CHAPTER 2.

LITERATURE REVIEW
2.0 REVIEW OF LITERATURE

2.1 The study drug: Metformin Hydrochloride

2.1.1 Description:
Metformin hydrochloride extended-release tablet is an oral antihyperglycemic agent used in the management of type 2 diabetes. Metformin hydrochloride (N,N-dimethyl imidodicarbonimidic diamide hydrochloride) is not chemically or pharmacologically related to other classes of oral antihyperglycemic agents. The structural formula is as shown:

\[
\text{H}_3\text{C} \quad \text{N} - \text{C} - \text{NH} - \text{C} - \text{NH}_2 \cdot \text{HCl}
\]

Metformin hydrochloride is a white to off-white crystalline compound with a molecular formula of \( \text{C}_4 \text{H}_{11} \text{N}_5 \cdot \text{HCl} \) and a molecular weight of 165.63. Metformin hydrochloride is freely soluble in water and is practically insoluble in acetone, ether, and chloroform. The \( pK_a \) of metformin is 12.4. The pH of a 1% aqueous solution of metformin hydrochloride is 6.68.

2.1.2 CLINICAL PHARMACOLOGY

Metformin is an antihyperglycemic agent, which improves glucose tolerance in patients with type 2 diabetes, lowering both basal and postprandial plasma glucose. Its pharmacologic mechanisms of action are different from other classes of oral antihyperglycemic agents. Metformin decreases hepatic glucose production, decreases intestinal absorption of glucose, and improves insulin sensitivity by increasing peripheral glucose uptake and utilization. Unlike sulfonylureas, metformin does not produce hypoglycemia in either patients with type 2 diabetes or normal subjects (except in special circumstances) and does not cause hyperinsulinemia. With metformin therapy, insulin secretion remains unchanged while fasting insulin levels and day-long plasma insulin response may actually decrease.
2.1.3 Pharmacokinetics

2.1.3.1 Absorption and Bioavailability

The absolute bioavailability of a Metformin 500mg tablet given under fasting conditions is approximately 50-60%. Studies using single oral doses of metformin 500 mg to 1500 mg, and 850 mg to 2550 mg, indicate that there is a lack of dose proportionality with increasing doses, which is due to decreased absorption rather than an alteration in elimination. Food decreases the extent of and slightly delays the absorption of metformin, as shown by approximately a 40% lower mean peak plasma concentration (C_{max}), a 25% lower area under the plasma concentration versus time curve (AUC), and a 35 minute prolongation of time to peak plasma concentration (T_{max}) following administration of a single 850-mg tablet of metformin with food, compared to the same tablet strength administered fasting. The clinical relevance of these decreases is unknown.

Following a single oral dose of extended release tablet of metformin, C_{max} is achieved with a median value of 7 hours and a range of 4 hours to 8 hours. Peak plasma levels are approximately 20% lower compared to the same dose of metformin, however, the extent of absorption (as measured by AUC) is similar to metformin.

At steady state, the AUC and C_{max} are less than dose proportional for metformin extended release tablet within the range of 500 mg to 2000 mg administered once daily. Peak plasma levels are approximately 0.6, 1.1, 1.4, and 1.8 μg/mL for 500, 1000, 1500, and 2000 mg once-daily doses, respectively. The extent of metformin absorption (as measured by AUC) from extended release metformin at a 2000 mg once-daily dose is similar to the same total daily dose administered as metformin tablets 1000 mg twice daily. After repeated administration of extended release metformin, metformin did not accumulate in plasma.

Within-subject variability in C_{max} and AUC of metformin from extended release metformin is comparable to that with metformin.

Although the extent of metformin absorption (as measured by AUC) from the extended release tablet increased by approximately 50% when given with food, there was no effect of food on C_{max} and T_{max} of metformin. Both high and low fat meals had the same effect on the pharmacokinetics of extended release metformin.
2.1.3.2 Distribution

The apparent volume of distribution (V/F) of metformin following single oral doses of METFORMIN 850 mg averaged 654±358 L. Metformin is negligibly bound to plasma proteins, in contrast to sulfonylureas, which are more than 90% protein bound. Metformin partitions into erythrocytes, most likely as a function of time. At usual clinical doses and dosing schedules of metformin, steady state plasma concentrations of metformin are reached within 24-48 hours and are generally < 1 μg/mL. During controlled clinical trials of metformin, maximum metformin plasma levels did not exceed 5 μg/mL, even at maximum doses.

2.1.3.3 Metabolism and Elimination

Intravenous single-dose studies in normal subjects demonstrate that metformin is excreted unchanged in the urine and does not undergo hepatic metabolism (no metabolites have been identified in humans) nor biliary excretion. Renal clearance is approximately 3.5 times greater than creatinine clearance, which indicates that tubular secretion is the major route of metformin elimination. Following oral administration, approximately 90% of the absorbed drug is eliminated via the renal route within the first 24 hours, with a plasma elimination half-life of approximately 6.2 hours. In blood, the elimination half-life is approximately 17.6 hours, suggesting that the erythrocyte mass may be a compartment of distribution.

<table>
<thead>
<tr>
<th>Subject Groups: METFORMIN dose (number of subjects)</th>
<th>C_{max} (μg/mL)</th>
<th>T_{max} (hrs)</th>
<th>Clearance (mL/min)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Healthy, non-diabetic adults: 500 mg single dose (24)</td>
<td>1.03 (±0.33)</td>
<td>2.75 (±0.81)</td>
<td>600 (±132)</td>
</tr>
<tr>
<td>850 mg single dose (74)</td>
<td>1.60 (±0.38)</td>
<td>2.64 (±0.82)</td>
<td>552 (±139)</td>
</tr>
<tr>
<td>850 mg three times daily for 19 doses (9)</td>
<td>2.01 (±0.42)</td>
<td>1.79 (±0.94)</td>
<td>642 (±173)</td>
</tr>
<tr>
<td>Adults with type 2 diabetes: 850 mg single dose (23)</td>
<td>1.48 (±0.5)</td>
<td>3.32 (±1.08)</td>
<td>491 (±138)</td>
</tr>
<tr>
<td>850 mg three times daily for 19 doses (9)</td>
<td>1.90 (±0.62)</td>
<td>2.01 (±1.22)</td>
<td>550 (±160)</td>
</tr>
</tbody>
</table>

Table 2.1: Mean (±S.D.) Metformin pharmacokinetic parameters following single or multiple oral doses of metformin
850 mg single dose (12) | 2.45 (±0.70) | 2.71 (±1.05) | 412 (±98)
---|---|---|---
Renal-impaired adults:
850 mg single dose

<table>
<thead>
<tr>
<th>Mild (CL_cr 61-90 mL/min) (5)</th>
<th>1.86 (±0.52)</th>
<th>3.20 (±0.45)</th>
<th>384 (±122)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Moderate (CL_cr 31-60 mL/min) (4)</td>
<td>4.12 (±1.83)</td>
<td>3.75 (±0.50)</td>
<td>108 (±57)</td>
</tr>
<tr>
<td>Severe (CL_cr 10-30 mL/min) (6)</td>
<td>3.93 (±0.92)</td>
<td>4.01 (±1.10)</td>
<td>130 (±90)</td>
</tr>
</tbody>
</table>

\* All doses given fasting except the first 18 doses of the multiple dose studies

\^ Peak plasma concentration

\^ Time to peak plasma concentration

\^ Combined results (average means) of five studies: mean age 32 years (range 23-59 years)

\^ Kinetic study done following dose 19, given fasting

\^ Elderly subjects, mean age 71 years (range 65-81 years)

\^ CL_cr = creatinine clearance normalized to body surface area of 1.73 m^2

2.1.4 Adverse Effects

In worldwide clinical trials over 900 patients with type 2 diabetes have been treated with Extended release Metformin tablets in placebo- and active-controlled studies. In placebo-controlled trials, 781 patients were administered Extended release metformin and 195 patients received placebo. Adverse reactions reported in greater than 5% of the extended release arm and that were more common in Metformin than placebo-treated patients were diarrhea 9.6% and 2.6% and Nausea/Vomiting 6.5% and 1.5% for the metformin group and placebo arms respectively.

Diarrhea led to discontinuation of study medication in 0.6% of patients treated with extended release metformin. Additionally, abdominal pain, constipation, distention
2.1.5 Therapeutic indications
Metformin hydrochloride extended-release tablets, as monotherapy, are indicated as an adjunct to diet and exercise to improve glycemic control in patients with type 2 diabetes. And is indicated in patients 17 years of age and older. It may be used concomitantly with a sulfonylurea or insulin to improve glycemic control in adults (17 years of age and older). (Therapeutic drugs 1999)

2.1.6 Dosage
There is no fixed dosage regimen for the management of hyperglycemia in patients with type 2 diabetes with extended release Metformin or any other pharmacologic agent. Dosage of metformin extended release must be individualized on the basis of both effectiveness and tolerance, while not exceeding the maximum recommended daily doses. The maximum recommended daily dose of Metformin extended release in adults is 2000 mg. It should generally be given once daily with the evening meal, should be started at a low dose, with gradual dose escalation, both to reduce gastrointestinal side effects and to permit identification of the minimum dose required for adequate glycemic control of the patient. During treatment initiation and dose titration, fasting plasma glucose should be used to determine the therapeutic response and identify the minimum effective dose for the patient. Thereafter, glycosylated hemoglobin should be measured at intervals of approximately three months. The therapeutic goal should be to decrease both fasting plasma glucose and glycosylated hemoglobin levels to normal or near normal by using the lowest effective dose, either when used as monotherapy or in combination with sulfonylurea or insulin. The tablets must be swallowed whole and never crushed or chewed. Occasionally, the inactive ingredients of the tablets will be eliminated in the feces as a soft, hydrated mass.

