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ORIGINAL ARTICLE

Combining strategies to optimize a gel formulation containing miconazole: the influence of modified cyclodextrin on textural properties and drug release

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Abstract

Background: Miconazol, an antimycotic drug, is commonly formulated into semisolid formulations designed to be applied in the oral cavity to treat oral candidiasis. However, given its limited aqueous solubility, permeation through the biological membranes is low and therefore its activity is also limited. Cyclodextrins (CDs) have been widely used to increase the solubility and stability of poorly water-soluble drugs. *Aim*: The aim of this study is to formulate a gel containing an inclusion complex between a modified CD, methyl-β-cyclodextrin (MβCD), and miconazole (MCZ). The influence of the CD on the textural properties of the prepared gel and the drug release from formulation were evaluated. *Methods*: The gels were prepared using two polymers, Carbopol 71G and Pluronic F127, which were selected taking into account their bioadhesiveness and thermal-sensitive gelling properties, respectively. Texture profile analyses were performed at two different temperatures to ascertain the influence of the temperature on the gel texture properties. The in vitro MCZ release profiles from the prepared gel and the commercial gel formulations were evaluated and compared using modified Franz diffusion cells. *Results*: The addition of MβCD to the gel resulted in a decrease of the gel adhesiveness and firmness, and the MCZ release profile through *f*1 and *f*2 proved to be similar to the commercial product. *Conclusions*: A gel comprising miconazol in the form of an inclusion complex with MβCD showed suitable textural properties to be applied to the buccal mucosa. The MβCD enhanced the solubility of the MCZ in the gel formulation resulting in adequate in vitro drug release profiles.

Key words: *Carbopol; gel; methyl-β-cyclodextrin; miconazole; textural properties; pluronic*

Introduction

Polymeric gels have demonstrated extensive applications in the pharmaceutical field[1]. More specifically, gels have been successfully used for the local delivery of therapeutic agents to, for example, the buccal mucosa[2–4]. Oral candidiasis can be treated with gels comprising antifungal drugs for application on the buccal mucosa[5]. The major drawback associated with the use of formulations such as ointments, solutions, and creams on the buccal mucosa is the difficulty to maintain their effects for a significant period of time because they are very easily removed by salivation, temperature, tongue movement, and swallowing. Polymeric gels with an appropriate mucoadhesiveness offer the prospects of prolonging the residence time of controlled-release systems at the site of drug absorption and ensure an optimal contact[6]. It is accepted that the clinical and nonclinical performances of polymeric gels are dependent on their mechanical/rheological properties. Manipulation of the type and concentration of polymer, the state of the polymer, the formation of gel networks, the pH of the formulation, and the concentration of chemical materials are strategies that may be successfully used to engineer a defined product performance[7].

Miconazole (MCZ) is a drug with antimycotic activity, which exhibits very poor aqueous solubility, thus limiting its permeation through biological membranes and therefore its activity when formulated into oral semi-solid formulations[8,9].

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Cyclodextrins (CDs) have been widely used in semisolid formulations to increase the solubility and the stability of therapeutic substances in water, as well as to promote the release of drugs through biological membranes. Whereas conventional penetration promoters, such as alcohols and fatty acids, act by disrupting the lipid layers of the biological barrier; CDs act by increasing the availability of the drug on the surface of the biological membrane[10,11]. Because of the penetration enhancer properties of CDs, these compounds have been increasingly used in the preparation of gels for various applications, that is, vaginal[12–14], ocular[15,16], and buccal[17].

The formation of complexes between the MCZ and two different CDs, hydroxypropyl-β-cyclodextrin (HPβCD) and methyl-β-cyclodextrin (MβCD), was previously demonstrated by Ribeiro et al.[18] It was found that the complexation of MCZ with both CDs significantly increased its aqueous solubility as seen by the very high efficiency of solubilization values attained. However, the concentration of MβCD was nearly half of that of HPβCD, suggesting that the former CD can be used to form inclusion complexes with this drug in a more efficient manner[18]. The solubility of the solid complexes formed with the MβCD was increased 17-fold when in phosphate-buffered solution (PBS) of pH 6.8.

