

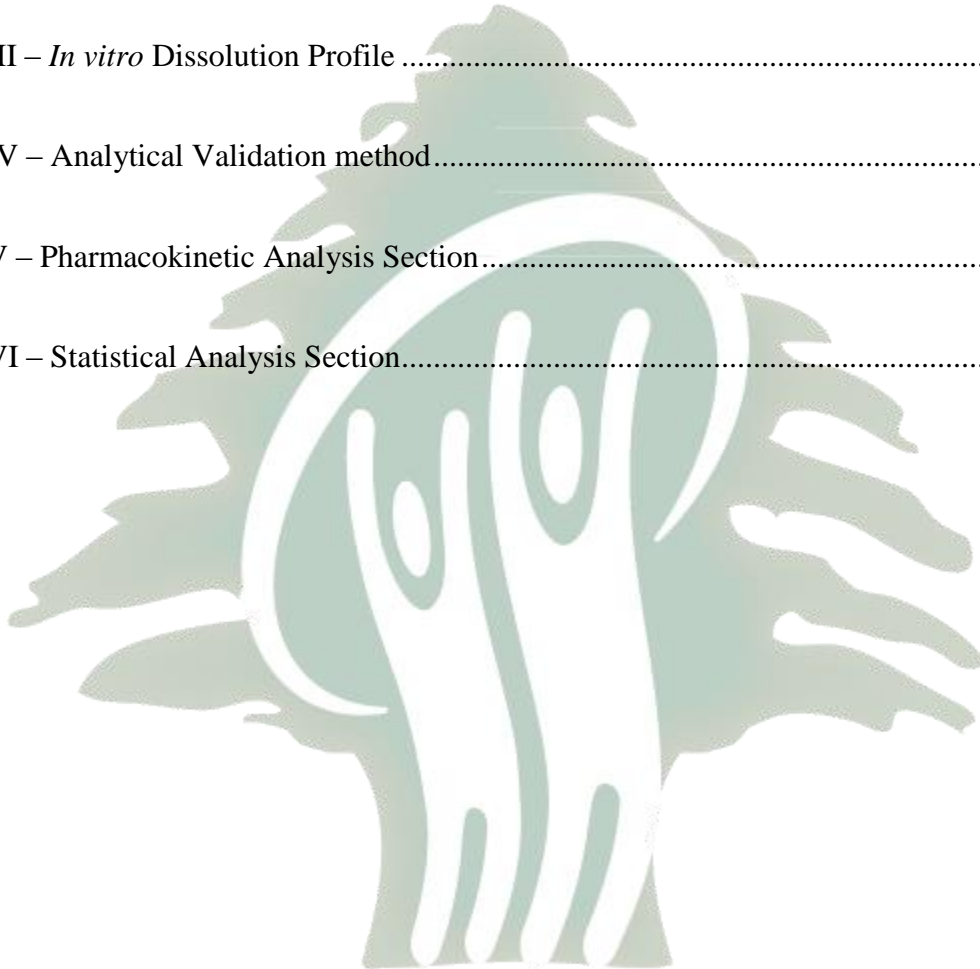


Guidelines for the Clinical Part Module 5 Bioequivalence

January 2017

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Definitions

Bioavailability

Bioavailability means the rate and extent to which the active drug substance or therapeutic moiety is absorbed from a pharmaceutical form and becomes available at the site of action. For drugs intended to exhibit a systemic therapeutic effect, bioavailability can be more simply understood as the rate and extent to which a substance or its therapeutic moiety is delivered from a pharmaceutical form into the general circulation. Indeed, in the case of such drugs, the substance in the general circulation is in exchange with the substance at the site of action.

Pharmaceutical equivalent

It refers to drug products, which contain the same active ingredient in the same strength (concentration) and dosage form, and is intended for the same route of administration. In general, it has the same labeling and meets compendial and other standards of strength, quality, purity, and identity. Pharmaceutical equivalent does not necessarily imply therapeutic equivalence as differences in the excipients and/or the manufacturing process can lead to differences in product performance.

