previous Lecture

next Lecture

Bioavailability / Bioequivalence

- Selection of CROs
- Selection of a Reference Product
- Metrics (AUC, C_{max}/t_{max}, Shape of Profile)
- Acceptance Ranges (0.80 – 1.25 and beyond)
- Sample Size Planning (Literature References, Pilot Studies)
- Steps in bioanalytical Validation (Validation Plan, Pre-Study Validation, In-Study Validation)
- **Study Designs**
- Protocol Issues
- Evaluation of Studies
- Advanced Topics
- Avoiding Pitfalls

Bioavailability / Bioequivalence

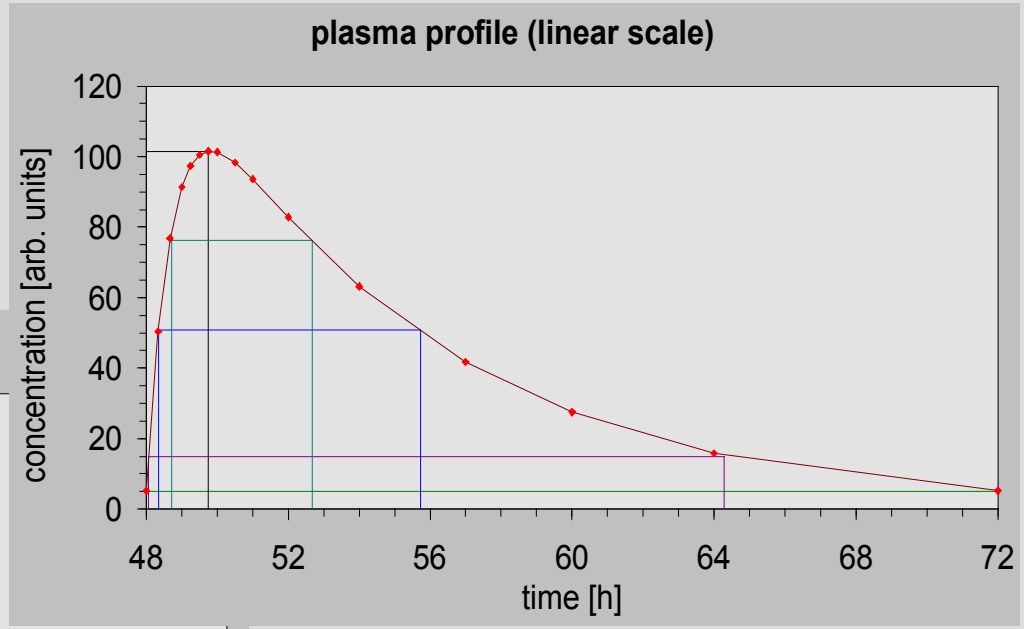
- **Study Designs**
  - Single Dose / Multiple Dose
  - Standard 2×2 Cross-over
  - Parallel Groups
  - for more than 2 Formulations