The aim of this study was to formulate a MCZ gel intended for buccal administration. The gel consisting of MCZ complexed with MβCD, Pluronic F127 (PLUR), and Carbopol® 71G (CARB) were prepared. The CD was used to improve the solubility of the MCZ[18]. PLUR was used in association with CARB to obtain a gel with mucoadhesive properties[19]. PLUR is a polyoxyethylene–polyoxypropylene–polyoxyethylene-type block copolymer consisting of 70% polyoxyethylene units. It has the ability to form a clear gel when in aqueous media at a concentration greater than 20% (w/w), and it exhibits the property of reversible thermal gelation, gelling at higher temperature (e.g., body temperature), and yielding a low viscosity solution upon cooling (e.g., at refrigerator temperature)[19]. CARB is a highly hydrophilic polyacrylic acid polymer and has been used to prepare gel formulations for topical administration because of high viscosity and good bioadhesiveness at low concentrations[6].

The influence of the modified CD, MβCD, on the gel mechanical behavior and MCZ release was investigated. The presence of interactions between different gel constituents was evaluated by thermal analysis. The gel firmness and the adhesiveness properties were evaluated on a texture profile analyzer at 25°C and 37°C. MCZ release from the gel was evaluated using modified Franz cells and compared with a commercially available formulation (Daktarin®), which is a MCZ gel suspension that does not contain CD.

Materials

MβCD (lot 781144; MW = 1190; average degree of substitution = 0.5) was kindly donated by Roquette (Lestrem, France). MCZ base (lot 00478343; MW = 416.13) was kindly donated by Janssen Pharmaceutica (Beerse, Belgium). CARB (lot TW56GAJ062) was purchased from Noveon, Inc. (Cleveland, OH, USA). PLUR (lot no. 046k0029) was obtained from Sigma (Steinheim, Germany). Daktarin® (lot no. 7CB5J00) was obtained at a drugstore. Milliq® ultrapure water obtained by reverse osmosis (Millipore, Madrid, Spain) was used. All other reagents were of analytical grade.

Methods

Preparation of solid MCZ/MβCD inclusion complex

The solid MCZ/MβCD system was prepared by freeze-drying method as described in our previous study by Ribeiro et al.[18] Briefly, equimolar amounts of the MCZ and MβCD were dissolved in ultrapure water. The solution was acidified with a few drops of hydrochloric acid (0.1 M) until attaining a clear solution. The mixture was agitated in orbital shaker for 24 hours at room temperature and then filtered through a 0.45-μm membrane filter (Millipore). The filtrate was frozen by immersion in an ethanol bath at −50°C (Benchtop shell freezer, Freezone® 79490, Kansas City, MS, EUA) and then lyophilized in a freeze-dryer (Lyph-lock 6 apparatus, Labconco, Kansas City, MS, EUA) for 48 hours.

The obtained powder was sieved (125 μm) and kept in a desiccator until use, and the content of drug was determined by UV assay.

The physical mixture (PM) was prepared by blending MCZ and previously sieved (125 μm) MβCD (1:1 molar ratio) in a ceramic mortar to be used as a reference.

Aqueous solubility of MCZ/MβCD inclusion complex

Fifteen milligrams of MCZ or an amount of PM or MCZ/MβCD inclusion complex equivalent to 15 mg of MCZ was added to 10 mL of the PBS (pH 6.8) in amber flasks with stoppers. Flasks were vortex-mixed for 5 minutes, sonicated in ultrasound bath for 30 minutes, and then continuously agitated in a water bath at 25 ± 1.0°C for 48 hours[20]. The solutions were filtered with 0.45-μm membrane (Millipore) and adequately diluted with ethanol. The drug concentration was determined by UV spectrophotometer at 273 nm[21] (Shimadzu UV–Visible 1603, Kyoto, Japan). The solubility results are the mean and SD of three replicates. For the PM and MCZ/MβCD inclusion complex, the solubility results were also

expressed as the increment of solubility when compared with the solubility of the drug alone.

Thermal analysis

Differential scanning calorimetry (DSC) analysis of the pure materials and binary systems were carried out using a Shimadzu DSC-50 System (Kyoto, Japan) equipped with a computerized data station TA-50WS/PC. The thermal behavior was studied by heating the samples (5–10 mg) in a sealed aluminum pan from 30°C to 250°C at a rate of 10°C/min and under a nitrogen flow of 20 cm^3/min. An empty sealed pan was used as a reference. Indium (99.98%, mp 156.65°C; Aldrich® Milwaukee, WI, USA) was used for the calibration of the equipment's temperature.

Preparation of gel formulations

CARB-PLUR gel was prepared by the cold method mixing different concentrations of CARB and PLUR (%, w/w)[22]. CARB and then PLUR were (Table 1) added to 70% of cooled water (3–5°C) under stirring conditions until a homogeneous solution is obtained. Ultrapure water was then added to make up the volume to the total amount. Gels were left overnight at 4°C until clear solutions were obtained. On the following day, the preparations were neutralized to pH 5.5–6.5 using NaOH 10% (w/v) solution.

