Bioavailability and Bioequivalence Studies

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Introduction

- ✓Essential to ensure uniformity in standards of quality, efficacy & safety of Pharmaceutical products
- ✓ Reasonable assurance is to be provided that various products containing same active ingredient, marketed by different licensees are clinically equivalent & interchangeable
- ✓Release of an active substance should be known & reproducible
- ✓Both Bioavailability & Bioequivalence focus on release of drug substance from its dosage form & subsequent absorption in circulation
- ✓Similar approaches to measure Bioavailability should be followed in demonstrating Bioequivalence

Bioavailability

Measurement of the relative amount & rate at which,
the drug from administered dosage form,
reaches the systemic circulation &
becomes available at the site of action

Bioavailable fraction (F), refers to the fraction of administered dose that enters the systemic circulation

$$F = \frac{\text{Bioavailable dose}}{\text{Administered dose}}$$

Therapeutic Relevance

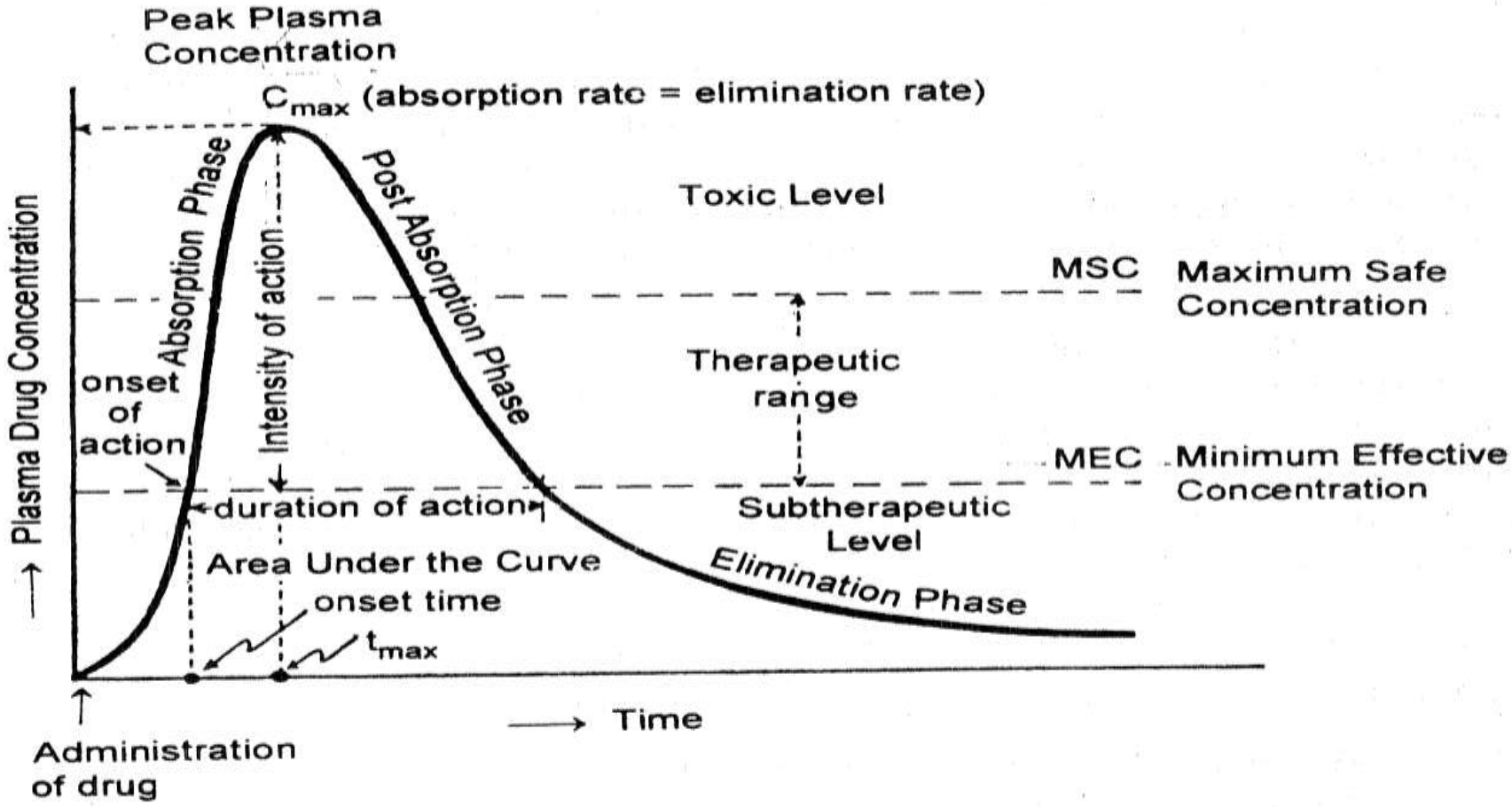


Fig. 9.1 A typical plasma concentration-time profile showing pharmacokinetic and pharmacodynamic parameters. obtained after oral administration of single dose of a drug.

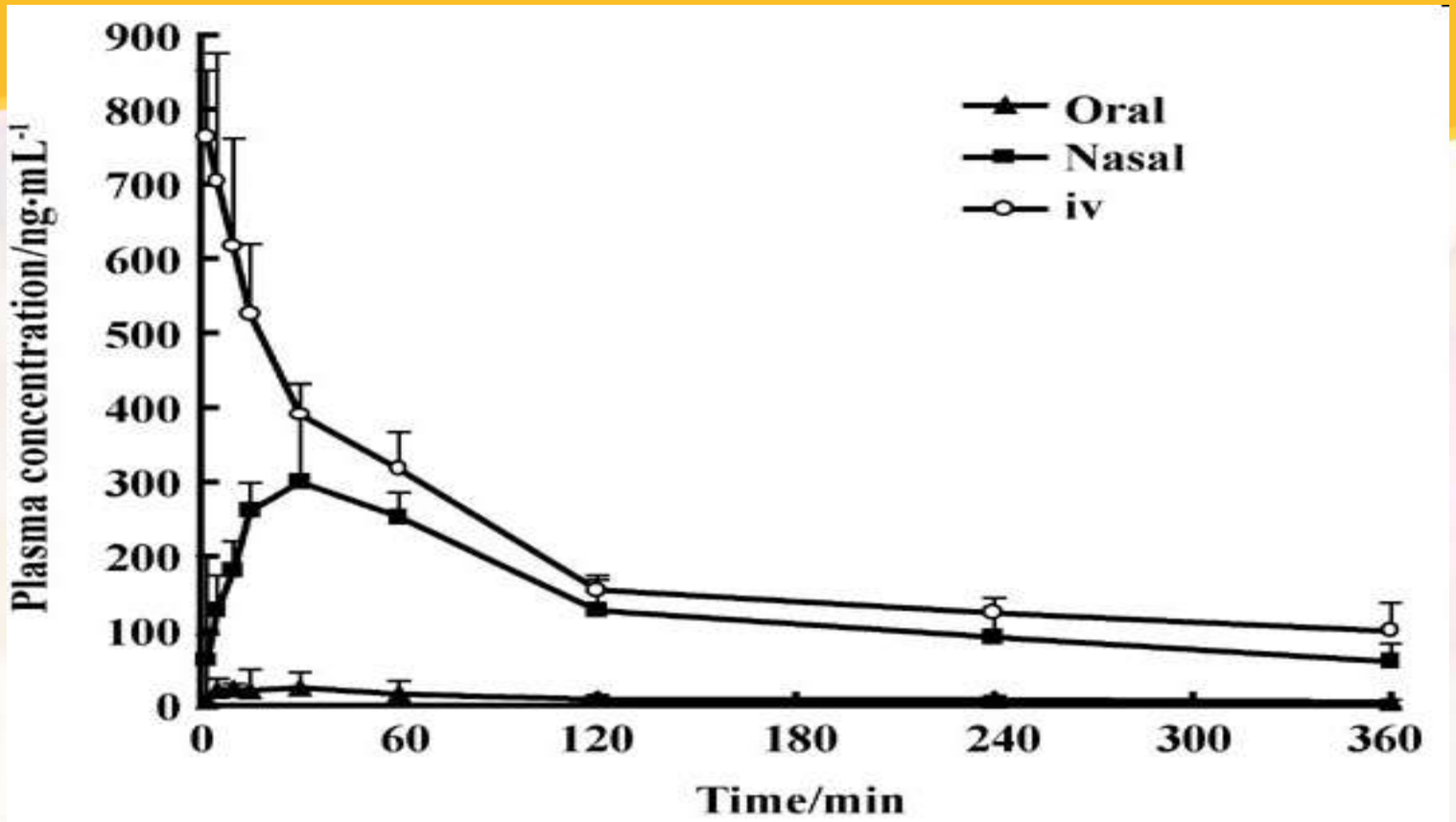
➢ Absolute Bioavailability

Compares the bioavailability of the active drug in systemic circulation following non-intravenous administration with the same drug following intravenous administration

✓ For drugs administered intravenously, bioavailability is 100%

✓ Determination of the best administration route

$$F_{ab} = \frac{(AUC)_{drug}}{(AUC)_{IV}}$$



Absolute Bioavailability of Nimodipine for different routes:

Oral : 1.17 %

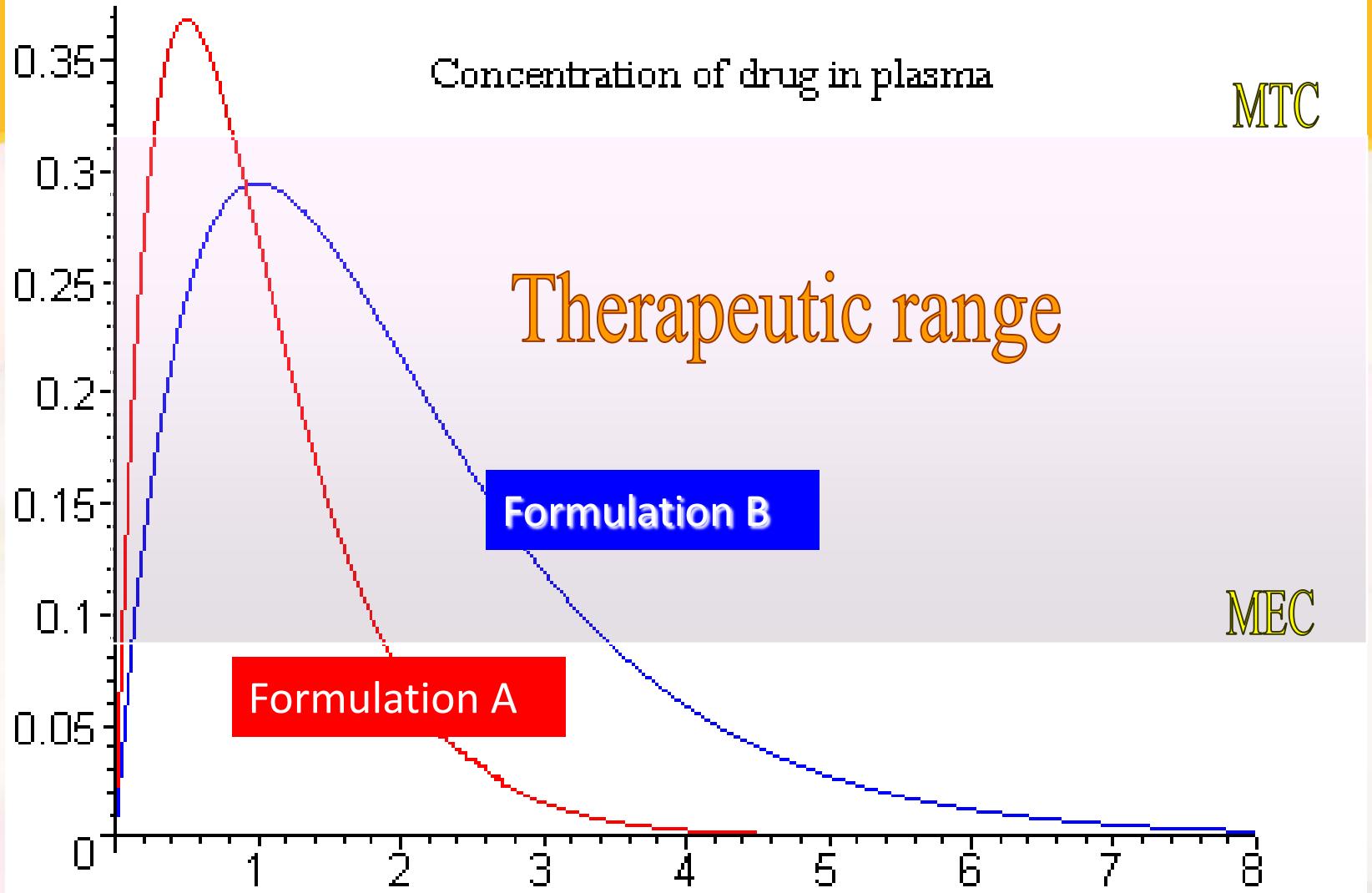
Nasal : 67.4 %

Intravenous: 100%

➢ Relative Bioavailability

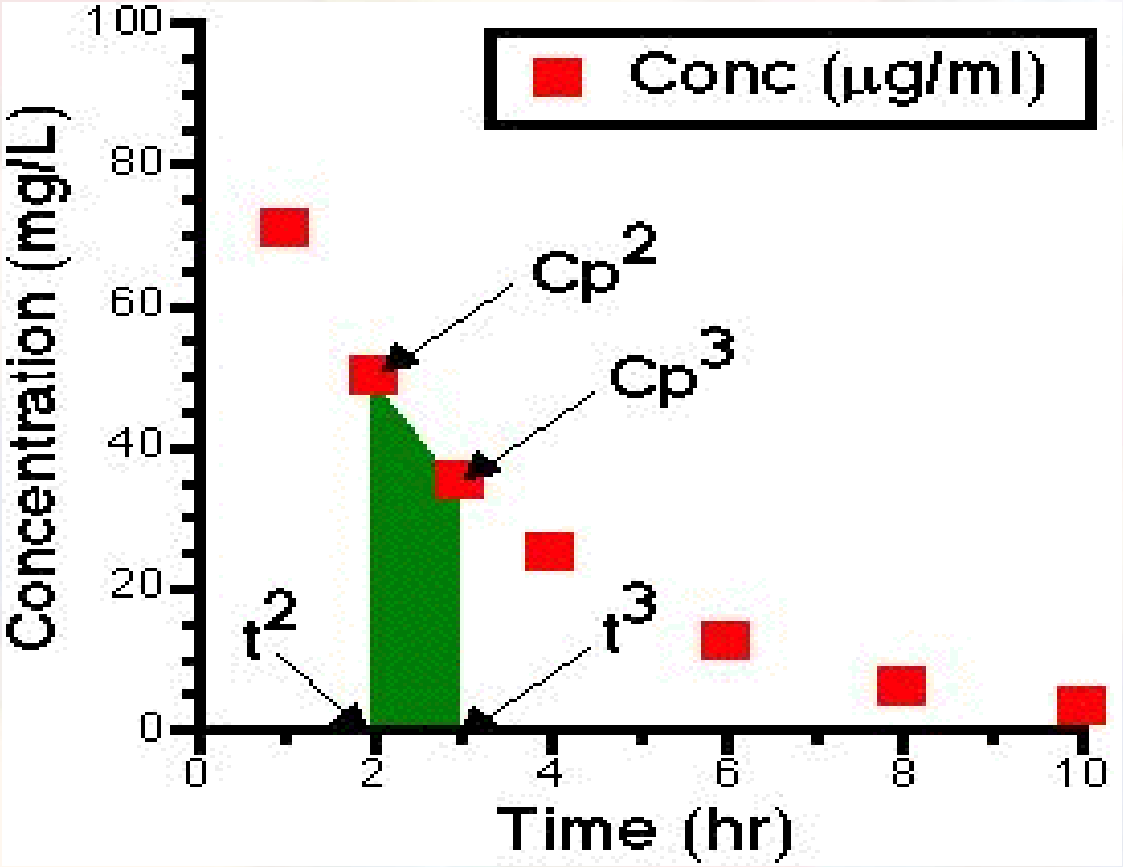
Compares the bioavailability of a *formulation (A)* of a certain drug when compared with another *formulation (B)* of the same drug, usually an established standard

$$F_{rel} = \frac{(AUC)_{drug}}{(AUC)_{standard}}$$



Plasma concentration versus time curve

AUC: Trapezoidal Rule



$$AUC_{2\text{-}3} = \frac{Cp^2 + Cp^3}{2} \times (t_3 - t_2)$$

Factors affecting Bioavailability of a Drug

➢Physical properties of a drug

✓**Physical state**:

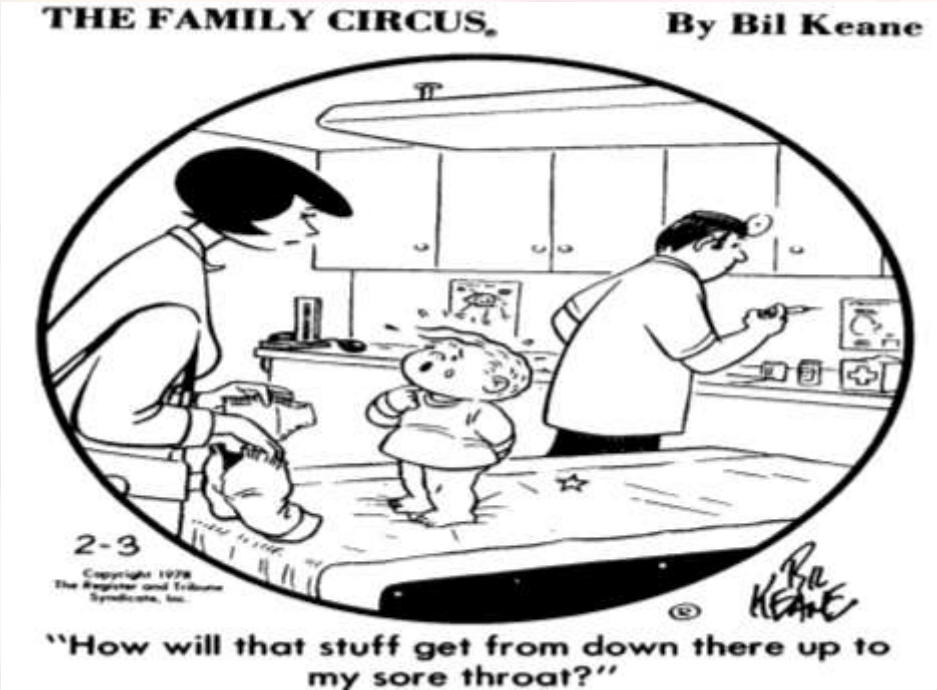
• Liquids > Solids
[Solution > Suspension > Capsule > Tablet > Coated tablet]

• Crystalloids > Colloids

✓**Lipid or water solubility**:

• Aqueous phase at absorption site

• Passage across Cell surface



➢Dosage forms

✓ **Particle size**:

- Important for sparingly soluble drugs
- ↓ the size, ↑ the absorption, ↓ the dose
- Nano-crystalline formulations of Saquinavir
- If ↓ absorption needed (local action on GIT), ↑ the size

➢Physiological factors

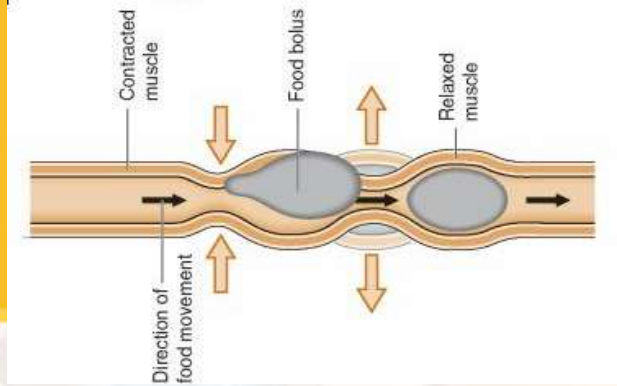
✓**Ionization**:

- Unionized form penetrates the GI mucosal lining quickly

✓**pH of the fluid**:

- Weakly acidic drugs: Aspirin, Barbiturates→ Stomach, duodenum
- Weakly basic drugs: Pethidine, Ephedrine→ Small intestine
- Strongly acidic / basic drugs: highly ionized & poorly absorbed

➢GI transit time



✓**Prolonged gastric emptying**:

• Delays absorption due to stasis
(e.g. with anticholinergics / Diabetic neuropathy)

✓**Increased peristaltic activity**:
(e.g. Metoclopramide→ speeds up the absorption of analgesics)

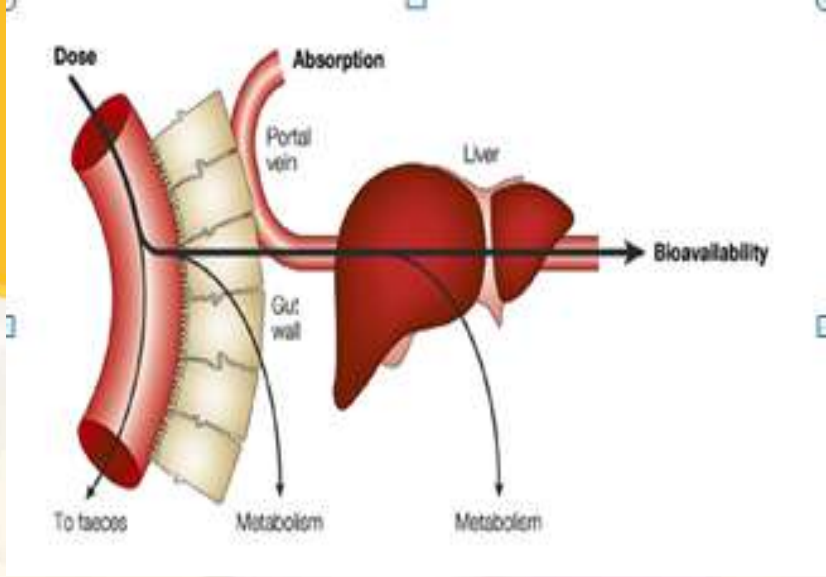
✓**Excessive peristaltic activity** (as in Diarrhoea) impairs absorption

✓**Fed state**:

• impairs progress of drug to intestine→ ↓ absorption (Indinavir)
• ↑ splanchnic blood flow→ ↑ absorption (Propranolol)

✓**First pass metabolism**:

- Gut wall (e.g. Isoprenaline)
- Liver (e.g. Opoids, ß-blockers, Nitrates)



✓**Presence of other agents**:

- Vitamin C ↑ Iron absorption, Phytates retard it
- Calcium ↓ absorption of Tetracyclines

✓**Disease states**:

- Malabsorption, Achlorhydria, Cirrhosis, Biliary obstruction can hamper absorption

✓**Entero-hepatic cycling**:

- Increases bioavailability (e.g. Morphine, OC pills)

Concept of Equivalents

➢Pharmaceutical equivalents

✓ equal amounts of the identical active drug ingredient,
(i.e. the same salt or ester of the therapeutic moiety)

✓ identical dosage forms

✓ not necessarily containing the same inactive ingredients

➢Pharmaceutical alternatives

✓ identical therapeutic moiety, or its precursor

✓ not necessarily the same:

- salt or ester of the therapeutic moiety
- amount
- dosage form

➢Bioequivalence

✓Pharmaceutical equivalent / alternative of the test product,

✓when administered at the same molar dose,

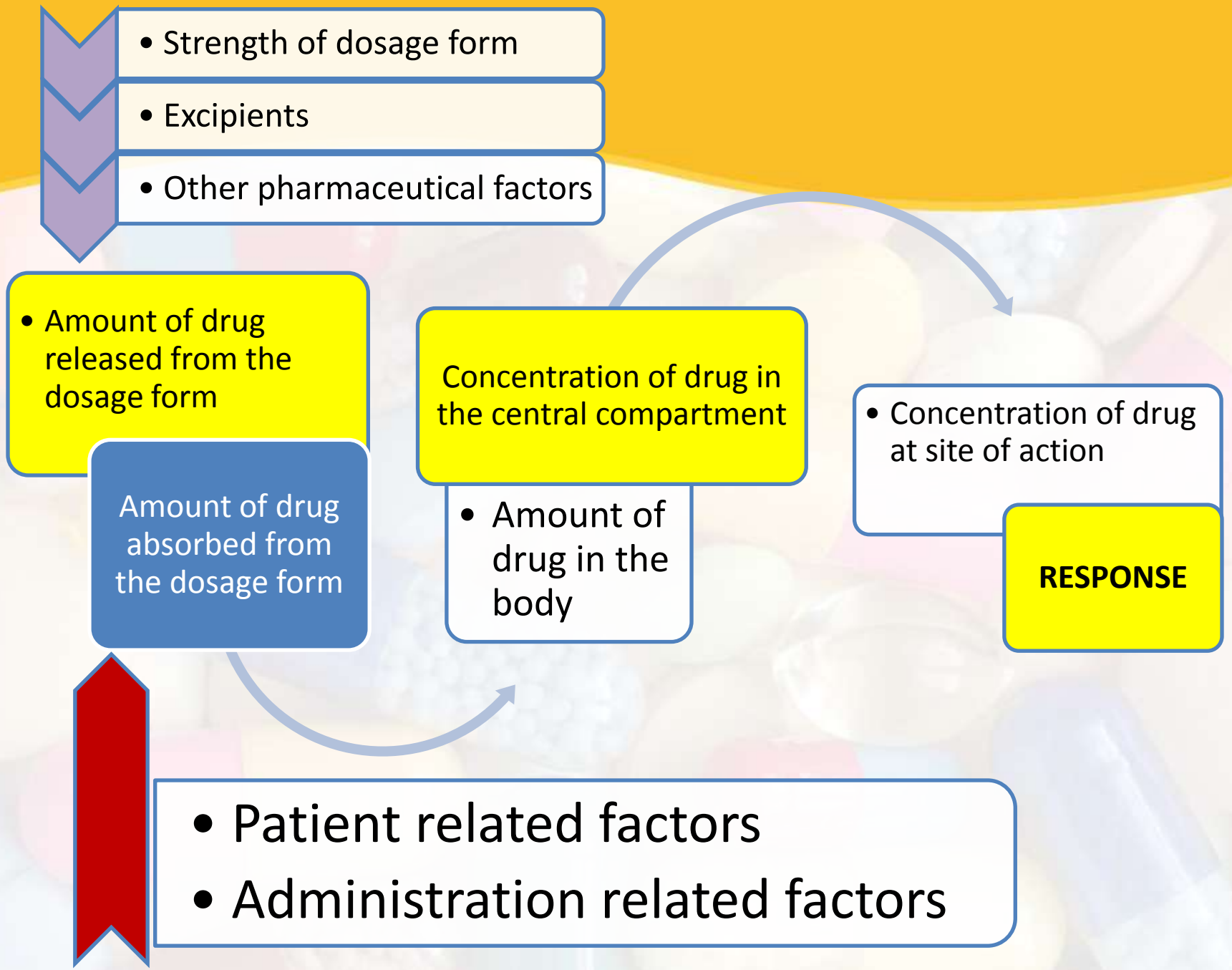
✓has the rate and extent of absorption

✓ not statistically significantly different from that of the reference product

➢Therapeutic equivalence

✓Same active substance or therapeutic moiety

✓Clinically show the same efficacy & safety profile


• Strength of dosage form
• Excipients
• Other pharmaceutical factors
• Amount of drug released from the dosage form
Amount of drug absorbed from the dosage form
Concentration of drug in the central compartment
• Amount of drug in the body
• Concentration of drug at site of action
RESPONSE
• Patient related factors
• Administration related factors

- Strength of dosage form
- Excipients
- Other pharmaceutical factors

In vitro Quality Control testing

Amount of drug absorbed from the dosage form

In vivo Bioequivalence studies

- Amount of drug in the body

- Concentration of drug at site of action

PD studies/ Clinical Trials

- Patient related factors
- Administration related factors

Reference Product

- ✓ Identified by the Regulatory Authorities as "Designated Reference Product"
- ✓ Usually the Global Innovator's Product
- ✓Protected by a patent
- ✓Marketed under manufacturers brand name
- ✓ Clinical efficacy & safety profile is well documented in extensive trials
- ✓ All generics must be Bioequivalent to it
- ✓ In India, CDSCO may approve another product as Reference product

Generic Drug

✓Drug product which is identical or bioequivalent to Brand/ Reference drug in:

- Active ingredient (s)
- Route of administration
- Dosage form
- Strength
- Indications
- Safety

✓<u>May have different</u>:

- Inactive ingredients
- Colour
- Shape

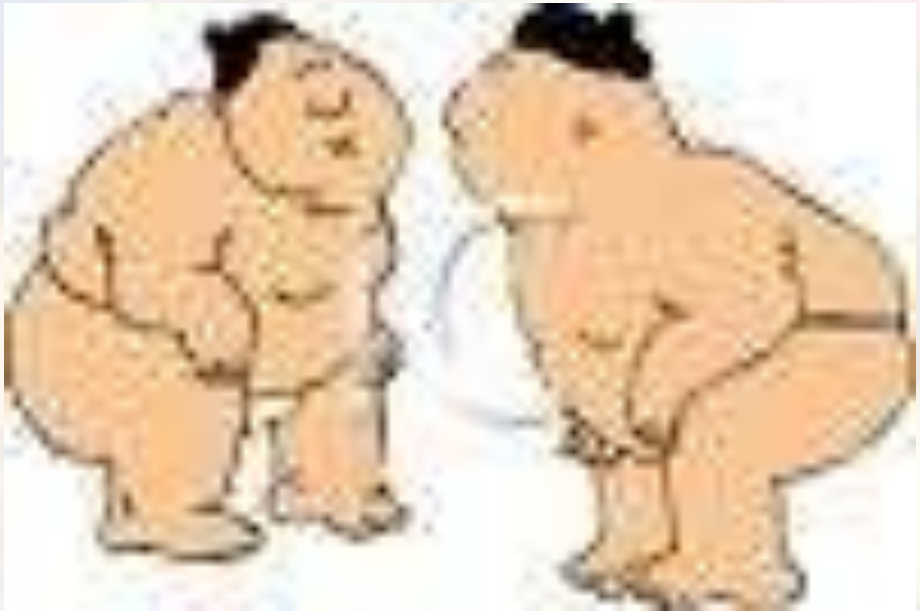
✓ Almost half of drugs in market have Generics

Price difference between Reference & Generic Drugs

Reference Drug	Generic Drug
• Expensive	• 30-80% cheaper
• 5/5000 new drug candidates tested in humans & 1 approved • Takes 12-15 yrs • Costs around 1 billion $	• Since already tested & approved, cost of simply manufacturing • Fraction of the cost of testing & development
• Drug Patents of 20yrs, applied before clinical trials begin • Effectively 7-12 yrs	• Approved for sale after drug patent protection expires

Fundamental Bioequivalence Assumption

When a generic drug is claimed **bioequivalent** to a Reference drug, it is assumed that they are **therapeutically equivalent**



Bioequivalence Background

✓ Using bioequivalence as the basis for approving generic copies in US "Drug Price Competition and Patent Term Restoration Act of 1984," also known as the Waxman-Hatch Act

✓ Created Generic Industry & ↑ their availability

✓ Most successful legislation

✓ Benefited Brand & Generic firms

- Generic firms→ Rely on findings of safety & efficacy of Innovator drug after Patent expiration

- Innovator firms→ Patent extensions of 5yrs to make up for time lost while their products were going through FDA's approval process

Indian Legislation

- ✓ In India, CDSCO provides "**Guidelines for Bioavailability & Bioequivalence Studies**" mentioned in Schedule Y

- ✓ As per the Drugs & Cosmetic Rules (IInd Amendment) 2005, all bioavailability and bioequivalence studies should be conducted in accordance to these Guidelines

News:

Ranbaxy faces possibility of a permanent injunction in US

CNBC; January 27, 2012

Requirement of BA & BE Studies

✓ For IND/NDAs:

To establish equivalence between:

- *Early & late* clinical trial formulations
- Formulations used in clinical trial & stability studies
- *Clinical trial formulations & to-be-marketed* drug product
- Any *other comparisons*, if appropriate

✓ ANDA for a generic drug product

✓Change in components, composition, &/or manufacturing process

✓Change in dosage form (capsules to tablet)

Objectives of BA & BE Studies

- ✓ Development of suitable dosage form for a New Drug Entity
- ✓ Determination of influence of excipients, patient related factors & possible interactions with other drugs
- ✓ Development of new drug formulations of existing drugs
- ✓ Control of quality of drug products, influence of → processing factors, storage & stability
- ✓ Comparison of availability of a drug substance from different form or same dosage form produced by different manufacturers

When is Bioequivalence not necessary (Biowaivers)

a) Parental Solution; same active substance with same concentration, same excipient

b) Oral Solution; same active substance with same concentration, excipient not affecting GI transit or absorption

c) Gas

d) Powder for reconstitution as solution; meets criterion (a) or (b)

e) Otic/Ophthalmic/Topical Solution; same active substance with same concentration, same excipient

f) Inhalational Product/ Nasal Spray; administered with or w/o same device as reference product ; prepared as aqueous solution ; same active substance with same concentration, same excipient

NDA vs ANDA Review Process

NDA Requirements

1. Chemistry
2. Manufacturing
3. Controls
4. Labeling
5. Testing
6. Animal Studies
7. Clinical Studies
8. Bioavailability

ANDA Requirements

1. Chemistry
2. Manufacturing
3. Controls
4. Labeling
5. Testing
6. Bioequivalence

Orange Book

- ✓ All FDA approved drugs listed (NDA's, ANDA's & OTC's)
- ✓ Expiration of patent dates
- ✓ Drug, Price and Competition Act (1984)
  FDA required to publish Approved Drug Products with Therapeutic Equivalence & Evaluations

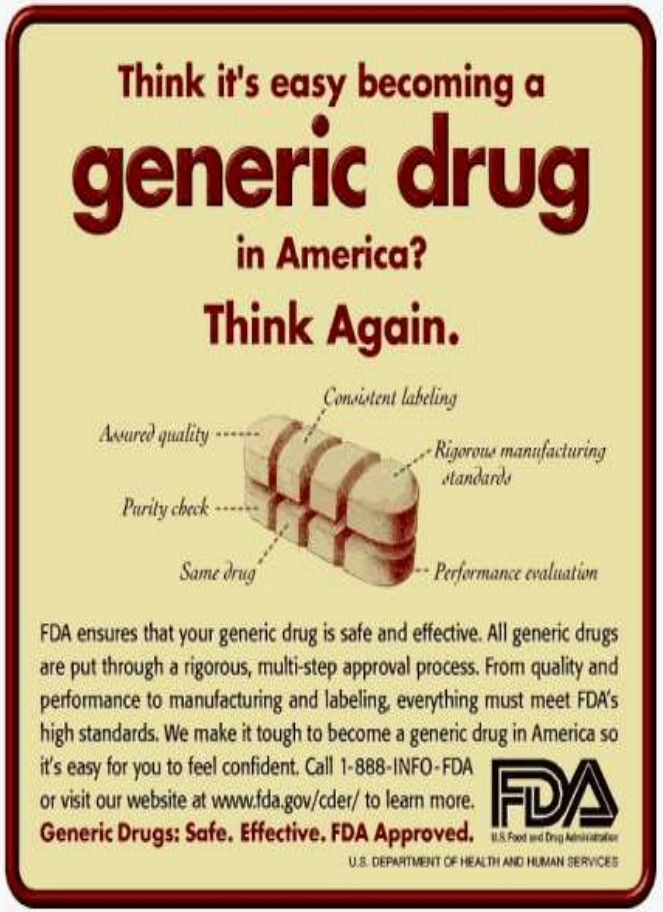
Methods used to assess Equivalence

I. Pharmacokinetic Studies

II. Pharmacodynamic Studies

III. Comparative Clinical Studies

IV. Dissolution Studies



Pharmacokinetic Studies

Study Design

✓Good experimental design, enhances the power of the study

✓ Depends on: question to be answered, nature of reference drug/ dosage form, benefit-risk ratio

✓ As far as possible, the study should be of crossover design & suitably randomized

✓Ideal design: Randomized two-period, two-sequence, Crossover design with adequate washout period

✓If the half-life is long: Parallel design

✓For hig

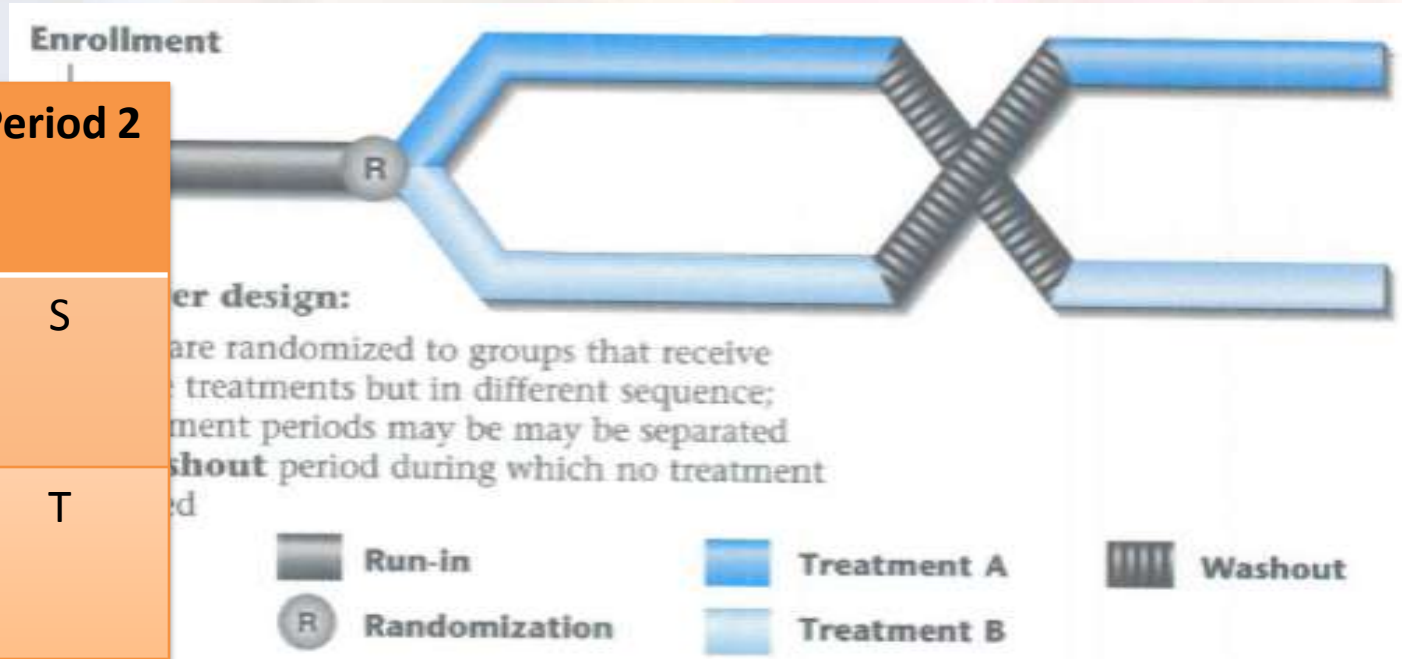
Any drug whose rate and extent of absorption shows large dose-to-dose variability within the same patient

