Chapter 7

Materials & Method.......
7.A. MATERIAL

7.A.1. ACTIVE DRUG SUBSTANCE – METOPROLOL TARTRATE

The active substance MT is suitable candidate for formulation in CDDS\(^1\); administered in 100 to 450 mg daily, in divided doses\(^2\). The elimination half-life is about 3-7 hours in most patients and is independent of the dose and duration of therapy\(^3\). The drug is readily and completely absorbed from the g.i.t. Peak plasma concentrations vary widely and occur about 1.5 to 2 hours after single oral dose. It belongs to Class 1\(^4-5\) based on Biopharmaceutical Classification System (BCS) based on solubility in aqueous media and intestinal permeability.

The free gift sample was obtained from AstraZenca Limited Bangalore, which had following batch details mentioned for the active drug MT.

<table>
<thead>
<tr>
<th>Product</th>
<th>Metoprolol Tartrate</th>
</tr>
</thead>
<tbody>
<tr>
<td>Batch No.</td>
<td>MTB AO 58</td>
</tr>
<tr>
<td>Manufacturing Date</td>
<td>May/2000</td>
</tr>
<tr>
<td>Expiry Date</td>
<td>April/2005</td>
</tr>
<tr>
<td>Assay</td>
<td>99.18%</td>
</tr>
</tbody>
</table>

Table 7.1. Pharmacopoeial Specifications for Metoprolol Tartrate\(^6\).

<table>
<thead>
<tr>
<th>Test parameter</th>
<th>Compendia Limits</th>
</tr>
</thead>
<tbody>
<tr>
<td>Description</td>
<td>White, crystalline powder or colorless crystals</td>
</tr>
<tr>
<td>Identification</td>
<td>Infrared Absorption</td>
</tr>
<tr>
<td>Melting point</td>
<td>121 –124 °C</td>
</tr>
<tr>
<td>pH</td>
<td>6.0 - 7.0 (in 2.0% w/v solution).</td>
</tr>
<tr>
<td>Clarity &amp; color of solution</td>
<td>A 2.0% w/v solution is clear.</td>
</tr>
<tr>
<td>Specific optical rotation</td>
<td>(+) 7.0(^0) – (+) 10.0(^0) at 20(^0) in 2.0% w/v solution.</td>
</tr>
<tr>
<td>Heavy metals</td>
<td>Not more than 10 ppm.</td>
</tr>
<tr>
<td>Sulphated ash</td>
<td>Not more than 0.1%.</td>
</tr>
<tr>
<td>Loss on drying</td>
<td>Not more than 0.5% determined on 1 gm by drying in an oven at 105(^0)C.</td>
</tr>
<tr>
<td>Chromatographic purity</td>
<td>Not more than RS spot.</td>
</tr>
<tr>
<td>Assay (titrimetry)</td>
<td>Between 99.0-101.0 % of (C(<em>{15})H(</em>{26})NO(_3))(_2),C(_4)H(_6)O(_8).</td>
</tr>
</tbody>
</table>
7. A. 2.  CONTROL RELEASE POLYMER – HPMC K4M and HPMC K100M

Free gift samples of hydroxypropyl methyl cellulose (HPMC) K4M and HPMC K100M were obtained from Colorcon Asia Pvt. Ltd., Charkop, Kandivali(W), Mumbai 400 0677. The manufacturer (Colorcon Asia Pvt. Ltd.) claims that its manufactured product meets the requirement of the USP-23 and European Pharmacopoiea 3rd Ed. and is certified Kosher. The product specifications are given in Table 7.2.

7. A. 2. 1.  Dow® Safety Data Sheet7-8

The material safety details provided by The Dow Chemical Company for the product HPMC K4M and K100M PREM are:

i. Product Name

METHOCEL® K4M/ or K100M PREMIUM HYDROXYPROPYL METHYLCELLULOSE EP (i.e., minimum 95 % < 100 mesh screen) GRADE.

ii. COMPOSITION/INFORMATION ON INGREDIENTS

Modified cellulose

iii. HAZARDS IDENTIFICATION

This product is not hazardous according to EC criteria.

iv. FIRST-AID MEASURES

Never give fluids or induce vomiting if patient is unconscious of is having convulsion.

- Inhalation: Remove to fresh air if effects occur. Consult a physician.
- Skin Contact: Wash off in flowing water or shower.
- Eye Contact: Irrigate immediately with water for at least 5 minutes.
- Ingestion: Consult a physician who will decide on need and method of emptying stomach.
Table 7.2. Hydroxypropyl methylcellulose product specifications as given by the manufacturer.1-10

<table>
<thead>
<tr>
<th>Details</th>
<th>Polymer Type</th>
<th>Methocel K4M</th>
<th>Methocel K100M</th>
</tr>
</thead>
<tbody>
<tr>
<td>Batch Number</td>
<td>NJ21012N12</td>
<td>NE14012NO2</td>
<td></td>
</tr>
<tr>
<td>Date of Manufacture</td>
<td>October 2000</td>
<td>August 2000</td>
<td></td>
</tr>
<tr>
<td>Drug Mfg. Lic. No.</td>
<td>1234</td>
<td>1234</td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Test Item</th>
<th>Specifications</th>
</tr>
</thead>
<tbody>
<tr>
<td>Description</td>
<td>White to slightly off white, fibrous or granular powder.</td>
</tr>
<tr>
<td>Identity</td>
<td>Meets the requirements of the USP and PhEUR.</td>
</tr>
<tr>
<td>Appearance of solution</td>
<td>Less colored than reference solution Y₆ and less opalescent than reference suspension III.</td>
</tr>
<tr>
<td>pH (1% solution)</td>
<td>5.5 - 8.0</td>
</tr>
<tr>
<td>Methoxy content</td>
<td>19.0 - 24.0%</td>
</tr>
<tr>
<td>Hydroxypropoxy content</td>
<td>7.0 - 12.0%</td>
</tr>
<tr>
<td>Apparent viscosity</td>
<td>2308 - 3755 mPa.s (nominal value 2903 mPa.s by rotation)</td>
</tr>
<tr>
<td>Apparent viscosity</td>
<td>3000 - 5600 cP (nominal value 4000 cP by Ubbelhode)</td>
</tr>
<tr>
<td>Chlorides</td>
<td>Maximum 0.5%</td>
</tr>
<tr>
<td>Heavy metals</td>
<td>Maximum 10 ppm as Pb</td>
</tr>
<tr>
<td>Loss on drying</td>
<td>Maximum 5.0%</td>
</tr>
<tr>
<td>Sulphated Ash</td>
<td>Maximum 1.0%</td>
</tr>
<tr>
<td>Organic Volatile Impurities</td>
<td>Will pass USP test &lt;467&gt;</td>
</tr>
<tr>
<td>Particle Size</td>
<td>Minimum 99.0% through No 40 US standard sieve.</td>
</tr>
<tr>
<td>Packaging</td>
<td>25 Kg polylined fibre drums.</td>
</tr>
<tr>
<td>Storage</td>
<td>The recommended storage temperature is 5 - 35 °C.</td>
</tr>
</tbody>
</table>

**Stability and Reactivity**

| Chemical Stability           | Stable under normal handling and storage conditions.                          |
| Materials to avoid            | Oxidizing agents.                                                             |