For gels containing other components besides CARB and PLUR, the extra components were previously dissolved in the cold PLUR solution containing 1.0% CARB, and then ultrapure water was added under gentle mixing conditions to the final volume.

Texture analysis

Texture profile analysis has been used to characterize the mechanical properties of gels and other semisolid systems[23]. This simple and rapid technique can provide information related to the gel mechanical parameters, such as hardness and adhesiveness. Hardness is the force required to attain a given deformation and can be obtained from the maximum force during the first compression cycle. Adhesiveness is the work required to overcome the attractive forces between the surface of the product and the surface of the probe with which the sample comes into contact and is represented by the negative area of the force–time curve[3,24,25]. Ideally, formulations designed for mucosal drug delivery should have low hardness yet high adhesiveness. Adhesiveness is an important parameter on the design of a mucosal gel, as a desirable gel contact and retention at the mucosal surface will ensure better clinical efficacy[26]. On the other hand, moderate gel firmness or strength is required for the easy application by spreading over the mucosa and greater adherence when in contact with the mucus.

Because PLUR has the property of thermo-sensitive gel[27], the temperature will be a parameter to be controlled. The thermal gelation of PLUR is able to form a depot by increasing the contact time, which produces a prolonged pharmacology action[28].

The textural analysis was performed in the compression mode using Texturometer apparatus (Stable Micro Systems TA-XT2*i*, Surrey, UK). Penetration test was carried out using a cylindrical probe of 10 mm diameter and a penetration depth of 5 mm at a speed of 3 mm/s. Formulations were kept in 50-mL glass jars during the study. After penetrating the sample, the probe returned to 40 mm above the gel surface. Measurements were performed at 25°C and 37°C to simulate the storage and the body temperatures, respectively. From the obtained graphics of force versus distance, the maximum force and the negative area were calculated to determine the gel firmness and the adhesiveness, respectively. Gel formulations were centrifuged at 1633 × *g* for 25 minutes to ensure removal of entrapped air and were stored at 4°C for at least 24 hours before the analysis. All samples were analyzed within 72 hours after gel preparation.

Statistical analysis

The statistical analysis of texture results was performed using SPSS for Windows (version 14.0). Comparison between two variables was performed using

Table 1. Aqueous solubility of MCZ in phosphate-buffered solution (pH 6.8) at 37 ± 0.5°C after 48 hours.

Gel formulation	Components amount (w/w) (g)							
CARB	—	0.5	1.0	1.5	1.0	1.0	1.0	1.0
PLUR	20	20	20	20	20	20	20	20
MCZ	—	—	—	—	2.0	—	—	—
MβCD	—	—	—	—	—	5.72	—	—
PM	—	—	—	—	—	—	7.72[a]	—
MCZ/MβCD inclusion complex	—	—	—	—	—	—	—	7.79[a]
Water	80	79.50	79.00	78.50	77.00	73.28	71.28	71.21

[a]Equivalent amount of 2% of MCZ.

Student's *t*-test ($P < 0.05$). Comparison between more than two variables was made with one-way analysis of variance (ANOVA) with a level of statistical significance $P < 0.05$. When the one-way analysis of variance analysis detected differences between samples, a post hoc test (Scheffé test) was conducted to identify those differences.

In vitro MCZ release studies

Modified Franz diffusion cells with an area of 1.327 cm^2 and a final volume of 16 mL were used to evaluate MCZ release profiles from gel formulations in a closed system. The diffusion cells consist of two compartments (donor and receptor) separated by a cellulose dialysis membrane (Visking Tubing 18/32; Visking Co., Chicago, IL, USA)[29,30] or a poly(ether)sulfone membrane (0.45 μm; Supor-450 Pall Corporation, Ann Arbor, MI, USA)[31,32] previously washed for 30 minutes in receptor solution. The donor compartment was filled with 200 mg of the gel containing the MCZ/MβCD inclusion complex, which corresponds to a drug content of 4 mg, using a syringe and covered with Parafilm® to prevent evaporation. The receptor compartment was filled with PBS/ethanol solution (50/50, v/v) to ensure pseudo-sink conditions by increasing MCZ solubility in the receptor solution[33–35]. The air bubbles were removed from the receptor solution using an ultrasound bath prior to testing. The receptor solution was stirred with a magnetic bar to ensure homogeneity of the system. The apparatus was maintained at 37 ± 0.5°C throughout the experiment.

MCZ release studies were carried out during 3 hours, and samples were withdrawn every 5 minutes. The exact volume of the receptor compartment was measured at the end of each study so as to accurately calculate the cumulative MCZ release from each gel formulation.