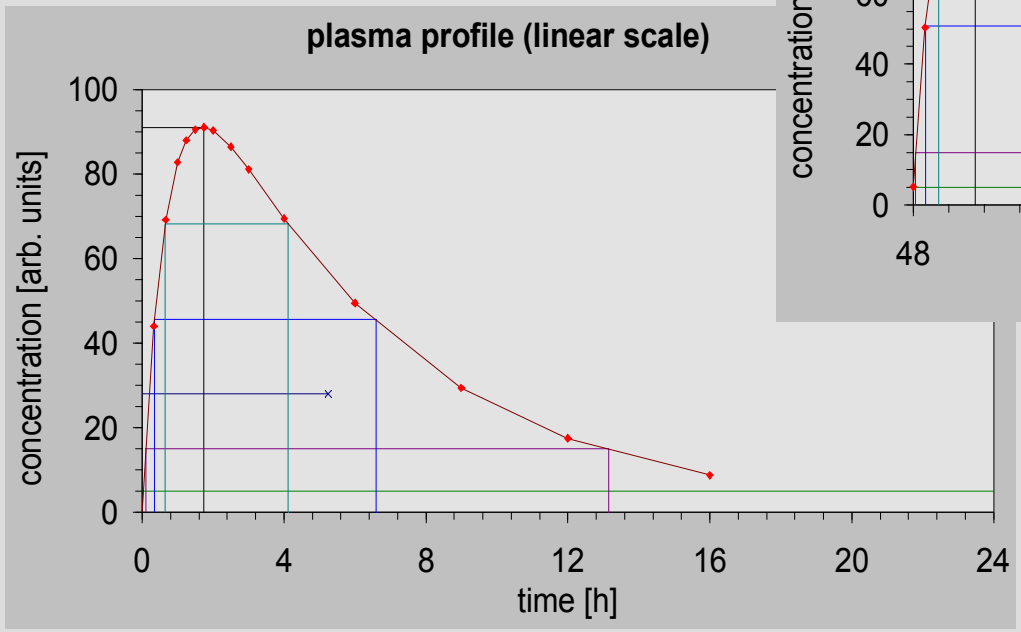
Bioavailability / Bioequivalence

- **Study Designs** (Single Dose / Multiple Dose)
  - Single Dose recommended in most international Guidelines, but steady-state studies:
    - may be required:
      - in the case of dose- or time-dependent pharmacokinetics,
      - for some modified release products (+ Single Dose BE).
    - may be considered:
      - if problems of sensitivity preclude sufficiently precise plasma concentration measurements after SD administration,
      - if the intra-individual variability in the plasma concentration or disposition precludes the possibility of demonstrating BE in a reasonably sized single dose study and this variability is reduced at steady state.

Bioavailability / Bioequivalence

- **Study Designs** (Single Dose / Multiple Dose)





Bioavailability / Bioequivalence

- **Study Designs** (Single Dose / Multiple Dose)
  - With the current developments in bioanalytical methodology (*e.g.*, LC-MS/MS), you should have strong evidence of infeasability if you claim the necessity of a Multiple Dose study based on lacking methods.

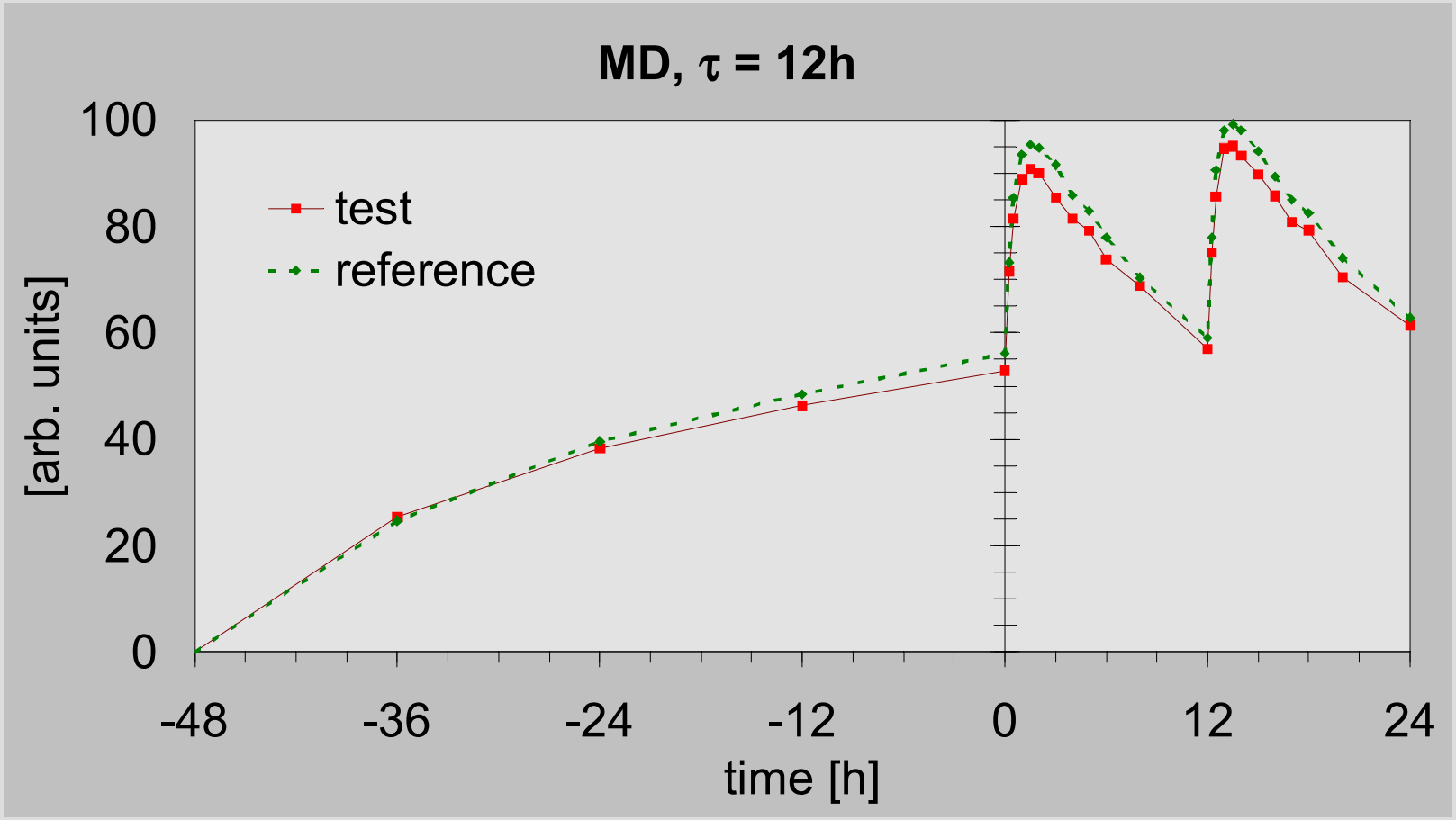
    Regulators are concerned with efficacy/safety issues and not with the budget of pharmaceutical companies.

