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# OPTIMIZATION AND CHARACTERIZATION OF MICROSPHERE FORMULATIONS FOR CONTROLLED DRUG DELIVERY OF CARVEDILOL

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**ABSTRACT:**

This comprehensive study focused on the fabrication, development and evaluation of six mucoadhesive gastroretentive formulations (CVF-1 to CVF-6) through various critical parameters, including encapsulation efficiency, drug loading capacity, mucoadhesion, yield percentage, swelling index, and kinetic modelling of drug release. The encapsulation efficiency revealed CVF-6 as the most effective, with a high percentage suggesting minimal drug loss during the encapsulation process. The drug loading capacity was notably high for CVF-4, indicating its potential for delivering a substantial amount of drug per unit weight of the microsphere. Furthermore, mucoadhesive properties and in vitro drug release studies at pH 1.2 provided insights into the interaction of microspheres with the gastric mucosa and their subsequent drug release behaviours. Kinetic modelling using Zero-order, First-order, Higuchi, and Korsmeyer-Peppas models validated the drug release mechanisms, highlighting the formulations' capacity for sustained and controlled drug release, particularly for CVF-2 and CVF-6 which demonstrated superior comprehensive profiles as gastroretentive mucoadhesive microspheres.

**Keywords:** Mucoadhesive, Gastroretentive microspheres, Carvedilol, Mucoadhesive microspheres

## INTRODUCTION

Gastroretentive drug delivery systems (GRDDS) have emerged as a pivotal strategy in the field of pharmaceutical sciences to enhance the bioavailability and therapeutic efficacy of drugs with narrow absorption windows in the upper gastrointestinal (GI) tract. Among various GRDDS technologies, mucoadhesive microspheres play a crucial role due to their ability to adhere to the gastric mucosa, thereby prolonging the gastric residence time of drugs. This prolongation allows for sustained drug release, improved bioavailability, and targeted delivery within the GI tract, which is particularly beneficial for drugs that are locally active in the stomach, or those that are absorbed primarily in the upper part of the small intestine (Mishra et al., 2018, Beg et al., 2019, Smart, 2005). Mucoadhesive microspheres are typically small, spherical polymeric particles that can bind to the mucosal surface of the stomach. The concept revolves around the microsphere's ability to adhere to the mucosal lining, a feature that prevents the dosage form from being emptied from the stomach during the gastric emptying process. This adhesion is facilitated by the presence of mucoadhesive polymers that interact with the glycoprotein content of mucus covering the epithelial cells lining the GI tract (Beg et al., 2019, Smart, 2005, Mansuri et al., 2016). The selection of polymers is critical for the formulation of effective mucoadhesive microspheres. Natural polymers such as chitosan, alginate, and pectin are frequently used due to their biocompatibility, biodegradability, and inherent mucoadhesive properties. Synthetic polymers, including polyacrylates and carbopol, are also employed for their strong adhesive forces and chemical versatility. These polymers can form hydrogen bonds and van der Waals forces with the mucin in the stomach lining, enhancing the mucoadhesive

characteristics of the microspheres (Mishra et al., 2018, Yeh et al., 2023, Rana et al., 2023, Wang et al., 2020, 2012, 2006, 1994).

The fabrication of mucoadhesive microspheres involves various techniques such as spray drying, emulsion solvent evaporation, ionotropic gelation, or hot-melt extrusion. Each method offers distinct advantages in terms of controlling particle size, drug loading, and release kinetics. For instance, the ionotropic gelation technique is particularly useful for encapsulating hydrophilic drugs and achieving controlled release profiles. The choice of method depends on the properties of the drug and polymer, as well as the desired therapeutic outcomes. The efficacy of mucoadhesive microspheres is influenced by several factors including particle size, surface morphology, polymer concentration, and the nature of the drug molecule. Smaller particles often provide a larger surface area for interaction with the mucosal surface but may exhibit quicker release rates. Thus, optimizing the size and surface properties of these microspheres is crucial for balancing adhesion with effective drug release (Smart, 2005, Khutoryanskiy, 2011, Mansuri et al., 2016).

In vitro and in vivo studies play a vital role in evaluating the performance of gastroretentive mucoadhesive formulations. Parameters such as swelling index, mucoadhesion strength, and in vitro drug release are critically assessed. The swelling index indicates the ability of the microspheres to swell in the presence of gastric fluids without dissolving, which is important for maintaining prolonged gastric residence. Mucoadhesion strength tests quantify the force required to detach the microspheres from the stomach lining, reflecting the potential of the formulation to stay in the stomach for extended periods. Moreover, drug release studies are conducted under simulated gastric conditions to understand the release kinetics and predict the in vivo behaviour. Gastroretentive mucoadhesive microspheres represent a significant advancement in targeted drug delivery systems, offering a promising approach for improving the pharmacokinetic profiles of drugs (Stancil et al., 2024, Naderi et al., 2024, Yuan et al., 2023, Kharel et al., 2022, Athar et al., 2022, Rissardo and Caprara, 2020). Through sophisticated formulation techniques and rigorous testing, these systems continue to evolve, providing enhanced solutions for the treatment of various diseases where prolonged gastric retention is advantageous. As research progresses, these innovative systems are expected to play an increasingly important role in personalized medicine, particularly in the management of diseases that benefit from localized drug delivery in the gastrointestinal tract (Smart, 2005, Khutoryanskiy, 2011, Mansuri et al., 2016). (Nadpara et al., 2012, Mishra et al., 2018).

Carvedilol is a non-selective beta-blocker with additional alpha-1 adrenergic blocking properties, making it distinctive among beta-blockers. This dual-action makes Carvedilol particularly effective in treating high blood pressure and heart failure. It is commonly prescribed to improve survival following heart attacks and to manage hypertension and angina (Frishman, 1998, Ruffolo Jr et al., 1990). The mechanism of action of Carvedilol involves the blocking of beta-1 and beta-2 adrenergic receptors, which are responsible for the effects of adrenaline and noradrenaline. By inhibiting these receptors, Carvedilol reduces heart rate, blood pressure, and the strain on the heart. Additionally, its alpha-1 receptor blockade helps dilate blood vessels, further lowering blood pressure and improving blood flow. Carvedilol has been widely studied and is notable for its protective properties against oxidative stress and apoptosis in cardiac tissues, which are beneficial in heart conditions. It also differs from other beta-blockers in that it does not significantly affect lipid metabolism or cause respiratory side effects, which are common drawbacks of other medications in this class. The drug is metabolized primarily in the liver and requires careful dose adjustments in individuals with hepatic impairment. Carvedilol's efficacy and safety profile have established it as a cornerstone therapy in the management of multiple cardiovascular conditions, offering a significant therapeutic advantage due to its dual blockade of adrenergic receptors (Dulin and Abraham, 2004, Stafylas and Sarafidis, 2008).