The amount of MCZ in the receptor solution was assayed at 273 nm by a UV spectrophotometer (Shimadzu UV-Visible 1603). Six replicates were performed for each sample. The amount of released MCZ was calculated using a validated calibration curve [$R^2 = 0.999$, repeatability coefficient of variation (CV) = 1.79%, and reproducibility (CV) = 2.4%].

Release data analysis

Drug release profiles were compared according to the FDA guidance using the mathematical comparison by applying f_1 (Equation 1) and f_2 (Equation 2) factors. According to the FDA guidance[36], values of f_1 between 0 and 15 and f_2 between 50 and 100 ensure sameness or equivalence of two dissolution profiles. In both equations, R and T represent the dissolution measurements at 'n' time points of the reference and test formulations, respectively:

$$f_1 = \left(\frac{\sum_{j=i}^{n} \left| R_j - T_j \right|}{\sum_{j=i}^{n} R_j} \right) \times 100 \qquad (1)$$

$$f_2 = 50 \times \log \left\{ \left[\frac{1}{\sqrt{1 + \frac{1}{n} \sum_{j=1}^{n} (R_j - T_j)^2}} \right] \times 100 \right\} . \qquad (2)$$

The main advantage of the calculation of f_1 (difference) and f_2 (similarity) factors is to provide a simple way of data comparison[37,38].

Results and discussion

Aqueous solubility of MCZ/MβCD inclusion complex

The intrinsic solubility of MCZ in water is less than 1.03 μg/mL[39]. The solubility of a guest molecule can be increased by complexation with CDs[40–43]. The aqueous solubility of MCZ in PBS was determined after 48 hours to be 7.2 ± 0.03 μg/mL, as seen in Table 2. The PM solubility increased by 3.4-fold (24.38 μg/mL) when compared with MCZ alone. The increase of aqueous solubility, observed for MCZ in the PM, was probably because of the formation of inclusion complex when in aqueous solution or can be attributed to the mixing process between the components and to the loss of crystallinity and purity of the components in the mixture. Moreover, in the complex form, MCZ showed an increase of aqueous solubility to 127.93 μg/mL. The solubility of a drug molecule can be increased by complexation with CDs[40–43].

Table 2. Gel formulations regarding composition and components amount.

Binary systems	Aqueous solubility (μg/mL)[a]	Increase (fold)
MCZ/MβCD (PM)	24.38 ± 0.03	3.40
MCZ/MβCD inclusion complex[b]	127.93 ± 0.04	17.68

[a]Solubility data at the end of 48 hours; expressed as mean ± SD for $n = 3$.
[b]Inclusion complex in solid state obtained by lyophilization.

Gel properties

Thermal analysis

DSC thermograms of polymers used in the gel formulations and of blends of those polymers can be seen in Figure 1. The thermogram of CARB presents two distinctive peaks, the peak at 100–105°C being attributed to the glass transition and the peak at 250°C caused by its thermal decomposition[44]. The glass transition of CARB at around 133°C reported by Kanis et al. was not observed[45]. The thermal profile of PLUR shows a melting point at 57.30°C.

In the polymer blend, a shift was noticed in the glass transition of the PLUR with increasing concentrations of CARB.

The Figure 2 shows the DSC thermograms of the pure MCZ, PM, blend of CARB/PLUR (1/20%) (w/w), the MCZ/MβCD inclusion complex, and blend of CARB/PLUR/(MCZ/MβCD inclusion complex). The MCZ thermogram is characterized by a single endothermic peak at 85.2°C, which corresponds to the melting point of the drug. In the MCZ/MβCD PM this endothermic peak is also present despite appearing reduced in intensity because of a dilution effect. However, the absence of the same peak in the MCZ/MβCD inclusion complex suggests the formation of an amorphous inclusion complex through molecular entrapment of the drug inside the MβCD cavity as previously confirmed[18].

The thermogram of the mixture of CARB/PLUR containing the MCZ/MβCD inclusion complex does not present any new peaks thus suggesting the lack of interaction between the various components of the formulation.

