Formulation and in vitro evaluation of mucoadhesive buccal tablets of Timolol maleate

Satyabrata Bhanja1*, P. Ellaiah1, Sujit Kumar Martha1, Pratit Kanchan Sahu1, Sandip Prasad Tiwari2, Bibhuti Bhusan Panigrahi3, Debajyoti Das4

1Department of Pharmaceutics, Jeypore College of Pharmacy, Jeypore (K), Odisha-764 002, Koraput, India
2Maharajhas College of Pharmacy, Pholbaugh, Vizianagaram, Andhra Pradesh, India
3HI-Tech College of Pharmacy, Pandara, Rasulgarh, Bhubaneswar-751 010, Odisha, India
4School of Pharmaceutical Sciences, SoA University, Bhubaneswar-751 030, Odisha, India

ABSTRACT

The present investigation is concerned with formulation and evaluation of mucoadhesive buccal tablets containing antihypertensive drug, Timolol maleate to circumvent the first pass effect and to improve its bioavailability with reduction in dosing frequency and dose related side effects. The tablets were prepared by direct compression method. Eight formulations were developed with varying concentrations of polymers like Carbopol 934, Polyethylene oxide and Hydroxy Propyl Methyl Cellulose. The tablets were tested for weight variation, hardness, surface pH, drug content uniformity, swelling index, and bioadhesive strength and in-vitro drug dissolution study. FTIR studies showed no evidence on interactions between drug, polymers, and excipients. The in vitro release of Timolol maleate was performed under sink conditions (Phosphate buffer PH 6.8, 37±0.5ºC, rpm 50) using USP-XXIV dissolution apparatus type II. The best in vitro drug release profile was achieved with the formulation F5 which contains the drug, Carbopol 934p and HPMC K4M in the ratio of 1:2.5:10. The surface pH, bioadhesive strength and swelling index of formulation F5 was found to be 6.34, 36.5 g and 80.3 %, respectively. The formulation F5, containing 10 mg of Timolol maleate exhibited 7 h sustained drug release i.e. 98.18 % with desired therapeutic concentration. The in vitro release kinetics studies reveal that all formulations fits well with zero order kinetics followed by Korsmeyer-Peppas, first order and then Higuchi’s model and the mechanism of drug release is non-Fickian diffusion.

Key words: Timolol maleate, mucoadhesive buccal tablet, Carbopol 934, Bioadhesive strength, In vitro drug release, Release kinetics

1. INTRODUCTION

Buccal delivery of drugs provides an attractive alternative to the oral route of drug administration, particularly in overcoming deficiencies associated with the latter mode of dosing [1]. Problems such as first pass metabolism and drug degradation in the GIT environment can be circumvented by administering the drug via buccal route. Moreover, the oral cavity is easily accessible for self medication and be promptly terminated in case of toxicity by removing the dosage form from buccal cavity. It is also possible to administer drugs to patients who cannot be dosed orally via this route [2,3].

Successful buccal drug delivery using buccal adhesive system requires at least three of the following (a) a bioadhesive to retain the system in the oral cavity and maximize the intimacy of contact with mucosa (b) a vehicle the release the drug at an appropriate rate under the conditions prevailing in the mouth and (c) strategies for overcoming the low permeability of the oral mucosa. Buccal adhesive drug delivery stem promote the residence time and act as controlled release dosage forms [4].

Buccal mucosa makes a more appropriate choice of site if prolonged drug delivery is desired because buccal site is less permeable than the sublingual site [5]. In addition, there is excellent acceptability and the drug can be applied,
localized and may be removed easily at any time during the treatment period [6]. It is beneficial in the case of Timolol to overcome the problem of frequent dosing due to its shorter half life (2.5-5 h). Prolonged release of the drug and increased bioavailability leads to the significant reduction in the dose and hence dose related side effects.

Hence, in the present work an attempt was made to formulate mucoadhesive buccal tablet for Timolol maleate using different mixtures of polymers in order to avoid extensive first pass metabolism, degradation in the stomach and prolonged effect.

2. MATERIALS AND METHODS

2.1 Materials

Timolol maleate was a gift sample from Micro Lab Pvt. Ltd., Bangalore. Polyethylene oxide was gift sample from Glenmark Pvt. Ltd, Mumbai. Hydroxy Propyl Methyl Cellulose and Carbopol 934 were purchased from Loba chemicals, Mumbai. All other reagents used were of analytical grade.