Bioavailability / Bioequivalence

- **Study Designs** (Single Dose / Multiple Dose)
  - Although using Multiple Dose studies in order to reduce variability for HVDs is proposed *'for consideration'* in the European NfG, such studies are not accepted in all EU countries, and this paragraph may be removed in the upcoming QA-document and/or the new NfG!

Bioavailability / Bioequivalence

- **Study Designs** (Single Dose / Multiple Dose)



Bioavailability / Bioequivalence

- **Study Designs** (Single Dose / Multiple Dose)
  - In order to fulfil the superposition principle of linear PK ($AUC_{\infty} = AUC_{\tau}$), you must demonstrate steady-state:
    - Linear-regression of pre-dose values in saturation phase:
      - ➔ slope (from at least the last three values) should not significantly differ from zero,
      - ➔ subjects not showing steady-state should be excluded from the evaluation.

Bioavailability / Bioequivalence

- **Study Designs** (Single Dose / Multiple Dose)
  - Demonstration of steady-state:
    - Multivariate method (simultaneous testing of all pre-dose values in all subjects):
      - → Hotellings T^2
      - → Drawback: if significant result, no possibility to exclude subjects (rendering the entire study worthless).
    - *t*-test of last two pre-dose values:
      - → Pro: most easy to perform, relatively insensitive to outliers.
      - → Con: as above.
  - No Wash-out between Periods (Switch-Over)!

Bioavailability / Bioequivalence

- **Study Designs** (Single Dose / Multiple Dose)
  - If your Drug shows Polymorphism (*e.g.*,

    CV_{inter} = 10fold*) of CV_{intra})
    - in metabolizing enzymes (*e.g.*, CYP450-3A), or
    - in transporters (PGP), which potentially may lead to
    - safety problems in Poor Metabolizers (PM),
      - → you should consider phenotyping in screening, and
      - → include only Extensive Metabolizers (EM) in the study (example: Paroxetine).

*) for most drugs CV_{inter} = 1.5fold – 2fold of CV_{intra}

Bioavailability / Bioequivalence

- **Study Designs**
  - Single Dose / Multiple Dose
  - Standard 2×2 Cross-over
  - Parallel Groups
  - for more than 2 Formulations