The material has a recommended shelf life of five years from the date of manufacture if stored in closed container.
v. FIRE-FIGHTING MEASURES
   • Extinguishing media: Water fog or fine spray. Carbon dioxide.
   • Extinguishing media to Avoid: DO NOT USE WATER JET. Dust explosion hazard may result from forceful application of fire extinguishing agents.
   • Hazardous Combustion Products: None known. Complete combustion will give carbon dioxide and water.
   • Protection of Firefighters: Wear positive-pressure self-contained breathing apparatus and protective fire fighting clothing (includes fire fighting helmet, coat, trousers, boots and gloves).
   • Specific Fire or Explosion Hazards: Dust of this product suspended in air is flammable and poses a definite explosion hazard if ignited.

vi. ACCIDENTAL RELEASE MEASURE
   • Methods to Cleaning up: Sweep up, recover if possible, or dispose of according to applicable regulations. Spills may cause very slippery surfaces when wet. If the spill is a viscous solution it should be further diluted with water before disposal.

vii. HANDLING AND STORAGE
   • Handling: Fine dust of this product can form explosive mixture with air and poses a definite fire and explosion hazard at all times; keep away from ignition sources. May cause very slippery surfaces when wet. The minimum dust explosion concentration is 30-160 g/m³.
   • Storage: The recommended storage temperature is 5-35 °C.

viii. EXPOSURE CONTROLS/ PERSONAL PROTECTION
   • Exposure Guidelines: The UK Health and Safety Executive has established an Occupational Exposure Standard (OES) of 10 mg/m³ 8-hour TWA total inhalable dust and 5 mg/m³ 8-hour TWA respirable dust.
- Engineering Controls: Good general ventilation should be sufficient.
- Personal Protective Equipment:
  - Respiratory Protection: No respiratory protection should be needed.
  - Skin Protection: No precautions other than clean body-covering clothing should be needed.
  - Eye/Face Protection: Use safety glasses.

ix. PHYSICAL AND CHEMICAL PROPERTIES
- Appearance: powder
- Color: white to off-white
- Odor: none
- Boiling point/range: not applicable
- Freezing point/range: not applicable
- Vapour pressure: swells in water; no solubility limit
- Relative vapour density (air=1): not applicable
- Specific gravity: not applicable
- pH: not applicable
- Log P (octanol/water): not applicable
- Flash point: not applicable
- Auto-ignition temperature: >350 °C
- Flammability: not applicable

x. STABILITY AND REACTIVITY
- Chemical Stability: Stable under normal handling and storage conditions.
- Materials to Avoid: Oxidizing agents.

xi. TOXICOLOGICAL INFORMATION
- Ingestion: Single dose oral toxicity is considered to be low. The oral LD_{50} for rats is >2000mg/kg.
- Skin Contact: Essentially non-irritating to skin. Skin absorption is unlikely due to physical properties.
- Eye Contact: Essentially non-irritating to eyes. Solid or dust may cause irritation or corneal injury due to mechanical action.
- Inhalation: No adverse effects are anticipated from inhalation.
- Other information: Based on available data, repeated exposures are not anticipated to cause significant adverse effects.

xii. ECOLOGICAL INFORMATION
- Degradation: Biodegradation under aerobic conditions is below detectable limits. Despite the very slow biodegradation rate the product should not present any environmental hazard in the water/soil compartment.
- Aquatic Toxicity: Modified cellulosics are generally non-harmful to aquatic organisms (LC50/EC50/IC50 greater than 100mg/L).

xiii. DISPOSAL CONSIDERATIONS
Any disposal practice must be in compliance with all local and national laws and regulations. Customers are advised to check their local legislation on governing the disposal of waste materials.

xiv. TRANSPORT INFORMATION
Product is not classified for any mode of transportation.

xv. REGULATORY INFORMATION
EC Classification and User Label Information

xvi. OTHER INFORMATION
No other information.

7.A.2.2. Particle Size Distribution of HPMC (Sieve Analysis)\textsuperscript{11-12}
Particle size of polymer can greatly influence polymer performance in the hydrophilic matrix. Fractions of polymers with smaller particle size have more surface area
relative to equivalent weights of fractions with larger particle size. The greater surface area provides for better polymer-water contact, thus increasing the overall rate at which complete polymer hydration and gelation occurs. This leads to more effective formation of the protective gel barrier so critical to the performance of hydrophilic matrix tablets. Hence, particle characterization of the polymer by sieve analysis was undertaken.

7.A.3. DILUENT – LACTOSE MONOHYDRATE (General Reagent Grade)

The product was purchased from Loba Chemicals, Mumbai. The specifications given on the container were

i. Molecular Formula: C\textsubscript{12}H\textsubscript{22}O\textsubscript{11}·H\textsubscript{2}O

ii. Molecular Weight: 360.31

iii. Mfg. By: Loba Chemie

iv. Batch No.: 56897

**Total Water Content: 4.8 – 5.4%**

**Maximum limits of impurities**

i. Free acid – limit 0.25 mL N%

ii. Insoluble matter: 0.005%

iii. Alcohol soluble impurities: 0.2%

iv. Nitrogen compounds (N): 0.02%

v. Arsenic (As): 0.001%

vi. Copper (Cu): 0.00005%

vii. Iron (Fe): 0.0002%

viii. Lead (Pb): 0.00005%

(Alcohol soluble impurities. For bacteriological purpose. Free from glucose)

7. A.4. LUBRICANT – MAGNESIUM STEARATE

*(precipitated fine powder, General Reagent Grade)*

The product was purchased from Loba Chemicals, Mumbai. The specifications given on the container were

i. Molecular Formula: C\textsubscript{36}H\textsubscript{70}MgO\textsubscript{4}

ii. Molecular Weight: 591.27

iii. Mfg. By: Loba Chemie

iv. Batch No.: 57168
v. Assay of Magnesium (calculated on dry substance): 4.5%

vi. Ash: 7.5%

vii. Loss on drying (105°C): 3.5%

viii. pH (saturated solution): 6.2 – 7.4

ix. Heavy metals (as Pb): 0.002%

x. Zinc stearate: 0.5%

xi. Chloride (Cl): 0.02%

xii. Sulphate (SO₄): 0.2%

xiii. Acid number of precipitated fatty acid: 195 - 210

7. B. METHODS

7. B.1. PREFORMULATION STUDIES

7. B.1.1. Spectral Scan of Active Drug Substance

The UV absorption spectrum of active substance MT in distilled water was performed to determine the spectrum peaks using 2 nm bandwidth Jasco V-530 UV-Spectrophotometer. A suitable quantity of MT was dissolved in distilled water and the spectrum measurement of the sample was performed against distilled water as the solvent reference blank. The scanning range was set 200-1000 nm at a speed, 40 nm/min and the data-collecting wavelength was 0.1 nm, with 3-cycle number.

Note: Any sample subjected for spectral absorption, during the entire period of experimental work was filtered using Millipore Millex-HV 13mm filter unit consisting of PVDF membrane filter with a pore size rating of 0.45 μm, so as to eliminate any suspended impurities interfering with the studies.

7. B.1.2. Melting Point

Melting point was determined as per IP 1996 method. The results both determined and taken from the literature are included.

7. B.1.3. pH

A 2% solution of MT distilled water was used to determine the pH using calibrated pH meter.
7. B.1.4. Dissociation Constant (pKa)

The values for MT reported in the literature taken.

