Research Paper

Preparation and Characterization of Gastroretentive Floating Microspheres of Ofloxacin Hydrochloride

Mona Semalty*, Shikha Yadav and Ajay Semalty
Department of Pharmaceutical Sciences, H.N.B. Garhwal University, India.

ABSTRACT: As Ofloxacin is preferably absorbed from the upper part of the gastrointestinal tract and is readily soluble in the acidic environment of the stomach, the floating microspheres of ofloxacin were formulated to develop gastroretentive formulation. These floating microspheres release the drug in the stomach and upper gastrointestinal tract and thereby improve the bioavailability. In the present study, six formulations of ofloxacin hydrochloride were prepared as floating microspheres by solvent diffusion technique using polymers such as ethyl cellulose, polyvinyl pyrrolidone K-90 and poly vinyl alcohol in different ratios. The prepared microspheres were evaluated for different physicochemical tests such as particle size, percent drug entrapment, drug content uniformity, SEM, buoyancy test, and in vitro drug release studies. The results of all the physicochemical tests of all formulations were found to be satisfactory. In vitro floatability studies revealed that most of the microspheres (52.5% to 95.5%) were floatable. The in vitro drug release was found to be in the range of 39.64 to 93.64 % at the end of 6 hours. It is concluded that these floating microspheres can be selected for the development of gastroretentive drug delivery system of ofloxacin hydrochloride for potential therapeutic uses.

KEYWORDS: Gastroretentive Floating Microspheres; Ofloxacin hydrochloride

Introduction
Oral drug delivery is the most used and preferred route of administration with the obvious advantage of ease of administration and patient acceptance. To develop a drug delivery system for oral administration, it is necessary to optimize not only the release rate of an active ingredient from the system but also the residence time of the system in the gastrointestinal tract (Lee et al., 1999).

Gastroretentive drug delivery is an approach to prolong gastric residence time, thereby targeting site-specific drug release in upper gastrointestinal (GI) tract. A gastroretentive dosage form (GRDF) releases medications in a controlled manner for extending the absorption phase of drugs which show a limited and narrow absorption window at the upper part of the gastrointestinal tract or drugs intended to treat local ailments in the gastroduodenum. This mode of administration may prolong the time period in which the blood drug concentrations are within the “therapeutic levels” and improve therapy. Besides being locally active in the stomach, these extended-release dosage forms with prolonged residence time in the stomach are also highly desirable for drugs that are unstable in the intestinal or colonic environment, and/or have low solubility at higher pH values (Streubel et al., 2003). Therefore, development of GRDFs has been a major pharmaceutical challenge during the past few decades (Eytan et al., 2003).

The gastroretentive dosage forms (GRDFs) has been designed in large part based on the following approaches (Chavanpatil et al., 2005): (a) low density form of the DF that causes buoyancy above gastric fluid; (b) high density DF that is retained in the bottom of the stomach; (c) bioadhesion to the stomach mucosa; (d) slowed motility of the gastrointestinal tract by concomitant administration of drugs or pharmaceutical excipients; (e) expansion by swelling or unfolding to a large size which limits emptying of the DF through the pyloric sphincter.

Floating drug delivery systems (FDDS) have a bulk density less than gastric fluid and therefore remain floating in the stomach without affecting gastric emptying rate for prolonged period. The drug is slowly released at the desired rate from the floating system. After release of drug, the residual system is expelled from the stomach. These floating dosage forms may have a number of advantages in oral drug delivery because they prolong retention in the gastrointestinal tract, particularly in the stomach. Gastroretentive delivery system facilitate sustained drug release and maintain high concentrations of drug within the gastric mucosa. This property may also be performed for treatment of Helicobacter pylori infection (Bardonnet et al., 2006; Blaser, 1992).
Floating delivery systems administered in a single-unit form [such as hydrodynamically balanced system (HBS)] are unreliable in prolonging the GRT owing to their ‘all-or-nothing’ emptying process and, thus, they may cause high variability in bioavailability and local irritation due to large amount of drug delivered at a particular site of the gastrointestinal tract (GIT). In contrast, multiple unit dosage forms (e.g. microspheres) enjoy the advantage since they pass uniformly through the GIT to avoid the vagaries of gastric emptying and provide an adjustable release, thereby, reducing the intersubject variability in absorption and risk of local irritation (Kawashima et al., 1991).

