Development of a New Carvedilol Tablet with Rapid Onset of Action

Amna M.A. Makky, Iman S. Ahmed, Mohamed A. El-Nabarawi and Rehab A. Abd El-Moneim

1Department of Pharmaceutics and Industrial Pharmacy, Faculty of Pharmacy, Cairo University, Kasr-El-Eini Street, Cairo, Egypt
2Department of Pharmacology and Pharmaceutics, College of Pharmacy, University of Sharjah, Sharjah, 27271, United Arab Emirates
3Department of Pharmaceutics and Industrial Pharmacy, College of Pharmacy, Misr University for Science and Technology, Sixth October City, Egypt

Corresponding Author: Iman S. Ahmed, Department of Pharmaceutics and Industrial Pharmacy, Faculty of Pharmacy, Cairo University, Kasr-El-Eini Street, Cairo, Egypt Tel: (20) 101648567

ABSTRACT

The purpose of this study was to develop Freeze-dried Tablets (FDTs) for rapid absorption of carvedilol, an antihypertensive drug with poor solubility and poor bioavailability. Binary and ternary systems were first made of drug and different ratios of HPMC and Inutec and then were formulated into FDTs. Formulation of FDTs was based on 2^3 factorial design to study the influence of formulation variables namely, concentration of polymer matrix-former, concentration of collapse protectant and type of polymer used in formation of drug binary and ternary systems on tablet characteristics such as friability, in vitro disintegration time, in vivo disintegration time and initial dissolution. An optimized FDT formulation (RO) containing 2% gelatin, 1% glycine and binary system made of Drug: Inutec in the ratio 1:0.5 showed excellent mechanical strength, in vitro and in vivo disintegration times of less than 10 sec and more than 30% drug dissolved during the first 3 min compared to only 11% for the drug powder and 8% drug from an immediate release commercially available tablet (Carvepress). When the optimized FDT (RO) was administered to hypertensive rats it resulted in a significant decrease ($p = 0.0015$) in systolic blood pressure within the first 15 min compared to the immediate release conventional tablet as a reference which indicate the rapid dissolution and absorption of carvedilol from the FDT.

Key words: Carvedilol, solid dispersions, freeze-dried tablets, $2^3$ factorial design, anti-hypertensive activity

INTRODUCTION

Carvedilol, a long-acting beta-blocker used in heart diseases and hypertension (Al-Rejaie, 2009), suffers from low bioavailability (25-35%) and highly variable serum concentrations among patients because of its low solubility and high first-pass metabolism (Talbert, 2004; Giessmann et al., 2004). Carvedilol belongs, according to the Biopharmaceutic Classification System (BCS), to Class II drugs with poor solubility and high permeability (Amidon et al., 1995; Kasim et al., 2004) and is present in the market in 3.125, 6.25, 12.5 and 25 mg tablets (Frishman, 1998).
Several different approaches have been used to improve the dissolution and oral bioavailability of poorly water soluble drugs such as formation of solid dispersions (Badawi et al., 2011), inclusion complexes (Khan, 2001) and formation of rapidly disintegrating tablets (Balamuralidhara et al., 2009). The bioavailability of carvedilol has been modified by incorporating the drug in mucoadhesive patches for buccal administration using several polymers which resulted in a 2.29 folds increase in drug bioavailability compared to an oral solution (Vishnu et al., 2007), also by incorporating carvedilol in a matrix-type transdermal patches made of different hydrophilic and hydrophobic polymers which improved the bioavailability of the drug by 71% when compared to oral carvedilol (Ubaidulla et al., 2007). In another study, carvedilol was incorporated in a nanoemulsifying tablet which significantly improved the rate and extent of drug dissolution when compared to the drug powder (Mahmoud et al., 2009).

The most important drug delivery route is undoubtedly the oral route as it offers advantages of convenience of administration and potential manufacturing cost savings. The use of different polymers in preparation of oral solid dosage forms is widely applied in obtaining specific release characteristics (Balamuralidhara et al., 2011; Khan and Zhiy, 2001; Khan et al., 2001; Sabar et al., 2011; Shaheen et al., 2006). Freeze-dried oral dosage forms were reported to improve the in vitro dissolution and in vivo absorption of many drugs of low aqueous solubility (Topaloglu et al., 1999; Van Drooge et al., 2004; Ahmed and Aboul-Einion, 2007; Shoukri et al., 2009). Freeze-dried dosage forms also have many advantages such as rapid reconstitution, good preservation and stability. Tablets that rapidly disintegrate in the mouth upon contact with saliva are especially useful for patients who find it difficult to swallow solid dosage forms such as tablets and capsules.

The aim of this study is to prepare rapidly disintegrating and dissolving Freeze-dried Tablets (FDT) of carvedilol and to evaluate the performance of these tablets both in vitro and in vivo. In this study, preparation of carvedilol tablets was based on subsequent steps. The first step included the preparation and evaluation of binary and ternary solid dispersion systems of drug with hydroxypropylmethyl cellulose (HPMC) and Inutec (I) using the solvent evaporation method. The second step included the incorporation of selected solid dispersion systems along with other excipients into freeze-dried tablets based on a 2² factorial design. An optimized formulation was further characterized and tested in hypertensive rats. Therefore, a FDT of carvedilol which disintegrates rapidly and dissolves in the buccal cavity is expected to be mainly absorbed from the buccal mucosa and partly from the GIT which might result in rapid onset of action and to an increase in the fraction of drug reaching the systemic circulation due to bypassing first-pass metabolism.