7. B.1.5. Moisture Content

Samples of MT (0.2 gm) were subjected to determination of their moisture content using Karl Fischer Titrimeter (AutoTitr....or, Labindia, Mumbai) using specially dried methanol (E. Merck, India) and combined Karl Fischer Reagent (E. Merck, India).

7. B.1.6. Density and Flowability

The samples (10 gms) of MT were subjected to evaluation of bulk density and tap density using a calibrated measuring cylinder (25 mL, Borosil) and Bulk Density Apparatus (Campbell Electronics, India) respectively; flowability was obtained from angle of repose obtained using funnel technique. Derived powder characteristics such as Bulkiness, Carr's Compressibility Index and Hausner's Ratio were calculated using standard equations described in detail in Chapter 6.

7. B.1.7. Drug-Excipient Compatibility Studies

7. B.1.7.i. Fourier transform infra red spectroscopic (FTIR)

For the consistent, reliable, and safe development of drug products, FTIR spectra were obtained to characterize pharmaceutical solids at the molecular level for complete characterization of materials used in the development of extended release formulation containing MT.

Method: The pre-dried (3 hours at 60 °C) samples were taken in the ratio of ~1:100 sample to KBr; triturated in the agate mortar with the pestle and placed on the holder. Spectra were obtained by diffuse reflectance technique using KBr on Jasco FT/IR 460 Plus with a resolution set for 4 cm⁻¹ and with a scanning speed of 2mm/sec over a wavelength of 4000 to 400 nm over the data gathered after 16
accumulation (repeats). The base line correction for KBr was performed and corrected for the sample.

7. B.1.7.ii. Chromatographic Studies

The drug polymer compatibility test was further analyzed by Chromatographic technique after storage under accelerated conditions of temperature and humidity (50 °C and 70% RH for 3 weeks)\textsuperscript{13}. The samples were powder-blended and compressed in the tablet machine to get the hardness of 8 kg/cm\textsuperscript{2} (to simulate tabletting); used in the study. The USP 27\textsuperscript{14} modified method was used for analysis of the sample. In place of Thin Layer Chromatographic Plate, Paper was used (Whatman Chromatographic Paper) and rest of the procedure remained the same. The details of which are stated below:

(i) The Test Solutions of drug MT and the mixtures of drug and polymer HPMC K4M, K100M, diluent Lactose and lubricant magnesium stearate were prepared in 90% ethanol, for spotting.

(ii) Chromatographic Chamber was lined with absorbent paper, and poured into the chamber containing 250 mL of a mixture of chloroform, methanol, ammonium hydroxide (80:15:2); the chamber was allowed to saturate for 1.5 hours.

(iii) About 5-µL portions of the Test Solution was applied at a distance of 2.5 cm from the bottom of the paper, the spots were dried and the paper was suspended in the Chromatographic chamber, closed the chamber, and allowed the chromatogram to develop until the solvent front moved about three-fourths of the length of the chromatogram. The chromatogram was removed, dried in a current of warm air until the odor of ammonia was no longer perceptible (about 45 minutes).

(iv) A beaker containing 0.5 gm of potassium permanganate was placed in the chamber and then 5 mL of 6 N hydrochloric acid was added to the beaker;
allowed to equilibrate for 5 minutes. Then the chromatogram was placed in the chamber for five minutes and then removed. The chromatogram was allowed to stand in a current of cool air for 1 hour before it was sprayed with detecting reagent.

(v) For detecting reagent, the solutions of potassium iodide (1 in 100) and soluble starch (prepared by triturating 3 gm in 10 mL of cold water and adding the mixture to 90 mL of boiling water with constant stirring) was prepared. Just prior to use, mixed 10 mL of each solution with 3 mL of alcohol.

- **Evaluation of Chromatogram**

  For the evaluation of the chromatogram, the number of spots and the \( R_f \) values were studied. \( R_f \) value is defined as the ratio of distance traveled by the solute to the solvent front and is characteristic for a particular substance under given set of eluting conditions.

\[
R_f = \frac{(Distance \ traveled \ on \ the \ medium \ by \ the \ compound)}{(Distance \ traveled \ by \ the \ front \ of \ the \ mobile \ phase)}
\]

Increase in number of spots means there the mixture has undergone degradation and change in \( R_f \) values indicate some chemical changes occurring in the substance under analysis. However, if the \( R_f \) value remains the same for the pure drug and the drug in the mixture(s), then it indicates the drug in particular mixture is compatible.

7.B.2. STANDARD CALIBRATION CURVE

7.B.2.1. **General Introduction**\(^{15}\)

Test procedures used for the assessment of various quality levels of different batches were strived to be in line with the Compendia. It consisted of the following sections (i) Rationale, (ii) Analytical Procedures and (iii) Data Elements, as described in USP.
Rationale: This section should identify the need for the method and describe the capability of the specific method proposed and why it is preferred, a comparison should be provided of limitations of the current Compendial method and advantages offered by the proposed.

Proposed Analytical Procedure: This section should contain a complete description of the analytical method sufficiently detailed to enable persons “skilled in the art” to replicate it. The write-up should include all important operational parameters and specific instructions such as preparation of reagents, performance of systems suitability tests, description of blanks used, precautions, and explicit formulas for calculation of test results.

Data Elements: This section should provide thorough and complete documentation of the validation of the analytical method. It should include summaries of experimental data and calculations substantiating each of the applicable analytical performance characteristics. These characteristics are described in the following Sections.

Validation: Validation of an analytical method is the process by which it is established, by laboratory studies, that the performance characteristics of the method meet the requirements for the intended analytical application. Typical analytical performance characteristics that should be considered in the validation of the types of method described in this document are (1) Accuracy, (2) Precision, (3) Specificity, (4) Detection Limit, (5) Quantitation Limit, (6) Linearity and (7) Range.

7.B.2.2. Analytical Performance Characteristics\textsuperscript{15}

Accuracy (definition): The accuracy of an analytical method is the closeness of test results obtained by that method to the true value. The accuracy of an analytical method should be established across its range.
• Accuracy is calculated as the percentage of recovery by the assay of the known added amount of analyte in the sample, or as the difference between the mean and the accepted true value, together with confidence intervals.

• The ICH documents recommend that accuracy should be assessed using a minimum of nine determinations over a minimum of three concentration levels, covering the specified range (i.e., three concentrations and three replicates of each concentration).

**Precision (definition):** The precision of an analytical method is the degree of agreement among individual test results when the method is applied repeatedly to multiple samples of a homogeneous sample.

• The precision of an analytical method is usually expressed as the standard deviation or relative standard deviation (coefficient of variation) of a series of measurements.

• Precision may be a measure of either the degree of reproducibility or of repeatability of the analytical method under normal operating conditions.

In this context, reproducibility refers to the used of the analytical procedure in different laboratories as in a collaborative study. Intermediate precision expresses within – laboratory variation as on different days, or with different analysts or equipment within the same laboratory.

Repeatability refers to the use of the analytical procedure within a laboratory over a short period of time using the same analyst with the same equipment.

• For most purposes, repeatability is the criterion of concern in USP analytical procedures, although reproducibility between laboratories or intermediate precision may well be considered during the standardizing of a procedure before it is submitted to the Pharmacopoeia.
The ICH documents recommend that repeatability should be assessed using a minimum of nine determinations covering the specified range for the procedure (i.e., three concentrations and three replicates of each concentration or using a minimum of six determinations at 100% of the test concentrations.