Ofloxacin hydrochloride is an anti-infective drug, used mainly in the treatment of lower respiratory infections, skin infection, urinary tract infections, and sexually transmitted diseases (except syphilis). Ofloxacin has broad activity against bacterial (Helicobacter pylori) infections and is used in combination with other drugs to treat tuberculosis. The bioavailability of ofloxacin is strongly dependent on the local physiology in the GI tract. Ofloxacin is preferably absorbed from the upper part of the gastrointestinal tract. Ofloxacin is readily soluble in the acidic environment of the stomach. In the intestine, where neutral to slightly alkaline pH conditions prevail, precipitation of the active compound occurs, which adversely affects absorption in the lower sections of the intestine. Therefore there is a need for systems that reside in the stomach over a relatively long time and release the drug there in a sustained manner (Sen and Kshirsagar, 2002). This can be achieved by the design and development of sustained release gastroretentive floating drug delivery system for ofloxacin (using suitable polymers) which would float and deliver the drug in the upper part of GIT in a sustained manner. Earlier gastroretentive drug delivery system for ofloxacin had been formulated as floating tablets (Chavanpatil et al., 2005).

The present work deals with the formulation and characterization of floating microspheres of ofloxacin hydrochloride using ethyl cellulose, polyvinyl pyrrolidone K-90 (PVP K-90) and poly vinyl alcohol.

**Materials and Methods**

Ofloxacin hydrochloride was obtained as a gift sample (Intas Pharmaceutical Ltd. Dehradun). Ethyl cellulose (EC), polyvinyl pyrrolidone K-90 (PVP K-90) and poly vinyl alcohol (PVA) were procured from Central Drug House, Mumbai. Other chemicals used were of analytical grade.

**General Method for Preparation of Floating microspheres**

Floating microspheres containing ofloxacin hydrochloride were prepared by an emulsification solvent evaporation technique (Soppimath et al., 2001) using EC, PVP K-90 and PVA polymers for microspheres (Table 1). Ofloxacin (500 mg) and polymer (500 mg) were weighed accurately and dissolved in 8ml ethanol, followed by the addition of 2ml isopropanol and 5 ml dichloromethane. The polymer solution was slowly introduced into 100ml of 1% poly (vinyl alcohol) aqueous solution while stirring at 250rpm using a mechanical stirrer equipped with a 3 blade propeller. The solution was stirred for 10min and the microspheres were collected by filtration. The floating microspheres were collected by decantation, while the non-floating microspheres were discarded along with any polymer precipitates. The microspheres were dried in an oven at 50 °C for 2h, weighed and then stored in a desiccator at room temperature till further use.

<table>
<thead>
<tr>
<th>Ingredient</th>
<th>F1</th>
<th>F2</th>
<th>F3</th>
<th>F4</th>
<th>F5</th>
<th>F6</th>
</tr>
</thead>
<tbody>
<tr>
<td>Drug</td>
<td>0.5%</td>
<td>0.5%</td>
<td>0.5%</td>
<td>0.5%</td>
<td>0.5%</td>
<td>0.5%</td>
</tr>
<tr>
<td>EC</td>
<td>0.5%</td>
<td>1%</td>
<td>1.5%</td>
<td>0.5%</td>
<td>1%</td>
<td>1%</td>
</tr>
<tr>
<td>PVP</td>
<td>------</td>
<td>------</td>
<td>------</td>
<td>1%</td>
<td>0.5%</td>
<td>1%</td>
</tr>
<tr>
<td>PVA</td>
<td>1%</td>
<td>1%</td>
<td>1%</td>
<td>1%</td>
<td>1%</td>
<td>1%</td>
</tr>
</tbody>
</table>

EC- Ethyl cellulose, PVP- Poly vinyl pyrrolidine, PVA- Poly (vinyl alcohol).