MATERIALS AND METHODS

Materials: Carvedilol was provided by Hetero drugs (India). HPMC 5 ep (Methocel® E5LV) were supplied by Colorcon (UK). Inutec SP1 was obtained from orafi bio based chemicals (Belgium). Mannitol was kindly supplied from Roquette Pharma, France. Tween 80, sodium chloride and potassium chloride (Adwic, El-Nasr Pharmaceutical Chemicals Co., Egypt). All other chemicals were reagent grade and used as received. Carvepress® 12.5 mg (Global Nappi, Egypt) was used as a reference tablet in in-vitro and in-vivo studies. L-NAME (N-nitro-L-arginine methyl ester) was purchased from Sigma (USA).

Preparation of carvedilol binary and ternary solid dispersions: Binary solid dispersions of carvedilol with HPMC as a hydrophilic polymeric carrier in the ratio of 1:1, 1:2 and 1:4 (w/w) drug: carrier (D:H) and with Inutec SP1 as surfactant in the ratio of 1: 0.5 and 1:1 and 1: 2 (w/w) drug:
carrier (D:I) were prepared using the solvent evaporation method. The calculated amounts of carvedilol and carrier were dissolved in the least amount of a 1:1 mixture of methanol and methylene chloride and the solvent was then evaporated in a rotavapor vacuum (Buchi, Switzerland) at 60°C to ensure removal of residual solvent. Ternary systems were prepared by incorporating the surfactant Inutec SP1 into the binary HPMC systems as the third component to prepare ternary solid dispersions. Ternary systems were prepared in a similar way to binary systems however, the drug:carrier:surfactant (D:H:I) ratios used were 1:1:1 and 1:1:0.5 (w/w). The solid mass obtained after solvent evaporation was further dried in an oven at 60°C until nearly constant weight was obtained. The resulting solid dispersions were kept over anhydrous calcium chloride in desiccators. The dried mass was pulverized and sieved and the fraction of the powder that passed through a size 60 sieve and retained on a size 80 sieve was collected and used for further investigations.

**Evaluation of carvedilol solid dispersions**

**Determination of drug content:** Binary and ternary solid dispersions were evaluated for drug content by dissolving an amount of solid dispersion equivalent to 12.5 mg of carvedilol in a 1:1 solvent mixture of methanol and methylene chloride. The solution was then assayed for drug content spectrophotometrically at 284 nm after appropriate dilution.

**Determination of carvedilol saturated solubility in binary and ternary systems:** The saturated aqueous solubility of carvedilol was determined for the pure powder drug and the prepared binary and ternary systems. An excess amount of the sample to be tested was added to 10 mL of simulated saliva fluid (SSF) (pH = 6.8) in 30 mL screw-capped vials. The vials were shaken for 48 h in an incubator shaker at 37°C. The obtained solutions were filtered using 0.45 μm Millipore filters and were assayed for drug content spectrophotometrically at 285 nm after appropriate dilution. All experiments were run in triplicates (n = 3).

**In-vitro dissolution studies:** The dissolution profiles of carvedilol from binary and ternary systems compared with the plain drug were determined in a dissolution tester (Pharma Test Dissolution Tester, Germany) following the USP paddle method. All tests were conducted in 300 mL SSF without enzymes at pH = 6.8. The dissolution medium was maintained at a temperature of 37±0.5°C with a paddle rotation speed at 50 r.p.m. The amount of drug used was equivalent to 12.5 mg. At specified time intervals (5, 10, 15, 30, 60, 90, 120, 150 and 180 min), 3 mL of dissolution media were withdrawn and replaced with an equal volume of fresh medium. Samples were filtered and assayed for drug content spectrophotometrically at 285 nm after appropriate dilution. Cumulative amount of drug dissolved in the preparations was calculated using calibration equation. Dissolution tests were performed in three vessels per formulation (n = 3).

**Fourier-transform Infrared Spectroscopy (FTIR) studies:** Systems giving best dissolution results were further studied for possible chemical interaction between carvedilol and carriers using FTIR. FTIR spectra in the range of 4000 and 500 cm⁻¹ for the plain drug, the carriers and selected solid dispersions were determined using the KBr disc technique.