The usual starting dose of Metformin hydrochloride extended release tablets is 500 mg once daily with the evening meal. Dosage increases should be made in increments of 500 mg weekly, up to a maximum of 2000 mg once daily with the evening meal.
2.1.7 Metformin Formulations

The pharmacokinetic characteristics of metformin IR are not conducive to the slow and controlled release of metformin desirable for a once daily formulation. Absorption of metformin IR in the gastrointestinal tract appears to be limited by permeability, with absorption occurring almost exclusively in the upper gastrointestinal tract and with poor permeability in the lower gastrointestinal tract (Timmins et. al, 2005). The absolute bioavailability of a 500mg dose of metformin IR is 50–60% and decreases as the dose increases, suggesting some form of saturable absorption or permeability/transit time-limited absorption. Food causes a reduction in the bioavailability of metformin IR. Additionally, metformin is highly soluble in water which usually results in rapid dissolution from a dosage form. These obstacles are compounded by the high unit dose of metformin IR, specifically, 500mg per tablet. Drugs that have limited absorption in the upper gastrointestinal tract, such as metformin, are usually regarded as poor candidates for incorporation into modified-release formulations. Most oral modified-release delivery systems function by delivering a drug for absorption over an extended period and along the length of the gastrointestinal tract following administration. However, such delivery devices might not be suitable for metformin because of the relatively narrow window of absorption. In addition, the high water solubility of metformin means that large amounts of polymer are required to control its release from conventional modified-release formulations. This makes the application of many existing extended-release (XR) technologies inappropriate for metformin. A number of mechanisms may be used to produce a dosage form that delivers a drug over an extended period in the upper gastrointestinal tract; however, many of these mechanisms have important limitations. For example, coadministration of metformin and propantheline – a drug that reduces gastrointestinal motility – has been shown to extend the period during which metformin plasma levels are maintained. However, administration of propantheline for the sole purpose of extending residence in the upper gastrointestinal tract has many disadvantages, including the potential for undesirable anticholinergic adverse effects. Various other delivery devices have been proposed that could extend the residence time of metformin in the upper gastrointestinal tract, including:

(i) floating or buoyant systems that float on gastric contents;
(ii) bioadhesive systems that adhere to the gastric mucosa/mucus layer;
systems with a large size, either intrinsically or through swelling/expanding systems designed to prevent passage through the pylorus.

To overcome these difficulties, a novel, biphasic, controlled-release delivery system – the Gel Shield Diffusion System, which can be included under the third category just described – has been developed and used for the XR formulation of metformin (Glucophage XR, US Prescribing information, 2006).

2.2 Bioavailability

Bioavailability is defined as the rate and extent to which the active ingredient or therapeutic moiety is absorbed from the drug product and becomes available at the site of action. For the drug products, which are not absorbed, or the drug levels in the biological matrix are too low to be reliably measured, bioavailability may be assessed by surrogate measurements that reflect the rate and extent to which the active ingredient or active moiety becomes available at the site of action.

Bioequivalence is a relative term. It is defined as the absence of significant difference in the rate and extent to which the active ingredient or active moiety in pharmaceutical equivalents or pharmaceutical alternatives becomes available at the site of drug action when administered at the same molar dose and under similar conditions in an appropriately designed study (CDER, 2003). In bioequivalence studies, the primary question is to compare measures of release of drug substance between the test and reference product. Hence bioequivalence is primarily a product quality question. Because product BA and BE are closely related, similar approaches for establishing BA and BE may be followed.

2.2.1 Historical perspective of BE

Law often becomes a necessary control mechanism when people could be exploited and there are large potential financial gains for businesses choosing to exploit. The society, by law, has removed much of the decisions making about new drug products from the manufacturers, investigators, and physicians and vested it in the government (the Drug Regulatory Agencies). The regulations require the Regulatory Agencies to assess safety, efficacy and quality of all new drug formulations, before they are marketed. The
The fundamental mission of the Drug Regulatory Agencies is protection of the consumers. The historical milestones of drug law are summarized in Table 2.2.

Table 2.2: List of major legislations, regulations and other milestones affecting drug development and marketing in the United States and other countries (Truman, 1992, updated).

<table>
<thead>
<tr>
<th>Year</th>
<th>Event</th>
</tr>
</thead>
<tbody>
<tr>
<td>1902</td>
<td>Biologics Control Act</td>
</tr>
<tr>
<td>1906</td>
<td>Pure Food and Drugs Act</td>
</tr>
<tr>
<td>1912</td>
<td>Sherley Amendment to Pure Food and Drugs Act</td>
</tr>
<tr>
<td>1938</td>
<td>Elixir Sulfanilamide Disaster. FDA control over safety of new drugs</td>
</tr>
<tr>
<td>1948</td>
<td>Miller Amendment</td>
</tr>
<tr>
<td>1951</td>
<td>Durham-Humphrey Amendments</td>
</tr>
<tr>
<td>1952</td>
<td>Hench: Brand substitution case report</td>
</tr>
<tr>
<td>1962</td>
<td>Thalidomide disaster in Europe FDA; Control over both safety and efficacy of drugs-Kefauver-Harris amendment</td>
</tr>
<tr>
<td>1963</td>
<td>Initial Good Manufacturing Practices (GMP) regulations</td>
</tr>
<tr>
<td>1974</td>
<td>World Health organization, recommendations for conduct of bioavailability studies</td>
</tr>
<tr>
<td>1974</td>
<td>Dissolution test adopted as standard for in vitro comparison of bioavailability in UK</td>
</tr>
<tr>
<td>1977</td>
<td>US FDA regulations for approval of BE. The ± 20% rule with p&lt;0.05</td>
</tr>
<tr>
<td>1983</td>
<td>Orphan drug act</td>
</tr>
<tr>
<td>1984</td>
<td>ANDA for generics approval-Waxman-Hatch act (Drug price competition and patent term restoration act)</td>
</tr>
<tr>
<td>1985</td>
<td>New 80-125% for CI law for approval of generic products</td>
</tr>
<tr>
<td>1987</td>
<td>Standard 2x2 crossover test design for BE studies</td>
</tr>
<tr>
<td>1989</td>
<td>Generics scandal in USA. Concern for adequate documentation and validation of BE studies</td>
</tr>
<tr>
<td>1992</td>
<td>90-111% CI for narrow therapeutic index drugs: Canadian FDA</td>
</tr>
<tr>
<td>1995</td>
<td>EEC-70-143% limit for Cmax only for drug with wide safety margin</td>
</tr>
<tr>
<td>1999</td>
<td>Draft regulations for BE studies: In India</td>
</tr>
<tr>
<td>2005</td>
<td>Schedule Y (amended) and Bioequivalence guidelines—in India</td>
</tr>
</tbody>
</table>
2.2.2 BE for first entry products

BE studies may be useful during drug development and registration for a first entry product during the Investigational New Drug (IND) or New Drug Application (NDA) period to establish links between (i) early and late clinical trial formulations (ii) formulations used in clinical trial and stability studies, if different (iii) Clinical trial formulations and to be marketed drug products (iv) other comparisons as appropriate. In each comparison, the new formulation or new method of manufacture is the test product and the prior formulation or method of manufacture is the reference product.

2.2.3 BE for interchangeable multi-source products

BE studies are a critical component of Abbreviated New Drug Applications (ANDA). The purpose of these studies is to compare relative BA measures between a pharmaceutically equivalent multi-source test product and the corresponding reference pioneer product. The pioneer product is termed as reference listed drug (RLD). Together with the determination of pharmaceutical equivalence, demonstrating BE allows a regulatory conclusion of therapeutic equivalence and interchangeability between the test and reference product (CDER, 1999).

2.2.4 BE for post approval changes

Generally specifications are adequate to assure product quality on the assumption that no important change occurs post-approval. In the presence of major changes in components and composition, and/or method of manufacture of a drug product after approval, BE may need to be re-demonstrated. For approved first-entry products, the drug product after the change should be compared to the drug product before change. For approved interchangeable multi-source products, the drug product after the change should be compared to the reference listed drug.

2.2.5 Types of bioavailability

Bioavailability can be classified into four different types (Ritschel and Kearns 1998), depending on the purpose of the study and scientific questions to be solved.
2.2.5.1 Absolute bioavailability

Absolute bioavailability is the ratio of the total area under the blood level time curve upon extra vascular route of administration to the area under the blood level time curve upon intravenous administration, corrected for the difference in the dose size.

\[
\text{Absolute bioavailability} = \frac{\text{AUC}_{\text{extravascular}} \times \text{dose}_{\text{i.v.}}}{\text{AUC}_{\text{i.v.}} \times \text{dose}_{\text{extravascular}}}
\]

2.2.5.2 Relative bioavailability

Relative bioavailability is the extent (EBA) and rate (RBA) of the bioavailability of a drug from two or more different dosage forms given by the same route of administration. For determination of EBA or RBA, blood level or urinary excretion data upon single or multiple dosing can be used. According to the FDA regulation the standard used in this procedure is an approved marketed drug product, a solution of the drug or suspension of the micronized drug.

\[
\text{Relative bioavailability} = \frac{\text{AUC of A}}{\text{AUC of B}}
\]

Where B is the reference standard.

2.2.6 Bioavailability in presence of first-pass effect

Drugs showing a first-pass effect may result in considerably lower blood level time curves. Even though the entire parent drug was absorbed from the site of administration, it did not reach systemic circulation in unchanged form.

The fraction of a peroral (po) or in part, rectal dose reaching systemic circulation \( F \), under the assumption of otherwise linear kinetics can be described by eqn.

\[
F = 1 - \frac{\text{Dose}_{\text{i.v}} \times f_m}{\text{LBF}} \times \frac{\text{AUC}_{\text{i.v}}}{\text{AUC}_{\text{i.v}}} \times 60 \times \lambda
\]

\( f_m \) - fraction of drug metabolised in liver
\( \text{LBF} \) - liver blood flow
\( \lambda \) - ratio of the concentration of the drug in whole blood to that in plasma
2.2.7 Relative optimal bioavailability

This term was suggested for optimizing extent and rate of bioavailability for a drug product during the development phase.

For determination of EBA \( \text{rel. opt.} \), the active drug is administered in aqueous solution without the addition of any further excipient by the same route which is intended for the drug product under development.

\[
\text{EBA rel. opt.} = \frac{\text{AUC (drug + vehicle; granules; tablets)}}{\text{AUC solution}} \times 100
\]

2.2.8 Different approaches used for measurement of bioavailability

There are several direct and indirect methods for the measurement of bioavailability in humans. The selection of method depends on the purpose of the study, analytical method and nature of the drug product. The methods useful in quantitative evaluation of bioavailability can be broadly divided into two categories: (a) Pharmacokinetic methods (b) Pharmacodynamic methods

2.2.8.1 Pharmacokinetic Methods

These are very widely used and are based on the assumption that the pharmacokinetic profile reflects the therapeutic effectiveness of a drug. Thus, these are indirect methods. The two major pharmacokinetic methods are:

2.2.8.1.1 Plasma level-time studies

Unless determination of plasma drug concentration is difficult or impossible, it is the most reliable method and method of choice in comparison to urine data. This method is based on the assumption that two dosage forms that exhibit superimposable plasma level-time profiles in a group of subjects should result in identical therapeutic activity. The three parameters of plasma level-time studies, which are considered important for determining bioavailability, are:
$C_{\text{max}}$: The peak plasma concentration that gives an indication whether the drug is sufficiently absorbed systemically to provide a therapeutic response.

