ABSTRACT

Transdermal drug delivery system (TDDS) delivers the drug through topical route for systemic effect at a predetermined and controlled rate. The drug safety, therapeutic efficacy and patient compliance can be gained by reducing both the size and number of doses which can ultimately be achieved through TDDS. In this study, transdermal films of Amitriptyline HCl have been formulated by solvent evaporation technique. Matrix type of film was prepared using polymers of Eudragit E100, hydroxy propyl cellulose (HPC) and polyvinyl pyrrolidone (PVP) in different compositions incorporating dibutyl phthalate as plasticizer. The films were evaluated for physicochemical properties, in vitro release, kinetics of drug release, skin permeation and skin irritation studies. Six translucent films (F1, F2, F3, F4, F5, F6) containing Amitriptyline HCl having uniform thickness, drug content and flexibility were prepared. Formulation F5 (Eudragit E100 & PVP at a ratio of 7:3) was selected as the optimized film considering the permeation and skin irritation studies. Six translucent films (F1, F2, F3, F4, F5, F6) containing Amitriptyline HCl having uniform thickness, drug content and flexibility were prepared. Formulation F5 (Eudragit E100 & PVP at a ratio of 7:3) was selected as the optimized film considering the highest drug release of 96.45 ± 1.24% in 24hrs. The release followed first order kinetics with diffusion based mechanism. The addition of PVP has resulted in much higher drug release than HPC. Formulation F5 showed maximum skin permeation of 49.27 ± 0.62% over period of 24hrs. The value of primary dermal irritation index was found to be zero. Formulation F5 had produced controlled release and permeation of Amitriptyline HCl up to 24hrs and thus suitable for transdermal drug delivery.

Keywords: Amitriptyline HCl, Eudragit E100, Transdermal delivery, In vitro release, Skin permeation, Skin irritation.

INTRODUCTION

Transdermal drug delivery is one of the most promising methods for systemic administration of drugs via the skin for variety of clinical indication. The adhesive dispersion type TDDS comi-patch®, fempatch®, climara patch® have received wide commercial acceptance. These systems deliver therapeutically effective concentration of the drug across the patient’s skin at a preprogrammed rate for prolonged period. TDDS offers advantages such as avoidance of gastro-intestinal disturbances, hepatic first pass metabolism, and pulsed entry into the systemic circulation, pain with injections, frequent dosing and rapid termination of drug input.

Amitriptyline HCl is a tricyclic antidepressant used to treat depression. The drug is often used to manage nerve pain resulting from cancer treatment and also chronic migraines. It undergoes first pass effect and produces side effects of weight gain, dry mouth, changes in appetite, drowsiness, muscle stiffness, nausea, constipation, nervousness, dizziness, blurred vision, urinary retention, insomnia and changes in sexual function. The adult dose for pain management is 10 mg to 150 mg. Longer duration of therapy and withdrawal due to side effects are the major concerns with existing oral route of delivery.

Eudragit® classes of polymers have been widely used in pharmaceutical formulations. The polymers are well tolerated by the skin and have a high capacity for loading drugs. Eudragit® E100 is an anionic cationic copolymer based on dimethyl aminoethyl methacrylate and neutral methacrylic esters. PVP is used as a binder, complexing agent, as an aid for increasing the solubility of drugs in liquid and semi-liquid dosage forms (syrups, soft gelatin capsules) and as an inhibitor of recrystallisation. Eudragit® E100 transdermal films have been prepared using either HPC or PVP, whereas the combination of PVP and HPC with eudragit E 100 has not yet been reported. Hence the present study was aimed to study the effect of PVP and HPC combination on eudragit E 100 film. This article summarizes the formulation of Amitriptyline HCl transdermal films and emphasizes the physicochemical, release, permeation and irritation properties of polymeric films.