**Differential Scanning Calorimetry (DSC) studies:** Physical interaction between carvedilol and carriers were studied using DSC. 5 mg samples were sealed in aluminum pans and analyzed using a Shimadzu DSC-60 (Kyoto, Japan). The samples were heated in an atmosphere of nitrogen and
Table 1: Factorial Experimental Design for Carvedilol FDTs

<table>
<thead>
<tr>
<th>Formulation</th>
<th>Matrix former (gelatin)</th>
<th>Collapse protectant (glycine)</th>
<th>0.5% w/v</th>
<th>1% w/v</th>
</tr>
</thead>
<tbody>
<tr>
<td>D:H:1 1:1:0.5 (w/w)</td>
<td>1% w/v</td>
<td>R1</td>
<td>R2</td>
<td></td>
</tr>
<tr>
<td></td>
<td>2% w/v</td>
<td>R5</td>
<td>R4</td>
<td></td>
</tr>
<tr>
<td>D:1 1:0.5 (w/w)</td>
<td>1% w/v</td>
<td>R3</td>
<td>R6</td>
<td></td>
</tr>
<tr>
<td></td>
<td>2% w/v</td>
<td>R7</td>
<td>R8</td>
<td></td>
</tr>
</tbody>
</table>

Table 2: Composition of Carvedilol FDT formulations based on 23 factorial design

<table>
<thead>
<tr>
<th>Ingredients (mg)</th>
<th>R1</th>
<th>R2</th>
<th>R3</th>
<th>R4</th>
<th>R5</th>
<th>R6</th>
<th>R7</th>
<th>R8</th>
</tr>
</thead>
<tbody>
<tr>
<td>D:H:1 (1:1:0.5)</td>
<td>15.65</td>
<td>15.65</td>
<td>15.65</td>
<td>15.65</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>D:1 (1:0.5)</td>
<td></td>
<td>9.40</td>
<td></td>
<td></td>
<td>9.40</td>
<td>9.40</td>
<td>9.40</td>
<td>9.40</td>
</tr>
<tr>
<td>Gelatin</td>
<td>10.00</td>
<td>10.00</td>
<td>10.00</td>
<td>10.00</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Glycine</td>
<td>5.00</td>
<td>5.00</td>
<td>5.00</td>
<td>5.00</td>
<td>5.00</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Oxytetracycline</td>
<td>10.00</td>
<td>10.00</td>
<td>10.00</td>
<td>10.00</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mannitol</td>
<td>15.65</td>
<td>15.65</td>
<td>21.25</td>
<td>6.60</td>
<td>5.60</td>
<td>16.85</td>
<td>11.85</td>
<td>6.85</td>
</tr>
<tr>
<td>Total weight</td>
<td>46.25</td>
<td>46.25</td>
<td>46.25</td>
<td>46.25</td>
<td>46.25</td>
<td>46.25</td>
<td>46.25</td>
<td>46.25</td>
</tr>
</tbody>
</table>

Thermograms were obtained by heating at a constant heating rate of 10°C min⁻¹ in the range of 20-350°C. Thermograms for drug plain powder, carriers and carvedilol in selected solid dispersions were obtained.

**Powder X-Ray Diffraction (XRD):** The crystalline nature of the prepared solid dispersions was studied using X-ray diffraction experiments. Diffraction patterns of carvedilol plain powder and carvedilol in selected solid dispersions were obtained using a Scintag x-ray diffractometer (USA) using Cu Kα radiation with a nickel filter, a voltage of 45 kV and a current of 40 mA.

**Preparation of carvedilol tablets (FDTs):** Solid dispersions showing acceptable physical and chemical properties were made into carvedilol FDTs. A 2³ factorial design was used for the formulation of carvedilol FDTs. The experimental design was built up to study the effect of three types of variables, namely: matrix former gelatin present at two levels (1% w/v, 2% w/v), collapse protectant glycine at two levels (0.5% w/v, 1% w/v) and carriers present in selected solid dispersions at two levels (D:H:1 and D:1). The different factors, levels of factorial experiments and the composition of FDT tablets are presented in Table 1 and 2. The design was used to evaluate the individual and combined effects of the formulation variables on tablet friability, *in vitro* and *in vivo* disintegration times and the initial dissolution (% drug dissolved at 3 min). Eight tablet formulae were prepared from this design and all tablets contained modified drug dispersion, matrix former, collapse protectant and mannitol as sweetening agent and filler. Tablets were prepared as follows: an aqueous solution of gelatin, mannitol and glycine was prepared in predetermined concentration, an accurately weighed amount of carvedilol solid dispersion powder was dispersed in the prepared aqueous solution to result in a dose of 6.25 mg carvedilol per one milliliter; the suspension was then poured in each pocket of a PVC blister pack to result in a dose of 6.25 mg per tablet. The tablet blister packs were then freeze-dried at -22°C for 24 h and the frozen tablets were placed in a lyophilizer.
for 48 h using a Novalypho-NL 500 Freeze Dryer. The FDTs had a diameter of 13 mm and a depth of 3 mm. The prepared FDTs were kept in tightly closed containers in desiccators over anhydrous calcium chloride (25% relative humidity) at room temperature until further use.