**Specificity (definition):** The ICH documents define specificity as the ability to assess unequivocally the analyte in the presence of components that may be expected to be present, such as impurities, degradation products, and matrix components. Lack of specificity of an individual analytical procedure may be compensated by other supporting analytical procedure.

**Detection Limit (definition):** The detection limit is a characteristic of limit tests. It is the lowest amount of analyte in a sample that can be detected, but not necessarily quantitated, under the stated experimental conditions.

**Quantitation Limit:** The quantitation limit is a characteristic of quantitative assays for low levels of compounds in sample matrices, such as impurities in bulk drug substances and degradation products in finished pharmaceuticals. It is the lowest amount of analyte in a sample that can be determined with acceptable precision and accuracy under the stated experimental conditions. The quantitation limit is expressed as the concentration of analyte (e.g., percentage, parts per billion) in the sample.

**Linearity:** The linearity of an analytical method is its ability to elicit test results that are directly, or by a well-defined mathematic transformation, proportional to the concentration of analyte in samples within a given range.

**Range:** The range of an analytical method is the interval between the upper and lower levels of analyte (including these levels) that have been demonstrated to be determined with a suitable level of precision, accuracy, and linearity using the
method as written. The range is normally expressed in the same units as test results (e.g., percentage, parts per million) obtained by the analytical method.

**Determination of Linearity and Range:** Linearity should be established across the range of the analytical procedure. It should be established initially by visual examination of a plot of signals as a function of analyte concentration of content. If there appears to be a linear relationship, test results should be established by appropriate statistical methods (e.g., by calculation of a regression line by the method of least square). In some cases, to obtain linearity between the response of an analyte and its concentration, the test data may have to be subjected to a mathematical transformation. Data from the regression line itself may be helpful to provide mathematical estimates of the degree of linearity. The correlation coefficient, y-intercept, slope of the regression line, and residual sum of squares should be submitted.

- The range of the method is validated by verifying that the analytical method provides acceptable precision, accuracy and linearity when applied to samples containing analyte at the extremes of the ranges as well as with the range.
- ICH recommends that, for the establishment of linearity, a minimum of five concentrations normally be used. It is also recommended that the following minimum specified range should be considered.

**ASSAY OF DRUG SUBSTANCE** (or a finished product): from 80% to 120% of the test concentration.

**DETERMINATION OF AN IMPURITY:** 50 - 120% of the specification.

**FOR CONTENT UNIFORMITY:** A minimum of 70% to 130% of the test concentration, unless a wider or more appropriate range, based on the dosage form (e.g., metered-dose inhalers) is justified.
FOR DISSOLUTION TESTING: ± 20% over the specified range, based on the specifications for a controlled-release product cover a region from 20%, after 1 hour, and upto 90%, after 24 hours, the validated range would be 0% to 110% of the label claim.

**Ruggedness:** The ruggedness of an analytical method is the degree of reproducibility of test results obtained by the analysis of the same samples under a variety of conditions, such as different laboratories, different analysts, different instruments, different lots of reagents, different elapsed assay times, different assay temperatures, different days, etc. ruggedness is normally expressed as the lack of influence of test results of operational and environmental variables of the analytical method. Ruggedness is a measure of reproducibility of test results under the variation in conditions normally expected from laboratory to laboratory and from analysts to analysts.

**Robustness:** The robustness of an analytical method is a measure of its capacity to remain unaffected by small but deliberate variations in method parameters and provides an indication of its reliability during normal use.

**7.B.2.3. Spectral Scan of Active Substance**

The UV absorption spectrum measurement of active substance MT in pH 6.8-phosphate buffer was performed to determine the spectrum peaks using 2 nm bandwidth Jasco V-530 UV-Spectrophotometer. A suitable quantity of MT was dissolved in pH 6.8 phosphate buffer and the spectrum measurement of the sample was performed against pH 6.8-phosphate buffer as the solvent reference blank. The scanning range was set between 200 to 1000 nm at a speed of 40 nm/min and the data-collecting wavelength was 0.1 nm, with 3-cycle number.
Note: Any sample subjected for spectral absorption, during the entire period of experimental work was filtered using Millipore Millex-HV 13mm filter unit consisting of PVDF membrane filter with a pore size rating of 0.45 μm, so as eliminate any suspended impurities interfering with the studies.

7.B.2.4. Standard Calibration Curve (CC) of Metoprolol Tartrate

A suitable quantity of MT was accurately weight\(^{16}\) using Afcoset Electronic Balance (sensitivity ± 0.1 mg so that measurement uncertainty (random plus systematic error) did not exceed 0.1% of the reading. The drug was dissolved in 100.0 mL pH 6.8-phosphate buffer to obtain the Stock Solution (SS). Aliquots of SS were suitably diluted with pH 6.8-phosphate buffer, to get series of MT. The spectrophotometric absorbances were determined at 275 nm against pH 6.8-phosphate buffer as the reference blank. Each sample measured was set to 10 cycles, at an interval of 5 seconds and with medium response. The Standard CC was obtained by plotting a graph of drug concentration versus absorbance. The statistical linear regression coefficient (slope), constant (intercept) and correlation coefficient values. The experiment was repeated for another 5 times and the corresponding data, linear regression values were calculated. From these replicated studies, the standard deviations were estimated.

7.B.2.5. Absorption Stability of Metoprolol Tartrate

During a particular analysis of the active drug, the analysis could for some reasons might not get completed immediately, but later on the next day. This might affect the UV spectral absorption values, if the active substance is prone to degradation in the medium. Hence the spectral absorbance stability of MT; at the same concentration was done over a period of 3 days in pH 6.8 phosphate buffer for 72 hours at 275 nm \(\lambda_{\text{max}}\) against pH 6.8 phosphate buffer as the blank. The percent deviation of change in the absorbance as a function of time was determined, for 6 sets
7.B.3. ANALYTICAL METHOD AND ITS VALIDATION FOR METOPROLOL TARTRATE EXTENDED RELEASE TABLETS

Analytical grade reagents and distilled water was used throughout the experimentation unless otherwise specified.

7.B.3.1. Preparation of Standard Solution

MT Reference Standard (RS) (99.18%), an accurately weighed quantity (60mg) of MT was dissolved in distilled water and diluted suitably to get the final drug concentration of 250 μgm/mL.

7.B.3.2. Preparation of Sample Solution

Twenty tablets were accurately weighed and their average weight was obtained. The tablets were finely crushed and powder equivalent to average weight of tablet was transferred to 100 ml conical flask and 30 ml of distilled water was added to it. The suspension was sonicated for about 30 mins followed by addition of water to make up the volume. The filtered portion of this solution (Millipore Membrane filter, 0.45μm) was used for spectral analysis.

7.B.3.3. Method validation

Excipient mixture (25 gm) without drug was prepared for use as placebo.

7.B.3.3.i. Accuracy

Accuracy of the analytical method was established by spiking MT RS (98.18%) on to the placebo. Accuracy was ascertained in three and over five concentration levels ranging between 135 μgm/mL and 410 μgm/mL (50-150% of drug concentration). MT tablets (average weight 250mg) contain drug and excipients (200mg). Accurately weighed drug (27-75 added to mg) was added to 30 mg powdered placebo tablets, mixed and transferred to 100 mL conical flask, 30 mL distilled water was added to it.
It was sonicated for about 30 mins followed by addition of distilled water to make up the volume. Filtered portions were taken for UV absorbance against distilled water as blank. Standard drug solution was prepared as described earlier Section 7.B.3.1 and its UV absorbance was taken.