MATERIALS AND METHODS

Amitriptyline HCl was obtained as a gift sample from Star Drugs Pharmaceutical Industries, Hosur (India). Eudragit® E100 was obtained from Rohm Pharma (Darmstadt, Germany). Polyvinyl pyrrolidone, Hydroxy propyl cellulose were purchased from SD fine chemicals Ltd, Mumbai (India).

Method of preparation of transdermal films

Amitriptyline hydrochloride films were prepared by solvent evaporation technique according to the formula given in Table 1. Weighed quantity of eudragit E100 was added to 5 ml ethanol and dissolved completely using mechanical stirrer (Remi, Mumbai). To the above solution, drug (10mg), PVP, HPC were added and mixed thoroughly followed by plasticizer (dibutyl phthalate) addition and mixed. The solution was poured on the thick aluminum foil (five cm² area) and covered by a funnel for 24 hrs for the solvent to evaporate. Total 6 formulations were prepared and stored for evaluation.

| Table 1: Formulation composition of Amitriptyline HCl transdermal films |
|-----------------------------|-----------------------------|-----------------------------|
| Formulation Code            | Ingredient(mg)              |                             |
| F1                          | Amitriptyline HCl 10        | Eudragit E100 210           | HPC 45 | PVP 45 |
| F2                          | 10                          | 210                         | 60    | 30    |
| F3                          | 10                          | 210                         | 30    | 60    |
| F4                          | 10                          | 210                         | 90    |       |
| F5                          | 10                          | 210                         |       | 90    |
| F6                          | 10                          | 300                         |       |       |

Physicochemical evaluation

The films were evaluated for the following physicochemical properties.

Thickness

The thickness of transdermal film was determined at different points in the film using screw gauge (Syracuse, Newyork).

Uniformity of weight

10 randomly selected films were weighed individually and their average weight was calculated. The standard deviation for each film weight was noted to check the uniformity of weight.
Drug content determination

Entire film was cut into pieces and dissolved in 100 ml of water. The solution was stirred continuously for 24 hrs in a shaker incubator (Daian Labtech Pvt Ltd, India) maintained at room temperature. The solutions were filtered and absorbance was measured at 239 nm in a UV-Visible spectrophotometer (Shimadzu, Japan).

Percentage moisture absorption

The films were weighed accurately and placed in the desiccator containing 100 ml of saturated solution of aluminum chloride, which maintains 79.50% relative humidity (RH). After three days, the films were taken out and weighed. The percentage moisture absorption was calculated using the formula:

\[
\text{Percentage moisture absorption} = \frac{\text{Final weight} - \text{Initial weight}}{\text{Initial weight}} \times 100
\]

Percentage moisture loss

The films were weighed accurately and kept in the desiccator containing one gram of anhydrous calcium chloride. After three days, the films were taken out and weighed. The moisture loss was calculated using the formula:

\[
\text{Percentage moisture loss} = \frac{\text{Initial weight} - \text{Final weight}}{\text{Initial weight}} \times 100
\]

Water vapor transmission rate (WVTR)\(^{17}\)

For the determination of WVTR, one gram of calcium chloride was weighed and placed in dried empty vials. The polymer films were pasted over the brim with the help of silicone adhesive grease and allowed to set for 5 minutes. The vials were weighed and placed in a humidity chamber (Bionik Innovation, India) maintained at 68% RH. The vials were taken out and weighed at the end of every 1st day, 2nd day, 3rd day and up to 7 consecutive days. The increase in vial weight was considered as a quantitative measure of moisture transmitted through the film.

\[
\text{WVTR} = \frac{W}{ST} \quad \text{-------- (3)}
\]

\(W\) is the increase in weight in 24 hrs; \(S\) is the area of the film exposed (cm\(^2\)); \(T\) is the exposure time (hrs)

In vitro drug release\(^{9,18}\)

The release study was conducted using the USP dissolution apparatus V (Veego Instruments Pvt. Ltd, India). The film was supported with a circular glass plate and covered by a mesh. This was then placed inside the dissolution basket that contains 500 ml of phosphate buffer (PH 7.4). The paddle was rotated at 100 rpm and the temperature was maintained at 32 ± 0.5°C. The samples were withdrawn at a regular time intervals and analysed using spectrophotometer at 239 nm. The dissolution medium was replaced with an equal volume of phosphate buffer at each sample withdrawal.