Carvedilol is primarily used in the treatment of cardiovascular diseases such as hypertension, heart failure, and angina, conditions that benefit from maintained consistent plasma drug levels to ensure therapeutic efficacy. The conventional oral dosage forms of carvedilol suffer from relatively low bioavailability, approximately 25-35%, primarily due to significant first-pass metabolism in the liver. Additionally, carvedilol has a short half-life of about 7-10 hours, necessitating multiple dosages throughout the day to maintain effective drug levels (Morgan, 1994).

The gastroretentive mucoadhesive microsphere approach can significantly mitigate these challenges. First, by localizing the drug's release in the stomach and upper part of the small intestine where carvedilol is absorbed, these microspheres can enhance the bioavailability of the drug. This localized, prolonged release not only ensures more consistent plasma concentrations but also reduces the frequency of dosing required, enhancing patient compliance. Moreover, the mucoadhesive properties of these microspheres allow for prolonged gastric retention time (Morgan, 1994, v Möllendorff et al., 1987). This is particularly advantageous for carvedilol, which, given its solubility and absorption characteristics, can benefit greatly from extended release in the upper gastrointestinal tract. The extended residence time in the stomach avoids the rapid transit to less favourable absorption sites in the lower gastrointestinal tract, optimizing the absorption window of the drug. In addition to pharmacokinetic benefits, the use of gastroretentive systems for carvedilol could also reduce the variability of plasma drug levels, a common issue with conventional formulations that can lead to inconsistent therapeutic outcomes. By moderating the drug release rate and extending the drug's presence at the absorption site, these systems can facilitate a more stable cardiovascular response, reducing the risks of peaks and troughs associated with conventional dosing (v Möllendorff et al., 1987). Thus, incorporating carvedilol into gastroretentive mucoadhesive microspheres presents a rational and innovative approach to overcome the limitations of existing formulations, aiming to improve the efficacy, safety, and patient adherence in the management of cardiovascular diseases (v Möllendorff et al., 1987). Therefore, considering all the above facts, this present study was designed to fabricate the development and evaluation of mucoadhesive gastroretentive microspheres of Carvedilol.

## MATERIAL AND METHODS

### Material

Several essential components were needed to prepare the mucoadhesive gastroretentive microspheres containing carvedilol. The active pharmaceutical ingredient (API), carvedilol, is well-known for its ability to control heart failure and hypertension. Newlife Pharma, Baddi, Himachal Pradesh, kindly supplied a gift sample of this medication. The main polymer, sodium alginate, was purchased from Loba Chem in Mumbai, India, coupled with chitosan, and hydroxypropyl methyl cellulose (HPMC K4M). Calcium chloride ($CaCl_2$) was acquired from Sigma Aldrich in Mumbai, India. Purchased from Himedia, India, The preparation technique required analytical-grade ethanol and water, both of which were provided by Loba Chem, India. The remaining substances, solvents, and reagents were all analytical grade and were exclusively purchased from reliable suppliers.

### Methods

### Preparation of microsphere formulations

The preparation of microsphere formulations containing Carvedilol involved a series of systematic steps. Initially, 2 g of Na-alginate was dissolved in 100 mL of distilled water under continuous stirring to achieve a homogeneous solution. Separately, the specified amounts of the additive polymers (either HPMC or Chitosan) were dissolved in distilled water. Subsequently, 0.1 g of Carvedilol was added to the Na-alginate solution and stirred continuously until the drug was uniformly dispersed. The additive polymer solution was then

combined with the drug-loaded Na-alginate solution, ensuring thorough mixing to form a uniform mixture. This polymer-drug mixture was then extruded using a syringe into a coagulation medium containing 1.5% $CaCl_2$ solution. The droplets formed were allowed to harden, resulting in the formation of microspheres through ionic gelation. The resulting microspheres were collected by filtration and washed with distilled water to remove any residual calcium chloride. Finally, the microspheres were dried at room temperature or in an oven until a constant weight was achieved. The dried microspheres were stored in airtight containers at room temperature for further characterization and analysis. The compositions of the different microsphere formulations are detailed in Table 1, with varying amounts of additive polymers and drug/Na-alginate ratios, aimed at optimizing the drug delivery properties of Carvedilol.

**Table 1.** The composition of the microsphere formulations

| Formulation | Amount of Na-alginate (g) | Amount of Carvedilol (g) | Polymer | Amount of Additive Polymer (g) | Drug/Na-Alginate Ratio | Coagulation Medium ($CaCl_2$ Solution) |
|---|---|---|---|---|---|---|
| CVF-1 | 2 | 0.1 | HPMC | 1 | 0.1:1 | 1.5% |
| CVF-2 | 2 | 0.1 | HPMC | 2 | 0.05:1 | 1.5% |
| CVF-3 | 2 | 0.1 | HPMC | 3 | 0.025:1 | 1.5% |
| CVF-4 | 2 | 0.1 | Chitosan | 1 | 0.1:1 | 1.5% |
| CVF-5 | 2 | 0.1 | Chitosan | 2 | 0.05:1 | 1.5% |
| CVF-6 | 2 | 0.1 | Chitosan | 3 | 0.025:1 | 1.5% |

**Characterization of the Carvedilol Mucoadhesive Gastroretentive Microspheres**

**Particle Size and Morphology**

Initially, the particle size distribution was determined using an optical microscope equipped with a calibrated eyepiece micrometer. Approximately 100 microspheres were randomly selected and measured to calculate the average particle size. The results were reported as mean diameter ± standard deviation. For the morphological analysis, SEM was employed. The microspheres were first dried and then mounted on metal stubs using double-sided adhesive tape. The samples were coated with a thin layer of gold under vacuum to make them conductive. The coated samples were then observed under the SEM, and images were captured at various magnifications. These images were analysed to assess the surface characteristics, shape, and overall morphology of the microspheres. This dual approach provided comprehensive data on the particle size and surface morphology, which are critical parameters for evaluating the quality and performance of the microsphere formulations (Hardenia et al., 2011, Das and Ng, 2010).