Bioavailability / Bioequivalence

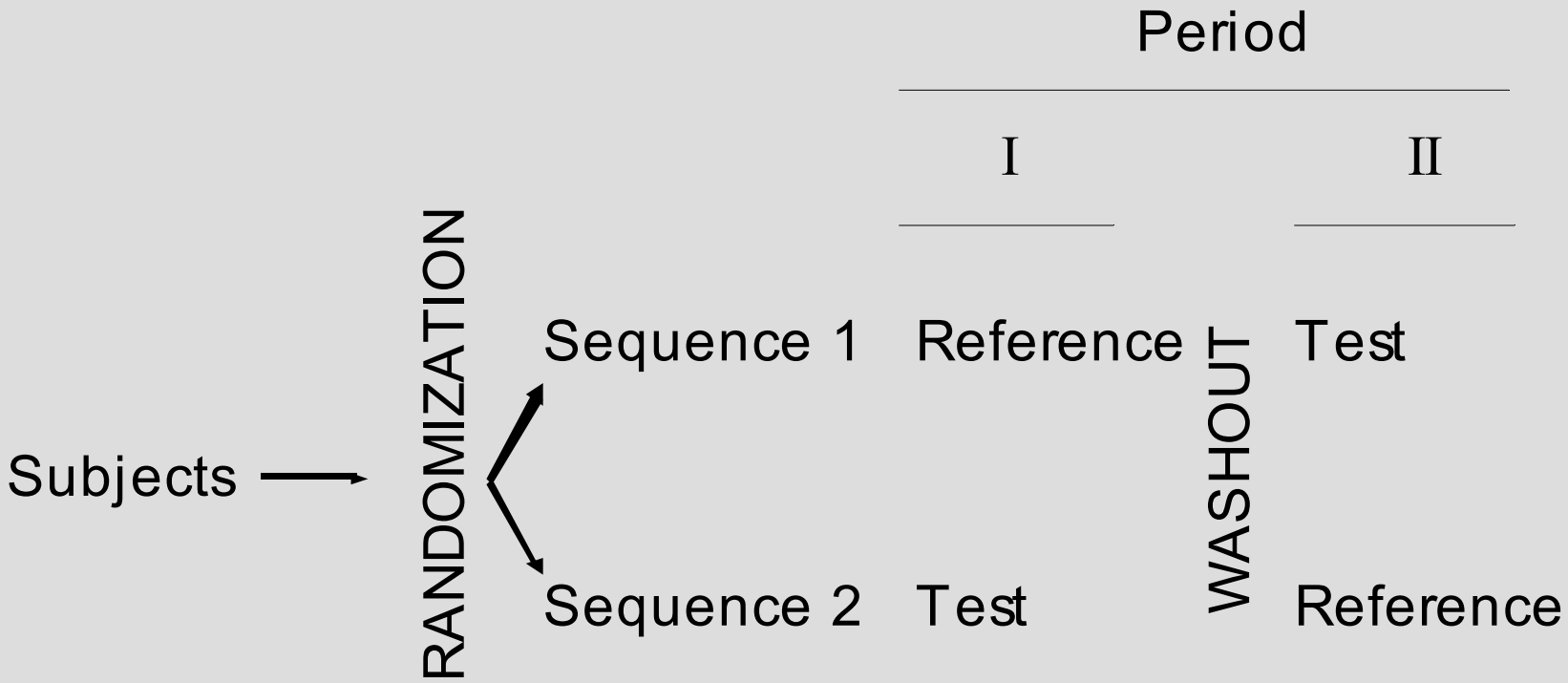
- **Study Designs** (Standard 2×2 Cross-over)
  - Suggested References
    - S.-C. Chow and J.-p. Liu;
      Design and Analysis of Bioavailability and Bioequivalence Studies.
      Marcel Dekker, New York (2nd ed. 2000)
    - B. Jones and M.G. Kenward;
      Design and Analysis of Cross-Over Trials.
      Chapman & Hall, Boca Raton (2nd ed. 2003)
    - S. Senn;
      Cross-over Trials in Clinical Research.
      Wiley, Chichester (2nd ed. 2002)

Bioavailability / Bioequivalence

- **Study Designs** (Standard 2×2 Cross-over)
  - Two-sequence, two-period, cross-over design
    - Each subject is randomly assigned to either sequence RT or sequence TR at two dosing periods.
    - Dosing periods are separated by a washout period of sufficient length for the drug received in the first period to be completely metabolized or excreted from the circulation.
    - Smaller subject numbers compared to a parallel design, since the *within-subject* variability determines sample size (rather than *between-subject* variability).

