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Release Profile of Lansoprazole Encapsulated in Sodium Alginate Beads

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Abstract: Symptomatic gastroesophageal reflux disease (GERD) affects 11% of the population worldwide. Lansoprazole was the second commercially available proton pump inhibitor, recommended for healing and symptomatic relief of reflux esophagitis. This work presents the encapsulation of lansoprazole in sodium alginate beads, generating a controlled release system of the encapsulated drug. The quantification of lansoprazole contents from the encapsulated raw material was carried out, using the methodology prescribed in the Brazilian Pharmacopoeia 5th edition, with adjustments, the encapsulation efficiency and the release profile. Experimental tests were performed using scanning UV-Vis spectrophotometry. Encapsulation efficiency was 100%, obtained from the measurement of the absorbance of the filtrate. Release profile shows the concentration of lansoprazole in the external environment was 4 μg/mL (16%) immediately after 10 min. Concentration of lansoprazole released by the beads follows a linear dynamic as a function of time, with a maximum release equivalent to 25 μg/mL after 210 min (100%). The rate of release is influenced by pH, being higher by 7.4. Beads subjected to heat treatment have a higher release rate at a temperature of 30 °C when compared to a temperature of 60 °C. Increase in temperature decreases the diameter of the beads. The presence of lansoprazole affects the release curve. The absorption rate of the microbeads with lansoprazole is 0.83, and the absorption rate of the microbeads without lansoprazole is 0.58. Sodium alginate beads have a good controlled release effect on lansoprazole.

Key words: Spectrophotometry, proton pump inhibitor, encapsulation, sodium alginate beads.

1. Introduction

Gastric acidity is an indispensable condition for good digestion of food. It destroys many ingested bacteria, provides the pH necessary for pepsin to begin digesting proteins, and stimulates the flow of bile and pancreatic juice [1]. However, imbalance of basic acids can lead to a series of digestive and pathological disorders, from heartburn to partial ulceration of gastric mucosa [2].

Among the existing gastric diseases, gastroesophageal reflux disease (GERD) has become quite common, especially in the first year after birth, and the prevalence rate in the first five months after birth is about 64% [3]. Consequently, other diseases, such as reflux esophagitis, have caused disorders and discomfort to the patient. Treatment involves changes in posture, diet, and use of specific medications.

Until the early 1990s, histamine receptor blockers (H2) had been the drugs of choice in the treatment, but from that time, it has been discovered that a slightly higher pH (pH > 3) is required within 16 to 24 hours for the healing of peptic ulcers, making proton pump inhibitors (PPIs) be the first choice in treatment [4].

Gastric acid secretion is inhibited in response to all stimulating agents until new pump molecules are synthesized. The potent action of PPIs, in addition to raising gastric pH, also results in a 24-hour reduction in intragastric volume, facilitating gastric emptying

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and reducing reflux volume [5, 6]. PPIs are unstable in acidic environments so when administered orally they are coated to protect them from stomach degradation, as they are absorbed in the duodenum. For this, they are found in several pharmaceutical forms such as: injectables, enteric coated tablets with prolonged release, fast disintegrating tablets, capsules with granules of enteric coating with normal and prolonged release [6, 7].

Lansoprazole belongs to a class of antisecretory compounds, which do not have the characteristics of anticholinergic or histamine H2 antagonists, but suppress gastric acid secretion by specific inhibition of the H^+/K^+ -ATPase enzyme system on the surface of gastric parietal cells [7]. As the enzyme system is considered as an acidic (proton) pump within the parietal cell, lansoprazole was characterized as an inhibitor of gastric acid pump, blocking the final step in acid production. It is recommended for the treatment of reflux esophagitis, Barrett's ulcer, duodenal ulcer, gastric ulcer, and Zollinger-Ellison syndrome [8].

The encapsulation process was developed approximately 60 years ago and is defined as a technology capable of encapsulating solid, liquid or gaseous materials in small sealed capsules, allowing the release of its contents at specific rates [9]. Several polymers have been used in the encapsulation of drugs, among which stands out, sodium alginate for having a biodegradable and biocompatible nature, with FDA permission for human use, being commonly administered orally for treatment of esophageal reflux [10].