$t_{\text{max}}$: The time of peak plasma concentration corresponds to the time required to reach maximum drug concentration after drug administration. At $t_{\text{max}}$, absorption is maximized and the rate of drug absorption equals the rate of drug elimination. When comparing drug products, $t_{\text{max}}$ can be used as an approximate indication of the drug absorption rate.

$\text{AUC}$: The area under the plasma level-time curve that gives a measure of the extent of absorption or the amount of drug that reaches the systemic circulation.

The extent of bioavailability can be determined by eqn.

$$F = \frac{\text{AUC}_{\text{oral}} D_{\text{iv}}}{\text{AUC}_{\text{iv}} D_{\text{oral}}}$$

2.2.8.1.2 Urinary excretion studies

This method of assessing bioavailability is based on the principle that the urinary excretion of unchanged drug is directly proportional to the plasma concentration of drug. This method is particularly useful for drugs extensively excreted unchanged in the urine. The method involves collection of urine at regular intervals for a time span equal to 7-10 biological half-lives, analysis of unchanged drug in the collected sample and determination of the amount of drug excreted in each interval and cumulative amount excreted. The three major parameters examined in urinary excretion data obtained with a single dose study are:

$$(\text{dx}/dt)_{\text{max}}$: The maximum urinary excretion rate, is obtained from the peak of plot between rate of excretion versus midpoint time of urine collection period. It is analogous to $C_{\text{max}}$ derived from plasma level studies since the rate of appearance of drug in the urine is proportional to its concentration in systemic circulation.

$(t_{u})_{\text{max}}$: The time for maximum excretion rate, is analogous to the $t_{\text{max}}$ of plasma level data. its value decreases as the absorption rate increases.
The cumulative amount of drug excreted in the urine, is related to the AUC of plasma level data and increases as the extent of absorption increases.

The extent of bioavailability can be calculated using eqn:

\[ F = \frac{(X_u)_{oral} D_{iv}}{(X_u)_{iv} D_{oral}} \]

2.2.8.2 Pharmacodynamic Methods

These methods are complimentary to pharmacokinetic approaches and involve direct measurement of drug effect on a physiologic process as a function of time. The two pharmacodynamic methods involve determination of bioavailability from: (a) Acute pharmacologic response (b) Therapeutic response

2.2.8.2.1 Acute pharmacologic response

In some cases quantitative measurement of a drug is difficult, inaccurate or non reproducible. In such cases an acute pharmacologic effect such as effect on pupil diameter, heart rate or blood pressure can be a useful index of drug bioavailability. Bioavailability can be determined by construction of pharmacologic effect-time curve as well as dose-response graphs. The method requires measurement of responses for at least 3 biological half-lives of drug in order to obtain a good estimate of AUC.

2.2.8.2.2 Therapeutic response

Theoretically the most definite, this method is based on observing the clinical response to a drug formulation given to patients suffering from disease for which it is intended to be used. Bioequivalent drug products should have the same systemic drug bioavailability and therefore the same predictable drug response. However, variable clinical responses among individuals that are unrelated to bioavailability might be due to differences in the pharmacodynamics of the drug. Various factors affecting pharmacodynamic drug
behaviour may include age, drug tolerance, drug interactions and unknown pathophysiologic factors.

2.2.8.3 In vitro Methods

Under certain circumstances, product quality BA and BE can be documented using in vitro approaches. For highly soluble, highly permeable, rapidly dissolving, orally administered drug products, documentation of BE using an in vitro approach (dissolution studies) is appropriate based on the biopharmaceutics classification system (BCS) (CDER, 2000). The preferred dissolution apparatus is USP apparatus I (basket) or II (paddle), used at compendially recognized rotation speeds (e.g., 100 rpm for the basket and 50-75 rpm for the paddle). In other cases, the dissolution properties of some ER formulations may be determined with USP apparatus III (reciprocating cylinder) or IV (flow through cell).

2.2.9 Factors Influencing Bioavailability (Lachman et al. 1998)

2.2.9.1 Pharmaceutical factors

- Physicochemical attributes of Drug substances
- Drug solubility and dissolution rate
- Particle size and effective surface area
- Bulk and tapped density, Powder flow characterization
- Polymorphism, amorphism and hygroscopicity
- Pseudopolymorphism (hydrates/solvates)
- Salt form of the drug
- Lipophilicity
- pKa of the drug and pH
- Drug stability (% volatile, LOD, moisture content)

2.2.9.2 Dosage Form Characteristics and Pharmaceutical Ingredients

- Disintegration time (tablets/capsules)
- Dissolution time
Manufacturing variables (method of granulation, compression force, intensity of packing of capsule contents)

- Pharmaceutical ingredient (excipients/adjuvants)
- Nature and type of dosage form
- Product age and storage condition

2.2.9.3 Barrier functions:
- Age
- Gastric emptying time & Intestinal transit time
- Gastrointestinal pH
- Diseases states
- Blood flow through the GIT

Physicochemical Attributes and Drug Permeability:
The bioavailability of oral drug administration is a function of Molecular characteristics, Dosage form design, and Barrier function of the organism. Barrier to oral delivery of a compound can involve both stability and transport. GIT is challenging environment for stability and transport of drug. Permeation depends on molecular size, aqueous solubility and lipophilicity. Drug of large molecular size transported through receptor mediated endocytosis (Transcytosis), and low molecular size (<400-500) through paracellular route (passive diffusion across the intestinal membrane). The Gastro intestinal tract pH from 1-8 reacts may cause acid or base catalysis hydrolysis or non-specific hydrolysis by enzymes includes pepsin, pancreatic enzyme, cytoplasm to drug and Gastric transit time. Drug bioavailability can be altered by the nature of food present. (Brahmankar et al. 1995).

2.2.9.4 Particle size:

Dissolution rate is typically influenced by particle size and wettability. The influence of wettability on the dissolution rate of pharmaceutical powder was studied by Lippold and ohm. Example: wetting agent: Polysorbate 80 (Lippold et. al., 1986)

Particle size and surface area of a solid drug are inversely related to each other. Particle size is of importance for drugs of low solubility. The critical point seems to be if the
solubility is less than 0.3 percent. With decreasing particle size, the surface area increases, thus increasing the area of solid matter being exposed to the dissolution media and, hence, dissolution rate increases. However, the actual solubility does not significantly change with particle size reduction (micronization) in the range used in pharmaceutical manufacture. The following equation describes the dissolution rate:

\[
\frac{dc}{dt} = k \cdot a \cdot (C_s - C_t)
\]

\( \frac{dc}{dt} \) = dissolution rate (amount per unit time) (Noyes Whitney equation)

\( k \) = constant depending on intensity of agitation, temperature, structure of solid surface, and diffusion coefficient

\( a \) = surface area of undissolved solute

\( C_s \) = solubility of drug in solvent

\( C_t \) = concentration of dissolved drug at time \( t \)

Examples of drugs for which therapeutic differences have been found depending on particle size are: amphotericin, aspirin, bishydroxycoumarin, chloramphenicol, digoxin, acetonide, griseofulvin, meprobamate, nitrofurantoin, Phenobarbital, phenothiazine, prednisolone, procaine penicillin, reserpine, spironolactone, sulfadiazine and tolbutamide (Ritschel et. al., 1998).

2.2.9.5 Polymorphism:
New drug substances exist in different crystalline forms which differ in their physical properties. Polymorphism may also include salvation or hydration products (also know as pseudopolymorphs) and amorphous forms. Polymorphism has direct impact on solubility. Particle shape and powder density is depended on polymorphism (crystal forms). These two physical parameters can affect manufacturability resulting in poor flowability and compaction.

Example: Ibuprofen, acetaminophen because of crystal habit of drug tendency of poor flow and sticking. Bioequivalent product manufactured with control of the polymorphic form of the drug substance and the dissolution behaviour of the drug product. Polymorphism is the phenomenon that a drug may exist in different crystalline forms, polymorphs. Polymorphism exists only in solid state. The most stable form has highest
stability but lowest dissolution rate. The least stable form usually has the most rapid dissolution rate. The unstable (metastable) forms convert more or less slowly into the more stable form. e.g. chloramphenicol palmitate appears in three different polymorphs, but only polymorph B is biologically active, since the other forms do not dissolve and are not hydrolysed. The polymorphs differ from each other with respect to their physical properties such as solubility, melting point, density, hardness and compression characteristics.

Some drugs can exist in amorphous form (i.e. having no internal crystal structure). In general, the amorphous state is more soluble and has a higher dissolution rate than the crystalline form. The crystalline form requires a higher amount of energy to free a molecule of drug from it than does the amorphous form. e.g. amorphous novobiocin and amorphous chloramphenicol esters are biologically active while their crystalline forms are inactive.

2.2.9.6 Ionization:
An acid in acid solution will not ionize; an acid in basic solution will ionize. A base in a basic solution will not ionize; a base in acid solution will ionize. The amount of drug that exists in unionized form is a function of dissociation constant (pK<sub>a</sub>) of the drug and pH of the fluid at the absorption site.

The negative log of the acid ionization constant (pKa) is defined as the ability of an ionizable group of an organic compound to donate a proton (H<sup>+</sup>) in aqueous media normally at 25°C. Henderson–Hasselbach equations are used to identify the percent of drug ionized at gastrointestinal pH. i.e.,

Acids: pH=pK<sub>a</sub>+log ionized drug concentration/Unionized drug concentration

Bases: pH=pK<sub>a</sub>+log unionized drug concentration/ionized drug concentration

When the concentration of ionized and unionized drug becomes equal, and thus pH=pK<sub>a</sub>. When the concentration of ionized and unionized drugs are not equal, the percent of ionization calculated by following formula:

Percent of Ionization = \[ \frac{100}{1+10^{x(pH-PKA)}} \]

Where x = -1 for acid drug, 1 for basic drug.
2.2.9.7 Partition coefficient:

Partition coefficient of a drug substance can provide useful information about its permeability characteristics. The partition coefficient is the ratio of concentrations of un-ionized compound between the two solutions. The logarithm of the ratio of the concentrations of the un-ionized solute in the solvents is called log P.