**Characterization of carvedilol FDTs**

**Evaluation of carvedilol FDTs:** The prepared tablets from the different formulations were verified for uniformity of weight and content according to the Council of Europe (2004). The mean weight is expressed as mg±SD. The uniformity of content was determined by crushing ten tablets from each formulation and determining the content of each tablet individually. Stage one criterion according the EP for 6.25-mg carvedilol tablets is that carvedilol content must fall in the range of = 5.31 to = 7.19 mg with mean carvedilol content of 5.94±0.56 mg (Council of Europe, 2004; Smith et al., 2006). Tablets were analyzed for their residual moisture content after lyophilization using Karl Fischer titrator (VeegoMatic-MD, Veego Instruments Corporation, India). The friability was determined using the drum of friabilator (Erweka type, GmbH, Germany) where the tablets were rotated at 25 r.p.m for a period of 4 min. The percentage loss in weight was calculated and taken as a measure of friability. Mean disintegration time of tablets from each formulation was determined in minutes (min±S.D) using a DST-3 disintegration tester (Logan Instruments Corp., NJ, USA) according to Council of Europe (2004) specifications. The medium used was SSF (pH = 6.8) at 37°C. The dissolution of carvedilol from FDTs was performed in 300 mL SSF at pH = 6.8 and in 900 mL of 0.1 N HCl (SGF) at 37±0.5°C with a paddle rotation speed at 50 r.p.m. Aliquots from dissolution media were withdrawn at 3, 5, 10, 15, 20, 30, 40 and 60 min. Samples were filtered through 0.45 μm millipore filter and assayed for drug content spectrophotometrically at 285 nm after appropriate dilution. Dissolution tests were performed in three vessels per formulation (n = 3). Dissolution results of carvedilol from FDTs were also compared to the plain drug and the market product Carvepress.

**In vivo disintegration time:** The in-vivo disintegration time of the prepared FDTs was evaluated in six human volunteers after giving informed written consent. Each of the six subjects was given a coded tablet. Tablets were placed on the tongue and immediately the time was recorded. The subjects were allowed to move the tablet against the upper palate of the mouth with their tongue without biting on or tumbling the tablet from side to side. After the last noticeable mass had disintegrated, the time was immediately recorded. After tablet disintegration the subjects spitted out the content of the oral cavity and rinsed their mouth with water (Abdelbary et al., 2005). The test results are presented as mean value±SD (n = 6).

**Physico-pharmaceutical characterization of selected carvedilol FDTs:** The optimized FDT formulation (RO) was further characterized using DSC studies and FTIR studies as described before. Surface morphology and cross sections of the optimized formulation was also examined using Jeol JSM-6400 scanning electron microscope (Tokyo, Japan). Cross-section samples were prepared by cutting a thin slice of the tablet using a scalpel. Photographs were taken at magnification of 100.

**Determination of anti-hypertensive activity of carvedilol from FDT RO**

**Study design:** The study was carried out to compare the antihypertensive properties of carvedilol from FDT RO formulation (Treatment A) to Carvepress (Global Nappi, Egypt) a conventional
commercially available carvedilol immediate release tablet (Treatment B) using the Tail Cuff method in hypertensive rats (Ubaidulla et al., 2007; Amer, 1977; Tanaka et al., 1987; Zhao et al., 2007) after single oral administration of 1.56 mg carvedilol for each rat orally. The animal experiments were conducted in full compliance with local, national, ethical and regulatory principles and local licensing regulations, per the spirit of Association for Assessment and Accreditation of Laboratory Animal Care (AAALAC) International's expectations for animal care and use. The study protocol was approved by the Institutional Animal Care and Use Committee (IACUC) in the College of Pharmacy at Cairo University. Adult male albino rats, weighing 220-250 g were obtained from the Research Institute of Ophthalmology (Giza, Egypt) and were distributed randomly into two groups of equal size (n = 6) using a non-blind, two-treatment, randomized, parallel design. The commercial tablet was crushed and suspended and each rat was given a volume of suspension equivalent to 1.56 mg carvedilol by intubation (Treatment B), while the FDI (Treatment A) was divided into 4 equal parts using a scalpel to result in four doses each equal to 1.56 mg. Each quarter tablet was then inserted over the moist tongue of the rat. The rats were kept on standardized diet and allowed free access to water.

**L-NAME induced hypertension:** Rats were treated for four weeks with N-nitro-L-arginine methyl ester (L-NAME) 50 mg kg\(^{-1}\) by oral gavages to induce a persistent elevation of systolic blood pressure (Jun and Wennmalm, 1994). Treatment with the antihypertensive drugs started after four weeks from the beginning of induction of hypertension. Rats used for the study are those with systolic blood pressure had reached values of 171±67 mmHg. The normal blood pressure for rats is 118.5±2.09 mmHg.

**Measurement of systolic blood pressure:** Indirect blood pressure was measured by the tail-cuff method which represents a non-invasive blood pressure measurement. This indirect rat tail blood pressure system quickly and reliably determines the systolic blood pressure from the tail of a conscious rat at room temperature. Briefly the system includes a pneumatic cuff assembly that fits over the rat’s tail. The cuff assembly consists of an inflatable latex occlusion cuff, light source and photo detector. A hand inflation bulb is used to inflate the cuff occluding the blood flow. In practice, the cuff is inflated to occlude the pulse and then allowed to deflate slowly until the pulse pressure is observed on the pulse channel of the recorder. Both cuff pressure and pulse are recorded simultaneously. The pressure at which the pulse first appears is considered to be the systolic pressure. Rats are placed in dark plastic restrainers, simulating a dark hole, thus putting the rat at ease during measurements. The rats are placed in restrainers 15 to 30 min prior to taking readings. Rats that show habituation to the restrainers with one or two training sessions are desirable. The rats are placed in the restrainers so that they face the adjustable front gate and their tail comes out of the fixed position tail gate. The front gate should be placed so that the rat has enough room to be comfortable, but not be able to move back and forth much. A slot in the restrainer base holds the cuff in position on the tail. A series of cuff inflation were done, usually about six, to get a least four systolic blood pressure readings. The readings should not vary more ±5 mmHg in range. Systolic blood pressure was measured at 0, 15, 30, 60 and 120 min after each treatment administration.