Derived from seaweed, alginate is a linear and anionic polysaccharide used as a thickener, stabilizer and gelling agent in the food and pharmaceutical industry. In molecular terms, alginate is a linear copolymer composed of alternative blocks of α-D-mannuronic acid (M) and α-L-glucuronic acid (G) units joined by glycosidic bonds of type (1-4). Homopolymer blocks of M and G, and their alternating sequence are coexisting in the alginate molecule [11, 12].

Alginate gelling property depends on the following factors: order and composition of the mannuronic and guluronic acid residues, molecular weight of the biopolymer and ion concentration in the sequential solution [13]. Alginates are capable of forming water-insoluble gels by cross-linking with divalent cations such as Ca^{2+}. Due to the smooth gelation process, an aqueous base relatively inert in the matrix and its high biocompatibility, hydrogel systems containing alginate have also been widely used as matrix for microencapsulation of bioactive peptides, proteins and for living cells [13].

This method has been applied to obtain sodium alginate microparticles and was initially developed for the encapsulation of phenols. The method is based on an ionic gel of alginate including encapsulating the material in an alginate solution, and then squeezing the mixture dropwise into the calcium chloride solution through a syringe [14].

One of the properties of alginate is its ability to form irreversible and heat-stable hydrogels when treated with divalent cations. They are also biocompatible, hydrophilic and biodegradable under normal physiological conditions. Therefore, sodium alginate has been used in several technological areas such as food, pharmaceutical, and agricultural industry and for the formation of hybrid materials combined with cationic polymers (such as chitosan) through polymerization or ionic interaction [15].

Considering the great interest in developing new encapsulated pharmaceutical forms to control the release of active ingredients in sodium alginate beads, the aim of this work was to evaluate the efficiency in the development of beads, using the polysaccharide alginate of sodium, providing a new alternative of manipulation and therapeutic application, opening perspectives for innumerable production possibilities. The quantification of levels of lansoprazole from encapsulated raw material, efficiency of encapsulation, release profile and degree of imbibition were carried

out. Experimental tests with scanning UV-VIS spectrophotometry.

2. Materials and Methods

2.1 Materials

Sodium alginate (AG; 200.0 kg/mol) was obtained from Sigma-Aldrich (Steinheim, Germany). Lansoprazole from compounding pharmacies (Rio Verde, Brazil) and previously analyzed for identity and purity was used as a reference standard (purity level 99.8%). Purified water was obtained through the Milli-Q system (Millipore Corporation®). The acetonitrile used was CLAE grade (Tedia®). Other solvents and reagents used were of analytical grade.

Analyzes were performed by UV-Vis spectroscopy using a scanning spectrophotometer (Quimis Q798UV2). The diameter of the beads was determined with the aid of a digital caliper. A high precision analytical balance was used to prepare the solutions.

2.2 Preparation of the Alginate Beads and Encapsulation

The preparation of the alginate beads containing lansoprazole was carried out as follows:

(1) $CaCl_2$ solution 1% w/v (0.09M) (SC): 2 g of anhydrous $CaCl_2$ was dissolved in 200 mL of distilled water and kept in the magnetic stirrer at 300 rpm for 20 min, the pH of this solution being adjusted to 3.5.

(2) Sodium alginate solution 1.5% w/v (AG): prepared by dissolving 0.75 g of sodium alginate in 50 ml of deionized water and pH adjusted to 6.5 with hydrochloric acid previously prepared at 0.1 mol/L. Solution was kept under stirring at 300 rpm at 40 °C for 24 h. Fig. 1 summarizes the preparation of solutions (AG) and (SC).