LogP is the octanol-water partition coefficient, P, is a measure of the differential solubility of a neutral substance between these immiscible liquids and thereby, a descriptor of hydrophobicity (or the lipophilicity) of a neutral substance. It is typically used in its logarithmic form, logP. Higher the value, more the hydrophilic and faster the dissolution in aqueous fluids. If log p>4 then the drug is very lipophilic, which is practically estimated by shake flask method. Usually intestinal permeability increases with lipophilicity but decreases with molecular weight or H-bonding properties. The formula to calculate log P is given below.

\[ \log P_{\text{oct/wat}} = \log \left( \frac{\text{Solute}_{\text{octanol}}}{\text{Solute}_{\text{un-ionized water}}} \right) \]

Ideally, for optimum absorption, a drug should have sufficient aqueous solubility to dissolve in the fluids at the absorption site and lipid solubility high enough to facilitate the partitioning of the drug in the lipoidal biomembrane and into systemic circulation. In other words, a perfect hydrophilic-lipophilic balance (HLB) should be there in the structure of the drug for optimum bioavailability. Gastro intestinal tract is a simple lipoidal barrier to the transport of drug. Larger the fraction of unionized drug, faster the absorption and greater the lipophilicity (K_{ow}) of the unionized drug, better the absorption.

Table 2.3: Factors affecting absorption of a drug from its dosage form (Brahmankar et al., 1995).

<table>
<thead>
<tr>
<th>PHARMACEUTICAL FACTORS</th>
<th>Dosage form related factors</th>
<th>PATIENT RELATED</th>
</tr>
</thead>
<tbody>
<tr>
<td>Physicochemical properties of drug substances</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Most drugs are either weak acids or weak bases. One of the easiest approaches to enhance the solubility and dissolution rate of such drugs is to convert them into their salt forms. At a given pH, the solubility of a drug, whether acidic/basic or its salt form, is a constant.

### Drug pKa and lipophilicity and GI pH-pH Partition hypothesis

The pH Partition theory explains in simple terms, the process of drug absorption from the GIT and its distribution across all biologic membranes. The theory states that for a drug...
compound of molecular weight greater than 100, which are primarily transported across
the biomembrane by passive diffusion, the process of absorption is governed by:

- The dissociation constant (pKa) of the drug.
- The lipid solubility of the unionised drug (a function of drug K_{o/w}).
- The pH at the absorption site.

2.2.9.10 Lipophilicity and drug absorption

The pKa of a drug determines the degree of ionisation at a particular pH and that only the
unionised drug, if sufficiently lipid soluble, is absorbed into the systemic circulation. Thus,
even if the drug exists in the unionised form, it will be poorly absorbed if it has poor lipid
solubility. Ideally, for optimum absorption, a drug should have sufficient aqueous solubility
to dissolve in the fluids at the absorption site and lipid solubility (K_{o/w}) high enough to
facilitate the partitioning of the drug in the lipoidal membrane and into the systemic
circulation. Hence, a perfect hydrophilic-lipophilic balance (HLB) should be there in the
structure of the drug for optimum bioavailability.

2.2.10 Patient related factors

2.2.10.1 Age

In infants, the gastric pH is high and intestinal surface and blood flow to the GIT is low
resulting in related absorption pattern in comparison to adults. In elderly persons, causes
of impaired drug absorption include altered gastric emptying, decreased intestinal surface
area and GI blood flow.

2.2.10.2 Gastric emptying

Apart from dissolution of a drug and its permeation through the biomembrane, the
passage from stomach to the small intestine, called as gastric emptying can also be rate
limiting step in drug absorption because the major site of drug absorption is intestine.
Thus generally speaking, rapid gastric emptying increases bioavailability of a drug.
2.2.10.3 Rapid gastric emptying is desired where:
A rapid onset of action is desired e.g. sedatives. Dissolution of drug occurs in the intestine e.g. enteric coated dosage forms. The drugs are not stable in the gastric fluids e.g. penicillin G, and erythromycin. The drugs are best absorbed from the distal part of the small intestine e.g. vitamin B₁₂.

2.2.10.4 Intestinal Transit
Since small intestine is the major site for absorption of most drugs, long intestinal transit time is desirable for complete drug absorption. The residence time depends upon the intestinal motility or contractions. The mixing movement of the intestine that occurs due to peristaltic contractions promote drug absorption, firstly, by increasing the drug-intestinal membrane contact, and secondly, by enhancing the drug dissolution especially of poorly soluble drugs, through induced agitation.

2.2.10.5 Blood flow to the GIT
GIT is extensively supplied by blood capillary network and the lymphatic system. The absorbed drug can thus be taken by the blood or the lymph. Since the blood flow rate to the GIT (splanchnic circulation) is 500 to 1000 times (28% of cardiac output) more than the lymph flow, most drugs reach the systemic circulation via blood whereas only a few drugs, especially low molecular weight, lipid soluble compounds are removed by lymphatic system. The high perfusion rate of GIT ensures that once the drug has crossed the membrane, it is rapidly removed from the absorption site thus maintaining the sink conditions and concentration gradient for continued drug absorption.

Table 2.4: Physiological factors affecting bioavailability (Ross and Wilson, 1997)

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Stomach</th>
<th>Small intestine</th>
<th>Large intestine</th>
<th>Rectum</th>
</tr>
</thead>
<tbody>
<tr>
<td>pH range</td>
<td>1-3</td>
<td>5-7.5</td>
<td>7.9-8.0</td>
<td>7.5-8.0</td>
</tr>
<tr>
<td>Length (cms)</td>
<td>20</td>
<td>285</td>
<td>110</td>
<td>20</td>
</tr>
<tr>
<td>Diameter (cms)</td>
<td>0.1-0.2</td>
<td>2.5</td>
<td>5</td>
<td>2.5</td>
</tr>
</tbody>
</table>
A drug with poor bioavailability is the one with
- Poor aqueous solubility and/or slow dissolution rate in the biologic fluids.
- Inadequate partition coefficient and thus poor permeation through the biomembrane
- Poor stability of the dissolved drug at the physiologic pH
- Extensive presystemic metabolism

2.2.11 Design and evaluation of BE study

The preferred approach is an in vivo study carried out in healthy volunteers to whom the 2 preparations (generic and innovator) are alternatively administered. The design and evaluation of well-controlled bioequivalence studies require the cooperative input from pharmacokineticists, statisticians, clinicians, bio-analytical chemists, and others.

2.2.11.1 Design

The design of a bioavailability and/or bioequivalence study is dependent upon the drug, dosage form and study objectives. For BE studies, both the test and reference drug formulations contain the pharmaceutical equivalent drug in the same dose and are given by the same route of administration. A pilot study in small number of subjects can be
carried out before proceeding with a full BE study. This study can be used to validate analytical methodology, assess variability, optimize sample collection time intervals or provide any other information. Non replicate crossover study designs are recommended by FDA (CDER, 2003) for immediate release and modified release dosage forms. However replicate designs can also be used. The recommended method for analysis to establish BE is average bioequivalence. The study should be of crossover designs and suitably randomized as far as possible. Some of the designs are discussed below.

2.2.11.2 Two-Period Crossover Design

In case of two formulations, an even number of subjects should be randomly divided into two equal groups. In the first period, each member of one group will receive a single dose of the test formulation and each member of the other group will receive standard formulation. After a suitable washout period (generally 5 half lives), in the second period, each member of the respective groups will receive a dose of an alternative formulation and the experiment will be repeated.

The design can be depicted as follows:

<table>
<thead>
<tr>
<th>Vol. No.</th>
<th>Period 1</th>
<th>Period 2</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>A</td>
<td>B</td>
</tr>
<tr>
<td>2</td>
<td>B</td>
<td>A</td>
</tr>
<tr>
<td>3</td>
<td>A</td>
<td>B</td>
</tr>
<tr>
<td>4</td>
<td>A</td>
<td>B</td>
</tr>
<tr>
<td>5</td>
<td>B</td>
<td>A</td>
</tr>
<tr>
<td>6</td>
<td>B</td>
<td>A</td>
</tr>
</tbody>
</table>

2.2.12 Latin Square Design

In case of more than two formulations, a latin square design should be used. For example in a bioequivalence study of 3 formulations, a group of volunteers will receive formulations in the sequence shown below:
The next group of 3 volunteers will receive formulations in the same sequence as shown above.

2.2.13 Balance Incomplete Block Design (BIBD)

In case there are more than 3 formulations, the Latin square design will not be ethically advisable, mainly because each volunteer may require the drawing of too many blood samples. However, if each volunteer is expected to receive at least two formulations, then such a study can be carried out using Balanced Incomplete Block Design. As per this design, if there are four formulations, six possible pairs or formulations can be chosen from four formulations. Then, the first 6 volunteers will receive these six pairs of formulations and the next six volunteers will receive the same six pairs in reverse order. The design is depicted below:

<table>
<thead>
<tr>
<th>Vol. No.</th>
<th>Period 1</th>
<th>Period 2</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>A</td>
<td>B</td>
</tr>
<tr>
<td>2</td>
<td>B</td>
<td>C</td>
</tr>
<tr>
<td>3</td>
<td>C</td>
<td>A</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Vol. No.</th>
<th>Period 1</th>
<th>Period 2</th>
</tr>
</thead>
<tbody>
<tr>
<td>7</td>
<td>B</td>
<td>A</td>
</tr>
<tr>
<td>8</td>
<td>C</td>
<td>A</td>
</tr>
<tr>
<td>9</td>
<td>D</td>
<td>A</td>
</tr>
<tr>
<td>10</td>
<td>C</td>
<td>B</td>
</tr>
<tr>
<td>11</td>
<td>D</td>
<td>B</td>
</tr>
<tr>
<td>12</td>
<td>D</td>
<td>C</td>
</tr>
</tbody>
</table>
The minimum acceptable number of volunteers will be 12.

\[ n \geq \frac{(\sigma^2)}{2D^2 [\alpha + t_{\beta}]^2} + 0.25 \alpha^2 \]

Where,

- \( n \) = no. of volunteers
- \( \alpha \) = Required level of significance (0.05)
- \( \beta \) = Required power of test (0.80)
- \( \sigma^2 \) = Error mean sum of squares from ANOVA (estimated/guess)
- \( D \) = Minimum difference between the means which if present, ought to be detected

The bioequivalence studies are conducted according to a well-defined protocol.