2.2 Compatibility studies

The drug-excipient compatibility studies were carried out using Fourier Transform Infrared Spectrophotometer (FTIR). Infra red spectra of pure drug and mixture of drug and excipients were recorded. A base line correction was made using dried potassium bromide and then the spectra of the dried mixture of drug, formulation mixture and potassium bromide were recorded on FTIR.

2.3 Formulation of mucoadhesive buccal tablets

The drug, polymers and excipients were mixed homogeneously in a glass mortar for 15 min. The mixture (150 mg) was then compressed using an 8 mm, biconcave punch in a single-stroke using 8-station rotary machine (The Rimek Mini Press-1). The results are shown in Table 1.

2.4 Evaluation of mucoadhesive buccal tablets

2.4.1 Weight variation

Eight tablets from each formulation (F1 to F8) were weighed using an electronic balance and the average weight was calculated.

2.4.2 Hardness

Tablets require a certain amount of strength or hardness and resistance to friability, to withstand mechanical shocks of handling in manufacture, packaging and shipping. The hardness of the tablets was determined using Monsanto hardness tester. It is expressed in Kg/cm². Three tablets were randomly picked from each formulation and the mean and standard deviation values were calculated and the results are shown in Table 2.

2.4.3 Friability

Friability is the measure of tablet strength. Roche type friabilator was used for testing the friability using the following procedure. Twenty tablets were weighed accurately and placed in the tumbling apparatus that revolves at 25 rpm dropping the tablets through a distance of six inches with each revolution. After 4 min, the tablets were weighed and the percentage loss was determined.

% loss = \frac{\text{Initial weight} - \text{Final weight}}{\text{Initial weight}} \times 100

2.4.4 Thickness

The thickness of three randomly selected tablets from each formulation was determined in mm using a vernier caliper (Pico India). The average values were calculated.

2.4.5 Content uniformity

Ten tablets from each formulation were taken, crushed and mixed. From the mixture 10 mg of Timolol Maleate equivalent of mixture was extracted thoroughly with 100 mL of pH 6.8 phosphate buffer. The amount of drug present in each extract was determined using UV spectrophotometer at 296 nm. This procedure was repeated thrice and this average was chosen.

2.4.6 Microenvironment pH

The microenvironment pH (surface pH) of the buccal tablets was determined in order to investigate the possibility of any side effects in vivo. As an acidic or alkaline pH may cause irritation to the buccal mucosa, it was determined to keep the surface pH as close to neutral as possible. The method adopted by Battenberg et al [7] was used to determine the surface pH of the tablet. A combined glass electrode was used for this purpose. The tablet was allowed to swell by keeping it in contact with 5 mL of distilled water (pH 6.5 ± 0.05) for 2 h at room temperature. The pH was measured by bringing the electrode in contact with the surface of the tablets and allowing it to equilibrate for 1 min.

2.4.7 Bioadhesion studies

In evaluation of adhesion, it is important to use uniform surfaces that allow the formation of reproducible adhesive bonds. In present study, sheep buccal mucosa was used as a model mucosal surface for bioadhesion testing [8]. Immediately after slaughter, the buccal mucosa was removed from the sheep and transported to laboratory in tyrode...
solution and kept it at 40°C. The composition of tyrode solution (g/L) is sodium chloride 8, potassium chloride 0.2, calcium chloride dihydrate 0.134, sodium bicarbonate 1.0, sodium dihydrogen phosphate 0.05 and glucose 1.0.

2.4.8 Fabrication of assembly

The Mucoadhesive forces of the tablets were determined by means of mucoadhesive measuring device shown in Fig. 1. The sheep buccal mucosa was cut into strips/pieces and washed with tyrode solution. At time of testing a section of sheep buccal mucosa (c) was secured keeping the mucosal side out, on the upper glass vial (B) using rubber band and aluminium cap. The diameter of each exposed mucosal membrane was 1 cm. The vial with the sheep buccal mucosa (C) was stored at 37°C for 10 min. Then one vial with section of sheep buccal mucosa (C) and another vial were fixed on height adjustable pan (E). To a lower vial a tablet (D) was placed with the help of bilayered adhesive tap, adhesive side facing downward. The height of the lower vial was adjusted so that a tablet could adhere to the sheep buccal mucosa on the upper vial. A constant force was applied on the upper vial for 2 min, after which it was removed and the upper vial was then connected to the balance. Then the weight on right side pan was slowly added in an increment of 0.5 g, till the two vials just separated from each other. The total weight (g) required to detach two vials was taken as a measure of Mucoadhesive strength. From this Mucoadhesive strength, the force of adhesive was calculated.