(3) Lansoprazole encapsulation: 100 μg/mL stock solution of lansoprazole (LZ) was prepared by dissolving 100 mg of the drug in 1 L of acetonitrile, then a solution of Lansoprazole-Alginate (LZ-AG) was prepared at proportion of 1: 4 (1 mL of LZ + 4 mL of AG) respectively. Beads containing encapsulated lansoprazole were obtained by the drip technique with the aid of 5 mL syringes over 200 mL of the solution (SC). BD 1.6 x 40 mm hypodermic needles were used. After gelation of the sodium alginate, the beads were kept under magnetic stirring at 300 rpm for 30 min. Finally, the solution with the beads went through filtration process for specific studies (Fig. 2).

2.3 Standard Calibration Curve

To measure the encapsulated lansoprazole content, a standard curve was obtained according to the methodology described in the Brazilian Pharmacopoeia 5th edition. They were diluted in volumetric flasks of 10 mL capacity containing acetonitrile, aliquots of 0.25; 0.5; 1; 2; 3; 4 and 5 ml lansoprazole solutions previously prepared with the same lansoprazole solvent at 50 μg/mL have purity of 99.9%. Absorbances of the resultingsolutions were obtained at 281 nm,

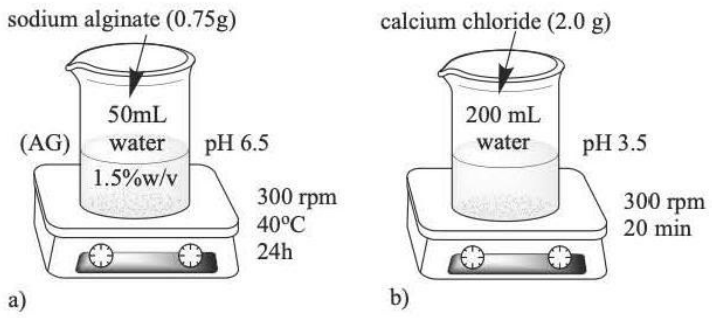

Fig. 1 Preparation of solutions for encapsulating lansoprazole: (a) 1.5% w/v sodium alginate solution (AG) and (b) 1% w/v calcium chloride solution (SC).

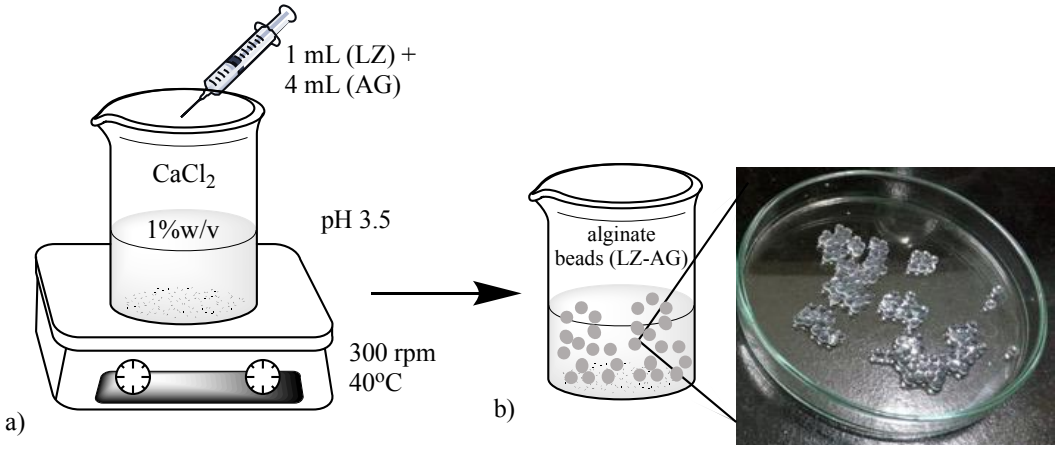

Fig. 2 Lansoprazole encapsulation process: (a) dripping the solution (LZ + AG) onto the solution (SC) and (b) Formation of lansoprazole-containing sodium alginate beads (LZ-AG).

using an acetonitrile solution as white. The calibration curve was prepared by plotting the absorbance as a function of the lansoprazole concentration.

2.4 Release Profile

The release profile of lansoprazole was evaluated under three conditions: (1) Immediately after encapsulation, (2) after drying the beads at room temperature (30 °C) and (3) after drying the beads at 60 °C in an oven. Drying was monitored until beads were constantly weighed.