**Drug Loading Efficiency and Encapsulation Efficiency**

To determine the drug loading efficiency and encapsulation efficiency of the prepared microspheres, a specific method was followed. Initially, a known quantity of dried microspheres, approximately 100 mg, was accurately weighed and transferred into a 100 mL volumetric flask. To extract the drug from the microspheres, 50 mL of phosphate buffer with a pH of 7.4 was added to the flask. This mixture was then sonicated for 30 minutes to ensure complete dissolution of the encapsulated drug. Following sonication, the solution was filtered through Whatman filter paper to remove any particulate matter. The filtrate was then analysed using a UV-Visible spectrophotometer set to a predetermined wavelength specific to Carvedilol. The absorbance of the solution was measured, and the concentration of Carvedilol was determined using a standard calibration curve. To calculate the Drug Loading Efficiency

(DLE), the following formula was used: DLE (%) = (Amount of drug in microspheres / Weight of microspheres) × 100. For the Encapsulation Efficiency (EE), the formula used was: EE (%) = (Practical drug content / Theoretical drug content) × 100. The practical drug content refers to the amount of drug actually found in the microspheres, as determined from the spectrophotometer analysis, while the theoretical drug content is the expected amount of drug based on the initial formulation. This detailed method ensured the accurate determination of both drug loading and encapsulation efficiencies, which are critical parameters for assessing the effectiveness of the microsphere formulations in drug delivery applications (Yadav and Jain, 2011).

**Percentage Yield**

The percentage yield of the prepared microspheres was calculated to evaluate the efficiency of the fabrication process. Initially, the weight of the dried microspheres was recorded. The percentage yield was then determined using the following formula:

$$\text{Yield (\%)} = \frac{Quantity\ of\ microspheres\ produced}{Theoretical\ content}\ x\ 100$$

Here, the total weight of initial materials includes the combined weights of Na-alginate, Carvedilol, and the additive polymers used in the formulation. This formula provided a measure of the efficiency and effectiveness of the microsphere production process. By comparing the actual weight of the final product to the theoretical weight of the initial components, the percentage yield offered insights into the overall success and reproducibility of the method. High percentage yield values indicated that the fabrication process was efficient, with minimal losses during production (Yadav and Jain, 2011)

**Swelling Index**

The swelling index of the prepared microspheres was determined to assess their ability to swell in an aqueous environment, which is crucial for their mucoadhesive properties and drug release behaviour (Shivanand et al., 2010). To measure the swelling index, the following procedure was used: Initially, a known weight of dried microspheres, approximately 100 mg, was accurately measured and recorded as W0W_0W0. These microspheres were then immersed in a beaker containing 100 mL of phosphate buffer with a pH of 7.4 at 37°C. The microspheres were allowed to swell for a predetermined period, typically 2 hours. After the swelling period, the microspheres were carefully removed from the buffer solution, blotted gently with filter paper to remove excess surface water, and weighed again. This weight was recorded as We.

**Swelling Index** = We -$W_O$/ $W_O$

Where, Wo = Initial weight of the dry microspheres,

We = weight of the swollen microspheres at equilibrium swelling in the media.

This formula provided a quantitative measure of the extent to which the microspheres could absorb water and swell. A higher swelling index indicated a greater capacity for water uptake, which is beneficial for enhancing the mucoadhesive properties and facilitating the controlled release of the encapsulated drug. This method ensured an accurate evaluation of the swelling behaviour of the microspheres, contributing to the overall assessment of their suitability for gastroretentive drug delivery applications.

***In Vitro* Mucoadhesion Study**

The in vitro mucoadhesion study was conducted to evaluate the adhesive properties of the prepared microspheres, which are essential for ensuring prolonged gastric retention. The study was performed using excised mucosal tissue from a freshly obtained porcine stomach (Hardenia et al., 2011). Initially, the mucosal tissue was thoroughly rinsed with phosphate buffer (pH 7.4) to remove any mucus and debris, and then mounted on a glass slide with the mucosal side facing up. A known quantity of dried microspheres, approximately 100 mg, was evenly spread over the surface of the mucosal tissue. The glass slide with the tissue and microspheres was then placed in a petri dish containing 50 mL of phosphate buffer (pH 7.4) at

37°C to simulate gastric conditions. The petri dish was then subjected to mild agitation using a mechanical shaker to mimic the peristaltic movements in the gastrointestinal tract. At predetermined time intervals (1, 2, 3, 4, 5, and 6 hours), the petri dish was gently swirled, and the number of microspheres still adhering to the mucosal surface was counted. This count provided a measure of the mucoadhesive strength of the microspheres over time. The mucoadhesion percentage was calculated using the following formula:

$$\text{Mucoadhesion } \% = \frac{\text{weight of adhered microspheres}}{\text{weight of applied microspheres}} \times 100$$

This formula quantified the proportion of microspheres that remained attached to the mucosal tissue compared to the total number initially applied. A higher mucoadhesion percentage indicated stronger adhesive properties, which are desirable for prolonged gastric retention and effective drug delivery. This in vitro study provided valuable insights into the adhesive characteristics of the microspheres, supporting their potential use in gastroretentive drug delivery systems.

***In Vitro* Drug Release Study**

The in vitro drug release study was conducted to evaluate the release profile of Carvedilol from the prepared mucoadhesive gastroretentive microspheres. This study aimed to determine the rate and extent of drug release over a specified period under simulated gastric conditions (Shivanand et al., 2010). To begin the study, a known quantity of dried microspheres containing Carvedilol, equivalent to 10 mg of the drug, was accurately weighed and placed in a dialysis bag. The dialysis bag was then submerged in a beaker containing 100 mL of phosphate buffer (pH 7.4) maintained at 37°C to simulate gastric fluid. The beaker was placed on a magnetic stirrer set at a moderate speed to ensure uniform mixing. At predetermined time intervals (e.g., 0.5, 1, 2, 3, 4, 5, 6, 8, 10, 12, and 24 hours), 5 mL aliquots of the dissolution medium were withdrawn using a pipette and replaced with an equal volume of fresh phosphate buffer to maintain sink conditions. Each withdrawn sample was filtered through a 0.45 µm filter to remove any particulate matter and analyzed using a UV-Visible spectrophotometer at a wavelength specific to Carvedilol. The concentration of Carvedilol in each sample was determined using a standard calibration curve. The cumulative percentage of drug release was then calculated using the following formula:

$$Cumulative\ Drug\ Release(\%) = \frac{Amount\ of\ drug\ released\ at\ each\ time\ poin}{Total\ amount\ of\ drug\ in\ the\ microspheres} * 100$$

The drug release data were plotted as cumulative percentage release versus time to visualize the release profile. This graph provided insights into the release kinetics and mechanism of drug release from the microspheres. The in vitro drug release study demonstrated how effectively the microspheres could control the release of Carvedilol over an extended period. A sustained release profile indicated the potential of the microspheres to maintain therapeutic drug levels in the gastrointestinal tract, enhancing the drug's efficacy and patient compliance. This method provided a comprehensive evaluation of the drug release characteristics of the microsphere formulations, supporting their application in gastroretentive drug delivery systems.