Kineti cs of drug release\(^{19}\)

The release kinetics of formulation (F5) was determined by fitting the drug release data with zero order kinetics (Eq. 1), first order kinetics (Eq. 2), higuchi equation (Eq. 3), korsmeyer-peppas equation (Eq. 4) and hixson crowell equation.

\[
\begin{align*}
Q_t &= K_0 t \\
\ln Q_t &= \ln Q_0 - K_1 t \\
Q_t &= K_f t^{1/2} \\
\ln \left( \frac{M_t}{M_{\infty}} \right) &= n \ln t
\end{align*}
\]

The following plots were made: Qt vs. t (zero order kinetic), ln(Q0 −Qt) vs. t (first order kinetic model) and Qt vs. t\(^{1/2}\) (higuchi model), where Qt is the percentage of drug released at time t, Q0 is the initial amount of drug present in the formulation and K0, K1 and Kf are equation constants. The mechanism of drug release was found using the korsmeyer-peppas model.

\[
\frac{M_t}{M_{\infty}} = K_p^n \quad \text{-------- (4)}
\]

Where \(M_t / M_{\infty}\) is the fraction of released drug at time t, \(K_p\) is the rate constant and \(n\) is the release exponent.

The value of “n” was calculated from the slope of the curve plotted between log fraction of drug released \(\left( \frac{M_t}{M_{\infty}} \right)\) vs. log time.

In-vitro skin penetration study\(^{20,21}\)

The permeation study was carried out for the film F5 using a locally fabricated keshary-chien type diffusion cell. Full thickness skin of rat was used for the study. Hair on the abdominal skin was removed and animal was sacrificed by cervical dislocation. The skin was cut by surgical procedure using scalpel and the subcutaneous tissue attached to the skin was peeled off. The film was applied to the stratum corneum side of the skin and then mounted in the diffusion cell with the dermal side in contact with the receptor fluid. The receptor compartment was maintained at a temperature of 32 ± 5°C and was continuously stirred at a constant rate of 500 rpm using a small magnetic bead rotated with the help of a magnetic stirrer (Remi, India). The receptor compartment was filled with 50 ml of saline phosphate buffer (pH 7.4). The samples were withdrawn from the receptor compartment at different time intervals and equal volume of receptor fluid was replaced at each sample withdrawal. The samples were diluted and then analyzed by UV – visible spectrophotometer at 239 nm.

Skin irritation study\(^{22}\)

Skin irritation test was performed for the film F5 to identify the presence of skin reactions erythema and edema. The test was carried out on healthy male rabbits. The dorsal surface of the skin was cleaned well and the hair was removed by shaving. The shaved skin was washed with rectified spirit and left overnight for any untoward reactions of shaving. The film F5 was fixed over the shaved area and observed for skin reactions after 1hr, 24 hrs, 48 hrs and 72 hrs. Scores were assigned according to the Draize scoring method. The scores assigned to the skin reactions and Primary Dermal Irritation Index (PDI) values are tabulated in Table 2 and Table 3.