Some elements of a bio-equivalence protocol are listed in Table 2.5:

Table 2.5: Elements of the bioavailability Protocol

| INVESTIGATORS' DECLARATION |
| FACILITIES               |
| 2.1 Clinical Services & Clinical Laboratory |
| 2.2 Analytical, Pharmacokinetics & Statistical Services |
| OBJECTIVE                |
| PRODUCTS TO BE EVALUATED |
| 4.1 REFERENCE (R) |
| 4.2 TEST (A) |
| 4.3 TEST (B) |
| INTRODUCTION            |
| PHARMACOLOGY            |
| 6.1 Absorption, Distribution, Metabolism and Excretion |
| 6.2 Adverse Effects     |
| 6.3 Dosage              |
| STUDY DESIGN            |
| 7.1 Summary             |
| 7.2 Number of Subjects  |
| 7.3 Admissions and Stay |
| 7.4 Fasting/Meals       |
| 7.5 Sampling Schedule   |
7.6 Blood Pressure
7.7 Washout Period

RESTRICTIONS
8.1 Medications
8.2 Diet
8.3 Activity

SELECTION OF SUBJECTS
9.1 Inclusion Criteria
9.2 Exclusion Criteria

10. SCHEDULE OF ASSESSMENTS

STUDY MEDICATION
11.1 Handling, Storage and Accountability Procedures
11.2 Dose
11.3 Assignment to Treatment Sequences
11.4 Assessment of Compliance

HAEMODYNAMIC MEASUREMENTS

PHARMACOKINETICS
13.1 Blood Sampling
13.2 Analytical Procedures
13.3 Pharmacokinetic Parameters

SAFETY
14.1 Clinical Safety Measurements

HANDLING OF SAFETY PARAMETERS
15.1 Adverse Events

STATISTICAL ANALYSIS

DEVIATIONS

ETHICAL CONSIDERATION
18.1 Basic Principles
18.2 Institutional Review Board
18.3 Informed Consent
18.4 Withdrawal/Drop-out of Subjects from Study
18.5 Volunteer Compensation
2.2.14 Statistical issues in BE studies

The pharmacokinetic parameters, C_{max}, T_{max} and AUC should be subjected to a three-way analysis of variance (3-way ANOVA) in order to test differences due to formulations, period and subjects. A more complex ANOVA may be appropriate in some circumstances, e.g. if treatments are replicated. The standard parametric ANOVA assumes homogeneity of variances, normality and additivity of independent variables.

In order to ensure homogeneity of variances between treatments, Bartlett's test or a similar test should be carried out prior to performing the ANOVA (DCGI, 2002). The primary comparison of interest in a bioequivalence study is the ratio of average parameter data (AUC or C_{max}) from the test and reference formulations rather than the difference between them. Log transformation of the data allows the general linear statistical model to draw inferences about the ratio of the two averages on the original scale. Log transformation thus achieves the general comparison based on the ratio rather than on the difference.

Moreover, plasma concentration data, including AUC and C_{max}, tend to be skewed and their variances tend to increase with the means. Log transformation corrects this situation and makes the variances independent of the mean.

Further, the frequency distribution skewed to the left, i.e., those with a log tail to the right is made symmetrical by log transformation.
In case no suitable transformation is available, the non-parametric method should be used. Tmax values being discrete, data on Tmax should be analysed using non-parametric methods.

2.2.15 Two one-sided tests procedures (TOST):

This procedure is also referred to as confidence interval approach. This method is used to demonstrate if the bioavailability of the drug from the test formulation is too high or low in comparison to the reference drug product. The 90% confidence limits are estimated for the sample means. In this test, presently required by the FDA, a 90% confidence interval about the ratio of means of the two drug products must be within $\pm 20\%$ for measurement of the rate and extent of drug bioavailability. The lower 90% CI for the ratio of means cannot be less than 0.8, and the upper 90% CI for the ratio of the means cannot be greater than 1.20. The 90% CI is a function of sample size and study variability, including inter and intra subject variability (CDER, 2003).

Current DCGI requirements for bio-equivalence approval is that 90% confidence interval should be within 80-125% for log transformed AUC and log transformed Cmax. For narrow therapeutic index drugs, the log transformed Cmax should be stricter.

Table 2.6 mentions the bioequivalence criteria followed by various regulatory agencies in the world.

<table>
<thead>
<tr>
<th>Agency</th>
<th>Log transformed Parameter</th>
<th>AUC&lt;sub&gt;θ-τ&lt;/sub&gt; using 90 % CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>USFDA</td>
<td>C&lt;sub&gt;max&lt;/sub&gt; using 90 % CI 80-125 % of reference</td>
<td>80-125 % of reference</td>
</tr>
<tr>
<td>CPMP (EU)</td>
<td>80-125 % of reference (if clinically acceptable)</td>
<td>80-125 % of reference</td>
</tr>
<tr>
<td>CANADIAN FDA (Health Canada)</td>
<td>80-125 % of reference</td>
<td>80-125 % of reference</td>
</tr>
<tr>
<td>DCGI</td>
<td>80-125 % of reference</td>
<td>80-125 % of reference</td>
</tr>
</tbody>
</table>

Table 2.6: Criteria of bio-equivalence of various regulatory agencies
Presently, bioequivalence testing compares products based on averages obtained in the AUC and C_{max} (Benet et al., 1995). However, the true purpose of bioequivalence testing should be to assure switchability of a patient's medication, (Wittkowsky, 1997).

### 2.3 Generic Drugs

A generic drug is the same as a reference-listed (i.e., brand name) drug with respect to conditions of use, active ingredient(s), route of administration, dosage form, strength, and labeling (CDER, 2003). In addition, the generic drug must be bioequivalent to (i.e., perform in the same manner as) the brand name drug.

A generic drug that is therapeutically equivalent is expected to have the same clinical effect and safety profile as the brand name drug when administered under the conditions specified in the labeling. If generic drugs are determined to be therapeutically equivalent, physicians and pharmacists can substitute them for brand name drugs.


In the early 1970s interest in generic drug products began to increase as various third party groups sought to reduce the cost of prescription medication. At that time there was no formal approval process that was routinely used by the USFDA to evaluate the safety and efficacy of generic drug products. In the intervening year, average bioequivalence testing has been adopted by FDA as a means to determine the equivalence of multisource drug products, but only after expiration of the patent (generally 17 years after the discovery of the original drug), can the competitors sell the generic version of drug (Meyer, 1998).

Because of the lower cost of development and competition in the market place, generic drug usually sell for less than the brand name drug products from the original manufacturers (innovators or pioneers). This fact has led many to believe that generic drugs are somehow inferior to brand-name products. This is obviously not true. The US
FDA mandate that the generic drug must be as safe and effective as the brand-name drugs

2.3.1 Generic drugs and product substitution (Interchangeability)

New multisource (generic) pharmaceutical products must be of good quality and at least as safe and efficacious as existing products. The need for interchangeability arises when a patient may change from one brand to another, for example in these circumstances:

- Physicians prescribe by generic name;
- Generic substitution by the pharmacist is permitted by national legislation;
- The same brand is not always available, for example in remote areas of the country;
- Patients in hospitals are given whatever brand the hospital has in stock, and often different brands are stocked on different occasions;
- Patients receive a different brand after discharge from hospital.

By their nature, different brands of modified (sustained, continuous, prolonged, slow) release products are more likely not to be equivalent than are different brands of immediate, conventional release products. Some DRAs take the view that such products should never be considered interchangeable, while others define a series of studies that should be conducted, including in some circumstances comparative clinical trials. For delayed release products, such as enteric-coated tablets, interchangeability is more readily demonstrated (WHO, 1998).

2.3.1.1 Considerations for modified-release products

Modified-release products include extended-release products and delayed release products. Extended-release products are variously known as controlled-release, prolonged-release and sustained-release products. To establish the bioequivalence of modified-release products, a single-dose, non-replicate cross-over, fasting study comparing the highest strength of the multisource and the comparator product should be performed. Single-dose studies are preferred to multiple-dose studies as single-dose studies are considered to provide more sensitive measurements of the release of API from the pharmaceutical product into the systemic circulation. Multiple-dose studies may need to be considered (in addition to a single dose study) for extended-release dosage
forms with a tendency to accumulate. The comparator product in this study should be a pharmaceutically equivalent modified-release product. The pharmacokinetic bioequivalence criteria for modified-release products are basically the same as for conventional release dosage forms.

Co-administration of food with oral pharmaceutical products may influence drug bioavailability and also in certain cases pharmacokinetic bioequivalence. In addition to physiological changes in the gastrointestinal tract, food can affect the release of the API from the formulation. A concern with modified-release products is the possibility that food may trigger a sudden and abrupt release of the API leading to "dose dumping". This would most likely be manifested as a premature and abrupt rise in plasma concentration time profile. Therefore, a pharmacokinetic bioequivalence study under fed conditions is a mandatory requirement for orally administered modified-release pharmaceutical products. A fed-state pharmacokinetic bioequivalence trial should be conducted after the administration of an appropriate standardized meal at a specified time (usually not more than 30 minutes) before taking the medicine. A high-fat meal often provides a maximal challenge to the robustness of release from the formulation with respect to prandial state. The composition of the meal should also take local diet and customs into consideration. The composition and caloric breakdown of the test meal should be provided in the study protocol and report.

Food-effect studies are necessary for all multisource modified-release formulations to ensure the absence of "dose dumping". The latter signals a formulation failure such that the dose is released all at once rather than over an extended period of time. This results in a premature and abrupt rise in the plasma concentration time profile. The composition of the meal should also take local diet and custom into consideration.

Generic substitution for drugs has become a widespread practice as the Food and Drug Administration (FDA) approves bioequivalent drugs, and patients and payors seek ways to trim healthcare costs. Formulation substitution (FS) can include proprietary to generic, one generic to another or generic to proprietary. In many cases, FS poses no problem to the health of the patient.