**Fourier Transform Infrared Spectroscopy (FTIR)**

Fourier Transform Infrared Spectroscopy (FTIR) was employed to analyse the chemical structure and identify any potential interactions between Carvedilol and the polymers used in the microsphere formulations. This analysis helped to confirm the integrity of the drug and detect any possible chemical interactions that might occur during the fabrication process. Initially, samples of pure Carvedilol, Na-alginate, HPMC, Chitosan, and the drug-loaded microspheres were prepared. Each sample was finely ground with potassium bromide (KBr) powder at a ratio of 1:100 (sample) to form a homogeneous mixture. This mixture was then

compressed into a transparent pellet using a hydraulic press.The FTIR spectra of these samples were recorded using an FTIR spectrophotometer. The scanning range was set from 4000 $cm^{-1}$ to 400 $cm^{-1}$ with a resolution of 4 $cm^{-1}$. The spectra of the pure components were first obtained to identify the characteristic peaks of Carvedilol and the polymers. Subsequently, the spectra of the drug-loaded microspheres were recorded and compared with the spectra of the pure components. This comparison was crucial to identify any shifts in the characteristic peaks or the appearance of new peaks, which would indicate potential interactions between the drug and the polymers. The FTIR analysis provided detailed information on the chemical structure and potential interactions in the microsphere formulations. The presence of characteristic peaks for Carvedilol in the spectra of the drug-loaded microspheres confirmed the retention of the drug's chemical integrity. Any shifts or changes in the peak positions were carefully analysed to assess the nature of the interactions between Carvedilol and the polymers. This method ensured a thorough understanding of the chemical compatibility and stability of the microsphere formulations, supporting their development as effective drug delivery systems.

**Statistical analysis**

GraphPad Prism (Version 8) was the statistical analysis programme used in this investigation for statistical analysis of the data. Tukey's tests were used as a post hoc analysis for particular group comparisons after a One-Way Analysis of Variance (ANOVA) was used to evaluate variability and significance among several groups. The data were displayed as averages ± standard deviation (SD). A cutoff point of as statistically significant $p < 0.05$ was established.

## RESULTS AND DISCUSSION

***Fabrication of microsphere formulations, Encapsulation efficiency and Drug loading***

The encapsulation efficiency (EE) and drug loading capacity (LC) are critical parameters in evaluating the effectiveness of the drug delivery system. The data in table 2 shows the encapsulation efficiency and drug loading capacity for six different formulations (CVF-1 to CVF-6). Encapsulation efficiency indicates the percentage of the initial drug amount that is successfully encapsulated within the microspheres. Higher EE values are desirable as they indicate minimal drug loss during the encapsulation process. Among the formulations, CVF-6 exhibits the highest encapsulation efficiency (96.65% ± 0.50), suggesting that this formulation process was the most effective in encapsulating the drug with minimal loss. CVF-1 and CVF-2 also demonstrate high encapsulation efficiencies of over 95%, indicating efficient drug encapsulation. Formulations CVF-3, CVF-4, and CVF-5 show slightly lower EE values, with CVF-4 having the lowest at 93.29%. These variations could be attributed to differences in formulation parameters such as polymer concentration, stirring rate, and solvent evaporation techniques.

Drug loading capacity reflects the amount of drug encapsulated within the microspheres relative to the total weight of the microspheres. Higher LC values indicate a higher concentration of drug per unit weight of microsphere. Formulation CVF-4 shows the highest drug loading capacity (45.76 ± 0.47), indicating that it contains the highest amount of drug per unit weight of microsphere. CVF-3 also has a high drug loading capacity (45.32 ± 0.44), followed closely by CVF-2 and CVF-6, both with LC values above 44. The formulations CVF-1 and CVF-5 have the lowest drug loading capacities at 42.87 ± 0.48. These differences in LC values could result from variations in the formulation process, such as the amount of polymer used, the drug-to-polymer ratio, and the efficiency of the encapsulation process. The encapsulation efficiency and drug loading capacity are crucial for optimizing the drug delivery system. Formulation CVF-6 appears to be the most efficient with the highest encapsulation efficiency and a high drug loading capacity. CVF-4, despite having the highest drug loading capacity, shows a slightly lower encapsulation efficiency compared to CVF-6. These results suggest that fine-tuning the formulation parameters can lead to an optimized drug delivery

system with high encapsulation efficiency and drug loading capacity, enhancing the therapeutic effectiveness and reducing dosing frequency (Table 2 and Figure 1).

**Table 2.** Encapsulation efficiency and drug loading capacity.

| Formulation Code | Drug Loading Capacity (LC) | Encapsulation Efficiency (EE) |
|---|---|---|
| CVF-1 | 42.87 ± 0.48 | 95.92 ± 0.53 |
| CVF-2 | 44.32 ± 0.45 | 95.34 ± 0.58 |
| CVF-3 | 45.32 ± 0.44 | 94.66 ± 0.42 |
| CVF-4 | 45.76 ± 0.47 | 93.29 ± 0.52 |
| CVF-5 | 42.87 ± 0.48 | 93.92 ± 0.42 |
| CVF-6 | 44.32 ± 0.60 | 96.65 ± 0.50 |