Bioavailability / Bioequivalence

- **Study Designs** (Standard 2×2 Cross-over)



Bioavailability / Bioequivalence

- **Study Designs** (Standard 2×2 Cross-over)
  - Multiplicative model (without carryover)

$$X_{ijk} = \mu \cdot \pi_k \cdot \Phi_l \cdot s_{ik} \cdot e_{ijk}$$

X_{ijk}: *ln*-transformed response of j-th subject (j=1,…,n_i) in i-th sequence (i=1,2) and k-th period (k=1,2), μ: global mean, μ_l: expected formulation means (l=1,2: μ_l=μ_T, μ_2= μ_R),

π_k: fixed period effects, Φ_l: fixed formulation effects (l=1,2: Φ_l=Φ_T, Φ_2= Φ_R), s_{ik}: random subject effect, e_{ijk}: random error.

Bioavailability / Bioequivalence

- **Study Designs** (Standard 2×2 Cross-over)
  - Multiplicative model (without carryover)
    - Main Assumptions
      - All $ln\{s_{ik}\}$ and $ln\{e_{ijk}\}$ are independently and normally distributed about unity with variances σ^2_s and σ^2_e,[1)]
      - All observations made on different subjects are independent.[2)]

1) This assumption may not hold true for all formulations; if the reference formulation shows higher variability than the test formulation, a ‘good’ test will be penalized for the ‘bad’ reference.

2) This assumption should not be a problem, unless you plan to include twins or triplets in your study.

Bioavailability / Bioequivalence

- **Study Designs** (Standard 2×2 Cross-over)

Transformations (*e.g.* […], logarithm) should be specified in the protocol and a rationale provided […]. The general principles guiding the use of transformations to ensure that the assumptions underlying the statistical methods are met are to be found in standard texts […].

In the choice of statistical methods due attention should be paid to the statistical distribution […]. When making this choice (for example between parametric and non-parametric methods) it is important to bear in mind the need to provide statistical estimates of the size of treatment effects together with confidence intervals […].

Anonymous [International Conference on Harmonisation];
Topic E 9: Statistical Principles for Clinical Trials.
http://www.ich.org/MediaServer.jser?@_ID=485&@_MODE=GLB
(5 February 1998)

Bioavailability / Bioequivalence

- **Study Designs** (Standard 2×2 Cross-over)

No analysis is complete until the assumptions that have been made in the modeling have been checked. Among the assumptions are that the repeated measurements on each subject are independent, normally distributed random variables with equal variances. Perhaps the most important advantage of formally fitting a linear model is that diagnostic information on the validity of the assumed model can be obtained. These assumptions can be most easily checked by analyzing the residuals.

B. Jones, B. and M.G. Kenward;
Design and Analysis of Cross-Over Trials.
Chapman & Hall, Boca Raton (2nd ed. 2003)

Bioavailability / Bioequivalence

- **Study Designs** (Standard 2×2 Cross-over)