It is critical that physicians have a better understanding of generic drugs, how they are
tested and the true comparative value of FS to better treat patients and avoid any possible adverse consequences. (Banahan et al., 1997)

Although 250 mg of a trade-name chemical is identical to 250 mg of the same generic chemical, a 250-mg generic pill containing that chemical may or may not have the same effect in the body as a 250-mg trade-name pill. That is because everything that is used in a particular product formulation affects how it is absorbed into the bloodstream. Inactive ingredients such as coatings, stabilizers, fillers, binders, flavorings, diluents, and others are necessary to turn a chemical into a usable drug product. These ingredients may be used to provide bulk so that a tablet is large enough to handle, to keep a tablet from crumbling between the time it is manufactured and the time it is used, to help a tablet dissolve in the stomach or intestine, or to provide a pleasant taste and color. Inactive ingredients are usually harmless substances that do not affect the body. However, because inactive ingredients can cause unusual and sometimes severe allergic reactions in a few people, one version, or brand, of a drug may be preferable to another. For example, chemicals called bisulfites (such as sodium metabisulfite), which are used as preservatives in many products, cause asthmatic allergic reactions in many people. Consequently, drug products containing bisulfites are prominently labeled as such. (Silverman, 2006)

Interchangeability and Substitution: Theoretically, any generic drug that is bioequivalent to its trade-name counterpart may be interchanged with it. For drugs that are off-patent, the generic drug may be the only form available. To limit costs, many doctors write prescriptions for generic drugs whenever possible. Even if the doctor has prescribed a trade-name drug, the pharmacist may dispense a generic drug unless the doctor wrote on the prescription that no substitution can be made. Also, insurance plans and managed care organizations may require that generic drugs be prescribed and dispensed whenever possible to save money.

Sometimes generic substitution may not be appropriate. For example, some available generic versions may not be bioequivalent to the trade-name drug. Such generic drugs may still be used, but they may not be substituted for the trade-name product. In cases in which small differences in the amount of drug in the bloodstream can make a very large difference in the drug's effectiveness, generic drugs are often not substituted for trade-
name drugs, although bioequivalent generic products are available. Warfarin, an
anticoagulant, and phenytoin, an anticonvulsant, are examples of such drugs. Finally, a
generic product may not be appropriate if it contains an inactive ingredient that the person
is allergic to.

Drugs that must be given in very precise amounts are less likely to be interchangeable,
because the difference between an effective dose and a harmful or an ineffective dose
(the margin of safety) is small. Digoxin, used to treat people with heart failure, is an
example.

The substitution of a generic drug can sometimes cause other problems for the consumer.
A doctor may write a prescription for a trade-name product and discuss the trade-name
product with the consumer. If a pharmacist dispenses an equivalent generic product and
the label does not also list the reference (trade-name product), the consumer may not
know how the generic product relates to the drug the doctor prescribed. To prevent this
confusion, pharmacists should include the reference trade name on the label when a
generic product is substituted.

The FDA has become much stricter on the approval of generic drugs since a 1980
scandal. This scandal tainted the generic drug industry in many people's eyes. Since
1990, the stricter regulations have helped ensure that generics are really as good as the
brand names.

Without much success, a number of studies have attempted to prove that particular brand
drugs are more efficacious than their generic counterparts. Of particular controversy is
one well-publicized story in which a brand name manufacturer paid a university
researcher to prove the brand drug (Synthroid) was superior to the generic
(levothyroxine). The researcher actually found the generic to be equivalent and even
somewhat better. What resulted was a long battle between the researcher and the
manufacturer, because the manufacturer refused to publish the data to support that the
generic was superior to their brand name drug. Seven years later, the brand manufacturer
published another study opposing the researcher's results. However, the FDA is now
requiring all manufacturers of that drug to demonstrate that their products are
bioequivalent (Dong et al, 1997).
While generic substitution is clearly defined in many cases, some medication categories require special consideration, i.e., critical dose and narrow therapeutic index drugs, products with special release mechanisms, bioengineered protein products, many hormonal products, older drugs marketed before 1938 that were not subject to FDA approval and others with limited bioequivalence data (Manolakis, 2007).

2.3.2 Clinical concerns about generic drugs

In 1995–1996, the UK Medicines Control Agency (MCA) tested 2427 generic product samples. A total of 228 deficiencies were discovered, and the MCA required 84 product quality improvements concerning labeling and packaging, analytic methods, and product specification. The report did not provide details of the findings, such as whether the deficiencies were related to branded or generic medicines and the gravity of the errors detected. The independent assessment of the quality of branded and generic products is hindered and limited. The FDA has recognized that with ~20% of drugs for which several branded or generic products are commercially available in the United States cannot be considered to be bioequivalent, and therefore are not freely interchangeable (Nightingale, 1987). However, to date, there is no documented evidence of failure of a generic formulation that is due to a bioequivalence determination (Henderson, 2001). Furthermore, the FDA Therapeutic Inequivalency Action Coordinating Committee maintained that a number of published reports of therapeutic inequivalence could not be supported when reviewed in greater depth and that, in many cases, therapeutic failures were the result of disease progression rather than the inequivalence of different formulations of the same drug (Rheinstein, 1990).

Although most generic products available today can be considered therapeutically equivalent to the corresponding brand-name products, a number of drug characteristics have been identified that can cause problems with the assessment of bioequivalence: relative insolubility in water, narrow therapeutic range, and nonlinear kinetics (Besag, 2000). Clinical practice also has revealed a number of different drug classes for which bioequivalence issues warrant caution in generic substitution: cardiovascular agents, anticonvulsant agents, psychotropic agents, nonsteroidal anti-inflammatory drugs (NSAIDs), and levothyroxine sodium. In addition, in several therapeutic categories, potential risks of generic substitution are thought to exist: modified-release formulations,
low-dose oral contraceptives and other hormonal therapies, and proton pump inhibitors (PPIs).

2.3.3 Therapeutic Equivalence and the Orange Book
A book published by the FDA each year and updated periodically also provides guidance about which drugs are interchangeable. This book, Approved Drug Products With Therapeutic Equivalence Evaluations (also known as "the orange book" because it has a bright orange cover), is available both in print and online to anyone but is intended for use by doctors and pharmacists.

The Orange Book is intended to provide public information and advice to health professionals and health agencies in order to promote public education in drug product selection and to foster containment of health care costs. The guide listed drugs classified as therapeutically equivalent to each other, and gives them an "A" rating. If the FDA does not consider a drug therapeutically equivalent, it is given a "B" rating. Most pharmacies purchase drugs with an "A" rating to dispense as generic.

2.3.4 Hatch-Waxman Act
The dual purpose of the Hatch-Waxman Act was to encourage the development of new innovator drugs by extending patent rights and to establish procedures facilitating the approval of low-cost generic drugs. These amendments to the FDCA codified in statute an abbreviated process (ANDA) for post-1962 drugs whereby a generic company could gain approval of its version of a drug without repeating the expensive and lengthy clinical trials used to establish safety and efficacy of the innovator drug (Drug Price Competition and Patent Term Restoration Act of 1984). Products approved under an ANDA must be pharmaceutical equivalents (i.e., have the same active ingredient(s), route of administration, dosage form, and strength) as the reference drug. They must also be bioequivalent and the manufacturer must supply other basic technical information related to manufacturing of the product that is normally required of an NDA [Federal Food, Drug and Cosmetic Act, section 505(j)(8)]. Generic drugs are pharmaceutical equivalents only
with respect to their active ingredients. The binders, diluents, and excipients (filler) in the formulation, as well as the method of manufacture, may vary.

2.3.5 Generic drug scandal and FDA reaction
The Waxman-Hatch Act, eased the testing requirements for generic versions of branded drugs. The elimination of this barrier to entry, along with the patent expiration for many very popular prescription drugs has led to a dramatic increase in the fraction of total prescriptions written for generics. Generic drugs' share of prescriptions sold by retail pharmacies roughly doubled in the 1980's (Frank and Salkever, 1997). In the 1990's, the generic share increased by an additional 30 percent, and in the year 2000, generics are expected to comprise 44 percent of all prescriptions. The generic industry is forecast to continue to expand as several blockbuster drugs will come off patent in the next few years. The $27 billion market for generic drugs is forecast to grow to $60 billion in 2010. The increased role of generic drugs has had an impact on the economics of the healthcare industry. Generic drugs are typically 30-60% cheaper than branded versions of the same product (Grabowski et al., 1996) and laws encouraging doctors to prescribe generic drugs when available are a part of the current effort to hold down the cost of healthcare.

In 1988, generic drug manufacturer Mylan Laboratories felt competitors' applications seemed to be moving ahead of its own, violating the OGD's policy of reviewing applications in the order received. Generic firms approved earlier in the queue have the advantage of less competition when they enter a market. Fewer generic versions for a particular drug means higher prevailing generic price and greater profits (Frank et al., 1994). The following year, Mylan's suspicions were confirmed. Officials at the OGD had been taking bribes to change the order of applications within the review queue. Also in 1989, FDA inspectors found that certain generic drug applicants were falsifying information in their ANDAs. One of the discoveries made by FDA inspectors was that some firms, when required to submit a sample of their product for the ANDA, were actually submitting a sample of innovator product. Needless to say, these samples did well in bioequivalance testing. These improprieties at the FDA and among some of the generic drug manufacturers posed a threat to the integrity of this blossoming industry. If the public were to lose confidence in the generic drug industry, manufacturers and
regulators would have a difficult time convincing people to accept these products as near-perfect substitutes for the more expensive branded drugs.

To bolster public confidence, the FDA initiated a crackdown on generic drug manufacturers and on its own reviewers in the OGD. The post-scandal crackdown was able to restore public confidence in the generic drug industry.

2.4 Biopharmaceutical Classification Of Drugs:
Based on aqueous solubility and intestinal permeability biopharmaceutical classification system classifies the drugs as

Class 1: High soluble, highly permeable
Class 2: Low soluble, highly permeable
Class 3: High soluble, low permeable
Class 4: Low soluble, low permeable

**High solubility:**
A drug substance is considered highly soluble when the highest dose strength is soluble in 250 ml or less of aqueous media over the pH range of 1-7.5. The pH solubility profile of the drug substance is determined at 37 ± 10°C in aqueous medium with pH in the range of 1-7.5. A sufficient number of pH conditions should be evaluated to accurately define the pH-solubility profile.

**High permeability:**
A drug substance is considered to be highly permeable when the extent of absorption in humans is determined to be 90% or more of an administered dose based on a mass balance determination or in comparison to an intravenous reference dose. (e.g., when the absolute bioavailability is 90% or more, or when 90% or more of the administered drug is recovered in urine). The methods used for determination of permeability include:

a. Mass balance studies, Absolute bioavailability studies and Intestinal perfusion methods in human
b. In vivo or in situ intestinal perfusion in a suitable animal model
c. In vitro permeability methods using excised intestinal tissues
d. Monolayers of suitable epithelial cells e.g. Caco-2 cells or TC-7 cells

Class I drugs exhibit a high dissolution and absorption. The rate limiting step is drug dissolution and if dissolution is very rapid then gastric emptying rate becomes the rate determining step.