Amount of MT (mg) was calculated using the following formula:

\[
\text{Amount of Metoprolol Tartrate} = \left( \frac{T_A}{S_A} \right) \cdot W_S \cdot \left( \frac{P_S}{10} \right)
\]

where

\[
T_A = \text{absorbance of Test Solution}
\]
\[
S_A = \text{absorbance of Standard Solution}
\]
\[
W_S = \text{weight of Standard (mg)}
\]
\[
P_S = \text{potency of Standard}
\]

\[
\% \text{ Recovery} = \left( \frac{\text{amount found}}{\text{amount added}} \right) 100
\]

7.B.3.3.ii. Precision of Assay

Precision of assay of the tablet was carried out using six determinations in three sets (two sets on one day and third on next day) at 100% of the test concentration (i.e., 280 µgm/mL). Procedure for standard and sample preparation was as reported above. UV absorbance of these solutions was taken at 275 nm on UV Spectrophotometer using water as blank.

Amount of MT was estimated using the formula:

\[
\text{Amount of Metoprolol Tartrate} = \left( \frac{T_A}{S_A} \right) \cdot W_S \cdot \left( \frac{P_S}{10} \right)
\]

where

\[
T_A = \text{absorbance of Test Solution}
\]
\[
S_A = \text{absorbance of Standard Solution}
\]
\[
W_S = \text{weight of Standard (mg)}
\]
\[
P_S = \text{potency of Standard}
\]

\[
\% \text{ Label Claim} = \left( \frac{\text{amount obtained (mg)}}{\text{Label Claim}} \right) 100
\]
7.B.3.3.i. Ruggedness for in vitro dissolution data
Ruggedness for dissolution (n=6) was carried out in the same laboratory, on different
days, with different formulations (check, pp 215). The dissolution was carried out on
USP Apparatus Type II with 500 mL of dissolution medium; the temperature of the
bath maintained at 37 °C ± 0.5 °C with paddle rotating at 50 rpm. Aliquots (10 mL)
were withdrawn after time intervals of 1, 4 and 8 hours respectively and replenished
with fresh medium maintained at 37 °C ± 0.5. The filtered aliquots were measured
spectrophotometrically to estimate the amount of drug released against medium as
blank. Standard solutions were prepared as described in Section 7.B.3.1 and UV
absorbance taken.

7.B.3.3.iv. Specificity
Specificity of the method was established by analyzing samples containing placebo
and other without placebo thereby demonstrating the ability of the Method to yield
reliable results without any interference of placebo. UV scan of (i) Placebo and (ii)
Placebo and Standard MT drug, were recorded in the range of 200 to 350 nm.
Standard preparation with Placebo was prepared by adding accurately weighed
amount of MT (99.18%, 50mg) to 30 mg powdered placebo in 100 mL volumetric
flask, 30 ml distilled water was added to it. The resulting solution was sonicated for
about 30 mins., the volume was made up with distilled water. The filtered portions of
the solution were taken for UV –absorbance against distilled water as blank at 275
nm. Standard preparation without placebo was prepared as described in Standard
Solution preparation above.
Amount of MT (mg) was estimated using the formula:

\[ \text{Amount of Metoprolol Tartrate} = \frac{T_A}{S_A} \cdot W_S \cdot \left( \frac{P_S}{10} \right) \]

where \( T_A \) = absorbance of Test Solution
\( S_A \) = absorbance of Standard Solution
\[ W_S = \text{weight of Standard (mg)} \]
\[ P_S = \text{potency of Standard} \]
\[ \% \text{Agreement} = \frac{\text{Test results with placebo}}{\text{Test results without placebo}} \times 100 \]

7.B.3.3.v. Limit of Detection (LOD) and Limit of Quantification

UV scans of standard drug solutions (conc. < 5 \( \mu \text{g/mL} \)) and the medium blank were recorded in the range of 200 to 400 nm. LOD was found to be 1\( \mu \text{g/mL} \) having the absorbance of 0.0036. Limit of Quantification was found to be 6\( \mu \text{g/mL} \) (6 times of the detection). Linearity at 50% and 150% of limit of Quantification Level (6\( \mu \text{g/mL} \)) was carried out. The results of concentration against absorbance were plotted.

7.B.3.3.vi. Linearity and Range

Linearity of MT was carried out in the range of 25 \( \mu \text{g/mL} \) to 300 as reported in Section 7.B.2.4. UV absorbance of the solutions was taken at 275 nm on UV spectrophotometer using pH 6.8 phosphate buffer solution as blank. The experiment was repeated six times. Plot of concentration against absorbance is drawn. After establishing linearity levels, range of the method was validated by verifying at the extremes of the range 25–300 \( \mu \text{g/mL} \).

7.B.4. LAB. SCALE FORMULATION AND FABRICATION OF ER DDS OF METOPROLOL TARTRATE – BY TABLETTING

(Wet-Granulation and Compression under High Pressure)

Taking into consideration drug-excipient compatibility studies, galenical development trials of extended release matrix tablets of MT employing wet granulation were undertaken with batch size of 50 gms. The fundamental principles of tablet manufacturing process have changed very little over the years\(^{17}\). Essentially an active is grouped up with binders, granulated, and then compressed to form a tablet\(^{18\text{-}19}\).
7.B.4.1. Processing Steps

7.B.4.1.i. Sifting

All the materials (polymer, drug and excipients − lactose monohydrate and magnesium stearate) were passed through sieve # 80 mesh screen so that the particle size of each kind were approximately of same size\(^20\).

7.B.4.1.ii. Dry Mixing /Blending

The drug, polymer, diluent and ½ of lubricant were blended by tumble-mix in a beaker for 20 minutes till the mixture was uniform (the method was standardized at the initial stage by scooping a sample of powder mass and then analyzing the amount of drug spectrophotometrically. The procedure was kept uniform for all the batches to minimize the error).

7.B.4.1.iii. Granulation

The pre-sieved powder mass is moistened with 90% V/V alcohol as a binder\(^21\) to render it coherent but by no means wet. It is then passed through a # 14 mesh screen and the material sifted to obtain granules.

- Level of Solvent: The amount of solvent employed was such that the mass was merely moist (rather than wet or pasty). 90% v/v ethanol was used as a granulating agent. Once the granulating liquid was added, mixing was continued until uniform dispersion is attained (~1 minute). The end point of mixing was determined by pressing a portion of the mass in the palm of the hand; and if the ball crumbled under the moderate pressure, then the mixture was ready for the next stage.

7.B.4.1.iv. Drying

The wet granules were dried at a temperature not exceeding 60 °C ± 2 °C for 2 hours till loss on drying (LOD) of the granules was less than 5% determined gravimetrically.
• Drying was done to remove the solvent that is used in forming the aggregates and to reduce the moisture content within the granules.

7.B.4.1.v. Milling and Shifting

The dried granules were passed sifted through sieve # 20. The granules retained were milled in glass mortar with the help of pestle and passed through # 20. LOD of granules was determined by gravimetric method. The percentage of granules below sieve # 60 (% fines) were determined.