I. Two-Period Crossover Design

✓2 formulations, even number of subjects, randomly divided into 2 equal groups

✓First period , each member of one group receive a single dose of the test formulation; each member of the other group receive the standard formulation

Subjects	Period 1	Period 2
1-8	T	S
9-16	S	T



II. Latin Square Design

- ✓ More than two formulations
- ✓ A group of volunteers will receive formulations in the sequence shown

Vol.No.	Period 1	Period 2	Period 3
1	A	B	C
2	B	C	A
3	C	A	B

III. Balance Incomplete Block Design (BIBD)

- ✓ More than 3 formulations, Latin square design will not be ethically advisable
- ✓ Because each volunteer may require drawing of too many blood samples
- ✓ If each volunteer expected to receive at least two formulation, then such a study can be carried out using BIBD

Vol. No.	Period 1	Period 2
1	A	B
2	A	C
3	A	D
4	B	C
5	B	D
6	C	D
7	B	A
8	C	A
9	D	A
10	C	B
11	D	B
12	D	C

IV. Parallel-Group Design

- ✓Even number of subjects in two groups
- ✓Each receive a different formulation
- ✓No washout necessary
- ✓For drugs with long half life

Treatment A	Treatment B
1	2
3	4
5	6
7	8
9	10
11	12

Parallel	Crossover
• Groups assigned different treatments	• Each patient receives both treatments
• Shorter duration	• Longer duration
• Larger sample size	• Smaller sample size
• No carryover effect	• Carryover effect
• Doesn't require stable disease & similar baseline	• Requires stable disease & similar baseline

V. Replicate Crossover-study design

✓For **highly variable drugs**

✓ Allows comparisons of within-subject variances

✓ Reduce the number of subjects needed

➢Four-period, two-sequence, two-formulation design (recommended)

OR

➢Three-sequence, three-period, single-dose, partially replicated

Period	1	2	3	4
Group 1	T	R	T	R
Group 2	R	T	R	T

VI. Pilot Study

✓ If the sponsor chooses, in a small number of subjects

✓ To assess variability, optimize sample collection time intervals & provide other information

✓ Example:

- Immediate-release products: careful timing of initial samples→ avoid a subsequent finding that the first sample collection, occured after the plasma concentration peak

- Modified-release products: determine the sampling schedule → assess *lag time* & *dose dumping*

✓ Can be appropriate, provided its design & execution are suitable & sufficient number of subjects have completed the study

Subject selection

- ✓Healthy adult volunteers
- ✓Age: 18-45 yrs
- ✓Age/Sex representation corresponding to therapeutic & safety profile
- ✓Weight within normal limits→ BMI
- ✓Women: Pregnancy test prior to 1st & last dose of study; OC pills C/I
- ✓Drug use intended in Elders (Age >60yrs)
- ✓Teratogenic Drugs→ Male volunteers
- ✓Highly toxic drugs: Patients with concerned disease (stable) eg. Cancer

Exclusion Criteria

- ✓H/o allergy to test drug
- ✓H/o liver or kidney dysfunction
- ✓H/o jaundice in past 6 months
- ✓Chronic diseases eg. Asthma, arthritis
- ✓Psychiatric illness
- ✓Chronic smoker, alcohol addiction, drug abuse
- ✓Intake of enzyme modifying drug in past 3 months
- ✓Intake of OTC/Prescription drugs past 2 weeks
- ✓HIV positive
- ✓BA & BE studies in past 3 months
- ✓H/o bleeding disorder

Selection of Number of Subjects

✓ Sample size is estimated by:

- Pilot experiment
- Previous studies
- Published data

✓ Significance level desired, usually 0.05

✓ Power of the study, normally 80% or more

✓ Expected deviation (Δ) from the reference product, as compatible with BE

✓ If no data available, reference ratio of 0.95 (Δ = 5%) used

- ✓ Minimum 16 subjects, unless ethical justification
- ✓ Allow for drop-outs
- ✓ Replace drop-outs→ substitute follow same protocol; similar environment
- ✓ Sequential/ Add-on Studies→ large no. of subjects required, results of study do not convey adequate significance

Genetic Phenotyping

✓Drug is know to be subject to genetic polymorphism

✓Cross-over design→ Safety & Pharmacokinetic reasons

✓All Parallel group design

✓Indian population:

- Captures genetic diversity of the world
- Forms continuum of genetic spectrum
- >1000 medically relevant genes

✓ Diverse patient/ volunteer pool for conducting BA & BE studies

Characteristics to be measured

✓ Accessible biological fluids like blood, plasma &/or serum to indicate release of the drug substance from the drug product into the systemic circulation

✓Mostly: Active drug substance

✓Active / Inactive metabolite maybe measured in cases of:

- Concentration of drug too low
- Limitation of analytical method
- Unstable drug
- Drug with very short half life
- Pro-drugs

✓ Excretion of drug & its metabolites in urine→ Non-linear kinetics

✓ Measure individual enantiomers when they exhibit:

- Different pharmacokinetic/ pharmacodynamic properties
- Non-linear absorption
- Safety/Efficacy purposes

✓ Drugs that are not absorbed systemically from site of application *surrogate marker* needed for BA & BE determination

Surrogate Markers

Drug product	Drug	Possible surrogate marker for bioequivalence
MDI	Albuterol	FEV1
Topical steroid	Hydrocortisone	Skin blanching
Anion exchange resin	Cholestryamine	Binding to bile acids
Antacids	Mg & Al hydroxide gel	Neutralization of acid
Topical antifungal	Ketoconazole	Drug uptake into stratum corneum

Blood Sampling points/ Schedule

Single-dose study of an immediate release product:

- ✓ For at least three elimination half-lives (cover >80% of AUC)
  - Absorption phase : 3-4 points
  - Around T_{max} : 3-4 points
  - During elimination : 4 points
- ✓ Intervals not longer than the half-life of the drug
- ✓ If urine tested, collect it for at least 7 half-lives

Method Validation

✓ **Accuracy/ Relative Recovery**

Closeness of determined value to the true Value

✓ **Precision**

Closeness of agreement obtained from the multiple sampling of the same homogeneous samples under certain prescribed conditions

• Repeatability

Precision under **same conditions**

same analyst, same apparatus, same interval of time, identical reagents

• Reproducibility

Precision under **different conditions**

different analysts, apparatus from different manufacturers, different days, reagents from different sources

✓ **Sensitivity**

Capacity of the test procedure to record small variations in concentration

• Limit of detection (LOD):

Lowest concentration of drug that will yield an assay response significantly different from that of a sample blank

• Limit of quantitation (LOQ/ sensitivity limit):

Lowest concentration of drug that can be determined with acceptable precision & accuracy under the stated experimental conditions

✓ **Selectivity/ Specificity**

Ability of the method to measure only what it is intended to measure

✓ Calibration of Instruments

- Should be done regularly & as per standard procedures in USP/BP/IP

- Done before starting the analysis at the development phase & during the study phase

✓ Predetermined SOPs

✓ Accreditation of analyzing laboratory

Parameters to be measured

✓Pharmacokinetic Parameters measured are:

- C_{max}
- T_{max}
- AUC_{0-t}
- $AUC_{0-\infty}$

$$AUC_{0-\infty} = AUC_{0-t} + C_{last}/k$$

For steady state studies:

- AUC_{0-t}
- C_{max}
- C_{min}
- Degree of fluctuation

Fasting & Fed State Conditions

➢ Fasting Conditions:

✓ Single dose study:

- Overnight fast (10 hrs) and subsequent fast of 4 hrs

✓ Multiple dose study:

- Two hours fasting before and after the dose

➢ Fed State Studies

✓Required when:

- Drug recommended with food
- Modified release product
- Assessment of C_{max} and T_{max} difficult with fasting state study