Pharmaceutical alternatives

Drug products are considered pharmaceutical alternatives if they contain the same therapeutic moiety, but are different salts, esters, or complexes of that moiety, or are different dosage forms or strengths. Different dosage forms and strengths within a product line by a single manufacturer are thus pharmaceutical alternatives, as are extended-release products when compared with immediate or standard-release formulations of the same active ingredients.

Generic product

A **generic medicinal product** is a product which has the same qualitative and quantitative composition in active substances and the same pharmaceutical form as the reference medicinal product (list of References can be found on FDA or EMEA sites), and whose bioequivalence with the reference medicinal product has been demonstrated by appropriate bioavailability studies.

The different salts, esters, ethers, isomers, mixtures of isomers, complexes or derivatives of an active substance are considered to be the same active substance, unless they differ significantly in properties with regard to safety and/or efficacy. Furthermore, the various immediate-release oral pharmaceutical forms shall be considered to be one and the same pharmaceutical form.

Reference product

A reference product is a pharmaceutical product with which the new product is intended to be interchangeable in clinical practice. The reference product would normally be the innovator product for which efficacy, safety and quality have been established. When the innovator product is not available the product which is the word market leader may be used as a reference product, provided that it has been authorized for marketing and its efficacy, safety and quality have been established and documented.

Bioequivalence

Is defined as “the absence of a 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 under similar conditions in an appropriately designed study”.

Two medicinal products containing the **same active substance** are considered bioequivalent if they are pharmaceutically equivalent or pharmaceutical alternatives and their bio availabilities (rate and extent) after administration in the **same molar dose** lie within acceptable predefined limits. These limits are set to ensure comparable *in vivo* performance, i.e. similarity in terms of safety and efficacy (EMA).

Although the concept of bioequivalence possibly could be considered applicable for herbal medicinal products, the general principles outlined in this guideline are not applicable to herbal medicinal products, for which active constituents are less well defined than for chemical entities.

Based on these definitions, MOPH will not accept any application for a generic drug:

1. If the bioequivalence study is not aimed to compare the generic (test) to a brand (reference) containing the same active substance(s).

2. If the reference brand drug is not registered in any of the countries (approved by the technical committee of the MOPH as referenced countries).

In addition, the bioequivalence study should not be prepared by the same pharmaceutical industry producing the generic drug. An independent CRO should realize the study (all sections of the study).

Standard design

If two formulations are compared, a randomized, two-period, two-sequence, single dose, crossover design is recommended. The treatment periods should be separated by a wash out period sufficient to ensure that drug concentrations are below the lower limit of bioanalytical quantification in all subjects at the beginning of the second period. Normally at least 5 elimination half-lives are necessary to achieve this (7 half-lives are required to eliminate 99 % of the drug).

Under certain circumstances an alternative well-established designs could be considered if the company provide a scientifically reasonable study design, statistical analyses.

Whenever possible and applicable, both sex (male and females) should be included in the study design in order for the sample to be representative of the general population.

Pilot studies

A pilot study in a small number of subjects can be carried out before proceeding with a full bioequivalence study. The study can be used to validate analytical methodology, assess variability, optimize sample collection time intervals, and provide other information. For example, for conventional immediate-release products, careful timing of initial samples may avoid a subsequent finding in a full-scale study that the first sample collection occurs after the plasma concentration peak. For modified-release products, a pilot study can help determine the sampling schedule to assess lag time and dose dumping. A pilot study that documents bioequivalence may be acceptable, provided that its design and execution are suitable and a sufficient number of subjects (e.g., 12) have completed the study.

Food-effect studies

Food-effect bioequivalence studies focus on demonstrating comparable bioavailability between test and reference products when administered with meals. Usually, a single-dose, two-period, two-treatment, two-sequence crossover study is recommended for food-effect bioequivalence study. Food-effect bioequivalence studies are generally recommended for modified release products. Food-effect bioequivalence studies are also recommended for certain conventional release drug products. Selection of conventional release drug products that require food studies is based upon certain considerations, such as:

- (i) Documented evidence of effect of food on drug absorption (e.g., cefaclor);
- (ii) The drug is recommended to be administered with food;
- (iii) The drug may produce gastric irritation under fasting conditions, thus may be taken with food (e.g., NSAIDs).