7.B.4.1.vi. Lubrication

Magnesium stearate was bolted over the granules and tumbled mixed for uniform distribution.

7.B.4.1.vii. Compression

The lubricated granules were compressed on 16-station D-tooling single rotary compression machine (make) using single set of punch and die with each batch granules weighed to 250mg, except for the optimized batch. The pressure was so adjusted to obtain tablet hardness of ~ 5 kg/cm² determined 1 hour after tableting. A flow diagram of wet granulation process in hydrophilic tablet is shown in Fig 7.1.

• Note: All the operations were carried out in a dehumidified area at a relative humidity of less than 20% at 25 °C.

7.B.4.2. Formulation of ER Matrix Compressed Tablets

Initially, formulation variables were designed based on the clues obtained from the literature (details in Chapter 5 and 6). This involved three stages:

Fig. 7.1. The wet granulation process of hydrophilic matrix tablet preparation.

(ii) 3 batches: B-1, B-2 and B-3 with high density HPMC K100M polymer at 30, 40 and 50 % concentration (w.r.t. total tablet weight).

(iii) 2 batches: C-1 and C-2 using blends of HPMC K4M and HPMC K100M at 20:30 and 30:20 (at 50% concentration level w.r.t. total tablet weight).

- Concentration of drug MT was kept constant to 50 mg in all the formulations. The lubricant magnesium stearate was added at 1% of the tablet weight and the filler
lactose monohydrate was used to make up to constant weight of the tablet to 250 mg.

- A total of 11 batches were prepared with the drug. Placebo tablets were compressed without the drug for each batch.

7.B.4.2.i. Justification for Batch Design

- Initially, the low density polymer concentration was gradually increased (from 5 to 50% at 6 levels) to know the polymer concentration level required to obtain desired in vitro drug release profile for the material chosen.

- Based on the analysis of results obtained from the earlier batch studies, it was desired to vary the polymer concentration to only 3 effective concentration levels, to study the effect of molecular weight on the drug release, within the purview of defined objective.

- Lastly, it was thought to study the polymers blend for: (a) its effect on in vitro drug release profile; and (b) its possible implication and a way to control batch reproducibility in this type of formulations which uses the rheological properties governing the drug release mechanism from the dosage form. This thought was based on: (i) polymers are characterized with their average molecular weights, (ii) no two batches of polymer is in molecular weight and its solution properties, (iii) molecular weight of polymer is directly related to polymer rheological properties and thixotropy, (iv) the drug release from non-disintegrating controlled release matrix is mostly governed by two phenomena namely (a) drug diffusion rate of from the swollen matrix and (b) polymer erosion rate of gelled matrix. Rather going for one polymer, if polymer-blend is used, it would give better flexibility, control and reproducibility over the critical properties controlling the drug release properties. However, no experiments were undertaken to testify this theoretical analogy. The mathematical relationship describing viscosity for the polymer blend can be obtained using the following equation
(\eta_0)^k = F_1(\eta_1)^k \eta + F(\eta_1)^k

where \( F_1 \) and \( F_2 \) are weight fractions of polymer 1 and 2, \( \eta_1 \) and \( \eta_2 \) are viscosity of polymer 1 and 2 respectively.

The equation that expresses the approximate relationship between polymer solution viscosity and polymer concentration is

\[ \eta = (1 + KC)^k \]

where \( \eta \) is the solution viscosity in mPa.s, \( C \) is the polymer concentration in solution (expressed in percent), and \( K \) is a constant specific to molecular weight.

7.B.4.2.ii. Justification for Batch Selecting a Batch for Lab. Pilot Scale Up and Further Studies

The USP 27 NF 22, 2004 Edition specifies the qualifying limits for Metoprolol as ER dosage form. Hence, from the prepared batches which had this qualifying \textit{in vitro} release profile under specified \textit{in vitro} dissolution test conditions. With the foregoing object the problem for optimized batch could be well defined expecting to have come to arrive at.

Defining the Problem

Make a controlled release tablet, which would not release more than 25 % of drug at the end of 1 hour and not less than 80 % at the end of 20th hour and between 20-40 % at 4th hour and between 40-60% at 8th hour under specific dissolution conditions. Mathematically, it can be stated as:

\[ \text{“Amount of drug dissolved} = \]

\[ T_{1 \, \text{hour}} \geq 25\% \), \, T_{4 \, \text{th hour}} = 20-40\% \), \, T_{8 \, \text{th hour}} = 40-60\% \), \, T_{20 \, \text{th hour}} \leq 80\% \)"
7. B. 5. EXTENDED RELEASE TABLET EVALUATION

7. B. 5.1. Tests As Described Earlier

The compressed ER matrix DDS were evaluated for (i) shape & color, (ii) size, (iii) friability, (iv) hardness, (v) weight variation, (vi) content uniformity. Details of which are already discussed in Chapter 3, Section 3.3. Further, (vii) disintegration and (viii) in vitro drug release tests were also performed.

7. B. 5.2. Disintegration Test (DT) Method

By virtue of its non-disintegrating hydrophilic matrix form, these system show controlled release over time. Hence, in place of distilled water, pH 6.8 phosphate
buffer was used as the test medium. At every time interval (0.25, 0.5, 0.75, 1, 1.5, 2, 2.5, 3, 3.5, 4, 5, 6, 7, 8, 10 and 24) the basket assembly was lifted to observe the tablets. The time was recorded when there was no mass left on the screen and the time was recorded as DT time. The other parameters remain as described in Section 3.3 of chapter 3.

7.B.5.3. *In Vitro* Dissolution Studies

Compendial Test Conditions For ER Metoprolol Succinate

In USP 27, there is an addition of drug release test for Metoprolol Succinate ER dosage form (pp 1230-1231). The test specifications are:

- **Test Specifications**
  - **Dissolution Medium:** 500 mL pH 6.8 phosphate buffer.
  - **Apparatus 2:** 50 rpm
  - **Temperature:** 37°C (±0.5)
  - **Time:** 1, 4, 8 and 20 hours

- **Analytical method for the estimation of drug released**
  Determine the amount of \((C_{18}H_{25}NO_3)_2\) \(C_4H_6O_4\) dissolved by employing HPLC method.

- **Tolerance limits are given below:**

<table>
<thead>
<tr>
<th>Time (hours)</th>
<th>Amount Dissolved</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Not more than 25%</td>
</tr>
<tr>
<td>4</td>
<td>Between 20% and 40%</td>
</tr>
<tr>
<td>8</td>
<td>Between 40% and 60%</td>
</tr>
<tr>
<td>20</td>
<td>Not less than 80%</td>
</tr>
</tbody>
</table>

Not less than 75% (Q) of the labeled amount \((C_{18}H_{25}NO_3)_2\) \(C_4H_6O_6\) is dissolved in 30 mins.

- **Criteria for Acceptance Level**: The individual monograph requirement is met if the quantities of active ingredient (Q) are dissolved from the units tested conform to Acceptance Table, given in Section 6.9.4.xii.i., Chapter 6.
- Experimental Test Conditions For Metoprolol Tartrate

The laboratory experimental test conditions for developed ER formulations of metoprolol tartrate salt, based on above guidelines were set which are as follows:

- **Test Conditions**
  
  **Equipment:** Electrolab TDT-08L Dissolution Tester, USP, Mumbai.
  
  **Medium:** 500 mL pH 6.8 phosphate buffer.
  
  **Apparatus 2:** 50 rpm
  
  **Temperature:** 37 °C (±0.5)
  
  **Time:** 0, 0.25, 0.5, 0.75, 1, 1.5, 2, 2.5, 3, 3.5, 4, 5, 6, 7, 8, 10
  
  and 24 hours = 17 readings.
  