Table 2: Dermal reactions and the score assigned in accordance with the Draize scoring criteria

<table>
<thead>
<tr>
<th>S. No</th>
<th>Erythema</th>
<th>Edema</th>
<th>Score</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>No Erythema</td>
<td>No Edema</td>
<td>0</td>
</tr>
<tr>
<td>2</td>
<td>Very slight Erythema</td>
<td>Very slight Edema</td>
<td>1</td>
</tr>
<tr>
<td>3</td>
<td>Well defined Erythema</td>
<td>Slight Edema</td>
<td>2</td>
</tr>
<tr>
<td>4</td>
<td>Moderate to severe Erythema</td>
<td>Moderate Edema</td>
<td>3</td>
</tr>
<tr>
<td>5</td>
<td>Severe Erythema to slight Eschar formation</td>
<td>Severe Edema</td>
<td>4</td>
</tr>
</tbody>
</table>

Table 3: Classification of irritation according to Primary dermal irritation index

<table>
<thead>
<tr>
<th>PDI</th>
<th>Classification</th>
</tr>
</thead>
<tbody>
<tr>
<td>Less than 0.5</td>
<td>No irritation</td>
</tr>
<tr>
<td>0.5-2.0</td>
<td>Irritation barely perceptible</td>
</tr>
<tr>
<td>2-5.0</td>
<td>Moderate irritation</td>
</tr>
<tr>
<td>More than 5</td>
<td>Severe irritation</td>
</tr>
</tbody>
</table>

Statistical analysis of data

Data were expressed as mean ± SD. Statistical evaluation was performed by one way analysis of variance (ANOVA) at a significance level of p<0.05 by Dunnett’s multiple comparison test using GraphPad prism software version 4.03.

RESULTS

The physicochemical properties of the matrix type transdermal films of Amitriptyline HCl are recorded in Table 4.

Drug content determination

The drug content of 2 cm\(^2\) film of F1 to F6 was found to be in the range of 9.87±0.31 mg to 9.95±0.24 mg. The results of the drug content analysis are given in Table 5.
Table 4: Physicochemical properties of amitriptyline HCl transdermal films

<table>
<thead>
<tr>
<th>Formulation Code</th>
<th>1WVTR</th>
<th>2% MA</th>
<th>3% ML</th>
<th>Thickness</th>
<th>Weight (mg)</th>
</tr>
</thead>
<tbody>
<tr>
<td>F1</td>
<td>0.049±0.002</td>
<td>4.58±1.12</td>
<td>1.60±0.51</td>
<td>0.57±0.042</td>
<td>35±1.56</td>
</tr>
<tr>
<td>F2</td>
<td>0.012±0.003</td>
<td>3.21±1.59</td>
<td>0.96±0.41</td>
<td>0.51±0.06</td>
<td>34±1.65</td>
</tr>
<tr>
<td>F3</td>
<td>0.01±0.002</td>
<td>4.15±0.98</td>
<td>0.86±0.42</td>
<td>0.55±0.047</td>
<td>34±1.25</td>
</tr>
<tr>
<td>F4</td>
<td>0.014±0.004</td>
<td>3.56±1.42</td>
<td>1.21±0.52</td>
<td>0.51±0.008</td>
<td>34±1.11</td>
</tr>
<tr>
<td>F5</td>
<td>0.017±0.002</td>
<td>6.41±1.98</td>
<td>2.31±0.61</td>
<td>0.54±0.008</td>
<td>35±1.62</td>
</tr>
<tr>
<td>F6</td>
<td>0.01±0.003</td>
<td>3.06±1.32</td>
<td>0.75±0.43</td>
<td>0.52±0.007</td>
<td>35±1.21</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD (n=6); 1 Water vapour transmission rate; 2 Percentage moisture absorption; 3 Percentage moisture loss.

Table 5: Drug content of amitriptyline HCl films

<table>
<thead>
<tr>
<th>S. No</th>
<th>Formulation code</th>
<th>Drug content (mg)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>F1</td>
<td>9.95±0.24</td>
</tr>
<tr>
<td>2</td>
<td>F2</td>
<td>9.87±0.31</td>
</tr>
<tr>
<td>3</td>
<td>F3</td>
<td>9.86±0.48</td>
</tr>
<tr>
<td>4</td>
<td>F4</td>
<td>9.91±0.26</td>
</tr>
<tr>
<td>5</td>
<td>F5</td>
<td>9.94±0.31</td>
</tr>
<tr>
<td>6</td>
<td>F6</td>
<td>9.88±0.26</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD (n=3)