Subjects

For a sound bioequivalence study the sponsor should enroll a number of subjects sufficient to ensure adequate statistical results, which is based on the power function of the parametric statistical test procedure applied. The number of subjects “should be not less than 12” (sometimes more than 24 are needed as in case of highly variable drugs) and should be determined using appropriate methods taking into account the error variance associated with the primary parameters to be studied (as estimated for a pilot experiment, from previous studies, pilot studies or published data), the significance level desired ($\alpha = 0.05$), and the deviation from the reference product compatible with bioequivalence ($\pm 20\%$) and compatible with safety and efficacy. In most of the cases 18-24 normal healthy subjects (sometimes more than 24) preferably non-smoking, between 18-50 years in age and within 10% of ideal body weight for height and body build (Metropolitan Life Insurance Company Statistical Bulletin, 1983) are enrolled in a crossover bioequivalence study. For a parallel design study a greater number of subjects may be required to achieve sufficient study power. Sponsors should enter a sufficient number of subjects in the study to allow for dropouts. Because replacement of subjects could complicate the statistical model and analysis, dropouts generally should not be replaced. If the final number of subjects who completed the study is inferior to the minimum number required that was calculated prior to beginning the study, then results are rejected.

The subjects are not permitted to take any prescription or over-the-counter drug products within two weeks of the start of the study. Ingestion of alcohol or caffeine or related xanthines containing food or beverages is not allowed within 48 hours.

For ethical reasons, some drugs should not be tested on normal subject. Bioequivalence is then to be performed on adult patients who are on stable regimens of the drug for the treatment of their disease.

Choosing the reference batch

Unless otherwise justified, the assayed content of the batch used as test product should not differ more than 5% from that of the batch used as reference product determined with the test procedure proposed for routine quality testing of the test product. The Applicant should document how a representative batch of the reference product with regards to dissolution and assay content has been selected. It is advisable to investigate more than one single batch of the reference product when selecting reference product batch for the bioequivalence study.

Choosing the test product units to be tested in the BE:

The test product used in the study should be representative of the product to be marketed and this should be discussed and justified by the applicant.

For example, for oral solid forms for systemic action:

- a) The test product should usually originate from a batch of at least 1/10 of production scale or 100,000 units, whichever is greater, unless otherwise justified.
- b) The production of batches used should provide a high level of assurance that the product and process will be feasible on an industrial scale. In case of a production batch smaller than 100,000 units, a full production batch will be required.
- c) The characterization and specification of critical quality attributes of the drug product, such as dissolution, should be established from the test batch, i.e. the clinical batch for which bioequivalence has been demonstrated.
- d) Samples of the product from additional pilot and/or full scale production batches, submitted to support the application, should be compared with those of the bioequivalence study test batch,

and should show similar *in vitro* dissolution profiles when employing suitable dissolution test conditions.

Comparative dissolution profile testing should be undertaken on the first three production batches. The dissolution testing should be performed on the same lot (or batch) number of test and reference used in the bioequivalence analysis.

If full scale production batches are not available at the time of submission, the applicant should not market a batch until comparative dissolution profile testing has been completed.

The results should be provided at a Competent Authority's request or if the dissolution profiles are not similar together with proposed action to be taken.

For other immediate release pharmaceutical forms for systemic action, justification of the representative nature of the test batch should be similarly established.

Which Dose to be tested?

If several strengths of a test product are applied for, it may be sufficient to establish bioequivalence at only one (**in general the highest one unless justified**) or two strengths, depending on the proportionality in composition between the different strengths and other product related issues. The strength(s) to evaluate depends on the linearity in pharmacokinetics of the active substance.

In case of non-linear pharmacokinetics (i.e. not proportional increase in AUC with increased dose) there may be a difference between different strengths in the sensitivity to detect potential differences between formulations. In the context of this guideline, pharmacokinetics is considered to be linear if the difference in dose-adjusted mean AUCs is no more than 25% when comparing the studied strength (or strength in the planned bioequivalence study) and the strength(s) for which a waiver is considered.