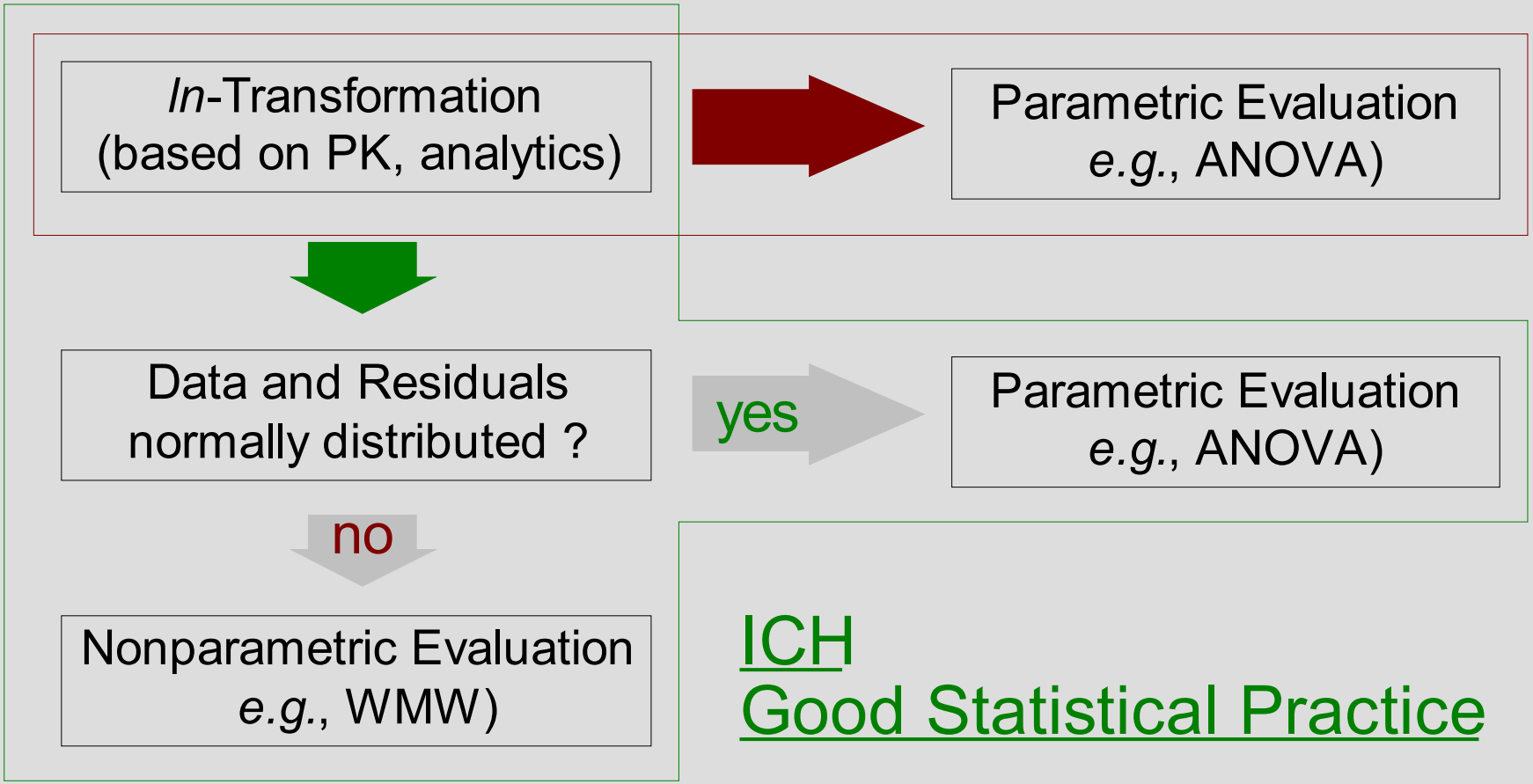
The limited sample size in a typical BE study precludes a reliable determination of the distribution of the data set. Sponsors and/or applicants are not encouraged to test for normality of error distribution after log-transformation […].

Anonymous [FDA, Center for Drug Evaluation and Research (CDER)]; Guidance for Industry: Statistical Approaches to Establishing Bioequivalence. http://www.fda.gov/cder/guidance/3616fnl.pdf (January 2001)

Bioavailability / Bioequivalence

- **Study Designs** (Standard 2×2 Cross-over)

FDA



ICH
Good Statistical Practice

Bioavailability / Bioequivalence

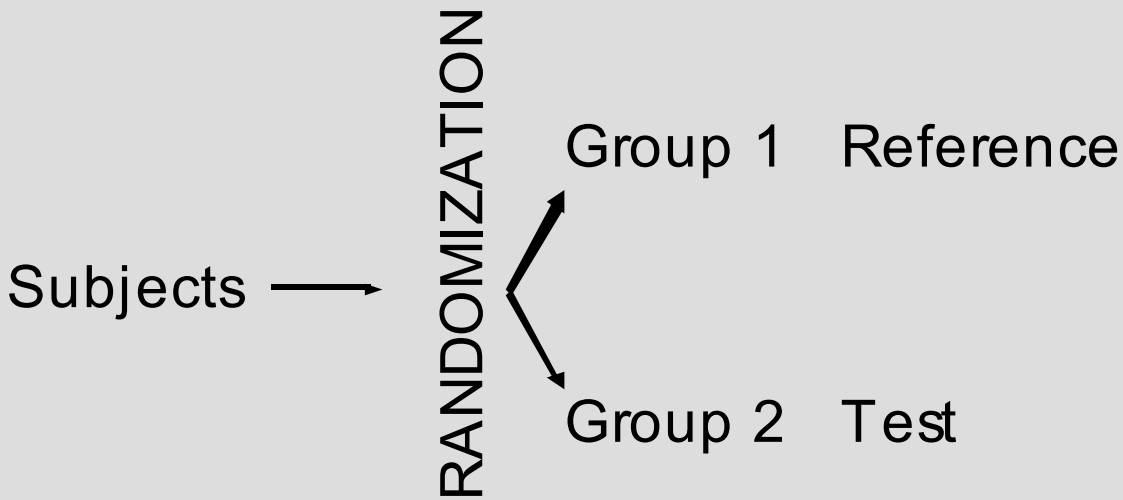
- **Study Designs** (Standard 2×2 Cross-over)
  - Advantages
    - Globally applied standard protocol for BE.
    - Straigthforward statistical analysis.
    - Scaled average bioequivalence for *'bad'* reference formulations (acceptance?).
  - Disadvantages
    - Not suitable for drugs with long half life (→ parallel groups).
    - Not optimal for studies in patients (→ parallel groups).
    - Not optimal for HVDs (→ replicate designs).
    - If carryover observed, study most likely fails.

