. This implies that it will adopt a zero-request interaction. In the Peppas plot, n > 0.5 was observed for all features, suggesting that drug transport may follow unusual diffusion. The Higuchi plot was determined to be straight. The optimum analog for drug discharge, the zero-request plot for the F6 description, was discovered to be direct in both disintegration media (Fig. 4).

The table indicates that for Higuchi's model, the R^2 values were higher than for all the microspheres. As a result, the release of Nateglinide from all films followed the diffusion rate-regulated process. The drug release in vitro has a variety of kinetic variations. The coefficient of regression (R2) for formulations F6 and (R2) was determined to be 0.989, 0.943, 0.934, and 0.001 for zero order, first order, Higuchi, and Pepas, respectively. The kinetics of zero-order paralleled the kinetics of drug release (Table 3).

Table3:Drug Release Kinetics of Formulation F6

TIME	%CDR	SQUARE T	LOG T	LOG%CDR	ARA	LOG%ARA
0	0	0	0	0	0	0
1	18.62	1	0	1.270	81.38	1.911
2	26.6	1.414	0.301	1.425	73.4	1.866
3	38.29	1.732	0.477	1.583	61.71	1.790
4	51.74	2.000	0.602	1.714	48.26	1.684
5	69.92	2.236	0.699	1.845	30.08	1.478
6	77.63	2.449	0.778	1.890	22.37	1.350
7	83.33	2.646	0.845	1.921	16.67	1.222
8	97.18	2.828	0.903	1.988	2.82	0.450

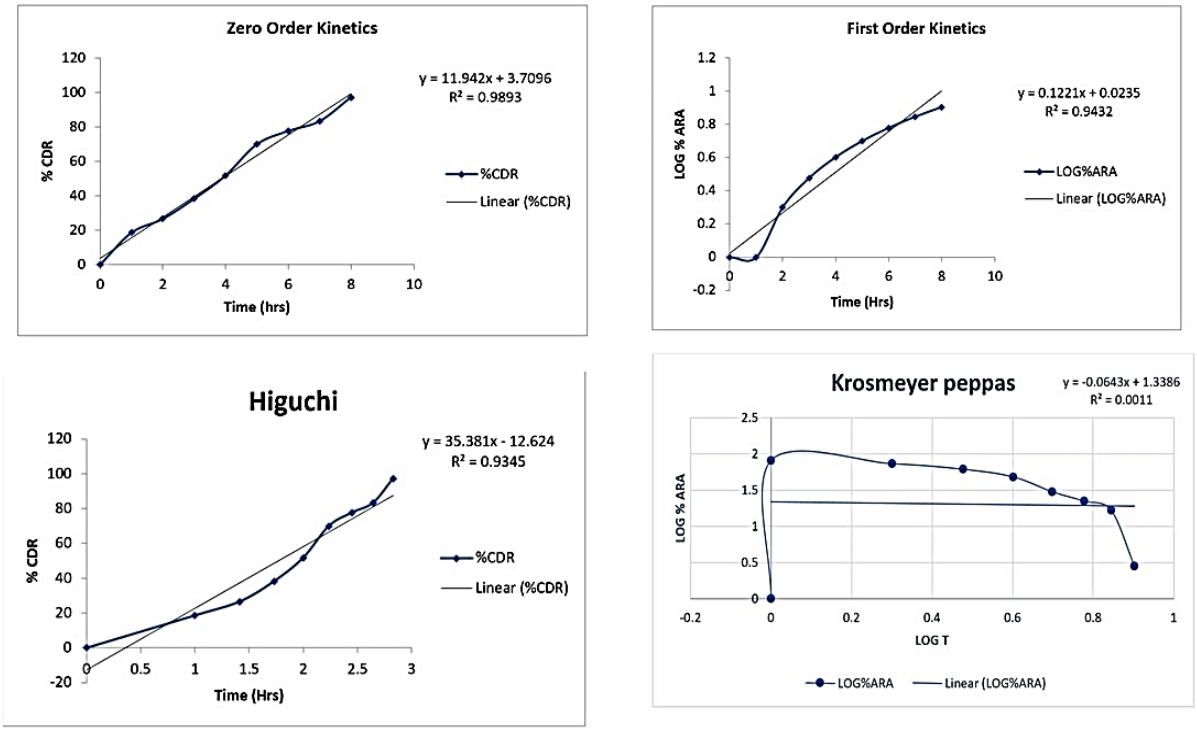

Fig 4: Drug Release Kinetics Optimized formulation (F6)

3.6 Stability Study

The F6 formulation underwent a stability test, heated to 25, 30, and 40 degrees Celsius for 90 days. The sample's drug content was checked at set intervals. Table 4 shows that the F6 formulation did not considerably improve the drug's quality. F6 was stable at the following temperatures.

Table 4: Results of stability studies of optimized formulation F-6

S.NO	Time in days	Physical changes	Mean % drug release		
			Nateglinide		
			25°C/60%	30°C/75%	40°C/75%
1.	01	No Change	97.18	97.18	97.18
2.	30	No Change	96.83	96.82	95.81
3.	60	No Change	96.79	96.79	96.75
4.	90	No Change	96.64	96.68	96.69

4. Conclusion

A Solvent diffusion emulsion technique created hollow microspheres since the material was soluble in the interior organic phase. This was important for weakly water-soluble medicines. The approach effectively synthesized nateglinide-loaded microspheres, which exhibited favorable micrometric characteristics. Encapsulation efficiency percentages for microspheres showed that medication loading was maximized and increased with polymer count. Ethylcellulose was discovered to have the best drug-keeping and releasing qualities of all the polymers tested. The drug was accurately and evenly dispersed throughout the hollow microsphere, as determined by drug content. Particle size analysis confirmed that all process variables fell within acceptable ranges and that the procedure could be repeated reliably. Therefore, the microspheres may show improved bioavailability of Nateglinide in diabetic individuals.

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