Where LC= Loading capacity, EE= Encapsulation efficiency

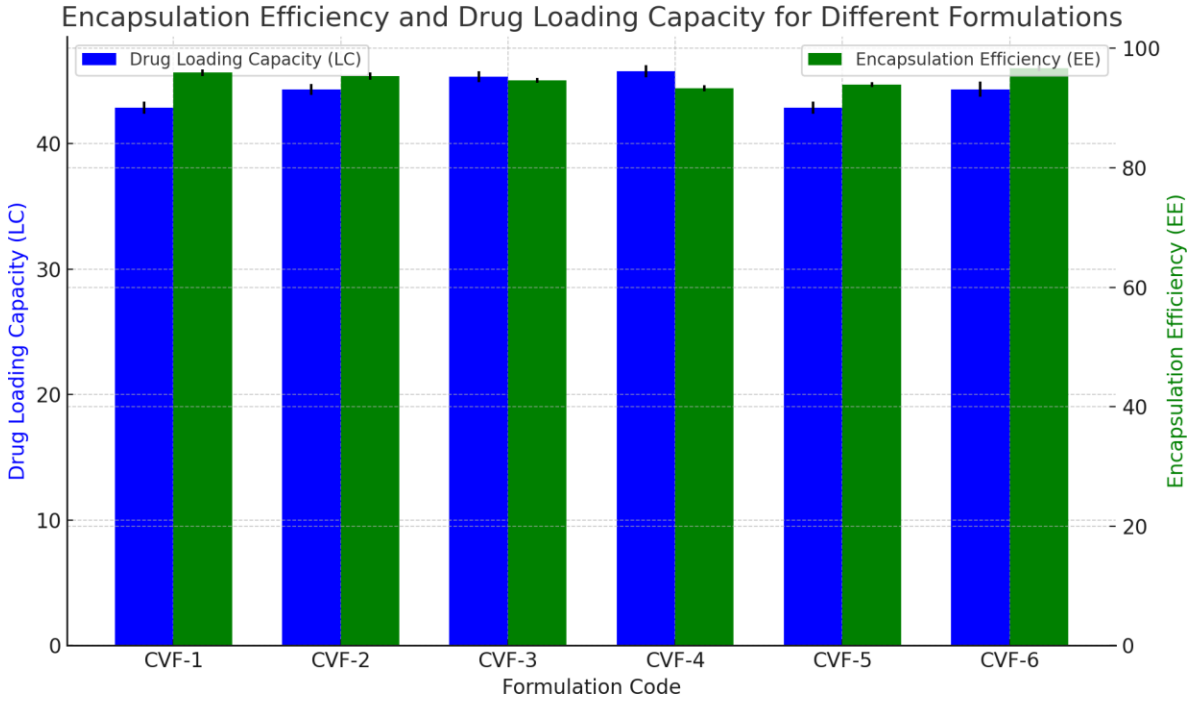


**Figure 1.** Depicting the Encapsulation efficiency and drug loading capacity.

***Characterization of the formulated microspheres***

***Scanning Electron Microscopy (SEM) & Particle size analysis***

The SEM images in figure 2 showcase the morphology of mucoadhesive microspheres. The left image reveals microspheres with a smooth, spherical shape, indicating a uniform and controlled fabrication process. The smooth surface suggests that the microspheres have a consistent coating, which is beneficial for controlled drug release and enhanced mucoadhesion properties. The spherical shape is ideal for gastric retention as it facilitates smoother interaction with the gastric mucosa, promoting prolonged adherence and sustained drug release. In contrast, the right image displays a microsphere with an irregular surface and rough texture. This could be due to any process-related factors such as solvent evaporation, polymer concentration etc. The irregularities and rough surface may affect the release profile, potentially leading to an initial burst release followed by a sustained release phase. Additionally, the rough surface could enhance mucoadhesion by providing more contact points for interaction with the mucosal layer. Overall, the images highlight the importance of optimizing fabrication

parameters to achieve the desired microsphere morphology, ensuring consistent drug release and effective mucoadhesion.

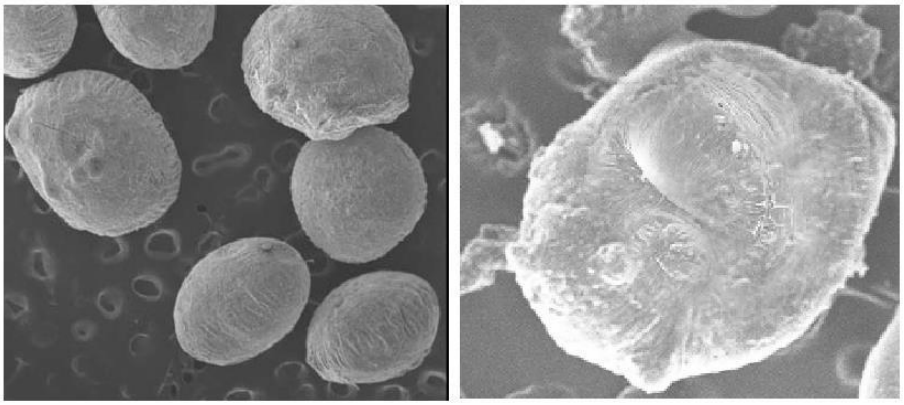

**Figure 2.** SEM Photograph of formulation.

The particle sizes range from 5.22 µm to 7.25 µm. CVF-1 has the largest average particle size (7.25 ± 0.42 µm), while CVF-5 has the smallest (5.22 ± 0.42 µm). The variations in particle size could impact the drug release rate and mucoadhesion properties, with larger particles potentially providing a slower release and enhanced mucoadhesion due to increased surface area. The formulations exhibit both spherical and non-spherical shapes. CVF-1, CVF-4, and CVF-5 are spherical, while CVF-2, CVF-3, and CVF-6 are non-spherical. Spherical particles generally have better flow properties and uniform drug release profiles compared to non-spherical ones. The non-spherical shapes might indicate variations in the manufacturing process. The differences in particle size and shape suggest that the manufacturing process needs optimization to achieve consistent particle morphology. Spherical microspheres like CVF-1, CVF-4, and CVF-5 are preferable for their predictable behaviour in drug release and mucoadhesion. In contrast, the non-spherical shapes in CVF-2, CVF-3, and CVF-6 might lead to irregular drug release profiles, potentially affecting the therapeutic efficacy. Understanding these variations is crucial for optimizing formulation parameters to ensure consistent and effective drug delivery systems.

**Table 3.** Particle size and shape of microspheres

| Formulation Code | Particle size (µm) ± SD | Shape |
|---|---|---|
| CVF-1 | 7.25 ± 0.42 | Spherical |
| CVF-2 | 6.32 ± 0.44 | Non spherical |
| CVF-3 | 6.29 ± 0.41 | Non spherical |
| CVF-4 | 7.16 ± 0.39 | Spherical |
| CVF-5 | 5.22 ± 0.42 | Spherical |
| CVF-6 | 6.82 ± 0.41 | Non spherical |

***Yield percentage and Swelling index***

The yield percentages range from 84.45% to 93.57%. CVF-2 and CVF-6 have the highest yields (93.57% ± 3.42), indicating an efficient fabrication process with minimal material loss. Conversely, CVF-3 has the lowest yield (84.45% ± 3.54), suggesting potential areas for process improvement to reduce material wastage. The swelling index measures the ability of the microspheres to absorb water, which is crucial for their mucoadhesive and drug release properties. CVF-1, CVF-3, and CVF-5 show high swelling indices (1.04 ± 0.0021, 1.02 ± 0.0021, and 1.03 ± 0.0021, respectively), indicating a high capacity for water uptake. In contrast, CVF-2, CVF-4, and CVF-6 have significantly lower swelling indices (0.29 ± 0.0011,

0.30 ± 0.0011, and 0.28 ± 0.0011, respectively), which may affect their mucoadhesive properties and the rate of drug release.

The formulations with higher swelling indices (CVF-1, CVF-3, and CVF-5) are likely to exhibit better mucoadhesion and sustained drug release profiles due to their greater water absorption capacity. However, the formulations with lower swelling indices (CVF-2, CVF-4, and CVF-6), despite their high yield percentages, may require modifications to enhance their swelling properties. Balancing the yield percentage with an optimal swelling index is crucial for developing effective mucoadhesive drug delivery systems. This data highlights the need for process optimization to achieve both high yield and desirable swelling characteristics.