Lansoprazole was encapsulated as follows: Alginate beads containing lansoprazole were immersed in 3 mL of HCl at pH 1.5 and phosphate buffer at pH 7.4 (release medium). About 50 mg beads containing lansoprazole were placed in the release medium at a concentration of 25 µg/mL and stirred at a speed of 400 rpm. Absorbance readings at 281 nm were performed in triplicate using 3 mL of solution in quartz cuvettes.

2.5 Swelling Properties

The swelling rate (Q) was performed by weighing (up to constant weight) on a precision scale, 50 mg of LZ-AG and AG beads dried at room temperature (30 °C), and placing them in a container with distilled water. At 5 min intervals, the beads were removed from the medium, excess water was removed by placing them on a thin absorbent paper and weighed on an analytical balance. The swelling rate was obtained according to Equation 1 [16].

$$Q = \frac{(Mt - Mo)}{Mo} \quad (1)$$

where Mo is mass of dry beads and Mt is the quality after soaking for 10 minutes.

2.6 Encapsulation Efficiency and Beads Size

The encapsulation efficiency was evaluated by measuring the absorbance of the filtrate at 281 nm. The average size of the beads was obtained by the arithmetic mean of the diameter of 10 beads were randomly selected using ImageJ software.

3. Results and Discussions

Drugs are designed to release their active ingredients quickly when ingested, but some drugs are formulated to have a slow and gradual release, which can control their absorption by the body and prevent the peaks and troughs of the drug concentration in the blood. Therefore, a system capable of releasing drugs in the body in a controlled manner increases the safety of the product and expands its effects.

To evaluate the release profile of lansoprazole encapsulated in sodium alginate beads, the calibration curve was obtained from previously known concentrations of lansoprazole using raw material of high purity content (99.8%) in accordance with the the requirements of Pharmacopoeia Brazilian 5th edition.

All tests were done in triplicate to verify that there is no significant difference between the three absorbance readings to ensure reproducibility and thus to ensure the reliability of the experiment. Eight points were taken with different concentrations of lansoprazole, as shown in Fig. 3.

The regression equation is obtained by the least square method through the calibration curve. The determination coefficient (R^2) was obtained as an important parameter in the calibration and indicates how much of the total variation is common to the elements that make up the analyzed pairs. The value obtained in the curve was 0.9997, which is a good coefficient. In order to evaluate the drug release of lansoprazole, the microspheres containing lansoprazole were first placed in hydrochloric acid with pH 1.5 and phosphate buffer with pH 7.4. The parameters were adjusted according to the law of stomach and intestinal tract, and the initial concentration was 25 μg/mL. Figure 4 shows the release profile of lansoprazole in the three conditions adopted: (1) immediately after encapsulation, (2) drying at room temperature (30 °C) and (3) drying at 60 °C, both in simulated release medium at pH = 7. 4.

In Fig. 4a release profile of lansoprazole is influenced by heat treatment. The curve related to the drying temperature of the beads at 60 °C shows the release rate is much lower when compared to the curve obtained immediately after the encapsulation of lansoprazole. In fig. 4b, the release curve of the drug immediately after encapsulation shows a linear trend (R^2 = 0.991), so the release rate of the drug and the absorption of the drug in the lumen can be better controlled during administration. However, curves obtained after drying the beads show a slow release. When beads passed through drying process, the average diameter significantly reduces what may have been influenced by the thermal effect. Beads diameter according to the drying temperature can be seen in table 1.

According to the study of Sacchetin and his collaborators [17], the particle size obtained with sodium alginate is directly related to the concentration and viscosity of the sodium alginate solution used, and it is observed that low concentrations is conducive to the reduction of particle size.