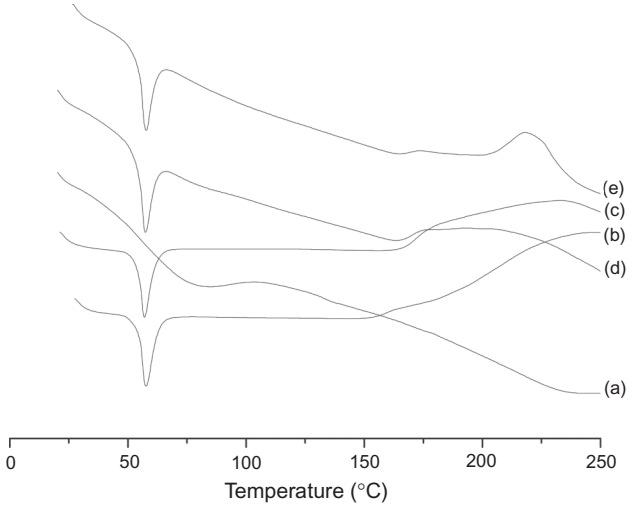

Figure 1. DSC thermograms of (a) pure CARB, (b) pure PLUR, (c) blend of CARB/PLUR (0.5/20%), (d) blend of CARB/PLUR (1.0/20%), and (e) blend of CARB/PLUR (1.5/20%).

Texture analysis

Textural parameters of maximum force (firmness) and negative area (adhesiveness) were obtained for the different gel formulations (Table 1). Figure 3 represents the firmness of gels formulated with PLUR 20% and different concentrations of CARB (0.5%, 1.0%, and 1.5%), and CARB/PLUR (1.0/20%) with MCZ, MβCD, PM, or MCZ/MβCD inclusion complex at 25°C and 37°C.

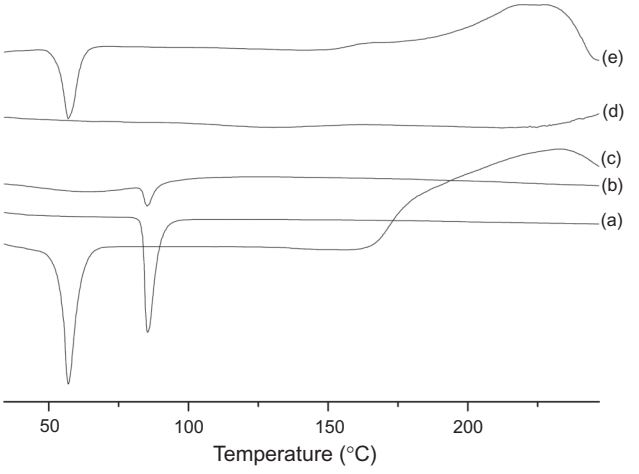

Figure 2. DSC thermograms of (a) pure MCZ, (b) PM, (c) blend of CARB/PLUR (1.0/20%), (d) MCZ/MβCD inclusion complex, (e) blend of CARB/PLUR/(MCZ/MβCD inclusion complex) (1.0/20/7.79%).

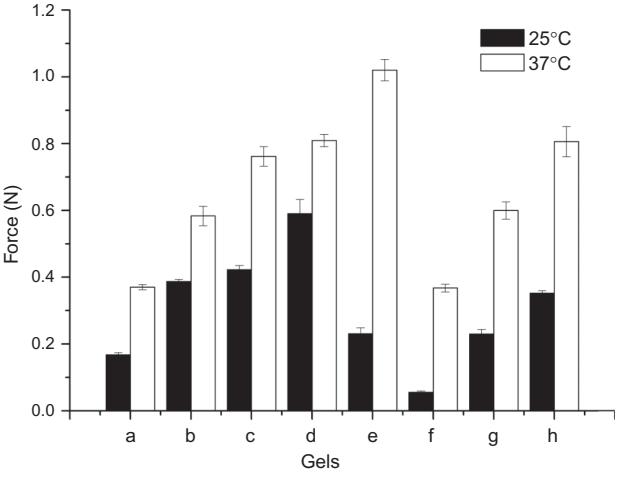

Figure 3. Plot of the measurements made for the interpretation of gel firmness at 25 (black column) and 37°C (white column) of formulations with (a) PLUR (20%), (b) CARB/PLUR (0.5/20%), (c) CARB/PLUR (1.0/20%), (d) CARB/PLUR (1.5/20%), (e) CARB/PLUR/MCZ (1.0/20/2.0%), (f) CARB/PLUR/MβCD (1.0/20/5.72%), (g) CARB/PLUR/PM (1.0/20/7.72%), and (h) CARB/PLUR/(MCZ/MβCD inclusion complex) (1.0/20/7.79%). Each column represents the mean ± SD ($n = 3$).

The addition of increasing concentrations of the CARB significantly altered the gel firmness ($F = 173.273$, $P < 0.0001$). The gel of PLUR 20% showed the lowest value of firmness by comparing with the gels of CARB/PLUR. When the concentrations of the CARB are 0.5% or 1.0%, the firmness is comparable; however, when the concentration of CARB was increased to 1.5%, the firmness increased considerably.