✓ Requires consumption of a high fat food, 15 minutes before dosing

✓ Provide 950-1000 kcals

✓ Fat- 50%, Proteins 15-20%, Carbohydrate- 30-35%

✓ Ethnic & cultural variation considered

✓ Specified in protocol

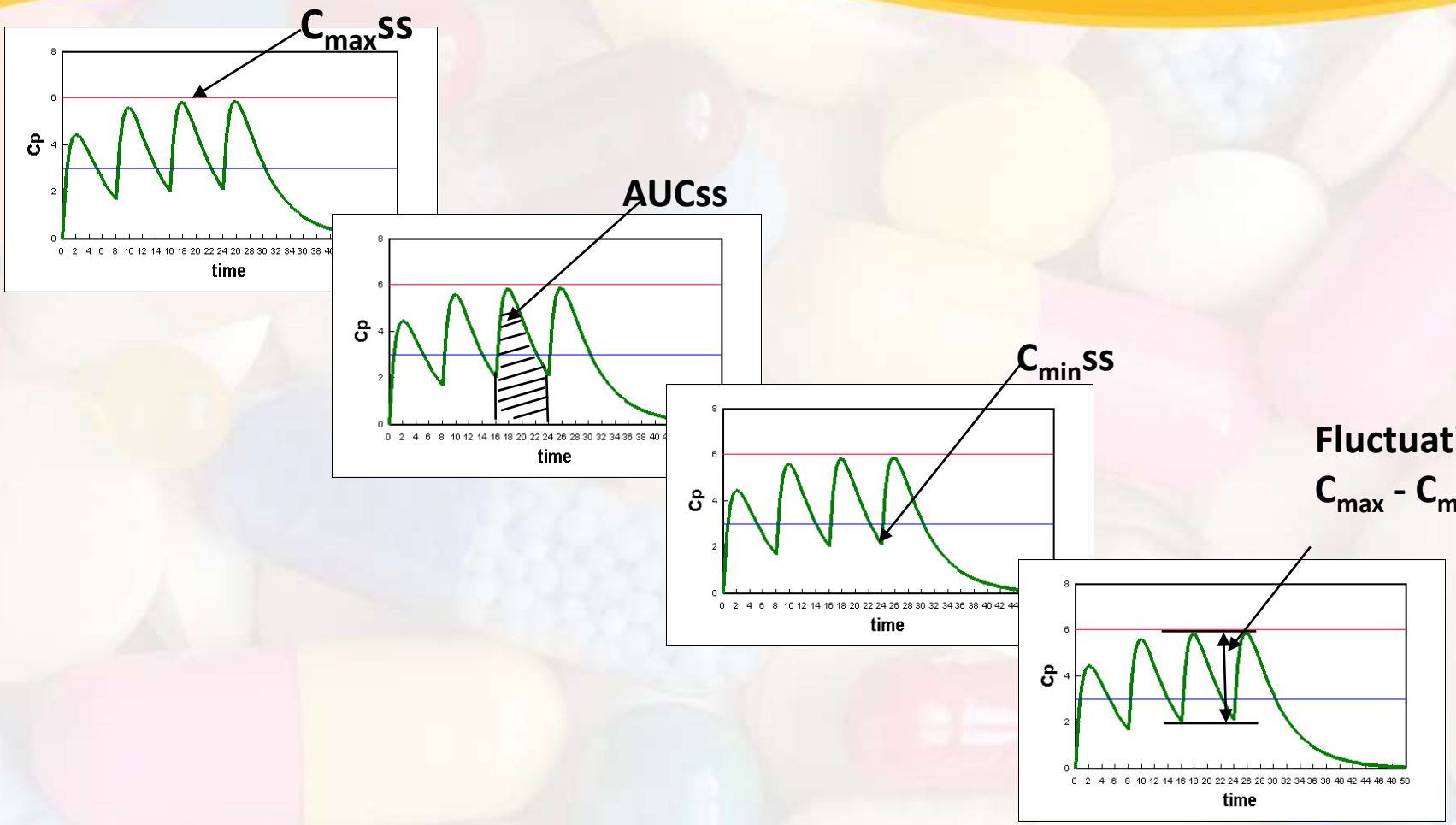
Steady State/ Multiple Dose Studies

- ✓ Long elimination half life→ Accumulation in the body
- ✓ Toxic drugs requiring multiple dose therapy
- ✓ Some Modified-release drugs
- ✓ Combination products
- ✓ Drugs inducing own metabolism
- ✓ Drugs showing non-linear pharmacokinetics

✓ Disadvantages:

- Difficult to conduct
- Costly
- Longer monitoring
- Longer exposure to drug

Parameters in Multiple dosing studies



Reporting for products likely to accumulate: Steady State studies

- **Acetaminophen** accumulation in pediatric patients after repeated therapeutic doses

- 10 patients studied at steady-state after repeated doses

Total AUC_{ss} for Acetaminophen was as:

0.181 $(ml/min/kg)^{-1}$ after the first dose

0.202 $(ml/min/kg)^{-1}$ at steady-state (**$p < 0.05$**)

- There was no evidence of hepatotoxicity

- These data suggest that acetaminophen may accumulate after repeated therapeutic doses in children with fever

Statistical Evaluation

✓ Primary concern of bioequivalence is to limit Consumer's & Manufacturer's risk

- C_{max} & AUC analysed using ANOVA
- T_{max} analysed by non-parametric methods

✓ Use *natural log transformation* of C_{max} and AUC

- Calculate Geometric means of C_{max} of Test [C_{max}'t]
- Calculate Geometric means of C_{max} of Reference [C_{max}'r]
- Calculate **Geometric Mean Ratio**= [C_{max}'t] / [C_{max}'r]

✓ Calculate 90% confidence interval for this GMR for C_{max}

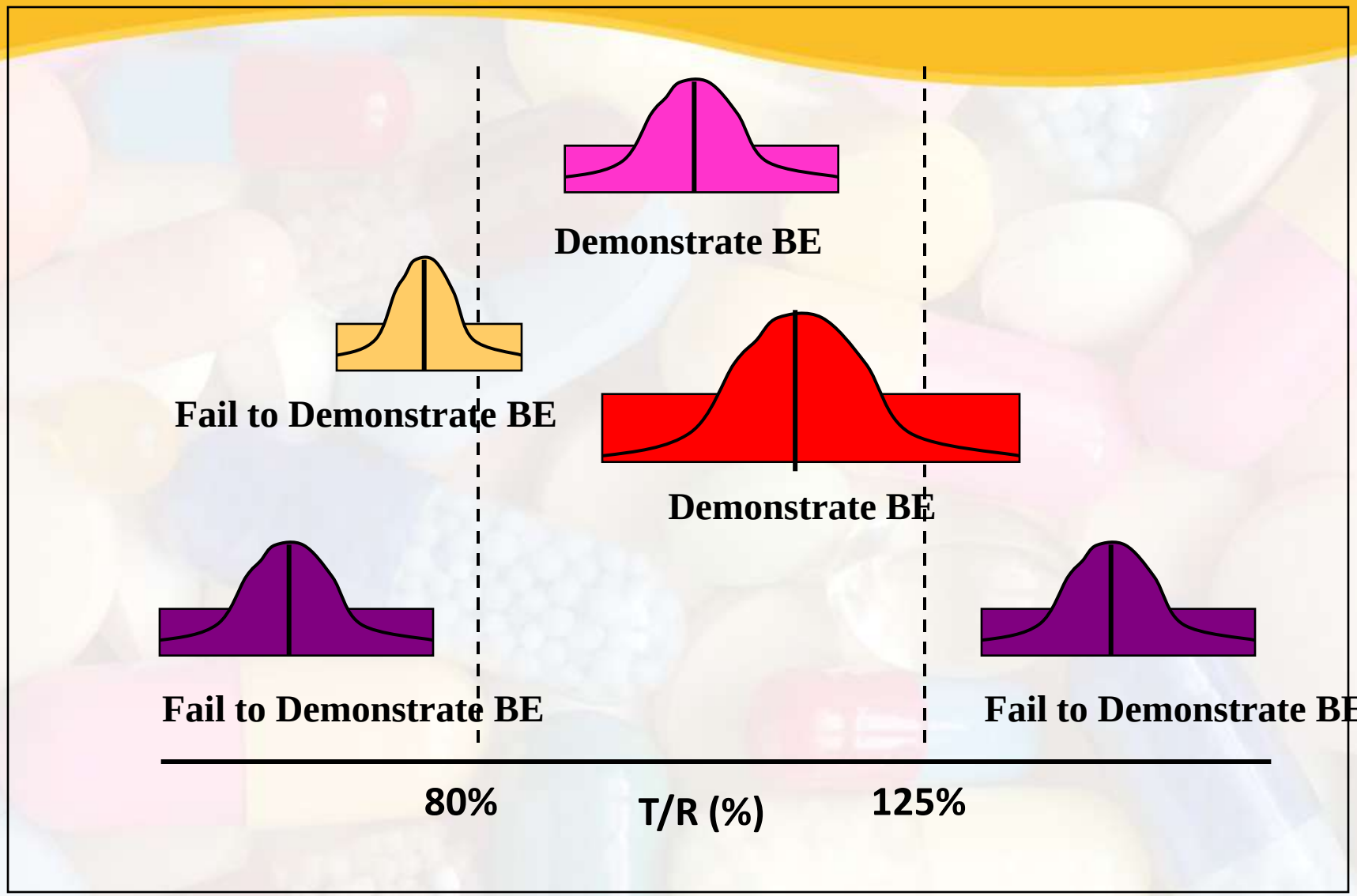
✓ Similarly calculate GMR for AUC

To establish BE:

- ✓ The calculated 90% CI for **C_{max} & AUC**, should fall within range:
  80-125% (Range of Bioequivalence)

- ✓ Non-parametric data 90% CI for **T_{max}** should lie within clinical acceptable range

BE Results


Demonstrate BE
Fail to Demonstrate BE
Demonstrate BE
Fail to Demonstrate BE
Fail to Demonstrate BE
80%
T/R (%)
125%

✓ Tighter limits may be required for drugs which have:

- A narrow therapeutic index
- A serious dose-related toxicity
- A steep dose-response curve
- Non-linear pharmacokinetics within therapeutic range

✓ Wider range maybe acceptable, based on sound clinical justification

✓ Suprabioavailability

- New product displays an extent of absorption, larger than approved product
- Reformulation to lower dosage f/b fresh BA & BE study
- Otherwise, clinical data required

Bioequivalence assessment of two formulations of ibuprofen

Table 5 Pharmacokinetic parameters over eight hours with two formulations, Brufen® (reference) and Dolaraz® (test) after a single oral dose of 100 mg formulation in 24 healthy adult male volunteers

Parameter	Reference mean (±SD)	Test (Mean ± SD)
$AUC_{0-\infty}$ µg/mL/hour	31.79 (10.60)	29.69 (9.79)
AUC_{0-t} µg/mL/hour	28.17 (8.12)	27.21 (9.01)
C_{max} µg/mL	9.92 (2.13)	10.05 (1.84)
T_{max}, hours	0.80 (0.42)	0.90 (0.58)
K_e	0.31 (0.22)	0.36 (0.23)
$T_{1/2}$, hours	2.98 (1.37)	2.44 (1.19)

Table 6 Statistical results and ratios of means of test and reference products and 90% confidence intervals

Pharmacokinetic parameters	Doloraz® mean	Brufen® mean	Ratio of means	90% CI
$LnAUC_{0-\infty}$	3.34	3.41	0.981	0.807–1.092
$LnAUC_{0-t}$	3.26	3.30	0.987	0.838–1.098
LnC_{max}	2.29	2.27	1.009	0.914–1.138

Modified-release drug products

- ✓ Drug release characteristics of time course &/or release location
- ✓ Chosen to achieve therapeutic &/or convenience objectives not offered by immediate release forms

Includes:

- Delayed release
- Sustained release
- Mixed immediate & sustained release
- Mixed delayed & sustained release
- Mixed immediate & delayed release

Should meet following criteria:

✓ Meet the label claims

✓ Preclude any dose-dumping

✓ Provide therapeutic equivalence with:

- Multiple doses of reference product

OR

- Reference modified release formulation

✓ Produce plasma levels within therapeutic range

Study Design for Modified Release formulation

- **Unlikely to accumulate**:

✓ First market entry

Comparison between Single dose of Modified release preparation & Immediate release formulation as per established dose regimen

✓ Subsequent market entry

Comparison with Reference Modified release product

- **Likely to accumulate**:

✓ Both single & steady state doses of Modified Release formulation compared with immediate release formulation as per established dose regimen

➢ Effect of food:

✓ Not known/ Known that food affects absorption:

Two way cross over studies both in Fasting & Fed state

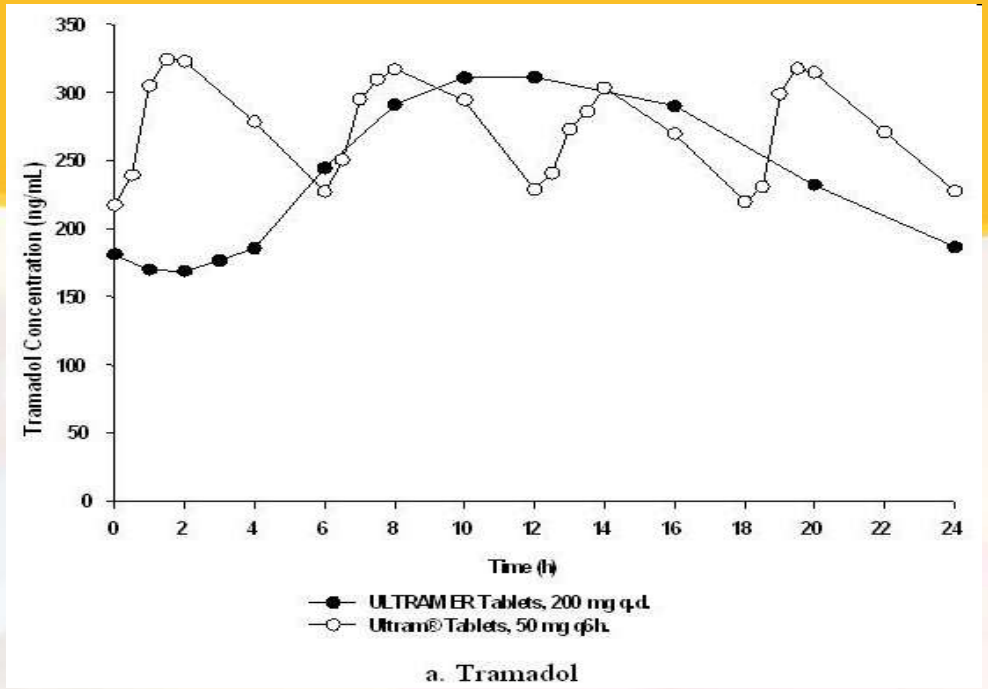
✓ Known that it not affected by food:

Three way cross over study done with

- Reference product in Fasting state
- Test product in Fasting state
- Test product in Fed state

Tramadol : modified release

Mean Steady-State Pharmacokinetic Parameter Values (n=32)



a. Tramadol

Pharmacokinetic Parameter	**ULTRAM - ER (200- mg OD)**	**ULTRAM (50-mg QID)**
AUC_{0-24} (ng.h/mL)	5975 (85-90%)	6613
C_{max} (ng/mL)	335	383
C_{min} (ng/mL)	187	228
T_{max} (h)	12	1.5
% Fluctuation	61	59

Conduct of Study

➢ Pre-study Requirements

- ✓ IEC approved protocol
- ✓ Written procedure (SOPs) for all the study related activities
- ✓ In accordance with ICH-GCP Guidelines
- ✓ Adequate infrastructure- Clinical facility
- ✓ Trained Study personnel
- ✓ Healthy Volunteers

➢ Screening of Healthy volunteers

- ✓ Recruitment through advertisements
- ✓ Written consent for Screening & Consent for HIV testing
- ✓ Height & weight
- ✓ Medical History
- ✓ Physical examination, ECG & vital signs examination
- ✓ Blood & Urine sample

(Lab testing,; tests for HIV, Hepatitis A, B & C; UPT→ females)

➢ Volunteer Selection & Recruitment

- ✓ Volunteers called 1 day before study & admitted
- ✓ Written ICF taken

➢ During the Study

- ✓ Standardized study environment
- ✓ Vital signs examination at scheduled times
- ✓ Standardised amount of water [~240ml]
- ✓ No concomitant medications [including herbal remedies]

- ✓ Administration of the study medication is supervised by the investigator
- ✓ Same time of dosing (multiple dosing)
- ✓ Sampling time with deviation of 2 mins allowed
- ✓ Uniform & identical meals at identical times in all periods
- ✓ Restriction of xanthines, grapefruit, citrus fruits, smoking, alcohol
- ✓ Physical activity & posture standardized→ limit effects on GI flow & motility
- ✓ All activities recorded in CRFs with time & date

➢ End of Study

- ✓ Post-study examination for safety assessment
- ✓ Compensation to subjects as per agreed terms
- ✓ Clinical part of study completed

Documentation

- Signed detailed protocol
- Approval by Ethics Committee
- Volunteer Information sheet
- Informed Consent Form (ICF)
- Case Record Form (CRF)
- Undertaking by investigator
- CV of investigator
- Randomization chart
- Laboratory certification
- Analytical method validation details
- Chromatograms of all volunteers including any aberrant ones
- Tabulated Raw Data of volunteers

Maintenance of Records & Retention of Study Samples

- ✓ All **Records** of in vivo tests on any marketed batch of a drug product should be maintained by the Sponsor for atleast 2 years after expiry date of the batch

- ✓ All **Drug samples** to be retained for a period of atleast 3 years after conduct of the study

OR

1year after expiry of the batch
[Stored in conditions consistent with the product labeling]

Reporting of a BA/BE Study

SURROGATE PARAMETERS	Drug plasma level to indicate clinical activity
PRIMARY PK PARAMETERS	F, MAT, Cmax, AUC(0->t) and AUC(0-> ∞)
SECONDARY PK PARAMETERS	Ke, tmax, t1/2e and [AUC(0->t)/ AUC(0-> ∞)]%
CONFIDENCE INTERVALS	Cmax, AUC(0->t) and AUC(0-> ∞) e.g. Cmax – 108.07 (100.61-116.09)%
CONCLUSIONS	Point estimates and the 90% con. Interval for the log transformed ratios for AUC were within 80.00-125.00%. Cmax to be within 75.00-133.00%. And Mean Absorption Time for Test Product is 84min. Therfore the absolute bioavailability of Furosen® (furosemide), F>1 proves that systemically bioavailable, can be concluded
FINAL REPORT DATE	19 APRIL 2009

Pharmacodynamic Studies

➢ Measurement of effect on a Patho-physiological process as a function of time, after administration of 2 different products

✓Necessity:

1. Quantitative analysis in plasma or urine not possible with sufficient accuracy & sensitivity

2. Drug concentrations are not surrogate endpoints
   e.g. Topical formulations without systemic absorption

3. In situations of 'Superiority Claims'

✓ In case only Pharmacodynamic data is collected→ other methods tried & why they were unsuitable

✓ Special considerations while conducting this study:

- Response measured→ Pharmacological/ Therapeutic effect→ relevant to Efficacy/ Safety of drug

- Methodology validated→ Precision, accuracy, reproducibility, specificity

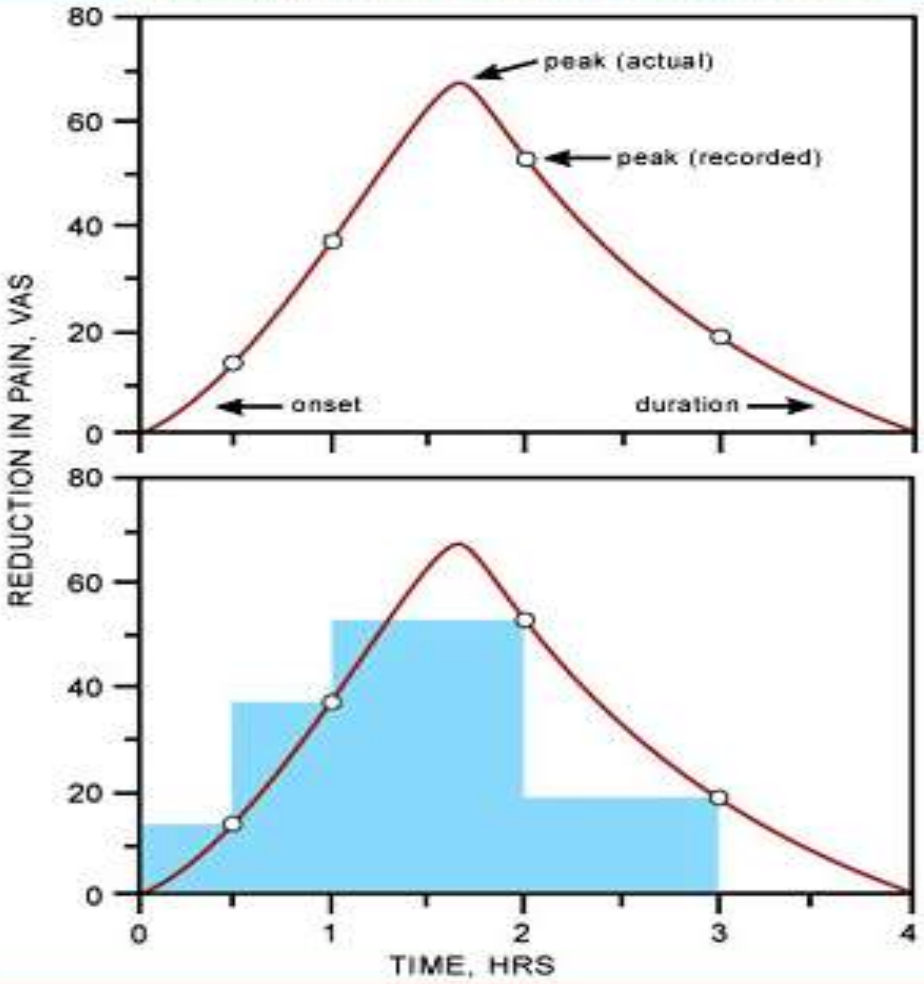
✓ Neither should produce a maximal response→
not possible to distinguish differences between formulations given in those doses

✓ Response measured "quantitatively" under double-blind conditions, on repetitive basis, to record pharmacodynamic events→
Pharmacodynamic effect curve
Eg: Heart rate, pupil diameter, BP

Parameters studied:

- Area under the curve
- Maximum response
- Time for maximum response

Figure 1.2: Approximating the Analgesic Time-Effect Curve with Intermittent Pain and Relief Measures



Reduction in pain is plotted against time elapsed since administration of study medication. *Open circles* correspond to the patient's pain assessment at scheduled interviews. Onset and duration of meaningful analgesia are indicated by *arrows (top panel)*; in this example, onset occurs well before the first scheduled interview. The area under this hypothetical curve *(bottom panel, shaded)* is often approximated by the sum of the products of each pain relief reading at a scheduled interview and the time since the previous interview.