2.4.9 Swelling study

Six Buccal tablets were individually weighed (W₁) and placed separately in Petri dishes with 5 mL of phosphate buffer of pH 6.8. At the time interval of 1, 2, 4, 6 and 8 h, tablet was removed from the Petri dish and excess water was removed carefully using the filter paper. The swollen tablet was then reweighed (W₂) and the percentage hydration was calculated using the following formula [9,10]:

\[
\text{Percentage hydration} = \left(\frac{W₂ - W₁}{W₁}\right) \times 100
\]

2.4.10 In-vitro dissolution studies

The In-vitro dissolution study was conducted as per the United States Pharmacopoeia (USP) XXIV. The rotating paddle method was used to study the drug release from the

---

**Table 1**

<table>
<thead>
<tr>
<th>Ingredients</th>
<th>Formulation code (mg)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>F1</td>
</tr>
<tr>
<td>Timolol maleate</td>
<td>10</td>
</tr>
<tr>
<td>Carbopol 934p</td>
<td>25</td>
</tr>
<tr>
<td>Polyethylene Oxide</td>
<td>100</td>
</tr>
<tr>
<td>HPMC K4M</td>
<td>-</td>
</tr>
<tr>
<td>Mannitol</td>
<td>10</td>
</tr>
<tr>
<td>Magnesium Stearate</td>
<td>3</td>
</tr>
<tr>
<td>Talc</td>
<td>2</td>
</tr>
<tr>
<td>Average weight</td>
<td>150</td>
</tr>
</tbody>
</table>
Table 2
Physico-chemical parameters of Timolol maleate buccal tablets

<table>
<thead>
<tr>
<th>Formulation code</th>
<th>Average weight (mg)</th>
<th>Hardness (Kg/cm²)</th>
<th>Friability (%)</th>
<th>Thickness (mm)</th>
<th>Drug content (%)</th>
<th>Surface pH</th>
<th>Bioadhesive strength (g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>F1</td>
<td>148.89±0.04</td>
<td>3.0±0.14</td>
<td>0.51±0.03</td>
<td>2.7</td>
<td>97.00</td>
<td>5.8</td>
<td>34.5</td>
</tr>
<tr>
<td>F2</td>
<td>150.00±0.19</td>
<td>4.5±0.17</td>
<td>0.31±0.07</td>
<td>2.9</td>
<td>98.78</td>
<td>5.9</td>
<td>31.4</td>
</tr>
<tr>
<td>F3</td>
<td>149.37±0.14</td>
<td>5.0±0.15</td>
<td>0.75±0.02</td>
<td>2.5</td>
<td>99.00</td>
<td>6.12</td>
<td>29.5</td>
</tr>
<tr>
<td>F4</td>
<td>148.48±0.31</td>
<td>6.5±0.12</td>
<td>0.55±0.01</td>
<td>2.6</td>
<td>98.25</td>
<td>5.69</td>
<td>27.6</td>
</tr>
<tr>
<td>F5</td>
<td>149.21±0.22</td>
<td>4.0±0.24</td>
<td>0.65±0.03</td>
<td>2.7</td>
<td>101.00</td>
<td>6.34</td>
<td>36.5</td>
</tr>
<tr>
<td>F6</td>
<td>150.13±0.23</td>
<td>4.5±0.23</td>
<td>0.35±0.06</td>
<td>2.9</td>
<td>96.00</td>
<td>6.15</td>
<td>34.1</td>
</tr>
<tr>
<td>F7</td>
<td>151.88±0.26</td>
<td>5.5±0.25</td>
<td>0.45±0.5</td>
<td>3.0</td>
<td>95.00</td>
<td>6.19</td>
<td>33.5</td>
</tr>
<tr>
<td>F8</td>
<td>149.38±0.19</td>
<td>7.0±0.11</td>
<td>0.49±0.07</td>
<td>2.6</td>
<td>95.65</td>
<td>6.16</td>
<td>31.5</td>
</tr>
</tbody>
</table>

Table 3
Percentage hydration of Timolol buccal tablets

<table>
<thead>
<tr>
<th>Formulation code</th>
<th>Percentage hydration</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>1 h</td>
</tr>
<tr>
<td>F1</td>
<td>46.2</td>
</tr>
<tr>
<td>F2</td>
<td>43.9</td>
</tr>
<tr>
<td>F3</td>
<td>40.8</td>
</tr>
<tr>
<td>F4</td>
<td>38.3</td>
</tr>
<tr>
<td>F5</td>
<td>47.6</td>
</tr>
<tr>
<td>F6</td>
<td>47.9</td>
</tr>
<tr>
<td>F7</td>
<td>47.8</td>
</tr>
<tr>
<td>F8</td>
<td>48.9</td>
</tr>
</tbody>
</table>

The dissolution medium consisted of 900 mL of phosphate buffer (pH 6.8). The release was performed at 37°C ± 0.5°C, at a rotation of speed of 50 rpm. 5 mL samples were withdrawn at predetermined time intervals (1 to 7 h) and the volume was replaced with fresh medium. The samples were filtered through Whitman filter paper No.40 and analyzed for Timolol after appropriate dilution by UV spectrophotometer at 296 nm. The % drug release was calculated using the calibration curve of the drug in phosphate buffer pH 6.8.