In order to assess linearity, the applicant should consider all data available in the public domain with regard to the dose proportionality and review the data critically. Assessment of linearity will consider whether differences in dose-adjusted AUC meet a criterion of $\pm 25\%$.

If bioequivalence has been demonstrated at the strength(s) that are most sensitive to detect a potential difference between products, *in vivo* bioequivalence studies for the other strength(s) can be waived.



BE report should include the following documents

When you submit the BE application to the Lebanese MOPH, make sure that you provide the following documents/items:

- Letter from the company (signed and dated) explaining your application and describing all submitted documents.
- BE application (attached with this requirement: section I to Section VI). All questions in each section should be answered and completed in order to review the application.
- In addition to a soft copy, a CD including the complete BE study should be submitted.
- Copy of the signed informed consents **for all subjects** enrolled in the study.
- Copy of the CRF **for all subjects** enrolled in the study.
- Copy of subjects' chromatograms realized in analytical section.
- Copy of all investigators CV
- CD including **raw data** (as Excel sheet) for all analytical validation method.
- CD including **raw data** (as Excel sheet) for plasma drug concentration at each time point for each subject enrolled in the study.

Raw data on Excel sheets are required because the Lebanese MOPH has the right to repeat any analysis/statistical analysis to validate the data presented in the BE study.

Notes:

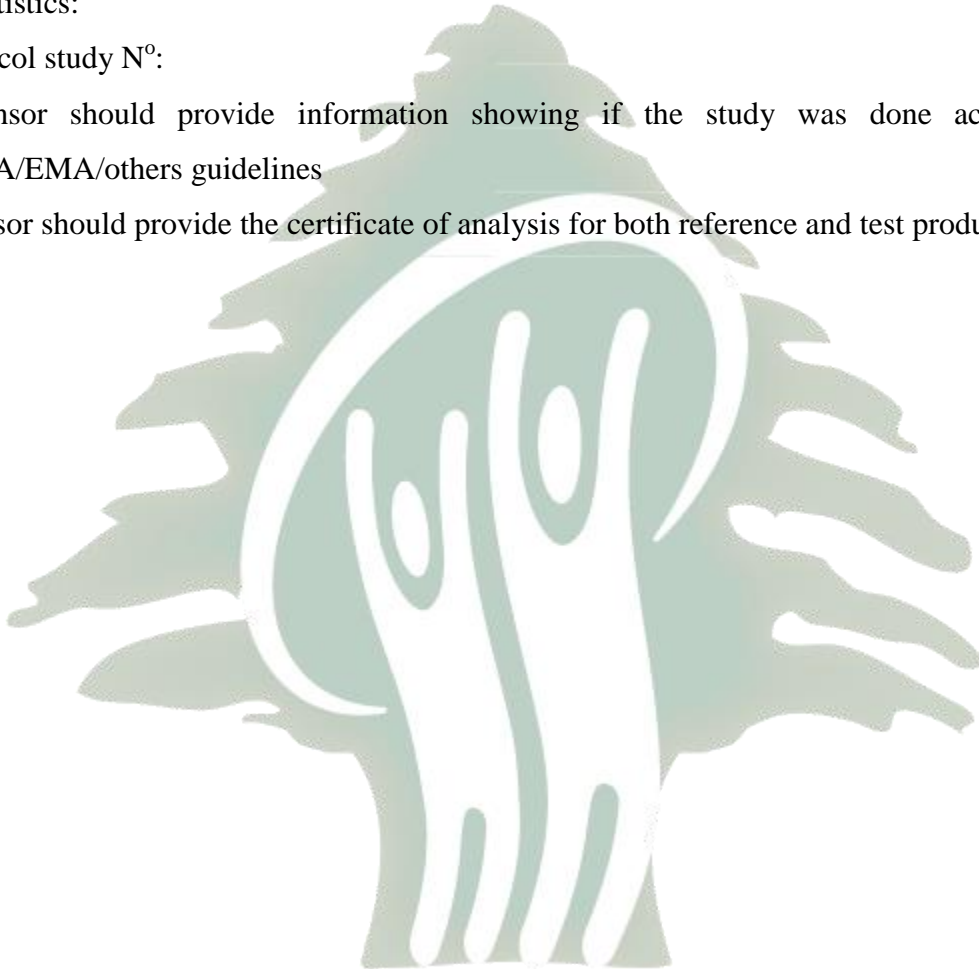
- 1- Any bad quality scanned document will be refused.
- 2- Correct scale for all individual plasma drug concentration *versus* time plots should be used and **no more than 2 plots per page** is allowed.
- 3- Plasma drug concentration *versus* time plots should be provided in both **linear and semi-logarithmic** scale.