The rise in the temperature from 25°C to 37°C resulted in a marked increase of firmness of all gels. At 37°C the firmness of the gels containing CARB 1.0% or 1.5% are comparable, suggesting that an amount of CARB 1% is sufficient to obtain a gel with an adequate firmness for buccal application.

Figure 4 represents the adhesiveness of the gels formulated with PLUR 20% and different concentrations of CARB (0.5%, 1.0%, and 1.5%) and CARB/PLUR (1.0/20%) with MCZ, MβCD, PM, or MCZ/MβCD inclusion complex at 25°C and 37°C. The adhesiveness of the different gel formulations at 25°C was statistically different ($F = 35.823$, $P < 0.0001$). This effect was also observed when the temperature of the study was 37°C ($F = 101.42$, $P < 0.0001$). As reported for the firmness, the adhesiveness of the gels also increased when the temperature increased.

Regarding the temperatures at which the study was conducted, the addition of increasing CARB concentrations increased the adhesiveness of the gel formulations. The adhesiveness of the gels with 1.0% or 1.5% CARB is similar, and therefore concentrations higher than 1.0% do not translate into better results for adhesiveness.

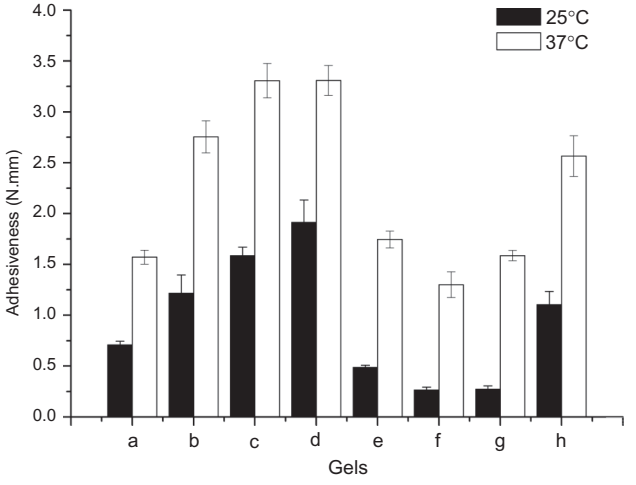

Figure 4. Plot of the measurements made for the interpretation of gel adhesiveness at 25 (black column) and 37°C (white column) of formulations with (a) PLUR (20%), (b) CARB/PLUR (0.5/20%), (c) CARB/PLUR (1.0/20%), (d) CARB/PLUR (1.5/20%), (e) CARB/PLUR/MCZ (1.0/20/2.0%), (f) CARB/PLUR/MβCD (1.0/20/5.72%), (g) CARB/PLUR/PM (1.0/20/7.72%), and (h) CARB/PLUR/(MCZ/MβCD inclusion complex) (1.0/20/7.79%). Each column represents the mean ± SD ($n = 3$).

In summary, all the gels showed a strong adhesiveness particularly at higher temperatures. Moreover, the addition of CARB considerably increased the adhesiveness as described by Singla et al.[46] and controlled the reversible thermal gelling properties of PLUR as previously reported by Dumortier et al.[22]

According to the results, the gel formulated with PLUR 20% and CARB 1.0% (w/w) showed the best compromise between firmness and adhesiveness for buccal application. The addition of MCZ, MβCD, PM, or MCZ/MβCD inclusion complex considerably changed the firmness of the various gels attained at 25°C ($F = 395.666$; $P < 0.0001$) and at 37°C ($F = 186.416$; $P < 0.0001$). A decrease in firmness with regard to the gel of CARB/PLUR (1.0/20%) was observed when the MCZ, MβCD, PM, and MCZ/MβCD inclusion complex were incorporated. The greatest effect on the decreasing firmness was observed with the CD alone. The value of firmness of the various gels at 25°C and 37°C follows the order: CARB/PLUR (1.0/20%) > MCZ/MβCD inclusion complex > MCZ > MF > MβCD.

The adhesiveness of the gels containing MCZ, MβCD, PM, or MCZ/MβCD inclusion complex was also statistically different at 25°C ($F = 188.624$; $P < 0.0001$) and at 37°C ($F = 108.173$; $P < 0.0001$). As previously discussed for firmness values, the adhesion decreased significantly when various constituents were incorporated, especially in the presence of the modified CD. Hydrophobic interactions could occur between the polymer chains and the MβCD resulting in a reduction of the polymer chains unfolding. Consequently, it may modify the polymer affinity for the hydration medium, decreasing its swelling and firmness[47]. Qi et al. demonstrated that the addition of HPβCD in a PLUR gel increased the length of gelling but drastically reduced the gel strength and adhesiveness[48]. Bonacucina et al. concluded that the presence of HPβCD also modified elastic character and micro-rheological structure of Poloxamer 407 gel samples[49]. In the complex form, the functional groups of the CD interact preferentially with the drug, therefore minimizing the interactions with the PLUR. For that reason, the mechanical properties of the gel are less affected.