  **Aliquot withdrawn:** 10 mL and replaced with same quantity of dissolution medium maintained at the same temperature.

- **Analytical method for the estimation of drug released**

  Spectrophotometric method was used instead of HPLC. The method used is described in section 7.B.2.4.of this chapter. The amount of drug released is estimated using the slope value of standard calibration curve of MT at 275 nm.

7.B.6. **PILOT BATCH**

- Taking into consideration drug-excipients compatibility study results, galenical development trials of extended release matrix tablets metoprolol tartrate employing wet granulation and compression technique, lab. pilot batch size was 200 gms.

7.B.6.1. **Materials**

- **Drug:** Metoprolol Tartrate (AstraZeneca)
  
  **Polymer:** HPMC K4M and HPMC K100M (Dow)
  
  **Diluent:** Lactose Monohydrate (Loba Chemicals)
  
  **Lubricant:** Magnesium Stearate (Loba Chemicals)
  
  **Binding Agent:** 90% V/V Alcohol (Ranbaxy)
7.B.6.2. **Working batch Formula:** The batch working formula is given below.

Table 7.4. Working batch formula for Pilot batch D.

<table>
<thead>
<tr>
<th>Material</th>
<th>Category</th>
<th>Quantity</th>
</tr>
</thead>
<tbody>
<tr>
<td>Metoprolol Tartrate</td>
<td>Active Drug</td>
<td>40 gms + 4gms</td>
</tr>
<tr>
<td>Hydroxypropyl methylcellulose</td>
<td>Control Release</td>
<td>60 gms</td>
</tr>
<tr>
<td>(HPMC K4M)</td>
<td>Polymer</td>
<td></td>
</tr>
<tr>
<td>Hydroxypropyl methylcellulose</td>
<td>Control Release</td>
<td>40 gms</td>
</tr>
<tr>
<td>(HPMC K100M)</td>
<td>Polymer</td>
<td></td>
</tr>
<tr>
<td>Lactose Monohydrate</td>
<td>Diluent</td>
<td>58 gms</td>
</tr>
<tr>
<td>Magnesium Stearate</td>
<td>Lubricant</td>
<td>02 gms</td>
</tr>
<tr>
<td>Alcohol (90% v/v)</td>
<td>Binding Agent</td>
<td>Quantity sufficient</td>
</tr>
</tbody>
</table>

7.B.6.3. **Method**

Wet granulation of metoprolol tartrate along with hydrophilic polymer, diluent and lubricant (half the quantity) was carried out in Kenwood Planetary mixer. The granules were dried at 60°C, lubricated (adding remaining half the quantity) and compressed.

- **Shifting:** Metoprolol tartrate and the excipients were sifted through sieve # 60.
- **Dry Mixing:** Metoprolol tartrate, lactose and polymers were mixed for 10 minutes in Kenwood mixer at 30 rpm.
- **Sample analysis (for mixing):** Sample were scooped from different places and analyzed spectroscopically for uniform distribution of drug in the powder blend.
- **Granulation:** The powder blend was granulated using 90 % V/V alcohol.
- **Drying:** The wet granules were dried in tray drier at 60°C (± 2) for 3-5 hours till loss of drying (LOD) of the granules was less than 5 % as determined by Karl Fisher Automatic Titration equipment.
- **Milling and Shifting:** The dried granules were sifted through sieve #18 and above mesh granules were milled in kitchen mixer. The milled granules were passed through sieve # 18 and mixed and sifted. LOD of granules was determined as mentioned earlier.
• **Lubrication:** The dried granules were blended with lubricantes in Kenwood mixer for 5.0 minutes at 20 rpm. Granule characteristic such as LOD, Particle Size Distribution, Density (tapped and untapped), Angle of Repose, Carr’s Index and Hausner Ratio were determined.

• **Compression:** The lubricated granules are compressed on 16 station D- tooling single rotatory machine (Cadmach) using singe set of punch and die with the weight adjusted to 250 mg.

7.B.6.4. **Process Optimization**

• **Effect of Particle Size:** First trial batch was carried using supplier’s material passed through sieve # 60. Remaining batches were carried out post-micronization of the drug (# 120).

• **Effect of Moisture:** The moisture content was varied between 0.5 – 5.0 % w/w and its effect on compressibility of Metoprolol tartrate granules was evaluated.

7.B.7. **PILOT BATCH EVALUATION**

7.B.7.1. **Tests As Described Earlier**

The evaluating tests for the pilot batch D were same as earlier batches with supplemented with few more tests described in the following sections based in line with the requirements of NDA and SUPAC; however only in vivo tests were not performed. This batch product was also compared with the Reference Listed Drug (RLD) in an attempt to compare the developed product with the innovators product.

7.B.7.2. **Reference Listed Drug:** For Metoprolol ER Dosage Form

The label details of the RLD of the strength available in the Indian market are given below.
R\textsubscript{x} Metoprolol Succinate Extended Release Tablets USP

\textbf{Seloken® XL 50 mg}

Each extended release film coated tablet contains Metoprolol Succinate USP 47.5 mg equivalent to Metoprolol Tartrate 50 mg. Color Titanium Dioxide

Dosage: As directed by the Physician.

Direction for use: The tablets or the divided halves of the tablets should be taken with water, do not crush or chew the tablets.

Precaution: Schedule H drug.

Warning: To be sold by retail on the prescription of a Registered Medical Practitioner only.

Storage: Do not store above 30 °C. Protect from light.

Manufacturing License Number: 30-141-23
Batch Number: SXF DO11
Manufacturing date: 07/2003 Expiry date: 06/2006
Retail Price: Not to exceed Rs. 59.60 for 7 tablets. Local Taxes Extra.
Mfg. by: AstraZeneca Pharma India Ltd, 12\textsuperscript{th} Mile, Bellary Rd, Bangalore – 560 063.

7.B.8. EXPOSURE STUDIES AND SHORT TERM STABILITY STUDY FOR EVALUATION OF LABORATORY SCALE DEVELOPED FORMULATION

7.B.8.1. Thermal Exposure of granules

MT lubricated granules (10 gms) were packed in an HDPE container and subjected to temperature of 60 ± 2 °C in an oven for 7 days. The granules post exposure, were compressed and the effect on tablet characteristics were compared with tablet characteristics observed for unexposed granules. The \textit{in vitro} dissolution data as % cumulative release versus time is plotted.

7.B.8.2. Exposure study of granules and tablets at 40 °C/ 75% RH and 8-10 °C/ 60% RH

MT granules (10 gms) and 20 tablets were directly exposed in an open petri-plate at 40 °C/ 75% RH and 8-10 °C/ 60% RH for 7 days to get a preview of its stability.

7.B.8.3. Accelerated Stability Study

Based on the results of initial exposure studies, the tablets were blister packed in clear PVC/PVDC 40 gsm (PVC/ PVDC 40 GSM – Bilcare) with a lidding membrane of Aluminium, 0.03 mm; using Pharmapack 240 Blister Packing Machine. In-process testing such as leak test under vacuum, physical observation of blister were
performed to ensure packing. The blisters were subjected to stability studies under different conditions (as per ICH Guidelines) described in Ch. 6, Section 6.12.2.