As can be seen from Table 1, the diameter of the beads decreased significantly under the drying condition of 60 °C. In the drip method, the size of the beads immediately after the drug is encapsulated depends on the diameter of the needle used, which in this work was 1.6 x 40 mm. Zang et al. [18] showed that the particle size of sodium alginate particles coated with berberine changed greatly after drying. Spherical geometry obtained with 1.5% sodium alginate has a smaller proportion of surface area per volume compared to other geometries, which in turn reduces contact between the trapped portions and

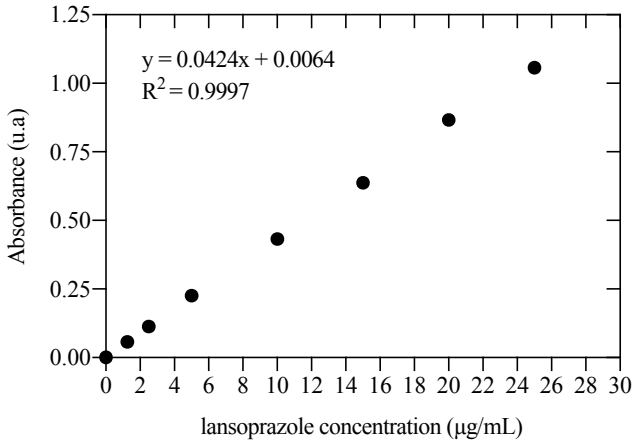

Fig. 3 Calibration curve obtained from lansoprazole with a purity of 99.8% in acetonitrile.

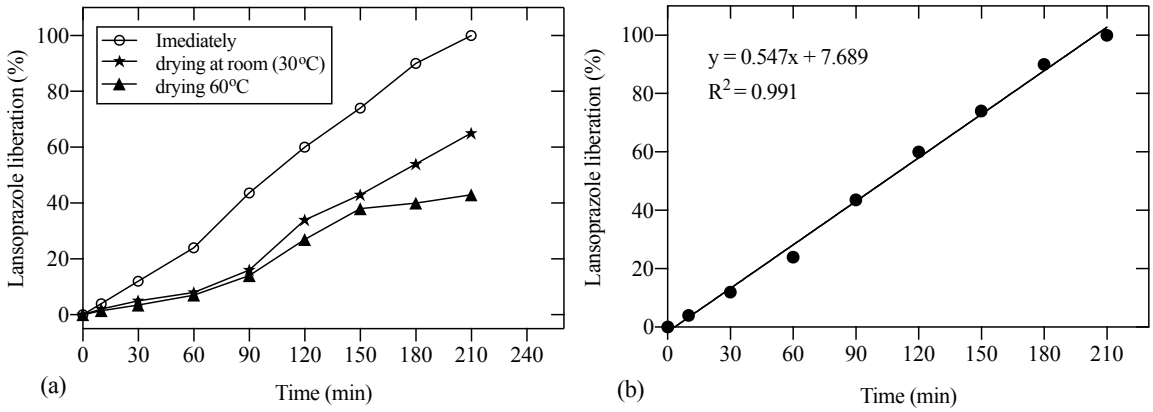

Fig. 4 Release profile of lansoprazole encapsulated in sodium alginate beads at pH 7.4: (a) in three conditions after immersion in simulated release medium and (b) linear adjustment of the release profile immediately after encapsulation.

Table 1 Average diameter of beads according to the treatment.

Treatment	Diameter (mm)
immediately	3.08 ± 0.092
30 °C	2.72 ± 0.034
60 °C	1.98 ± 0.016

external environment, therefore can increase encapsulation efficiency and reduce the release rate [19].

According to Fig. 4b, the concentration of lansoprazole measured in the solution was 4 µg/mL (16%) in the first 10 minutes, indicating that the active principle of the sphere was being transferred to the external environment. The dynamic release lasted for 210 min (3.5 h), and the concentration was recorded as 25 µg/mL (100%). The encapsulation efficiency, i.e. the actual content of lansoprazole in the microspheres during encapsulation, was also evaluated. The efficiency was 100%, determined by measuring the zero absorbance of 3 mL of the filtrate. Fig. 5 shows the effect of pH value on the release profile of lansoprazole immediately after encapsulation.