The ability of PLUR to interact with several drugs has rendered it useful as a drug delivery carrier[50,51]. MCZ also appears to interact with the PLUR micelles reducing the PLUR affinity for the hydration, thus decreasing the gel adhesiveness.

The gel containing MCZ/MβCD inclusion complex showed a value of adhesiveness and firmness close to the gel containing CARB/PLUR (1.0/20%), suggesting that a gel with suitable properties can be obtained by adding the MCZ/MβCD inclusion complex.

Release study

In vitro drug release is a basic parameter in the development of pharmaceutical dosage forms[52]. The total amount of drug at the mucosa layers and of the drug penetration through the mucosa will depend on the efficiency in which the drug is released from the semi-solid formulation. The MCZ belongs to class 2, low solubility and high permeability, in the biopharmaceutical classification[43], and it should be emphasized that the in vivo dissolution is the limiting step of drug absorption and may be variable because of factors related with the formulation.

In the evaluation of the in vitro drug release of pharmaceutical dosage forms containing liposoluble drugs such as MCZ, the use of solutions containing surfactants or organic solvents may be necessary to ensure the suitable solubility of these drugs for maintaining sink conditions[36]. Considering this, 50% of (v/v) ethanol was added in the receptor solution in the modified Franz diffusion cells to maintain the pseudo-sink conditions for the MCZ release studies. In addition, the selection of the artificial membrane to support the gel formulations was fundamental during establishment of suitable conditions of the in vitro release studies, where the porosity and physicochemical properties of the membrane must be controlled[29]. The artificial membrane should be an inert support for the drug and/or gel formulations, allowing the free circulation of the drug between cells compartments, the drug passage velocity across the membrane being greater than the velocity in which the drug is released from the gel formulation[53–55]. In addition, it is important to confirm that there are no physical or chemical interactions between the membrane and the formulation. The excipients present in the formulation may affect the physical integrity of the membrane, or in many cases the drug may bind to the membrane, being retained there. Additionally, the membrane should not contain any 'leachable' components that can cause interference to the drug assay[31].

Figures 5 and 6 represent the percentage of MCZ released over time for the gel formulated with MCZ/MβCD inclusion complex and the commercial gel formulation (Daktarin®), using a dialysis membrane or a poly(ether)sulfone membrane. The initial concentration of MCZ in the gel formulations was 2% (w/w).

It was found that this type of membrane has a significant effect on the passage of the drug to the receptor compartment. As can be seen, the dialyses membrane showed to be a limiting factor and influenced the process of drug diffusion possibly because of poor tolerance of the membrane to 50% (v/v) ethanol solution.

The f_1 and f_2 factors[38] were calculated and results can be seen in Table 3. In addition, the dissolution efficiency (DE) was calculated from the area under the curve of MCZ release profiles in the time interval (t) using the trapezium method[56]. Table 4 shows the DE values of MCZ for both formulations until 180 minutes, this parameter being more suitable for a quantitative comparison[57].

The MCZ release from the gel containing the drug in the complex form with the modified CD proved to be similar to that from the commercial formulation based on f_1 and f_2 factors, and it is also confirmed by the comparison between the DE values ($P > 0.05$). If the same behavior occurs in vivo, it is possible to

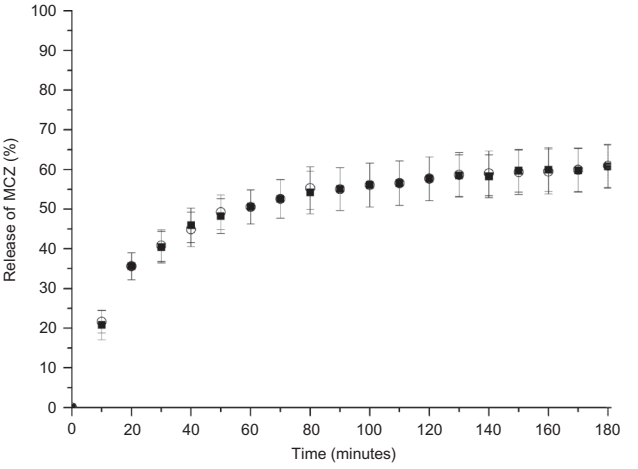

Figure 5. In vitro release profiles of MCZ from the proposed gel formulation containing the inclusion complex (■) and Daktarin® (○) carried out with dialysis membrane. The gel containing the inclusion complex comprises CARB/PLUR (1.0/20%) and MCZ/MβCD inclusion complex (7.79%). Each point represents the mean ± SD ($n = 6$).