Bioavailability / Bioequivalence

- **Study Designs** (Parallel Groups)
  - Two-group parallel design
    - Each subject receives one and only one formulation of a drug in a random fashion.
    - Usually each group contains the same number of subjects.
    - Higher subject numbers compared to a cross-over design, since the *between-subject* variability determines sample size (rather than *within-subject* variability).

Bioavailability / Bioequivalence

- **Study Designs** (Parallel Groups)



Bioavailability / Bioequivalence

- **Study Designs** (Parallel Groups)
  - Advantages
    - Clinical part (sometimes) faster than X-over.
    - Straigthforward statistical analysis.
    - Drugs with long half life.
    - Studies in patients.
  - Disadvantages
    - Lower statistical power than X-over (*rule of thumb:* subject number should at least be doubled).
    - Phenotyping mandatory for drugs showing polymorphism.

Bioavailability / Bioequivalence

- **Study Designs** (for more than two formulations)
  - Variance-Balanced Design
    - For *e.g.* three formulations there are three possible pairwise differences among formulation means (*i.e.*, form. 1 *vs.* form. 2., form 2 *vs.* form. 3, and form. 1 *vs.* form. 3).
    - It is desirable to estimate these pairwise effects with the same degree of precision (there is a common variance for each pair).
    - Such a design for three formulations is the six-sequence, three-period **Williams Design**.

Bioavailability / Bioequivalence

- **Study Designs** (for more than two formulations)

	Period		
Sequence	I	II	III
1	R	T_2	T_1
2	T_1	R	T_2
3	T_2	T_1	R
4	T_1	T_2	R
5	T_2	R	T_1
6	R	T_1	T_2

Bioavailability / Bioequivalence

- **Study Designs** (for more than two formulations)
  - Variance-Balanced Design
    - For *e.g.* four formulations there are six possible pairwise differences among formulation means.
    - Suitable is the four-sequence, four-period Williams Design.

	Period			
Sequence	I	II	III	IV
1	R	T_3	T_1	T_2
2	T_1	R	T_2	T_3
3	T_2	T_1	T_3	R
4	T_3	T_2	R	T_1

Bioavailability / Bioequivalence

- **Study Designs** (for more than two formulations)
  - Advantages
    - Allows to choose between two or more candidate test formulations.
    - Comparison of a test formulation with several references.
    - Standard design for establishment of Dose Proportionality.
  - Disadvantages
    - Statistical analysis more complicated (especially in the case of drop-outs).
    - May need measures against multiplicity (increasing the sample size).

Bioavailability / Bioequivalence

- **Study Designs** (for more than two formulations)
  - Disadvantages
    - Multiplicity:

      Bonferroni-correction needed if more than one formulation will be marketed (for 3 simultaneous comparisons without correction the patient's risk is increased from 5 % to 14 %).

k	$P_{\alpha=0.05}$	$P_{\alpha=0.10}$	α_{adj}	$P_{\alpha adj}$	α_{adj}	$P_{\alpha adj}$
1	5.00%	10.00%	0.0500	5.00%	0.100	10.00%
2	9.75%	19.00%	0.0250	4.94%	0.050	9.75%
3	14.26%	27.10%	0.0167	4.92%	0.033	9.67%
4	18.55%	34.39%	0.0125	4.91%	0.025	9.63%
5	22.62%	40.95%	0.0100	4.90%	0.020	9.61%
6	26.49%	46.86%	0.0083	4.90%	0.017	9.59%