- ✓ *Non Responders* excluded by prior screening

- ✓ If *Placebo effect* can occur→ 3rd Stage with Placebo treatment in study design

- ✓ In *Patients*→ Underlying Pathology & Natural-history considered

- ✓ Conventional acceptance range → defined in protocol, case to case basis

Orlistat capsule formulations

- Orlistat acts in the lumen of the stomach & small intestine, by binding lipases→ inhibits absorption of dietary fat
- 18 subjects were randomized for a parallel study
- Given different Orlistat formulations for 10 days, with high fat diet
- Fecal fat excretion over 24 hours→ endpoint for therapeutic equivalence
- **Ratio of FFE(24)** of the generic to the innovator formulation : **99.1%** with 90% confidence intervals of 83.8 -114.5%

Comparative Clinical Studies

✓Necessity:

- Both pharmacokinetic & pharmacodynamic parameters *not properly measurable or not feasible*
- Mention which methods were tried & found unsuitable

✓Statistical principles to be considered:

- *No. of patients*→ *Variability* of assessed parameters & *acceptance range*
- Much higher than BE studies

Following critical points need to be defined in advance, on case to case basis:

- ✓ Clinical end points (Target parameters)→ intensity & onset of response
- ✓ Size of equivalence range→ case-to-case basis
  (dependins on natural course of disease, efficacy of available treatments, target parameter)
- ✓ Statistical confidence interval approach:
  one-sided interval→ rule out inferiority
- ✓ Placebo included when appropriate
- ✓ Safety end-points in some cases

Comparative clinical study

Artesunate suppositories and oral Artesunate

- **Artesunate suppositories** (15 mg/kg/day for three days)
- **Oral Artesunate** (6 mg/kg/day for three days)

} with Mefloquine (25 mg/kg)

52 children participated (*large number*; BE studies : 16)

Mean times to fever subsidence : similar in two groups

Cure rates by day 28 : similar

} **Clinical parameters**

Time to parasite clearance : not significantly more in Suppository group

Safety profile : good in both groups

Dissolution Studies

1. Suitable to confirm unchanged product quality with minor changes in formulation / manufacturing after approval→

   SUPAC (Scale-Up & Post-Approval Changes)

2. Different strengths of drug manufactured by same manufacturer where:

- Qualitative composition is same
- Ratio of active ingredients & excipients is same
- Method of manufacture is same
- BE study has been performed on 1 strength
- Linear pharmacokinetics

3. Signal of bio-inequivalence

4. Assess batch-to-batch quality

✓ More than 1 batch of each formulation tested

Design should include:

- ✓ Individually testing of atleast 12 dosage units of each batch → Mean & Individual results with Sd or SE

- ✓ Measurement of percentage of content released at suitably spaced time points

(eg. At 10, 20 & 30 mins or appropriate for complete dissolution)

- ✓ Dissolution profile in atleast 3 aqueous media with pH range of 1.0-6.8 **Or** 1.0-8.0 wherever necessary

- ✓ Conduct tests on each batch with same apparatus & on same or consecutive days

Dissolution testing should be carried out in:

✓ USP Apparatus I at 100 rpm or Apparatus II at 50 rpm using 900 ml of the following dissolution media:

- 0.1N HCl or Simulated Gastric Fluid USP without enzymes
- a pH 4.5 buffer
- a pH 6.8 buffer or Simulated Intestinal Fluid USP without enzymes

✓ For capsules and tablets with gelatin coating

- Simulated Gastric and Intestinal Fluids USP (with enzymes) can be used

Advantages offered

✓ Reduced costs:

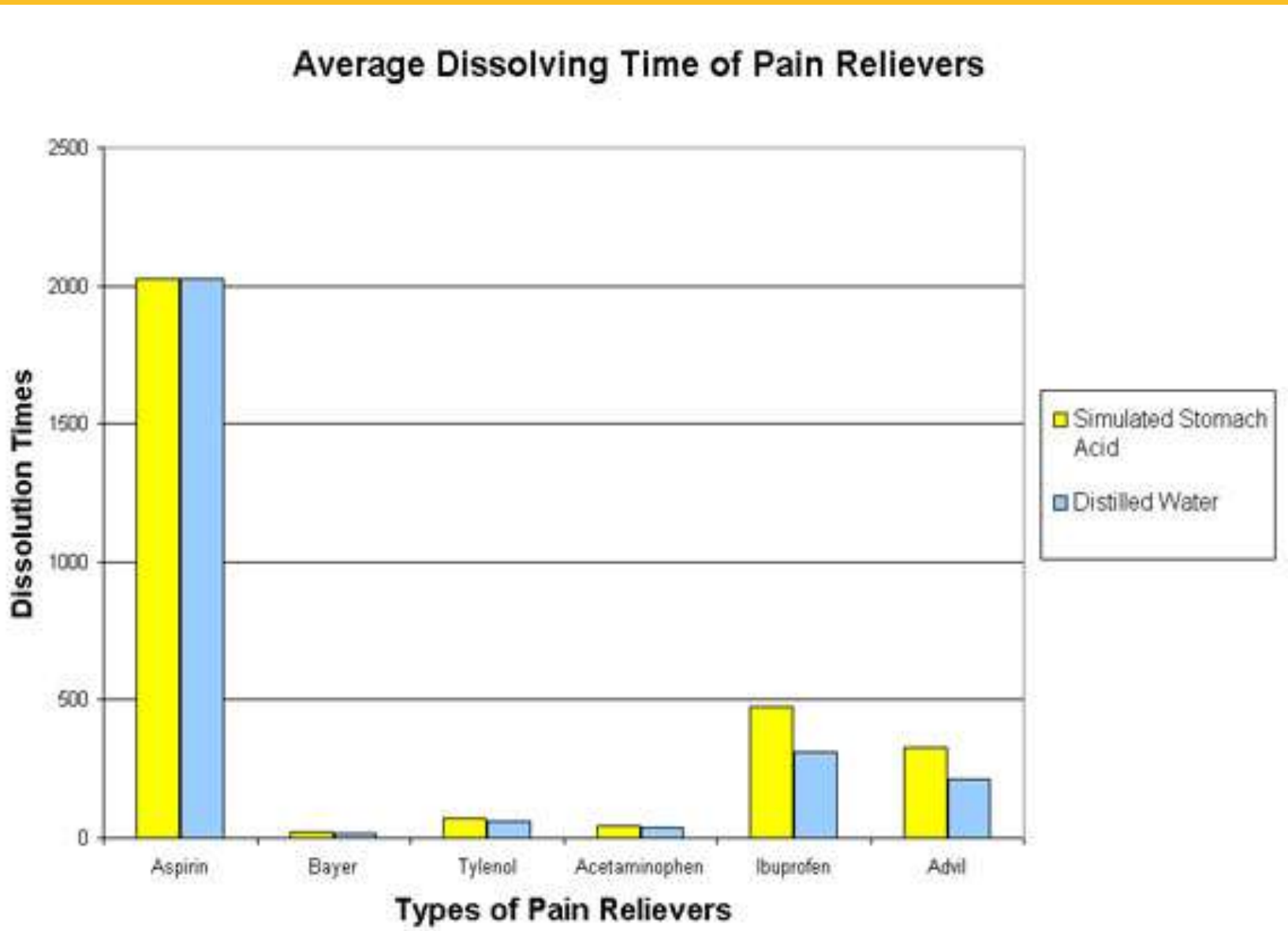
- Data anticipates Bioequivalence
- Type II error ++ for PK studies

✓More direct assessment:

- If complicated in-vivo assessment

✓ Ethical benefit:

- No unnecessary human study


Average Dissolving Time of Pain Relievers
Dissolution Times
2500
2000
1500
1000
500
0
Aspirin
Bayer
Tylenol
Acetaminophen
Ibuprofen
Advil
Types of Pain Relievers
Simulated Stomach Acid
Distilled Water

Conclusion

✓ Concept of BE has been adopted by the pharmaceutical industry & national regulatory authorities throughout the world for over 20 years

✓ There is a continuing attempt to understand & develop more efficient & scientifically valid approaches to assess bioequivalence of various dosage forms including some of the tough complex special dosage forms

✓ Bioequivalence industry always existed in India→ become more matured now

✓ Changes in patent laws has added tremendous fuel to this growth

✓ Many BA/BE CROs in India

- ✓ Generics help patients by making drugs available at affordable price while retaining their quality
- ✓ Balance public interests especially in diseases like Cancer & AIDs which have high prevalence in developing countries & patented drugs are steeply priced
- ✓ Value of drugs going off-patent in the regulated market is estimated at US $ 70-80 billions in next 5 years
- ✓ Translated into increased opportunities for Indian Pharmaceutical Industry → Export of generics to the regulated markets


Thank
You!