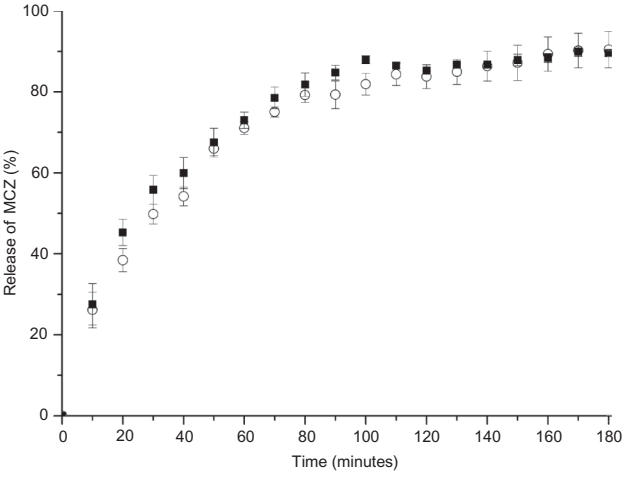

Figure 6. In vitro release profiles of MCZ from the proposed gel formulation containing the inclusion complex (■) and Daktarin® (○) carried out with poly(ether)sulfone membrane. The gel containing the inclusion complex comprises CARB/PLUR (1.0/20%) and MCZ/MβCD inclusion complex (7.79%). Each point represents the mean ± SD ($n = 6$).

Table 3. The f_1 and f_2 factors for comparison of MCZ release profiles from proposed gel formulation containing the inclusion complex and Daktarin® carried out with both artificial membranes.

	f_1		f_2	
Comparison	Dialysis membrane	Poly(ether)sulfone membrane	Dialysis membrane	Poly(ether)sulfone membrane
R/complex	1.33	7.14	95.84	66.57

R is a reference (Commercial Formulation Daktarin®).

Table 4. Dissolution efficiency values for in vitro release studies carried out with both artificial membranes.

	DE dialysis membrane (%)	DE poly(ether)sulfone membrane (%)
Gel containing MCZ/MβCD inclusion complex	50.02	73.24
Daktarin®	50.16	70.70

extrapolate that a larger amount of MCZ will be available for the penetration through the buccal mucosa. To better explain these results, the characteristics of the components of both formulations and the impact of these components on the MCZ release profile should also be considered. In the Daktarin® gel formulation, a nonionic surfactant, polysorbate, and ethyl alcohol are used to improve the solubility of MCZ. These compounds are also considered good absorption promoters that significantly modify the bioavailability and rate of penetration of a drug by modifying the content of the lipid membrane. CDs such as the modified one used in the proposed gel formulation can be considered as an alternative carrier to increase the dissolution of poorly water-soluble drugs, therefore enhancing drug release and permeation by increasing the availability of the drug at the surface of the biological membrane.

In the formulation proposed in this study the CD also has a taste-masking effect reducing the need for a flavoring agent. The Daktarin® gel formulation requires a flavoring agent to achieve a gel with the appropriate organoleptic properties to be administered in the buccal cavity. Moreover, CDs have an important role in the stability of the formulation, can provide taste masking, and are less irritant for the mucosa than surfactants and alcohol.

Conclusion

According to the results presented, the gel formulated with 20% PLUR and 1% CARB (w/w) presents the best compromise between firmness and adhesiveness for buccal application. It may appear that the rheological behavior and texture of various gel formulations have been influenced by the type and concentration of polymers and the temperature for the analysis.

Addition of MβCD to the CARB/PLUR gel reduced the firmness and adhesiveness compared with the gel formulated without this modified CD.

The determination of the in vitro MCZ release profile using modified Franz diffusion cells equipped with a synthetic membrane constituted an appropriate method for the evaluation of drug release from this type of formulation. The gel containing MCZ/MβCD inclusion complex showed a similar drug release profile when compared with the commercial gel formulation. Therefore, MβCD can be considered as an alternative to solubilizing agents like alcohol and polysorbate.

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Declaration of interest

The authors report no conflicts of interest. The authors alone are responsible for the content and writing of this paper.

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