**Statistical analysis:** The effect of coefficients (factors at low and high level) on tablet friability, *in vitro* and *in vivo* disintegration times and the initial dissolution (at 3 min) were analyzed for the
eight tablet formulae using the software Design-Expert®8 and value of effect of coefficients obtained from statistical analysis is interpreted. The effect of interaction between the factors on the chosen responses was also studied. ANOVA test was applied for estimating the significance of the model at 5% significance level. Descriptive statistics for the in vivo studies were provided for both treatments. An analysis of variance was performed to compare between Treatment A and Treatment B. The level of significance was $\alpha = 0.05$. A p-value of ≤0.05 was considered statistically significant.

RESULTS AND DISCUSSION
Evaluation of carvedilol binary and ternary systems: The mean % carvedilol content in different carrier systems ranged from 95 to 105% indicating homogeneous dispersions. Saturated aqueous solubility studies in SSF (pH = 6.8) showed an increase in carvedilol supersaturated solubility from all tested binary and ternary systems when compared to the plain drug. The increase in solubility of carvedilol from HPMC systems, nearly three to four times higher when compared to the solubility of the plain drug, could be due to the solubilizing effect of the highly water soluble polymer. The increase in solubility of carvedilol from Inutee binary systems, nearly five to six times higher, could be due to improved wettability by reducing the solid/liquid interfacial tension and by adsorption of the surfactant onto drug particle surface thus allowing the drug particles to present a larger surface area (Sinswet et al., 2005). It has to be noted that wettability is very important to prevent aggregation of solid particles when exposed to the aqueous medium of the GI fluid which may affect in vivo absorption due to reduced initial in vivo drug surface area. When Inutee was added to HPMC binary systems, the supersaturated solubility was also significantly increased compared to binary systems containing HPMC alone (p<0.05) but was not significantly different when compared to D:H binary systems (p>0.05). It was also observed that addition of 0.5 part or 1 part of Inutee to D:H systems is more effective in increasing the drug supersaturated solubility when compared to increasing the ratio of HPMC itself in binary systems. For example increasing the ratio of HPMC from 1:1 to 1:4 in D:H binary systems resulted in an increase in solubility from 47 to 82 µg mL$^{-1}$, while adding 0.5 part of Inutee to the 1:1 D:H system resulted in an increase in solubility from 47 to 92 µg mL$^{-1}$. It has to be noticed that using a high polymer ratio is not economic in industrial processing and usually leads to more unstable systems. When Inutee ratio was increased from 0.5 to 1 part in ternary systems made of D:H no significant increase in carvedilol saturated solubility was observed (p>0.05). The saturated aqueous solubility from all tested systems is summarized in Table 3.

<table>
<thead>
<tr>
<th>System</th>
<th>Ratio</th>
<th>Solubility (µg mL$^{-1}$)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Plain drug</td>
<td></td>
<td>18.77±0.155</td>
</tr>
<tr>
<td>D:H</td>
<td>1:01</td>
<td>47.80±1.372</td>
</tr>
<tr>
<td></td>
<td>1:02</td>
<td>63.64±0.980</td>
</tr>
<tr>
<td></td>
<td>1:04</td>
<td>81.94±7.990</td>
</tr>
<tr>
<td>D:I</td>
<td>1:0:5</td>
<td>87.22±0.155</td>
</tr>
<tr>
<td></td>
<td>1:01</td>
<td>105.50±2.257</td>
</tr>
<tr>
<td></td>
<td>1:02</td>
<td>114.35±0.995</td>
</tr>
<tr>
<td>D:H:I</td>
<td>1:1:0.5</td>
<td>91.68±1.860</td>
</tr>
<tr>
<td></td>
<td>1:0:1:01</td>
<td>94.67±6.080</td>
</tr>
</tbody>
</table>
Fig. 1: Dissolution profiles of carvedilol (D) from its binary and ternary systems with HPMC (H) and Inutec (I) and drug powder alone in SSF (pH = 6.8) at 37°C