Section I – Generalities

Sponsor should provide answers to all questions below and attach any required document:

1. Study Title:
2. Name of the active ingredient(s):
3. Manufacturer/Sponsor:
4. Therapeutic class:
5. Information about the Test Product
 - a. Brand Name:
 - b. Dosage:
 - c. Pharmaceutical form:
 - d. Manufacturer:
 - e. Batch number:
 - f. Manufacturing date:
 - g. Expiry date:
6. Information about the Reference Product
 - a. Brand Name:
 - b. Dosage:
 - c. Pharmaceutical form:
 - d. Manufacturer:
 - e. Batch number:
 - f. Manufacturing date:
 - g. Expiry date:
7. Provide evidence showing that it is a reference product according to FDA or EMEA lists.
8. Dose used in the BE study:
9. Information about the CRO
 - a. Name:
 - b. Country:
 - c. Address:
10. CRO study sites:
 - a. Clinical (hospital):

- b.** Clinical/medical Laboratory (for screening examination): should be certified by the authorities
 - c.** The Medical Laboratory (for screening examination) should meet GLPs and procedures as certified by an authoritative agency
 - d.** Analytical Facility:
 - e.** Pharmacokinetic studies:
 - f.** Statistics:
- 11.** Protocol study N°:
 - 12.** Sponsor should provide information showing if the study was done according to FDA/EMA/others guidelines
 - 13.** Sponsor should provide the certificate of analysis for both reference and test products.



Section II – Clinical Part

Sponsor should provide answers to all questions below and attach any required document:

1. Sponsor should provide a copy of the study protocol.
2. Sponsor should provide any amendment to the study protocol (if available).
3. Sponsor should include a copy of the informed consent form
Note: signed informed consent for all participants should be attached in BE report.
4. Sponsor should provide the IRB protocol approval.
Note: Sponsor should attach the IRB approval document (dated with all committee members signatures).
5. Official certificates of GCP and GLP compliances should be provided in the BE report.
Note: Date of certificates should fit the study period.
6. Quality assurance (QA) audits performed by the CRO with dates and signatures.
7. Official authorization (certificate) for laboratory where routine lab analysis had been done.
8. GMP (for the manufacturer).
9. **Sponsor should provide the following dates/period:**
 - a. Informed Consent was signed by applicants on:
 - b. Date of the screening examination:
 - c. Period I started on:
 - d. Period I ended on:
 - e. Period II started on:
 - f. Period II ended on:
 - g. First blood sample was taken on:
 - h. Last blood sample was taken on:
 - i. Study report released on:
10. Sponsor should provide the study duration:
 - a. For the clinical part:
 - b. For the bioanalytical part:
 - c. For the statistical part:
11. Wash out period:

Note: it should be at least 5 $t_{1/2}$ of the active ingredient(s).

12. Sponsor should indicate the drug half-life.
13. Sponsor should provide the sample size (number of subjects included and number of subjects who completed both periods of the study). Sponsor **should detail the method used to calculate the minimum number** (formula, and all numbers used in the formula) **of subjects** required to be enrolled in the study. Number of subjects who completed both periods should not be less than the minimum number of subjects calculated prior to beginning the study (as required above), or else the study will be rejected.
14. Number of blood sample per subject.
15. Sponsor should indicate the time period for each blood sample.
16. Sponsor should provide the total volume of blood drawn per subject and describe it.
17. Sponsor should provide the anticoagulant used in blood test tube.
18. Sponsor should indicate how samples were stored (at clinical site and analytical site) and transported to the analytical site. (i.e. samples were stored on ice/4 °C/-20 °C/-80 °C/etc ...)
19. All screening examination data and individual Case Report Form should be provided.
20. Sponsor should provide a table with all adverse events and discuss it.