2.4.11 Release kinetic studies

To find out the mechanism of drug release from hydrophilic matrices, the in vitro release data was treated with different kinetic models, namely zero order, first order, Higuchi and Korsemeyer-Peppas.

3. RESULTS AND DISCUSSION

3.1 Compatibility studies

The incompatibility between the drug and excipients were studied by FTIR spectroscopy. The spectral data of pure drug and various drug-excipient mixtures are presented in Fig. 2. The results indicate that there was no chemical incompatibility between drug and excipients used in the formulation.

3.2 Weight variation test

The weight variation test was conducted for each batch of all formulations F1 to F8 as per I.P and the results are shown in Table 2. The weight variation test for all the formulations complies with the IP limit (± 10%).

3.3 Hardness test

The adequate tablet hardness is necessary requisite for consumer acceptance and handling. The measured hardness of the tablets of each batch of all formulations i.e. F1 to F8 were ranged between 3.0 to 7.0 Kg/cm² and the results are shown in Table 2.

3.4 Friability test

The friability test for all the formulations were done as per the standard procedure I.P. The results of the friability test were tabulated in Table 2. The data indicates that the friability was less than 1% in all formulations ensuring that the tablets were mechanically stable.

3.5 Thickness

The thickness of the tablets was found to be almost uniform in all formulations F1 to F8. The thickness was found to be in the range of 2.5 to 3.0 mm. None of the formulations (F1 to F8) showed a deviation. Hence, it is concluded that all the formulations complied the thickness test and the results are shown in Table 2.

3.6 Drug content

The drug content of each batch of all the formulations (F1 to F8) was evaluated as per the standard protocol and the results are shown in the Table 2. The results indicate that the percentage of drug content was found to be 95.00% to 101.00%. Hence it is concluded that all the formulations are following acceptable limits as per Indian Pharmacopoeia i.e. ± 5%.

3.7 Surface pH

Surface pH of all the formulations F1 to F8 was found to be 5.8 to 6.38, which is well within the limit of acceptable salivary pH range of 5.69 to 6.34 (Table 2). Hence, it was concluded that all formulations could not produce any local irritation to the mucosal surface.
3.8 Bioadhesive strength

The *in vitro* bioadhesive strength study was performed and the results are shown in the Table 2. On the modified physical balance and measure the force (N) required detaching the tablet. The bioadhesion characteristics were affected by the concentration of the bioadhesive polymers. Increase in concentration of polymer increases bioadhesive strength of formulation. The formulations (F1, F2, F3 and F4) with Carbopol 934p and Polyethylene oxide showed the bioadhesive strengths of 34.5, 31.4, 29.5 and 27.6 g respectively. The formulations (F5, F6, F7 and F8) with Carbopol 934p and HPMC K4M showed the bioadhesive strengths of 36.5, 34.1, 33.5 and 31.5 g, respectively.

3.9 Swelling study

The swelling studies were conducted for all formulations i.e. F1 to F8 and the results were shown in Table 3. All the formulations were hydrated generally by keeping the tablets in contact with water for 1 h to 8 h.

The highest hydration (swelling) i.e. 80.3% was observed with the formulation F5. This may be due to quick hydration of polymers (Carbopol 934p and HPMC K4M). The swelling rate of tablets increased in the case of formulation F5 containing Carbopol 934p and HPMC K4M in the ratio of 1:2.5:10.

3.10 In-vitro release studies

The formulations F1, F2, F3 and F4 containing drug, Carbopol 934p and Polyethylene oxide polymers in the ratios of 1:2.5:10, 1:3.5:9, 1:4.5:8 and 1:5.5:7, respectively. The *in vitro* cumulative drug release profile of formulations F1, F2, F3 and F4 showed 85.94%, 80.65%, 75.30% and 73.14%, respectively. Among these four formulations, F1 was found to be highest percentage drug release. During the study it was observed that the tablets were initially swell and no erodible over the period of 7 h (Fig. 3).