**Table 4.** Swelling Index and yield percentage

| Formulation Code | Yield % | Swelling Index |
|---|---|---|
| CVF-1 | 86.53 ± 2.97 | 1.04 ± 0.0021 |
| CVF-2 | 93.57 ± 3.42 | 0.29 ± 0.0011 |
| CVF-3 | 84.45 ± 3.54 | 1.02 ± 0.0021 |
| CVF-4 | 85.56 ± 3.84 | 0.30 ± 0.0011 |
| CVF-5 | 86.53 ± 2.97 | 1.03 ± 0.0021 |
| CVF-6 | 93.57 ± 3.42 | 0.28 ± 0.0011 |

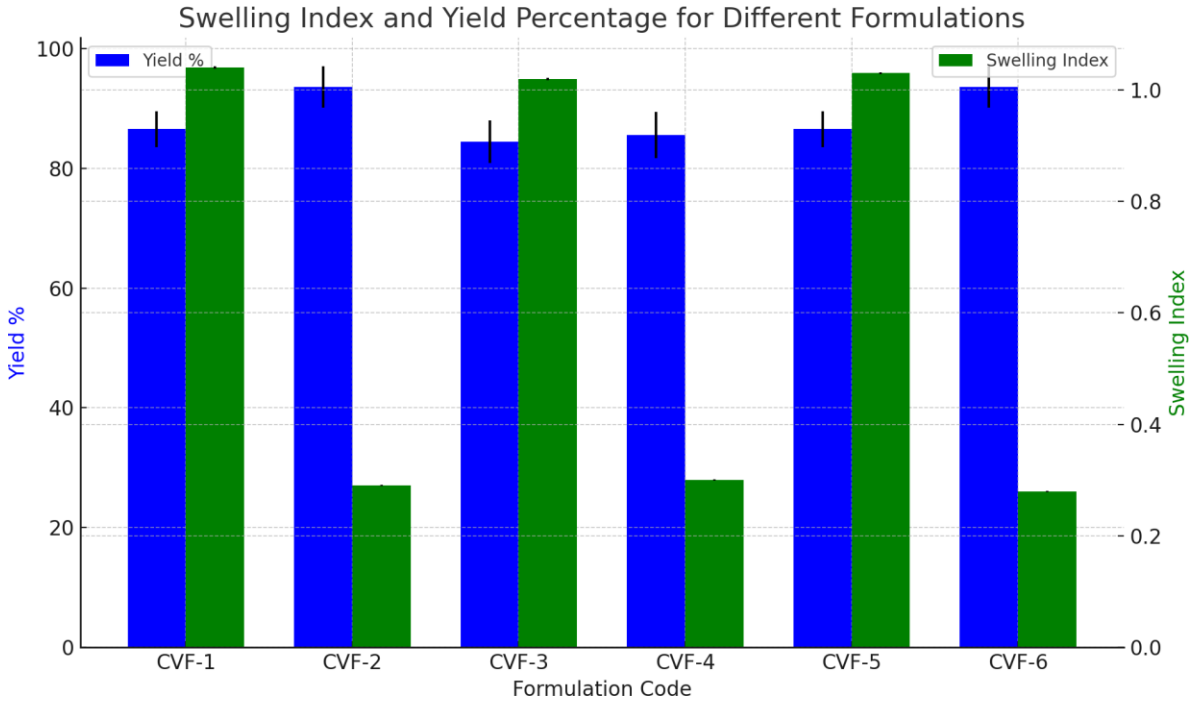


**Figure 3.** Depicting the comparison of Swelling Index and yield percentage

***Mucoadhesive property***

Table 5 presents the mucoadhesive properties of various formulations by measuring the percentage of mucoadhesion at the 10-hour mark. This parameter indicates the ability of the microspheres to adhere to the mucosal surface, which is crucial for prolonged drug release and retention in the gastric environment. Among the formulations, CVF-6 demonstrates the highest mucoadhesion percentage (79.43% ± 1.11), suggesting superior adhesive properties that could lead to more effective and sustained drug release in the gastrointestinal tract. CVF-2 also shows high mucoadhesion (78.82% ± 1.10), making it another strong candidate for prolonged drug retention. On the other hand, CVF-4 has the lowest mucoadhesion percentage (69.76% ± 1.09),

which might indicate a less effective interaction with the mucosal surface and potentially shorter retention time. The variations in mucoadhesive properties can be attributed to differences in polymer composition, particle size, and surface characteristics of the microspheres. Formulations CVF-2 and CVF-6, with their high mucoadhesion percentages, are likely to offer better therapeutic outcomes by ensuring longer retention and sustained drug release. In contrast, formulations with lower mucoadhesion percentages, such as CVF-4, may require further optimization to enhance their adhesive properties. Overall, understanding the mucoadhesive behaviour is essential for developing effective gastroretentive drug delivery systems (Table 5 and Figure 4).

**Table 5.** Mucoadhesive property

| **Formulation code** | **% Mucoadhesion at 10 hour** |
|---|---|
| CVF-1 | 73.91±1.02 |
| CVF-2 | 78.82±1.10 |
| CVF-3 | 72.63±1.01 |
| CVF-4 | 69.76±1.09 |
| CVF-5 | 71.91±1.07 |
| CVF-6 | 79.43±1.11 |