**In vitro drug release study**

The results obtained from the in vitro release study are shown in the Figure 1. The cumulative percent of drug released from F5 (96.45±1.24% in 24 hrs) was significantly (p<0.05) high when compared with other formulations. Film F6 exhibited a lowest drug release of 78.87% ±1.32%. All the formulations showed an initial burst release followed by controlled release that has been extended for 24 hours. Highest Regression-coefficient ($R^2$) value was obtained for the zero order kinetic model for F5 and the results are given in table 6. The calculated “n” value from the slope of korsmeyer-peppas equation was 0.441 (i.e. n<0.5).

**In vitro skin permeation study**

The formulation F5 was selected for the permeation study based on its highest in vitro release. The drug permeation from the film F5 and aqueous solution of the drug were 49.27%±0.62% in 24hrs and 94.30%±0.58% in 14 hrs respectively. In vitro skin permeation of film F5 is shown in Figure 2.

**Skin irritation study**

The results of the skin irritation studies are shown in Table 7. The calculated primary dermal irritation index value was zero.
Animal no drug across the matrix may relax the polymeric network enabling rapid diffusion of the drug. Higher uptake of water allows water to diffuse more easily into the film which results in the more hydrophilic property of PVP when compared to HPC. PVP increase with increase in PVP concentration. This might be due to remained uniform and the WVTR, % MA, % ML were found to formulations F1 to F6. Thickness, weight of all the formulations indicated that there is no much difference among the ML which might be attributed to the hydrophobic nature of eudragit E 100. However, a small percentage of moisture present in the film may help in retaining the flexibility, reduces brittleness during their storage. The uniform drug content in all the formulations showed the homogenous dispersion of the drug in the entire film. The miscibility of drug, polymer and the plasticizer in the chosen solvents may be responsible for the homogenous dispersion of the drug in the film.

The results of the in vitro drug release clearly indicates that the hydrophilic polymer Eudragit E100 resists the release of hydrophilic drug Amitriptyline HCl whereas the addition of hydrophilic polymers PVP and HPC increases the release of Amitriptyline HCl from the transdermal films. Initial burst release followed by slow release was observed for the formulations containing PVP and HPC. The burst release might be due to the hydrophilic polymers which do not have interaction with the hydrophilic drug. Whereas the eudragit E 100 that account for the slow release of drug may be due to the interaction of this hydrophobic polymer with the hydrophilic drug.The burst release obtained may also be due to the migration of the drug towards the surface of the polymeric matrix when it comes into contact with the dissolution medium. Here the crystallization of drug substances might have been prevented by the addition of PVP. The initial burst release helps to achieve minimum effective plasma drug concentration quickly. However PVP shows higher drug release effect than HPC. This may be due to the raise in solubility of the drug by PVP. It has been suggested that the PVP might have interrupted with the polymer chain continuity greatly than the HPC, hence offering much matrix relaxation for rapid diffusion of the drug. It has been observed that the increase in concentration of PVP increases the drug release rate. The drug release from the transdermal film F5 followed the fickian (diffusion controlled) transport mechanism. There was no lag time observed with respect to skin permeation of diclofenac diethanolamine and chlorpheniramine transdermal film. Further the combination of eudragit E 100/PVP has been reported as the best dissolution medium. Here the crystallization of drug substances obtained may also be due to the migration of the drug towards the surface of the polymeric matrix when it comes into contact with the dissolution medium. Here the crystallization of drug substances might have been prevented by the addition of PVP. The initial burst release helps to achieve minimum effective plasma drug concentration quickly. However PVP shows higher drug release effect than HPC. This may be due to the raise in solubility of the drug by PVP. It has been suggested that the PVP might have interrupted with the polymer chain continuity greatly than the HPC, hence offering much matrix relaxation for rapid diffusion of the drug. It has been observed that the increase in concentration of PVP increases the drug release rate. The drug release from the transdermal film F5 followed the fickian (diffusion controlled) transport mechanism. There was no lag time observed with respect to skin permeation of diclofenac diethanolamine and chlorpheniramine transdermal film. Further the combination of eudragit E 100/PVP has been reported as the best from blends of HPC, PVP and Eudragit E 100 using dibutyl phthalate as plasticizer was thin, flexible and smooth. The film showed satisfactory physicochemical performance. From the in vitro release and skin permeation results it can be concluded that the controlled release of Amitriptyline HCl across the skin could be achieved for prolonged period. No skin irritation was observed for the optimized formulation F5. The result clearly indicates that hydrophobic eudragit E100 resists the release of hydrophilic drug Amitriptyline HCl whereas the addition of hydrophilic polymers PVP and HPC.