Class 2 drugs have a high absorption but a low dissolution therefore absorption is limited primarily by drug dissolution in the gastrointestinal tract. In vivo drug dissolution is then a rate limiting step for absorption except at a very high dose.


Class 3 drugs, have high dissolution, low absorption. In vivo permeability is rate limiting step for drug absorption. These drugs exhibit a high variation in the rate and extent of drug absorption. Since the dissolution is rapid, the variation is attributable to alteration of physiology and membrane permeability rather than the dosage form factors.


Class 4 drugs exhibit a lot of problems for effective oral administration. The route of choice for administering is parenteral with the formulation containing solubility enhancers.

2.5 Invitro Dissolution Studies:

In the pharmaceutical industry, dissolution is defined as the amount of drug substance that goes into solution per unit time under standardized conditions of liquid/solid interface, temperature and solvent composition, i.e. mass transfer from the solid surface to the liquid phase. Intrinsic dissolution rate can be defined as rate of dissolution pure pharmaceutical active when conditions such as pH, surface area, temperature, agitation, rate and ionic strength of dissolution media kept constant.

Important dissolution factors can be identified by Noyes Whitney equation:

\[
\frac{dC}{dt} = \frac{D \cdot A \times (C_s - C_b)}{h}
\]

Where \( \frac{dC}{dt} \) is a rate of drug dissolution at time 't'

A = surface area of the particle, \( h \) = thickness of the stagnant film layer

\( C_s \) = Saturated solubility of compound at the particle media interface

\( C_b \) = Concentration of compound in the bulk medium \( C_b << C_s \)

\( D = 1/N \times (VA) \); \( D \) = diffusion coefficient of compound in the medium

Where \( N \) = solvent viscosity; \( VA \) = Solute molecular volume;

In vivo tests are extremely costly, tedious and time consuming moreover exposing the healthy subjects to hazards of drugs. So need to reduce our reliance on in vivo studies. Dissolution is a prerequisite for bioequivalence since the drug must first dissolve before it can be absorbed by the gastrointestinal tract\(^9\). Dissolution tests are now designed to mimic the general conditions encountered in the physiological environment of the GIT. The dissolution of drugs from orally administered solid dosage forms in vivo and in vitro is influenced by variations in the natural or simulated gastrointestinal fluid (and physical variables such as hydrodynamic flow, and mechanical stress. The physiological conditions that can affect drug release include the following: Intestinal transit time, gastric emptying and variable pH.
The dissolution test methods are also now designed to mimic the general conditions encountered in the physiological environment of the GIT and they are desirable alternate for in vivo tests as well as quality control tests.

2.5.1 Dissolution Method Development:
There are several factors that must be considered in the design of a dissolution test. Selection of apparatus, Nature of agitation, Speed of agitation (50/75/100 rpm), Performance precision of the apparatus, media composition, Viscosity, Volume (500/900/1000/2000ml), Temperature and 'sink conditions' to be maintained, since in vivo 'sink condition' created due to intestinal permeability and in vivo dissolution is a complex process, method of introduction of the dosage form, location of dosage unit, sampling techniques, changing the dissolution fluid, pH of the media and Time points to get discrimination etc.

Systematic Approach involves:
- Literature information: Reference listed drugs, summary basis of approval, Physicians desk reference, Pharmacokinetic data, BCS class, food affects, particle size, crystal form, bulk density of API.

2.5.2 Sink condition
Maintenance of large volumes of solution defined as 'sink conditions' (drug concentration in solution maintained constant at a low level) 3 times the unit dose to be taken for studies, NLT 1.5 times the unit dose is the acceptance criteria; at 25°C using drug, at 37±0.5°C using drug; At 25°C using drug + process placebo; at 37±0.5°C using drug + process placebo.

Dissolution media selection is based on physiological conditions, pH solubility profile, pH stability profile, pKa of the drug substance, partition coefficient.

Medias: 0.1N HCl, 0.01N HCl, pH 4.5 Acetate/Phosphate buffer, pH 6.8 Phosphate buffer, pH 7.2 /7.4 Phosphate buffer.

2.5.3 Selection of apparatus:
49
<table>
<thead>
<tr>
<th>Tablets/Capsules</th>
<th>Apparatus 1</th>
<th>Rotating basket</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tablets/Capsules</td>
<td>Apparatus 2</td>
<td>Paddle assembly</td>
</tr>
<tr>
<td>Escalating pH media</td>
<td>Apparatus 3</td>
<td>Reciprocating cylinder</td>
</tr>
<tr>
<td>Low soluble drugs</td>
<td>Apparatus 4</td>
<td>Flow-through cell</td>
</tr>
<tr>
<td>Semisolids and transdermal</td>
<td>Apparatus 5</td>
<td>Paddle over disk</td>
</tr>
<tr>
<td>Transdermal patches</td>
<td>Apparatus 6</td>
<td>Cylinder</td>
</tr>
<tr>
<td>Transdermal patches</td>
<td>Apparatus 7</td>
<td>Reciprocating holder</td>
</tr>
</tbody>
</table>

**European Pharmacopoeia (Pharm Europa, 6th ed., 2007)**

<table>
<thead>
<tr>
<th>For solid dosage forms</th>
<th>Paddle/basket/flow through cell</th>
</tr>
</thead>
<tbody>
<tr>
<td>For transdermal patches</td>
<td>Disk assembly method /rotating cylinder</td>
</tr>
<tr>
<td>For special dosage forms</td>
<td>Chewing apparatus/Flow through apparatus.</td>
</tr>
</tbody>
</table>

### 2.5.4 Limitations of dissolution testing

**Invitro dissolution testing can be non–discriminate:** Example; Mebandazole polymers A, B, and C dissolution profiles met the specification of 75% dissolved in 120 minutes although they exhibited different therapeutic effects. (Zhang et. al, 2004)

Invitro dissolution testing can be over –discriminate: Example; FDA sponsored studies with manufactured fast, medium and slow dissolving tablets of metoprolol and propranolol. Slow dissolving tablets of metoprolol failed in USP dissolution test. However the in vivo pharmacokinetic a study demonstrates the bioequivalence of fast, medium and slow dissolving tablets with their corresponding formulations. The IVIVC suggests that in vivo dissolutions are not rate limiting step for this formulation so that difference in dissolution rate does not make any difference.

Formulation specific IVIVC: IVIVC is only valid for one particular type of dosage form containing certain rate controlling excipients with the same release mechanism. If a drug is formulated in the same type of a solid dosage form, such as tablets, formulations with different drug release mechanisms would require the development of separate IVIVC with different in vitro dissolution methodology.
2.5.5 Setting of Dissolution Specifications:
A specification is defined as a list of tests, references to analytical procedures, and appropriate acceptance criteria which are numerical limits, ranges, or other criteria for the tests described. When a specification is first proposed, justification should be presented for each procedure and each acceptance criterion included. (Elkoshi, 1999 ; ICH Topic Q 6)

The justification should refer to relevant development data, pharmacopoeial standards, test data for drug substances and drug products used in toxicology and clinical studies, and results from accelerated and long term stability studies, as appropriate. Additionally, a reasonable range of expected analytical and manufacturing variability should be considered. It is important to consider all of this information.

2.5.5.1 Dissolution profile Comparison:
Moore and Planner proposed a model independent mathematical approach to compare the dissolution profiles using two factors, $f_1$ (difference factors) and $f_2$ (similarity factors). The formula used to calculate $f_1$ and $f_2$ are

$$f_1 = \frac{\sum_{t=1}^{n} |R_t - T_t|}{\sum_{t=1}^{n} R_t} \times 100$$

$$f_2 = 50 \times \log \left\{ \left[ 1 + \frac{1}{n} \sum_{t=1}^{n} (R_t - T_t)^2 \right]^{-0.5} \times 100 \right\}$$

Where $\log = \text{logarithm base 10}$, $n =$ number of sampling time points, $\Sigma =$ summation over all time points, $R_t =$ dissolution time point $t$ of the reference (pre-change batch), $T_t =$ dissolution at time point $t$ of the test (post change)

The $f_1$ value should be between 0-15 to indicate difference between two dissolution profiles. The $f_2$ value should be between 50-100 to indicate similarity between two dissolution profiles. When the two profiles are identical, $f_2 = 100$. If an average of 10% difference at all measured time points results then $f_2 = 50$. When both test and reference products dissolve 85% or more of the label amount of the drug in $\leq 15$ minutes using all three dissolution media recommended above, the profile comparison with an $f_2$ test is unnecessary.
2.5.5.2 Conditions for a Dissolution profile comparison

At least 12 units should be used for each profile determination. Mean dissolution values can be used to estimate the similarity factor, f2. To use mean data, the % coefficient of variation at the earlier point should not be more than 20% and at other time points should not be more than 10%.

For circumstances where wide variability is observed, or a statistical evaluation of f2 metric is desired, a bootstrap approach to calculate a confidence interval can be performed.

The dissolution measurements of the two products (test and reference, pre- and post-change, two strengths) should be made under the same test conditions. The dissolution time points for both the profiles should be the same, e.g., for immediate release products 15, 30, 45 and 60 minutes, for extended release products 1, 2, 3, 5 and 8 hours.

Because f2 values are sensitive to the number of dissolution time points, only one measurement should be considered after 85% dissolution of the product. For products which are rapidly dissolving, i.e., more than 85% in 15 minutes or less, a profile comparison is not necessary.

A f2 value of 50 or greater (50-100) ensures sameness or equivalence of the two curves and, thus, the performance of the two products.

Delayed release:
A modified release product in which the release of active substance is delayed for a finite "lag time", after which release is unhindered [e.g. enteric coated or "Gastro resistant" (Pharm.Eur., 2007) oral tablets or capsules which remain intact in the stomach and only disintegrate in the higher pH of the small intestine]. Delayed release results in a longer Tmax but with Tmax and elimination half life unchanged.

In delayed release component, the drug may not be sufficiently protected for residence time greater than 2 hours in the gastric pH of 1.2. Low pH may also alter the performance by causing chemical reactions of the materials used in the dosage for modifying the release of drug. Therefore, while the final dissolution test may only require a 1-2 hour
presoak at gastric pH, the dosage form should be thoroughly evaluated at gastric pH if there is potential for long gastric residence times. If the goal of the dosage form is to release the drug in the duodenum, e.g., target transport through tight junctions, then the dissolution test should reflect the possibility of a short residence time. This is especially true if the mechanism for targeting the release is enteric coating. Further hampering of drug release can occur if the enteric coating erodes at pH 6.5, since the pH at the proximal duodenum is closer to 5.5 than 6.5. Therefore, an appropriate dissolution test for pH sensitive release mechanism such as enteric-coated dosage forms may require several pH simultaneously taking into consideration the potential in vivo residence time at each pH.