**Evaluation of floating microspheres**

**Scanning electron microscopy**

To detect the surface morphology of the microspheres, SEM of the floating microspheres was performed at IIT Roorkee by Scanning electron Microscope (Jeol of Japan Model No.5600).

**True Density**

The microspheres were immersed in 0.02% tween 80 solutions for three days in a metal mesh basket. The submerged microspheres were used for density measurements. True density of floating microspheres was determined by liquid displacement method using relative density bottle.

**Particle size**

Particle Size of floating microspheres was performed with the help of optical microscope for randomly selected samples of all the formulation (Swarbrick and Martin, 1996).
Percent Drug Loading

Ofloxacin Hydrochloride content in the microspheres was estimated by a UV Spectrophotometric (Lambda 25, Perkin Elmer, US) method based on the measurement of absorbance at 294 nm in distilled water (Semalty and Semalty, 2007). Microspheres equivalent to 100 mg were weighed and added in 100 ml of distilled water. The volumetric flask was stirred continuously for 24 h on a magnetic stirrer. At the end of 24 h sample was withdrawn, diluted suitable and measured spectrophotometrically at 294 nm for the drug content. Quantitative estimation of ofloxacin hydrochloride was calculated by using equation obtained by linear regression analysis of the calibration curve of the drug in distilled water. The Drug Loading in microspheres was estimated using the formula, Percent Drug Loading (L) = (Qm/Wm)\times 100, where Wm is the weight of microspheres and the Qm is the quantity of drug present in Wm of microspheres (Semalty and Semalty, 2008). Percent Drug Loading of various formulations is shown in Table 2.

In vitro floatability studies

In vitro floatability studies on floating microspheres were carried out using USP apparatus II. To assess the floating properties, the microspheres were placed in 0.1 N hydrochloric acid containing 0.02 % v/v Tween 80 surfactant to gastric conditions. Tween (0.02% v/v) was used to impart wetting effect of the natural surfactants such as phospholipids in the GIT. The buoyancy was calculated by: Buoyancy (%) = Wf / (Wf + Ws) 100; Where Wf and Ws are weights of the floating and the settled microspheres, respectively (Kale et al., 2001).

Table 2 Physical evaluation of floating microspheres of Ofloxacin.

<table>
<thead>
<tr>
<th>Formulation Code</th>
<th>Average particle size (µm)</th>
<th>True Density</th>
<th>Percent Buoyancy</th>
<th>Percent Drug loading</th>
<th>Drug release after 6 hr</th>
</tr>
</thead>
<tbody>
<tr>
<td>F1</td>
<td>340.30 ± 2.608</td>
<td>0.789±0.003</td>
<td>95.50±1.46</td>
<td>86.56±1.45</td>
<td>70.67 ± 1.24</td>
</tr>
<tr>
<td>F2</td>
<td>367.25 ± 3.917</td>
<td>0.779±0.002</td>
<td>91.50±1.84</td>
<td>51.82±2.30</td>
<td>93.64 ± 3.05</td>
</tr>
<tr>
<td>F3</td>
<td>388.50 ± 4.142</td>
<td>0.806±0.011</td>
<td>92.50±0.098</td>
<td>53.94±2.92</td>
<td>79.13±1.44</td>
</tr>
<tr>
<td>F4</td>
<td>250.80 ± 0.364</td>
<td>0.812±0.010</td>
<td>59.50±1.54</td>
<td>93.80±2.40</td>
<td>67.25 ± 1.67</td>
</tr>
<tr>
<td>F5</td>
<td>254.07 ±1.856</td>
<td>0.774±0.006</td>
<td>52.51±1.42</td>
<td>39.72±3.30</td>
<td>39.64±1.09</td>
</tr>
<tr>
<td>F6</td>
<td>278.50 ± 2.588</td>
<td>0.803±0.002</td>
<td>61.51±2.68</td>
<td>75.60±3.50</td>
<td>58.66±1.07</td>
</tr>
</tbody>
</table>

Results are presented as mean ± standard deviation (n=3).