The release rate of lansoprazole is influenced by the pH value of the external medium (simulated). The rate at pH 1.5 is lower than that at pH 7.4. As can be seen from Figure 5, the release rate decreases as the pH value increases. Sadeghi et al. showed that the release rate of sodium alginate microspheres coated with simvastatin was proportional to the pH value [20]. This is due to the existence of carboxylic acid (-COO) group in the molecular structure of alginate. The ionization of these groups makes the release system respond to the stimulation mediated by the change of pH value. For biological applications this becomes interesting since the pH of biological fluids is varied,

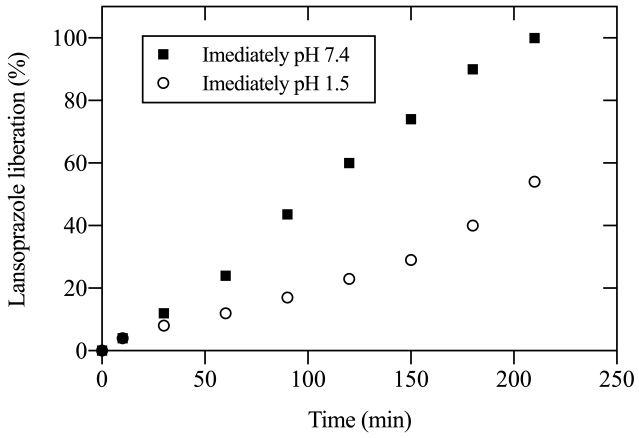

Fig. 5 Lansoprazole release profile immediately after encapsulation in two simulated release medium at pH 1.5 and 7.4.

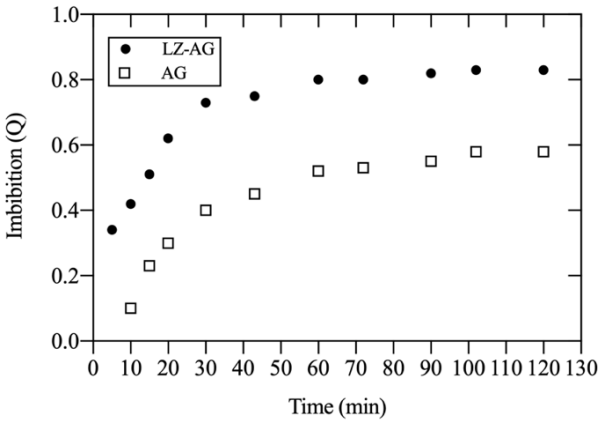

Fig. 6 Swelling rate of the sodium alginate beads with and without lansoprazole.

for example, stomach pH varies from 1 to 3, intestinal colon pH from 7 to 7.5.

When external pH is below 3.4, the polymer remains insoluble because the carboxylic groups are not ionized. However, when the pH value is higher than 4.4, the carboxylic acid groups become ionized and electrostatic repulsion of the negative charge occurs, causing expansion of the polymeric chain and swelling, which is pH 7.4 [11]. This implies that at low pH values, such as in the stomach, the release profile becomes slower because the polymer shrinks. Alginates become more soluble at higher pH, which justifies the need for rapid release, whereas the reverse is true at lower pH [12]. This feature allows the creation of pH-responsive delivery systems.

The swelling rate assesses the hydration capacity of the beads with time until equilibrium and reflects the diffusion rate of the encapsulated drug. Imbibition in the beads was evaluated with (LZ-AG) and without lansoprazole (AG) at pH 7.4, as shown in Fig. 6.

At equilibrium (120 min), the swelling value (Q) of LZ-AG beads was 0.83, and the swelling value (Q) of AG beads was 0.58. A possible explanation for the higher swelling rate shown by LZ-AG beads may be that the lack of lansoprazole allows calcium chloride to compress the alginate chain more tightly [21].

4. Conclusions

A new sustained-release model was established by encapsulating lansoprazole in sodium alginate microspheres. The encapsulation rate is 100%, and the encapsulation efficiency is extremely high. The release curve remains linear and could be evaluated for 210 minutes (3.5 hours). By adjusting the drying temperature and pH value, the dose released with time can be determined, and the treatment scheme can be put forward according to the encapsulation condition.

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Conflicts of Interest

The authors declare no conflicts of interest.

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