Table 7.5. Conditions for short term stability evaluation of MT ER tablets (batch D).

<table>
<thead>
<tr>
<th>Incubation Conditions</th>
<th>Withdrawal Period (in months)</th>
<th>Number of Packs</th>
</tr>
</thead>
<tbody>
<tr>
<td>30 °C/ 65% RH</td>
<td>1, 2 and 6</td>
<td>10 blisters /condition</td>
</tr>
<tr>
<td>40 °C/ 75% RH</td>
<td>1, 2 and 6</td>
<td>10 blisters /condition</td>
</tr>
<tr>
<td>8-10 °C/ 60% RH</td>
<td>1, 2 and 6</td>
<td>10 blisters /condition</td>
</tr>
<tr>
<td>Photo Stability Chamber</td>
<td>1, 3</td>
<td>10 blisters /condition</td>
</tr>
</tbody>
</table>

The tablets after the specified incubation periods were withdrawn and evaluated to ascertain their physical and chemical stability employing standard reported procedures.

Table 7.6. Stability evaluating parameters for Metoprolol Tartrate ER tablets (Batch D).

<table>
<thead>
<tr>
<th>Physical Parameters</th>
<th>Chemical Parameters</th>
</tr>
</thead>
<tbody>
<tr>
<td>Appearance of Tablet and the Pack</td>
<td>Assay</td>
</tr>
<tr>
<td>Thickness of Tablet</td>
<td>Dissolution</td>
</tr>
<tr>
<td>Average Weight of the Tablet</td>
<td></td>
</tr>
<tr>
<td>Hardness</td>
<td></td>
</tr>
<tr>
<td>Loss on Drying</td>
<td></td>
</tr>
<tr>
<td>Friability</td>
<td></td>
</tr>
</tbody>
</table>

7.B.9. **DIMENSIONAL SWELLING STUDIES (Under Static Conditions)**²¹

The tablet was glued to a glass plate and kept immersed in pH 6.8 phosphate buffer solution at room temperature. At periodic time intervals (0, 0.25, 0.5, 0.75, 1, 1.5, 2, 2.5, 3, 3.5, 4, 5, 6, 7, 8, and 10 hours), the change in the radial and axial dimensions of the swelling matrix were measured using Vernier Calipers, with minimal damages caused to the gelled matrix. The tablet was replaced again immersed in the medium. The experiment was repeated 6 times and the average of 6 readings was taken. The normalized tablet dimensions estimated using the following equation:

\[
\text{Normalized radial dimension} = \frac{R(t)}{R(0)}
\]

and

\[
\text{Normalized axial dimension} = \frac{A(t)}{A(0)}
\]

where \(R(0)\) and \(A(0)\) are the dimensions at time = 0; \(R(t)\) and \(A(t)\) are the dimensions at the time = \(t\).
7.B.10. MATHEMATICAL MODELS FOR RELEASE KINETICS

7.B.10.1. Zero Order Model
Mathematically, the zero order release rate is represented by the following equation

$$\frac{dc}{dt} = k_0$$

where $c =$ concentration of drug release, $t =$ time of release and $k_0 =$ Zero order release constant. The values of the % drug release and time were fitted in the above equation to determine the suitability of this model by plotting a graph of % cumulative drug release versus time.

7.B.10.2. First order model
Mathematically, the first order rate is represented by the following equation

$$\log W = \frac{\log W_0}{2.303} + \log W_0$$

where $W =$ amount of the drug left in the matrix, $W_0 =$ initial amount of the drug in the matrix, $k =$ first order release constant time$^{-1}$ and $t =$ time. The values of the % drug release and time were fitted in the above equation to determine the suitability of this model by plotting a graph of log of cumulative % drug release versus time.

7.B.10.3. Higuchi Model
Higuchi model to study release of drug is based on following equation

$$F_t = Q = [D (2C - C_s). C_s t]^{1/2}$$

where $F_t = Q =$ amount of drug release in time $t$; $C =$ initial drug concentration; $C_s =$ drug solubility in the matrix media; and $D =$ diffusion coefficient.

The values of % drug release and time were fitted into simplified Higuchi equation, which is as follows

$$Q = k t^{1/2}$$

where $k =$ Higuchi dissolution constant. A graph of amount of drug release in time $t$ versus square root of time was plotted to describe the model.

7.B.10.4. Hixon-Crowell Model
Hixon-Crowell model is described by the following equation

$$W_{0}^{1/3} - W_{t}^{1/3} = K_s \ t$$

where $W_0 =$ initial amount of drug in the dosage form, $W_t =$ drug remaining in dosage form at time $t$ and $K_s =$ constant incorporating surface - volume relation. The model
is described by plotting the graph of cube root of fraction drug unreleased versus time.

7.B.10.5. Korsmeyer - Peppas Model

Korsmeyer - Peppas model is described by the following equation

\[ \frac{M_t}{M_\infty} = a \cdot t^n \]

Where \( \frac{M_t}{M_\infty} \) = fraction drug released and \( n \) = release exponent indicative of drug release mechanism. The model is described by plotting the graph of log fraction drug release versus log time.

7.B.10.6. Similarity Factor

In recent years, FDA has placed emphasis of a dissolution profile comparison in the area of post-approval changes and biowaivers. Under appropriate test conditions, a dissolution profile can characterize the product more precisely than a single point dissolution test. A dissolution profile comparison between pre-change and post-change products for SUPAC related changes, or with different strengths, helps assure similarly in product performance and signals bioinequivalence. The similarity factor adopted by US FDA was calculated using the formula

\[ f_2 = 50 \cdot \log \left( 1 + \left( \frac{V}{n} \right) \cdot \left( \sum_{t=1}^{n} (R_t - T_t) \right)^{-1} \right) \cdot 100 \]

where \( R_t \) = % drug release of reference product at each time point \( t \); \( T_t \) = % drug release of test product at each time point \( t \) and \( n \) = sampling number.

7.B.10.7. Difference Factor

The difference factor adopted by US FDA was calculated using the formula

\[ f_1 = \left( \frac{\sum_{t=1}^{n} |R_t - T_t|}{\sum_{t=1}^{n} R_t} \right)^{0.10} \]

where \( R_t \) = % drug release of reference product at each time point \( t \); \( T_t \) = % drug release of test product at each time point \( t \) and \( n \) = sampling number.

The factor \( f_1 \) is proportional to the average difference between the two profiles; where as factor of \( f_2 \) is inversely proportional to the average squared difference between the two profiles, with emphasis on the larger difference among all the time points. The factor \( f_2 \) measures the closeness between the two profiles.
References


[9] Methocel K100M Premium CR EP, Batch QA15012NO1, Mfg. Date 15.01.02, MEPS/IF/10825/03/00, Colorcon Asica Pacific Pte Ltd., 51 Merchant Road, # 03-05, Merchant Square, Singapore 058283.

[10] Methocel K4M Premium CR EP, Batch QA03012NO31, Mfg. Date 03.01.02, MEPS/IF/10821/04/99, Colorcon Asica Pacific Pte Ltd., 51 Merchant Road, # 03-05, Merchant Square, Singapore 058283.