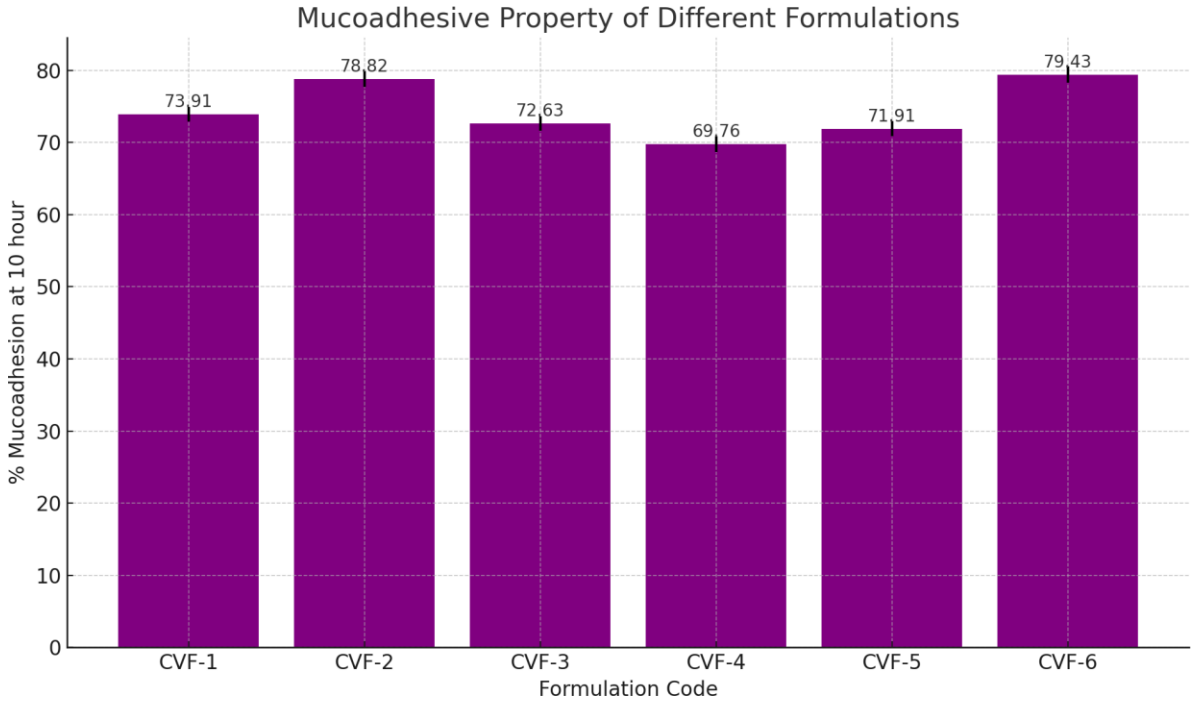


**Figure 4.** Percentage Mucoadhesion at 10 hours for different formulations

***In vitro drug release***

The results of the in vitro drug release study for various formulations (CVF-1 to CVF-6) at pH 1.2 over a 12-hour period reveal significant insights into their release profiles. At the 1-hour mark, the initial drug release percentages range from 17.01% for CVF-6 to 20.41% for CVF-1, with CVF-1 showing the highest initial release, indicating a faster initial dissolution rate. As time progresses to 2 hours, the drug release increases, with values ranging from 27.26% for CVF-6 to 31.98% for CVF-1, maintaining the trend of CVF-1 having the highest release percentage. By the 3-hour point, drug release percentages span from 32.34% for CVF-3 to 36.73% for CVF-1, continuing with CVF-1 showing the highest release. At 4 hours, the drug

release ranges from 37.16% for CVF-6 to 39.84% for CVF-1. The trend of CVF-1 leading in release percentages is consistent up to 5 hours, where release percentages vary more significantly, from 40.59% for CVF-6 to 49.16% for CVF-1. At 6 hours, drug release percentages range from 50.14% for CVF-6 to 57.83% for CVF-1, and at 7 hours, from 60.16% for CVF-6 to 62.72% for CVF-1. By 8 hours, the release ranges from 63.70% for CVF-6 to 67.30% for CVF-1. At 9 hours, drug release continues to increase, ranging from 69.05% for CVF-6 to 72.73% for CVF-1. By 10 hours, the release percentages span from 75.28% for CVF-6 to 80.41% for CVF-1, and at 11 hours, from 84.61% for CVF-6 to 88.84% for CVF-1. Finally, at 12 hours, the drug release percentages range from 93.59% for CVF-6 to 99.84% for CVF-1, with CVF-1 nearing complete release. In summary, CVF-1 consistently demonstrates the highest drug release at each time point, indicating a faster and more extensive release profile, which suggests that CVF-1 may have a formulation that allows for quicker drug dissolution and diffusion. CVF-2 and CVF-6 also show relatively high release percentages but are consistently lower than CVF-1, maintaining a steady release pattern beneficial for sustained drug delivery. CVF-3, CVF-4, and CVF-5 show lower drug release percentages, indicating a slower release mechanism, potentially due to differences in polymer composition or microsphere structure. Thus, CVF-1 is most effective for rapid drug release, making it suitable for applications requiring quick onset of action, while CVF-2 and CVF-6, with their steady and sustained release, are preferable for prolonged therapeutic effects. Understanding these release profiles is crucial for optimizing formulations to meet specific therapeutic needs, ensuring the drug is released at the desired rate and duration.

**Table 6.** Results of In vitro drug release study at pH 1.2

| **Time** | **CVF-1** | **CVF-2** | **CVF-3** | **CVF-4** | **CVF-5** | **CVF-6** |
|---|---|---|---|---|---|---|
| 1hr | 20.41 ± 0.45 | 18.05 ± 0.40 | 17.02 ± 0.35 | 17.20 ± 0.32 | 17.76 ± 0.37 | 17.01 ± 0.34 |
| 2hr | 31.98 ± 0.57 | 29.62 ± 0.50 | 28.65 ± 0.48 | 27.45 ± 0.42 | 28.01 ± 0.45 | 27.26 ± 0.44 |
| 3hr | 36.73 ± 0.62 | 33.95 ± 0.55 | 32.34 ± 0.53 | 32.55 ± 0.52 | 33.12 ± 0.56 | 32.37 ± 0.51 |
| 4hr | 39.84 ± 0.64 | 38.48 ± 0.60 | 38.12 ± 0.58 | 37.34 ± 0.56 | 37.91 ± 0.59 | 37.16 ± 0.54 |
| 5hr | 49.16 ± 0.75 | 46.95 ± 0.69 | 41.52 ± 0.62 | 40.77 ± 0.61 | 41.34 ± 0.64 | 40.59 ± 0.60 |
| 6hr | 57.83 ± 0.84 | 55.93 ± 0.80 | 54.00 ± 0.77 | 50.33 ± 0.71 | 50.89 ± 0.73 | 50.14 ± 0.70 |
| 7hr | 62.72 ± 0.91 | 61.27 ± 0.87 | 61.00 ± 0.86 | 60.36 ± 0.84 | 60.91 ± 0.85 | 60.16 ± 0.82 |
| 8hr | 67.30 ± 0.98 | 66.08 ± 0.96 | 66.12 ± 0.97 | 63.90 ± 0.92 | 64.45 ± 0.94 | 63.70 ± 0.91 |
| 9hr | 72.73 ± 1.04 | 70.06 ± 1.01 | 69.25 ± 1.00 | 69.23 ± 0.99 | 69.80 ± 1.01 | 69.05 ± 0.98 |
| 10hr | 80.41 ± 1.15 | 78.61 ± 1.12 | 75.58 ± 1.07 | 75.48 ± 1.06 | 76.03 ± 1.08 | 75.28 ± 1.05 |
| 11hr | 88.84 ± 1.27 | 86.06 ± 1.23 | 84.70 ± 1.21 | 84.80 ± 1.22 | 85.36 ± 1.24 | 84.61 ± 1.20 |
| 12hr | 99.84 ± 1.42 | 97.06 ± 1.38 | 95.23 ± 1.35 | 93.78 ± 1.33 | 94.34 ± 1.34 | 93.59 ± 1.32 |