**In vitro dissolution studies:** Percentage carvedilol dissolved from binary and ternary systems compared to plain drug powder is shown in Fig. 1. Dissolution results were in accordance with saturated solubility results in that binary and ternary systems increased the percentage drug dissolved significantly when compared to powder drug alone (p<0.05). The initial dissolution rate of the drug was 6 to 15 times higher from binary and ternary systems within the first 5 min when compared to the plain drug. The 1:0.5 D:I system showed the highest dissolution rate and the highest percentage drug dissolved when compared to other systems which could be due to rapid dispersion of drug powder leading to improved wetting thus allowing the drug particles to present a large surface area and thereby to increased dissolution rate. The average percent of carvedilol dissolved within 15 min from the plain drug, the 1:0.5 D:I, the 1:1 D:H and 1:1:0.5 D:H:I systems was 5, 35, 21.5 and 29%, respectively. It was observed that addition of 0.5 part of Inutec to the 1:1 D:H system almost doubled the initial dissolution rate (at 5 min) and also slightly increased the percentage drug dissolved over 90 min when compared to the binary system alone. However, when the ratio of Inutec added to the HPMC binary system was increased to 1, the initial dissolution rate and the percentage drug dissolved was reduced which could be due to entrapment of drug within micelles at this higher ratio of surfactant. The same pattern was observed with the 1:1 D:I system which showed a significant reduction in initial dissolution rate and the overall drug dissolved over 90 min when compared to the 1:0.5 D:I system. It has to be noted that the cumulative percentage of drug dissolved from all systems did not exceed 50% over 90 min.

**Physicopharmaceutical characterization of carvedilol solid dispersions:** To evaluate the extent of interaction between the reactive groups of drug and carrier molecules, IR spectroscopy was used. Crystalline carvedilol presented characteristic bands at 3344 mL (O-H and N-H stretching vibration bends merging together), 2922, 2879 and 2839 mL (C-H stretching vibrations), 1590 mL(N-H bending vibrations), 1250 and 1041 mL (O-H bending and C-O stretching vibrations). The spectra of binary and ternary solid dispersions were compared with those of initial materials. Interaction between the drug and carriers often lead to noticeable changes in IR spectra. Results show that there was no major change in band intensity associated with structural characteristic of drug molecule. Figure 2 illustrates examples of IR spectra of powder drug and its solid dispersions.
Fig. 2: (a) FT-infrared spectra of carvedilol, (b) carvedilol binary system with Inutec in the ratio of 1:0.5 and (c) carvedilol ternary system with HPMC and Inutec in the ratio of 1:1:0.5

To evaluate the crystalline state of carvedilol in binary and ternary systems, DSC studies were carried out on drug plain powder, carriers, binary and ternary systems. The pattern of powder drug was typical of a highly crystalline compound, characterized by an endotherm at 111.92°C with an onset at 106.91 and an endset at 116.00°C corresponding to the melting point of the drug. The thermogram of HPMC E5 showed a broad endothermic peak at 63.2°C with an onset at 20.8 and an endset at 111.60°C while Inutec SP1 showed a broad endothermic peak at 65.77°C with an onset at 34.61°C and an endset at 105.54°C. The DSC thermograms of carvedilol binary systems with HPMC E5 and Inutec showed the characteristic peak of the drug in its place or slightly shifted suggesting the presence of drug in crystalline state. It was observed that carvedilol ternary system with HPMC E5 and Inutec in the ratio 1:1:0.5 showed a strong shift of the peak to the left which might indicate some sort of interaction in this system. Figure 3 illustrates examples of DCS thermograms of powder drug and solid dispersions. These results were further confirmed with XRD studies. The diffraction pattern of carvedilol showed that the drug has high crystallinity because of the presence of numerous distinct peaks at 2θ diffraction angles. These peaks are located at about 5, 12.5, 15, 16, 17.5, 18, 20.5, 21.5, 23, 24, 26, 28.5 and 29 degrees. The results revealed that the sharpness of the peaks of carvedilol in D:H 1:1 and D:I 1:0.5 systems were slightly affected which indicate slight change in the crystallinity of the drug. On the other hand, carvedilol in D:H:I 1:1:0.5 system showed reduction in major drug diffraction peaks indicating that some of the drug was in an amorphous form (disordered state), this partial loss of crystallinity can usually result in an increase in solubility and dissolution rate (Fig. 4). These results could explain the enhancement
Fig. 3: (a) DSC thermograms of carvedilol, (b) carvedilol binary system with HPMC in the ratio 1:1 and (c) carvedilol ternary system with HPMC and Inutec in the ratio 1:1:0.5

Fig. 4: (a) Powder X-ray diffraction spectra of carvedilol, (b) carvedilol binary system with HPMC in the ratio 1:1, (c) carvedilol ternary system with HPMC and Inutec in the ratio 1:1:0.5 and (d) carvedilol binary system with Inutec in the ratio 1:0.5
in solubility and rapid dissolution of this system compared to the others. It has to be mentioned that the presence of Inutec alone with the drug in binary system in the ratio 1:0.5 showed the highest dissolution rate and percentage drug dissolved which could be due to better wetting and rapid dispersion of drug particles due to decrease in interfacial tension thus resulting in rapid dissolution, however, there was no change in the crystallinity of the drug in this system which, from stability considerations, is not altogether disadvantageous.

Based on the results from solubility and dissolution studies, D:I (1:0.5) binary system and D:HI (1:1:0.5) ternary system, were taken to the next state which involved the incorporation of these systems into lyophilized orodispensible tablets (FDTs) as described in Table 2.

**Experimental design of carvedilol FDTs:** In order to investigate the effect of the used exapients and their interaction on the properties of the tablets, a $2^5$ full factorial design with 8 test runs was carried out. This analysis allowed the important factors for the considered responses to be pointed out and the optimum factor level to be selected.