Similarly the formulations F5, F6, F7 and F8 drug, containing Carbopol 934p and HPMCK4M polymers in the ratios of 1:2.5:10, 1:3.5:9, 1:4.5:8 and 1:5.5:7 respectively. The *in vitro* cumulative drug release profile of formulations F5, F6, F7 and F8 showed 98.18%, 88.25%, 82.75% and 76.35%, respectively. Among these four formulations, F5 was found to be highest percentage drug release. During the study it was observed that the tablets were initially swell and non-erodible over the period of 7 h (Fig. 4).

It was concluded that by increasing the concentration of Carbopol 934p in the formulation, the drug release rate from the tablets was found to be decreased. But when the concentration of secondary polymers (Polyethylene Oxide and HPMC K4M) increased, the drug release rate was found to be increased. This may be due to increased hydration (or) swelling characteristics of polymers with increased concentrations. From the overall data it was found that the

---

Fig. 2. FT-IR spectra of A) Timolol maleate; B) Timolol maleate + Carbopol 934p; C) Timolol maleate + Polyethylene oxide; D) Timolol maleate + HPMC K4M.
formulation F5 showed the maximum percentage of drug release i.e. 98.18% at the end of 7 h.

### Table 4

<table>
<thead>
<tr>
<th>Formulations</th>
<th>Zero order R²</th>
<th>First order R²</th>
<th>Higuchi R²</th>
<th>Korsmeyer-Peppas R²</th>
<th>n</th>
</tr>
</thead>
<tbody>
<tr>
<td>F1</td>
<td>0.998</td>
<td>0.945</td>
<td>0.922</td>
<td>0.999</td>
<td>0.898</td>
</tr>
<tr>
<td>F2</td>
<td>0.997</td>
<td>0.937</td>
<td>0.886</td>
<td>0.997</td>
<td>0.958</td>
</tr>
<tr>
<td>F3</td>
<td>0.993</td>
<td>0.970</td>
<td>0.901</td>
<td>0.977</td>
<td>0.879</td>
</tr>
<tr>
<td>F4</td>
<td>0.984</td>
<td>0.956</td>
<td>0.865</td>
<td>0.992</td>
<td>0.939</td>
</tr>
<tr>
<td>F5</td>
<td>0.997</td>
<td>0.908</td>
<td>0.899</td>
<td>0.993</td>
<td>0.961</td>
</tr>
<tr>
<td>F6</td>
<td>0.996</td>
<td>0.933</td>
<td>0.930</td>
<td>0.993</td>
<td>0.841</td>
</tr>
<tr>
<td>F7</td>
<td>0.997</td>
<td>0.955</td>
<td>0.902</td>
<td>0.997</td>
<td>0.903</td>
</tr>
<tr>
<td>F8</td>
<td>0.991</td>
<td>0.975</td>
<td>0.885</td>
<td>0.994</td>
<td>0.935</td>
</tr>
</tbody>
</table>

#### 3.11 Drug release kinetics

In-vitro drug release data of F1 to F8 were fitted to zero order, first order, Higuchi and Korsmeyer-Peppas equations to ascertain the pattern of drug release (Table 4). The R² values were found to be higher in zero-order followed by Korsmeyer-Peppas, first order and then Higuchi, which indicates all the formulations followed zero-order release pattern. According to Korsmeyer-Peppas equation, the release exponent “n” value is > 0.5, which indicates the mechanism of drug release for all formulations is non-Fickian diffusion type.

### 4. CONCLUSIONS

Timolol maleate mucoadhesive buccal tablets could be formulated using the drug, Carbopol 934p and HPMC K4M with the ratios of 1:2.5:10. It can be seen that by increasing the concentration of Carbopol 934p in the formulation, the drug release rate from the tablets was found to be decreased. But when the concentration of HPMC K4M increased, the drug release rate was found to be increased. The in vitro release kinetics studies reveal that all formulations fits well with zero order kinetics followed by Korsmeyer-Peppas, first order and then Higuchi’s model and the mechanism of drug release is non-Fickian diffusion. Further, an elaborate in vivo study is to be carried out for the best formulation using a suitable animal model.

### ACKNOWLEDGEMENTS

Authors wish to thank Jeypore College of Pharmacy, Jeypore, Odisha, for providing research laboratory to carry out this project work. The authors also wish to show their deep gratitude to Micro Lab Pvt. Ltd., Bangalore and Glen mark Pvt. Ltd, Mumbai for providing the gift samples of Timolol maleate and Polyethylene oxide, respectively.

### REFERENCES