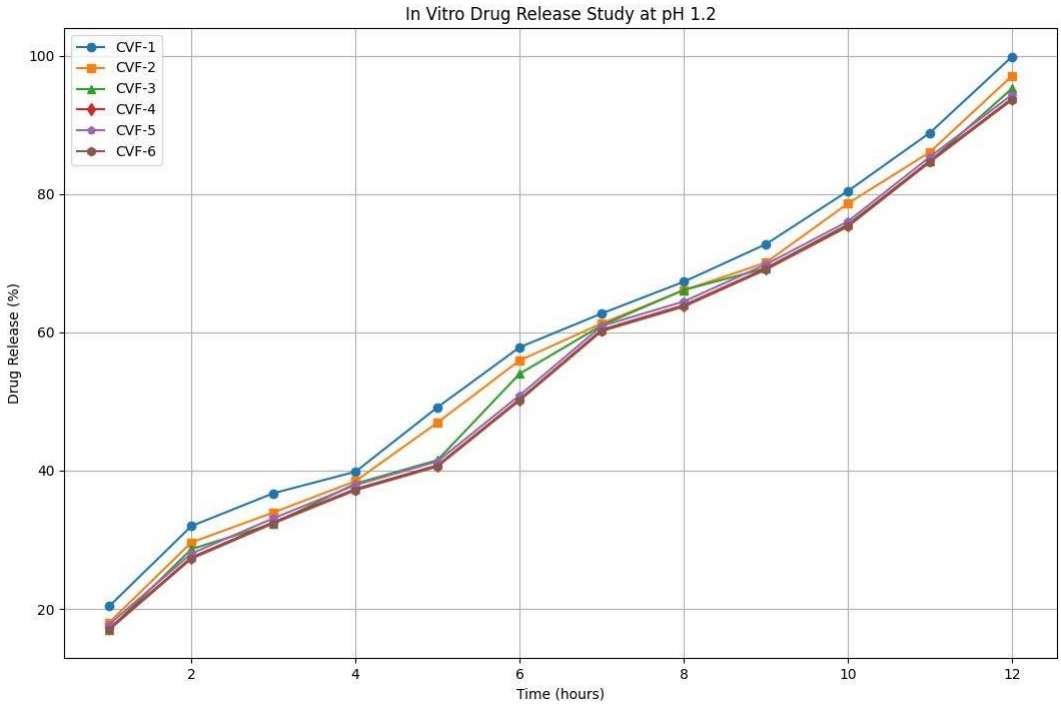


**Figure 5.** In vitro drug release at pH 1.2

**Kinetic Modelling**

Table 7 provides a detailed analysis of the kinetic modelling results for drug release data across six formulations (CVF-1 to CVF-6), highlighting the effectiveness of different release mechanisms. The Zero-order, First-order, Higuchi, and Korsmeyer-Peppas models offer insights into the release dynamics, with R-squared values indicating the fitness of each model to the actual data. In the Zero-order model, formulations like CVF-1 and CVF-4 demonstrate high R-squared values close to 0.992, suggesting a consistent and uniform drug release rate that is independent of the concentration. The First-order model, indicating release dependent on the drug concentration, also shows high R-squared values for these formulations, especially for CVF-4 and CVF-6, suggesting that the rate of release decreases over time as the drug concentration decreases. The Higuchi model, which describes release from an insoluble matrix as a function of the square root of time, shows slightly lower R-squared values across all formulations. This indicates that while diffusion is a significant release mechanism, it may not be the only one governing the release process. The Korsmeyer-Peppas model, crucial for understanding complex release mechanisms from polymeric systems, shows very high R-squared values for all formulations, particularly CVF-2 and CVF-6. This suggests that the release mechanisms are not purely Fickian but likely involve a combination of diffusion and erosion processes. The n-values greater than 0.5 in the Korsmeyer-Peppas model for all formulations suggest anomalous (non-Fickian) transport, where both the matrix swelling, and drug diffusion contribute to the drug release. Overall, the formulations CVF-2 and CVF-6 stand out with their superior fit in the Korsmeyer-Peppas model, indicating that they may offer the most controlled and extended-release profiles. These formulations are well-suited for therapeutic applications that require prolonged drug administration, making them optimal choices based on the kinetic modelling.

**Table 7.** Kinetic modelling of the drug release data

| Formulation | Zero-order R-squared | First-order R-squared | Higuchi R-squared | Korsmeyer-Peppas Parameters | Korsmeyer-Peppas R-squared |
|---|---|---|---|---|---|
| CVF-1 | 0.9911 | 0.9558 | 0.9271 | kK=16.55, n=0.70 | 0.9799 |
| CVF-2 | 0.9922 | 0.9676 | 0.9206 | kK=15.12, n=0.72 | 0.9849 |
| CVF-3 | 0.9896 | 0.9681 | 0.9085 | kK=14.03, n=0.75 | 0.9824 |
| CVF-4 | 0.9923 | 0.9691 | 0.9026 | kK=13.35, n=0.76 | 0.9831 |
| CVF-5 | 0.9924 | 0.9672 | 0.9061 | kK=13.76, n=0.75 | 0.9825 |
| CVF-6 | 0.9924 | 0.9697 | 0.9015 | kK=13.22, n=0.77 | 0.9833 |

**Optimum Formulation**

Based on the comprehensive analysis of all parameters, **CVF-2** appears to be the most optimal formulation. It combines high drug loading capacity, high encapsulation efficiency, excellent mucoadhesion, and a steady, near-complete drug release profile. CVF-2 demonstrates a balanced performance across all critical attributes, making it highly suitable for effective and sustained drug delivery. While CVF-1 shows the highest drug release, its mucoadhesion is slightly lower than CVF-2 and CVF-6. CVF-6 has excellent mucoadhesion and encapsulation efficiency, but a slightly lower drug release compared to CVF-2. Therefore, considering all factors, CVF-2 stands out as the best formulation overall.

## CONCLUSIONS

This study clearly demonstrated the successful fabrication and development of mucoadhesive gastroretentive microspheres for prolonged delivery of carvedilol. The findings from this investigation demonstrated the intricate balance between formulation parameters that dictate the performance of mucoadhesive gastroretentive drug delivery systems. CVF-2 emerged as the most optimal formulation, excelling in encapsulation efficiency, drug loading capacity, and mucoadhesion, coupled with a robust and controlled drug release profile as evidenced by kinetic modelling. This formulation not only ensured minimal drug loss during encapsulation but also provided sustained drug release, making it ideal for therapeutic applications requiring prolonged drug action. CVF-6 also showed promise with excellent mucoadhesion and encapsulation efficiency, suggesting its suitability for applications necessitating extended gastric retention. The study highlighted the necessity for meticulous optimization of formulation parameters to enhance therapeutic effectiveness and patient compliance. Future research should focus on refining and optimizing these formulations to further enhance their efficacy and utility in clinical settings, emphasizing the development of mucoadhesive drug delivery systems that can efficiently deliver therapeutic agents in a controlled and prolonged manner.

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