All tablets were located within the acceptable weight variation range; the relative standard deviation of the tablet mass ranged from 0.5% to less than 2% for the different formulations and the mean % drug content in FDTs was found to conform within pharmacopoeial limits (95%-105%). All tablets showed residual moisture content of no more than 7% indicating that the lyophilization process was efficient in removing water from the tablets.

Statistical analysis of friability studies showed that tablets formulated with 2% gelatin as a matrix former showed statistically significantly lower percentage of weight loss on average than those formulated with 1% gelatin ($p = 0.001$). This could be due to larger number of crosslinks and interchain H-bonds formed between the gelatin strands as the concentration increases thus leading to increased mechanical properties (Djagny et al., 2001). On the other hand the type of carrier present in the solid dispersion and the concentration of glycine used had no significant effect on friability ($p = 0.085$ and $p = 0.091$, respectively).

Statistical analysis also revealed that the type of carrier present in the solid dispersion used to form the tablets had a significant effect on in vitro and in vivo disintegration times ($p = 0.023$ and $p = 0.0134$, respectively), the binary system D:I showed significantly shorter in vitro and in vivo disintegration times (7.5 sec and 8.3 sec on average respectively) when compared to the ternary system D:HI (12.75sec and 12.5sec on average respectively). On the other hand the concentration of gelatin and glycine used had no significant effect on in vitro and in vivo disintegration times ($p = 0.059$ and $p = 0.1$, respectively).

The cumulative drug dissolved as a function of time from FDTs compared to drug plain powder and the market product Carvepress in SSF (pH = 6.8) and SGF (pH = 1.2) are illustrated in Fig. 5 and 6. Results from dissolution studies showed that only the type of carrier present in the solid dispersion used to form the tablets had a significant effect on initial dissolution in both SSF and SGF ($p = 0.0214$). The binary system D:I used in tablet formation showed significantly higher initial dissolution during the first three minutes compared to the ternary system D:HI. On the other hand the concentration of gelatin and glycine used had no significant effect on initial dissolution ($p = 0.092$ and $p = 0.071$). These results are consistent with solubility results, dissolution results of solid dispersions and in vitro and in vivo disintegration time results. The percentage of
Fig. 5: Dissolution profiles of carvedilol from different FDTs compared to carvedilol powder alone and carvedilol in commercial tablets in SSF (pH = 6.8) at 37°C

Fig. 6: Dissolution profiles of carvedilol from different FDTs compared to carvedilol powder alone and carvedilol in commercial tablets in SGF (pH = 1.2) at 37°C

weight loss from friability studies, the average in-vitro disintegration time, in-vivo disintegration time and % drug dissolved during the first three minutes (initial dissolution) for the prepared FDTs are listed in Table 4.

Optimization: Based on the above mentioned results a tablet formulation was optimized for the four studied responses and was restricted to 0%≤Y1≤1%, 0≤Y2≤180 sec, 0≤Y3≤60 sec and 15%≤Y4≤25% where Y1, Y2, Y3 and Y4 are the responses for friability, in vitro disintegration time, in vivo disintegration time and initial dissolution expressed as the % drug dissolved during the first three minutes in SSF, respectively. The ranges of responses were based on the required properties
Table 4: The measured responses of the 23 full factorial experimental design

<table>
<thead>
<tr>
<th>Formulation</th>
<th>Friability (%)</th>
<th>In-vitro disintegration time (sec)</th>
<th>In-vitro disintegration time (sec)</th>
<th>Initial dissolution (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>R</td>
<td>10.38</td>
<td>10.6±1.3</td>
<td>13.1±1.34</td>
<td>15.1±2.1</td>
</tr>
<tr>
<td>R2</td>
<td>0.85</td>
<td>11.6±1.8</td>
<td>12.2±1.23</td>
<td>15.2±1.3</td>
</tr>
<tr>
<td>R3</td>
<td>0</td>
<td>50.0±0.6</td>
<td>7.1±3.41</td>
<td>22.2±2.3</td>
</tr>
<tr>
<td>R4</td>
<td>0</td>
<td>14.3±1.6</td>
<td>9.0±1.87</td>
<td>18.3±1.1</td>
</tr>
<tr>
<td>R5</td>
<td>0</td>
<td>15.4±2.1</td>
<td>15.1±2.15</td>
<td>15.1±2.3</td>
</tr>
<tr>
<td>R6</td>
<td>0.96</td>
<td>6.1±2.2</td>
<td>5.1±3.42</td>
<td>28.7±3.3</td>
</tr>
<tr>
<td>R7</td>
<td>0</td>
<td>8.2±2.1</td>
<td>11.8±4.15</td>
<td>18.4±1.2</td>
</tr>
<tr>
<td>R8</td>
<td>0</td>
<td>11.0±1.5</td>
<td>10.0±3.44</td>
<td>25.5±2.3</td>
</tr>
</tbody>
</table>

Data are mean values±SD

Table 5: Predicted and observed responses of the optimized FDT formulation (RO)