**DISCUSSION**

Amitriptyline HCl transdermal film was designed using an adhesive polymer matrix of eudragit E 100 for the controlled release of the drug. Total of six different formulations were prepared by varying the composition of HPC and PVP. The eudragit E 100 (hydrophobic polymer) composition was fixed so as to study the effect of HPC and PVP (hydrophilic polymers) on drug release. The added plasticizer dibutyl phthalate at a concentration of 30% w/w of polymer, helps increasing the drug release in addition to impart flexibility to the film. Plasticizer act by binding with the polymer chains and thus may relax the polymeric network enabling rapid diffusion of the drug across the matrix. The physicochemical properties of the films indicated that there is no much difference among the formulations F1 to F6. Thickness, weight of all the formulations remained uniform and the WVTR, % MA, % ML were found to increase with increase in PVP concentration. This might be due to the more hydrophilic property of PVP when compared to HPC. PVP allows water to diffuse more easily into the film which results in higher uptake of water. The film F6 showed least WVTR, % MA, % ML which might be attributed to the hydrophobic nature of eudragit E 100. However, a small percentage of moisture present in the film may help in retaining the flexibility, reduces brittleness during their storage. The uniform drug content in all the formulations showed the homogenous dispersion of the drug in the entire film. The miscibility of drug, polymer and the plasticizer in the chosen solvents may be responsible for the homogenous dispersion of the drug in the film.

**Table 7:** Score obtained in the skin irritation test for F5

<table>
<thead>
<tr>
<th>Animal no</th>
<th>Sex</th>
<th>Score obtained after a period of 1hr</th>
<th>24hrs</th>
<th>48hrs</th>
<th>72hrs</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Male</td>
<td>0/0</td>
<td>0/0</td>
<td>0/0</td>
<td>0/0</td>
</tr>
<tr>
<td>2</td>
<td>Male</td>
<td>0/0</td>
<td>0/0</td>
<td>0/0</td>
<td>0/0</td>
</tr>
<tr>
<td>3</td>
<td>Male</td>
<td>0/0</td>
<td>0/0</td>
<td>0/0</td>
<td>0/0</td>
</tr>
</tbody>
</table>

**Fig. 2:** In vitro skin permeation of Amitriptyline HCl film (F5)

**CONCLUSION**

Amitriptyline HCl adhesive matrix type transdermal film prepared from blends of HPC, PVP and Eudragit E 100 using dibutyl phthalate as plasticizer was thin, flexible and smooth. The film showed satisfactory physicochemical performance. From the in vitro release and skin permeation results it can be concluded that the controlled release of Amitriptyline HCl across the skin could be achieved for prolonged period. No skin irritation was observed for the optimized formulation F5. The result clearly indicates that hydrophobic eudragit E100 resists the release of hydrophilic drug Amitriptyline HCl whereas the addition of hydrophilic polymers PVP and HPC.
increases the drug release. Amitriptyline HCl transdermal matrix film containing eudragit E100 and PVP has shown promising results than the film containing eudragit E100 and HPC.

REFERENCES