Extended Release:
The FIP -Guideline and European Pharmacopeia demand at least 3 specification points, the first after 1-2 hours (around 20-30% drug release) to provide assurance against premature drug release. The second specification point has to be around 50 % drug release to define the dissolution pattern. At the last point, the dissolution limit should be at least 80 % drug release to ensure almost quantitative release. Alternatively, a dissolution of <80% has to be justified and should be supported by a test duration of at least 24 hours. There are slight differences with regard to the United States Pharmacopeia, where only > 2 test points are demanded considering the individual monograph. (Moore 1996).

2.6 In-vitro-In-vivo Correlation
Invitro-invivo correlation is the demonstration of the direct relationship of in vitro dissolution rate of drugs and their in vivo bioavailability. Generally, the in vitro property is the rate or extent of drug dissolution or release while the in vivo response is the plasma drug concentration or amount of drug absorbed. Correlation is used to ensure batch-to-batch consistency in the physiologic performance of a drug product by use of such in vitro values and to serve as a tool in the development of a new dosage form with desired in vivo performance.

There are two basic approaches by which a correlation between dissolution testing and bioavailability can be developed.
1. By establishing a relationship, between the in vitro dissolution and the in vivo bioavailability parameters. If this relationship becomes linear with a slope of 1, then
curves are super imposable, and there is a 1:1 relationship which is defined as point-to-point or level A correlation.

2. By using the data from previous bioavailability studies to modify the dissolution methodology in order to arrive at meaningful in vitro-invivo correlation.

2.6.1 Levels of IVIVC:
Three correlation levels have been defined in IVIVC FDA guidance.

Level A: Represents a point to point relationship between in vitro dissolution rate and in vivo input rate of the drug from the dosage forms. Usually estimated by a two stage procedure (Example: deconvolution followed by comparison of the fraction absorbed to the fraction dissolved). Generally linear, but non-linear are also acceptable.

Level B: Correlation based on statistical moment analysis. Example: in vitro MDT vs. in vivo MRT or MAT

Level C: In this level of correlation, one dissolution time point (t50%, t90%, etc.) is compared to one mean pharmacokinetic parameter such as AUC, t_{max} or C_{max}. Therefore, it represents a single point correlation. Example: in vitro T50% vs. in vivo Tmax

Table 2.7: Establishment of In Vitro and In Vivo data

<table>
<thead>
<tr>
<th>Level</th>
<th>In Vitro</th>
<th>In Vivo</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>Dissolution curve</td>
<td>Input (absorption) curves</td>
</tr>
<tr>
<td>B</td>
<td>Statistical moments (MDT)</td>
<td>Statistical moments (MRT, MAT, etc.)</td>
</tr>
<tr>
<td>C</td>
<td>Disintegration time, time to have 10, 50, 90% dissolved in dissolution rate, dissolution efficiency</td>
<td>C_{max} – T_{max} – K_{a} time to have 10, 50, 90% absorbed AUC (total or cumulative)</td>
</tr>
</tbody>
</table>
2.6.2 Biowaivers:
Recent research has lead to the use of in-vitro tests to waive additional in vivo bioequivalency studies for some pharmaceutical products. The use of in vitro testing to achieve a waiver of in vivo studies is commonly referred to as a biowaiver. The FDA guidance outlines five categories of biowaivers: 1) biowaivers without an IVIVC, 2) biowaivers using an IVIVC: non-narrow therapeutic index drugs, 3) biowaivers using an IVIVC: narrow therapeutic index drugs, 4) biowaivers when in vitro dissolution is independent of dissolution test conditions and 5) situations for which an IVIVC is not recommended for biowaivers.

2.6.3 Biowaiver of Generic drug:

2.6.3.1 (i) Waiver of in vivo BE studies based on BCS: Recommended for a solid oral test product that exhibit rapid (85% in 30 mints) and similar in vitro dissolution under specified conditions to an approved reference product when the following conditions are satisfied:

- Products are pharmaceutical equivalent
- Drug substance is highly soluble and highly permeable and is not considered have a narrow therapeutic range
- Excipients used are not likely to affect drug absorption;

2.6.3.2 (ii). Waiver of invivo BE for IR oral dosage form: bioequivalence studies may be waived for compositionally similar strengths when one strength in a range has been studied, under these conditions the following conditions are satisfied

- Product are manufactured by the same manufacturer and process
- Linear pharmacokinetics
- The qualitative composition of the different strengths is the same; except in the case of
  - Flavours/colours
  - The ratio between amounts of drug and excipients is the same or in case of preparations containing a low concentration of the drug (less than 5%), the ratio between the amounts of excipients is similar.
the dissolution profile should be similar for additional strengths and the
strength of the
batch used in BE study

2.6.3.3 Waivers for Scale-up and Post approval changes: Biowaivers may be granted
for manufacturing site changes, equipment changes, manufacturing process
changes, and formulation composition changes according to a predictive and
reliable IVIVC.

2.7 Formulations of Extended release formulations available in India

Table 2.8: List of extended release tablets of Metformin available in India (Medclick,
2007)

<table>
<thead>
<tr>
<th>Brand Name</th>
<th>Manufacturer</th>
<th>Formulation</th>
<th>Strength</th>
<th>Strip of</th>
<th>Cost</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bigomet</td>
<td>Otsira Genetica (A Div. of Aristo Pharma Ltd.)</td>
<td>Tablet</td>
<td>500 mg</td>
<td>10</td>
<td>Rs 9.50</td>
</tr>
<tr>
<td>Dibimet</td>
<td>Novartis</td>
<td>Tablet</td>
<td>500 mg</td>
<td>10</td>
<td>Rs 9.50</td>
</tr>
<tr>
<td>Baymet</td>
<td>Bayer (India) Limited</td>
<td>Tablet</td>
<td>500 mg</td>
<td>10</td>
<td>Rs 8.80</td>
</tr>
<tr>
<td>Bigesens</td>
<td>Zydus Cadila</td>
<td>Tablet</td>
<td>500 mg</td>
<td>10</td>
<td>Rs 9.50</td>
</tr>
<tr>
<td>Bigesens-XR</td>
<td>Cadilla Pharmaceuticals Ltd.</td>
<td>Tablet</td>
<td>500 mg</td>
<td>10</td>
<td>Rs 9.50</td>
</tr>
<tr>
<td>Daomet SR</td>
<td>J.B Chemicals &amp; Pharmaceuticals Limited</td>
<td>Tablet</td>
<td>500 mg</td>
<td>10</td>
<td>Rs 15.00</td>
</tr>
<tr>
<td>Diomet-SR</td>
<td>Bal Pharma</td>
<td>Tablet</td>
<td>500 mg</td>
<td>10</td>
<td>Rs 18.00</td>
</tr>
<tr>
<td>Dibeta-SR</td>
<td>Torrent Pharmaceuticals Ltd.</td>
<td>Tablet</td>
<td>500 mg</td>
<td>10</td>
<td>Rs 15.00</td>
</tr>
<tr>
<td>Exermet</td>
<td>Cipla Limited</td>
<td>Tablet</td>
<td>500 mg</td>
<td>10</td>
<td>Rs 14.00</td>
</tr>
<tr>
<td>Forminal-SR</td>
<td>Alembic Ltd.</td>
<td>Tablet</td>
<td>500 mg</td>
<td>10</td>
<td>Rs 7.50</td>
</tr>
<tr>
<td>Product</td>
<td>Manufacturer</td>
<td>Formulation</td>
<td>Quantity</td>
<td>Price</td>
<td></td>
</tr>
<tr>
<td>------------------</td>
<td>-------------------------------</td>
<td>-------------</td>
<td>----------</td>
<td>--------</td>
<td></td>
</tr>
<tr>
<td>Getmet-SR</td>
<td>RAVENBHEL</td>
<td>Tablet</td>
<td>500 mg</td>
<td>10</td>
<td>Rs 15.00</td>
</tr>
<tr>
<td>Gluconorm-SR</td>
<td>Lupin Laboratories Ltd.</td>
<td>Tablet</td>
<td>500 mg</td>
<td>10</td>
<td>Rs 15.03</td>
</tr>
<tr>
<td>Gluformin XR</td>
<td>Nicholas Piramal India Ltd. (NPIL)</td>
<td>Tablet</td>
<td>500 mg</td>
<td>10</td>
<td>Rs 16.00</td>
</tr>
<tr>
<td>Glumet XR</td>
<td>Cipla Limited</td>
<td>Tablet</td>
<td>500 mg</td>
<td>10</td>
<td>Rs 7.50</td>
</tr>
<tr>
<td>Glumet-XR</td>
<td>Profic Organic Ltd.</td>
<td>Tablet</td>
<td>500 mg</td>
<td>10</td>
<td>Rs 7.75</td>
</tr>
<tr>
<td>Glyciphage</td>
<td>Franco Indian Pharmaceuticals Ltd.</td>
<td>Tablet</td>
<td>500 mg</td>
<td>10</td>
<td>Rs 15.87</td>
</tr>
<tr>
<td>Insuimet</td>
<td>Cadila Pharmaceuticals Ltd.</td>
<td>Tablet</td>
<td>500 mg</td>
<td>10</td>
<td>Rs 15.03</td>
</tr>
<tr>
<td>Mf-SR</td>
<td>Psychotropics India Limited (PIL)</td>
<td>Tablet</td>
<td>500 mg</td>
<td>10</td>
<td>Rs 10.00</td>
</tr>
<tr>
<td>Roftem-SR</td>
<td>Khandelwal Laboratories Ltd.</td>
<td>Tablet</td>
<td>500 mg</td>
<td>10</td>
<td>Rs 14.00</td>
</tr>
<tr>
<td>Walaphage-SR</td>
<td>Wallace Pharmaceuticals Ltd.</td>
<td>Tablet</td>
<td>500 mg</td>
<td>10</td>
<td>Rs 12.00</td>
</tr>
<tr>
<td>Cetapin XR</td>
<td>Aventis Pharma Limited.</td>
<td>Tablet</td>
<td>500 mg</td>
<td>10</td>
<td>Rs 24.44</td>
</tr>
</tbody>
</table>