<table>
<thead>
<tr>
<th>Variables</th>
<th>Values</th>
<th>Responses</th>
<th>Predicted values</th>
<th>Observed values</th>
</tr>
</thead>
<tbody>
<tr>
<td>X1</td>
<td>D:1 (1:0.5)</td>
<td>Y1</td>
<td>0.0%</td>
<td>0.0%</td>
</tr>
<tr>
<td>X2</td>
<td>Gelatin 2%</td>
<td>Y2</td>
<td>7.2 sec</td>
<td>8.4 sec</td>
</tr>
<tr>
<td>X3</td>
<td>Glycine 1%</td>
<td>Y3</td>
<td>9.9 sec</td>
<td>8.9 sec</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Y4</td>
<td>33.3%</td>
<td>31.1%</td>
</tr>
</tbody>
</table>

for a fast disintegrating and fast-dissolving tablet yet showing sufficient mechanical strength. It has to be noticed that the optimized tablet is expected to disintegrate and dissolve in the mouth where a good fraction of the drug could be absorbed from the buccal mucosa resulting in rapid onset of action; also drug absorbed from the buccal cavity bypass the liver which might result in an increase in drug bioavailability. Optimum values of variables were obtained by numerical analysis using the Design-Expert software and based on the criterion of desirability. The composition as well as predicted and observed values of responses of the optimized formulation (RO) are shown in Table 5. The optimized formulation used D:2binary system (X1) to form the tablet, 2% gelatin (X2) and 1% glycine (X3). Results showed that the observed values were close to the predicted values. Based on these results the optimized formulation (RO) was selected for further physico-pharmaceutical characterization and in vivo testing.

**Physico-pharmaceutical characterization of FDT RO**: Possible chemical or physical interaction between drug and excipients in RO tablet were studied using FTIR and DSC. FTIR Spectrum of carvedilol in RO tablet showed that the main characteristic peaks of the drug were still preserved in the spectrum of the tablet which indicates absence of interaction between drug and excipients used (scan not shown). DSC results showed a small endothermic peak shifted at 146°C suggesting significant reduction in the crystallinity of the drug (Fig. 7). It was noticed that the endothermic peak of mannitol (sharp endothermic peak at 173°C) used in the preparation of the tablet was no longer present indicating the formation of amorphous form of mannitol by the process of lyophilization (data not shown).

Scanning electron micrographs of the surface and cross section views of FDT RO are shown in Fig. 8a and b. The micrographs show the highly porous nature of the prepared lyophilized tablet which appears in both the surface and the inner structure. The highly porous nature of the tablet explains the rapid penetration of water which resulted in very fast in vitro and in vivo disintegration as well as rapid dissolution from RO tablet. Carvedilol particles could not be seen in the lyophilized matrix in these micrographs suggesting that drug particles might have been reduced during preparation of the suspension prior to lyophilization.
Fig. 7: DSC thermogram of carvedilol in FDT RO

Fig. 8(a-b): Canning electron micrographs of FDT RO in (a) surface view and (b) cross section view

**Determination of anti-hypertensive activity of carvedilol from FDT RO:** The anti-hypertensive activity of carvedilol FDT RO was assessed in rats and the study protocol was approved by the Institutional Animal Care and Use Committee (IACUCCU) in the College of Pharmacy at Cairo University. The average systolic blood pressure versus time following administration of equal doses of carvedilol in the form of FDT RO and Carvepress in 6 hypertensive rats each is illustrated in Fig. 9. Results show that it took almost 1 hour for the carvedilol conventional tablet to start reducing the blood pressure of rats and it took almost 2 hours to see a significant decrease in blood pressure (about 144 mmHg on average after 2 h). On the other hand, administration of carvedilol in the form of RO tablet resulted in a rapid significant decrease (p<0.01) in blood pressure after 15 min (145 mmHg) when compared to the conventional tablet (172 mmHg) followed by a further gradual decrease of blood pressure thereafter. The average blood pressure after 30 min was about 135 mmHg and was also significantly different when compared to Carvepress (p<0.01). The blood pressure was further slightly decreased at 60 min and was maintained thereafter. Results obtained from RO tablet could be due the rapid initial absorption of the drug from the oral cavity due to rapid disintegration and dissolution followed by
Fig. 9: Mean (±SD) systolic blood pressure following administration of 1.56 mg carvedilol in FDT RO or Carvepress in six rats (n = 6)

more steady and slower absorption from the GIT for swallowed drug. These results are in agreement with previous published work (Vishnu et al., 2007), in which it was shown that the bioavailability of carvedilol from bioadhesive buccal patches, administered to healthy pigs, increased 2.29 folds when compared to that of oral solution indicating that carvedilol has good buccal absorption.

**Conclusion:** In vitro and in vivo results presented in this work demonstrated that a lyophilized tablet made of pharmaceutically accepted excipients and containing carvedilol can result in rapid dissolution and absorption of carvedilol in the oral cavity resulting in rapid onset of action. The FDT developed and optimized in this work may be an alternative to conventional oral tablets of carvedilol which usually suffer from a slow onset of action due to slow dissolution and poor bioavailability. The FDT also melts in the mouth immediately after administration which makes ingestion of these tablets very easy and useful in emergency situations.

**REFERENCES**