In vitro Drug Release Study

The release study was carried out in a USP 24 dissolution apparatus type 1 (six-station dissolution apparatus, Veego 6DR, India), slightly modified in order to overcome the small volume of the dissolution medium. The dissolution medium was 100 mL IPB, pH 1.2 maintained at 37±0.5 °C and kept in a glass beaker fixed inside the USP dissolution flask. Microspheres equivalents to 100 mg of ofloxacin were filled in empty capsule shells. One capsule was used in each test and placed in the basket, which was rotated at 50 rpm. Filtered samples (5 mL) were manually collected at different intervals. The samples were compensated with an equal volume.

The concentration of drug released in the medium was assayed spectrophotometrically at 294 nm after suitable dilution with the dissolution medium when necessary. The experiment was carried out in triplicate.

Results and Discussion

The Floating microspheres of ofloxacin were prepared by Emulsion solvent evaporation Method using Ethyl cellulose, Poly vinyl pyrrolidine, and Poly (vinyl alcohol). The prepared Floating microspheres were evaluated for different physicochemical tests such as particle size, true density, flow properties, drug content, in vitro floatability and in vitro drug release studies.

Scanning Electron Microscopy showed that F1, F2, F4, F5, F6 formulation produced spherical microspheres compared to F3 (Fig. 1). The scanning electron microscopy confirmed the hollow nature of microspheres with pores on the surface of floating microspheres, which imparted floating properties to the prepared floating microspheres.
To assess flow properties of prepared floating microspheres micromeritic properties like particle size and true density were determined (Table 2). The densities of floating microspheres were found to be less than the density of gastric fluid, therefore tended to float over gastric fluid (Arora et al., 2005; Semalty and Semalty, 2008). So the prepared microspheres combine the advantages of multiple unit systems and good floating properties. However, like all floating systems, their efficacy is dependent on the presence of enough liquid in the stomach, requiring frequent drinking of water (Hwang et al., 1998).

Particle size analysis of different formulation was done by optical microscopy. The average particle size for ethyl cellulose microspheres was in the range between 340.30 µm and 388.50 µm, while the average particle size for ethyl cellulose and Poly vinyl pyrrolidine microspheres was in the range between 250.80 µm and 278.50 µm. The average particle size of microspheres was found to be increasing with the increase in concentration of the polymer.

Drug content in F1, F2, F3, F4, F5 and F6 Formulation were estimated by UV Spectrophotometric method. Percent loading efficiency were found in the range of 39.72 to 93.80 %. Formulation F4 containing ethyl cellulose (0.5%) and Poly vinyl pyrrolidine (1%) showed maximum percent loading of drug up to 93.80 %. Formulation F3 containing ethyl cellulose (1.5%) showed least percent loading (39.72 %) of drug. The rank order of Percent loading was found to be as followed F4 > F1 > F6 > F3 > F2 > F5.

In vitro floatability studied revealed that most of the microspheres (52.5% to 95.5%) were floatable. Rank order of In vitro floatability was found to be as followed: F1 > F3 > F2 > F6 > F4 > F5.

In-vitro drug release studies of all the formulations were performed in pH 1.2 acidic buffer at 294 nm. Significant difference was observed in the release pattern of Ofloxacin floating microspheres EC, PVP, PVA (Fig. 2). It was found that the drug release from the formulations were distinguishably different for the different polymers used in the formulations. The rank order of drug release after 6hr was found to be 70.67, 93.64, 79.13, 67.25, 39.64 and 58.66 percent of formulation F1, F2, F3, F4, F5 and F6 respectively. Formulation F2 containing ethyl cellulose (1%) showed the maximum release after the 6h.

**Conclusion**

The results of all the physicochemical tests of all formulations were found to be satisfactory. In vitro floatability studies revealed that most of the microspheres (52.5% to 95.5%) were floatable. The in vitro drug release was found to be in the range of 39.64 to 93.64 % at the end of 6 hour. It is concluded that these floating microspheres can be selected for the development of gastroretentive drug delivery system.
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References