Section III – *In vitro* Dissolution Profile

***In vitro* dissolution testing is an integral part of the assessment of bioequivalence.**

It should be performed on the same lot (or batch) numbers of both test and reference drugs used in the *in vivo* BE study.

Sponsor should provide answers to all questions below and attach any required document:

1. Dates (start and end of analysis)
2. Sponsor should provide dissolution data **as tables and figures** for each of the used medium.
3. Sponsor should provide the number of dissolution units used (for test and reference drugs).
The dissolution profiles can be compared only when number of **dissolution units used are equal to or greater than 12.**
4. Sponsor should also provide sampling time, method of assay of the active ingredient dissolved, limits used.
5. Sponsor should provide the composition of the medium used (3 media are required). For immediate release forms, the 3 media are **HCl 0.1N, Acetate buffer pH 4.5 and phosphate buffer pH 6.8. Any changes in the medium composition should be justified.**
6. For each medium used you have to provide the following:
 - a. Medium composition and pH:
 - b. Apparatus used:
 - c. Speed (rpm):
 - d. Temperature:
 - e. Volume used:
 - f. Duration:
 - g. Difference factor (f_1) =
 - h. Similarity factor (f_2) =

Notes:

- 1- Similarity factor of 50 – 100 ensures sameness of two products. i.e. f_2 value between 50 and 100 suggests that the two dissolution profiles are similar.
- 2- Difference factor of 0 – 15 ensures minor difference between two products. i.e. f_1 value between 0 and 15 is required.
- 3- For rapid dissolving products, that may dissolve 85% in 15 minutes, calculation of f_1 and f_2 is not required.



Section IV – Analytical Validation method

Reminder: Raw data should be saved on excel sheet and provided to MOPH on separated CD.

Sponsor should provide answers to all questions below and attach any required document:

1. Sample preparation and drug/metabolite extraction method used
2. The analytical method used to quantify the analyte.
3. Analyte (drug or metabolite) measured in plasma/urine.
4. Certificate of analysis for the reference standards used in the analysis
5. The internal standard used. **It's obligatory to use an internal standard.** The method rejected if no internal standard is included during samples preparation.
6. Provide information about the biological matrix used in the preparation of the standard curve.
7. Method used should be validated for the following parameters:

a. Linearity: provide the linearity zone, standard curve equation and R^2 . Only one range for the standard curve should be used for all sample calculations.

The range of the standard curve should cover the lowest quantifiable concentration and the highest concentration of the samples.

No sample could be used in the BE calculations if outside the linearity zone tested.

b. Recovery (extraction yield) for drug (metabolite) and for internal standard.

Note: The **recovery** of an analyte in an assay is the detector response obtained from an amount of the analyte added to and extracted from the biological matrix, compared to the detector response obtained for the true concentration of the pure authentic standard. Recovery pertains to the extraction efficiency of an analytical method within the limits of variability. **Recovery of the analyte need not be 100%**, but the extent of recovery of an analyte and of the internal standard should be consistent, precise, and reproducible. Recovery experiments should be performed by comparing the analytical results for extracted samples at three concentrations (low, medium, and high) with unextracted standards that represent 100% recovery.

c. Inter-day Accuracy

d. Intra-day Accuracy

Note: The **accuracy** of an analytical method describes the closeness of mean test results obtained by the method to the true value (concentration) of the analyte. Accuracy is determined by replicate analysis of samples containing known amounts of the analyte. Accuracy should be measured using a minimum of five determinations per concentration. A minimum of three concentrations in the range of expected concentrations is recommended. The mean value **should be within 15% of the actual value except at LLOQ, where it should not deviate by more than 20%**. The deviation of the mean from the true value serves as the measure of accuracy.

e. Inter-day Precision

f. Intra-day Precision

Note: The **precision** of an analytical method describes the closeness of individual measures of an analyte when the procedure is applied repeatedly to multiple aliquots of a single homogeneous volume of biological matrix. Precision should be measured using a minimum of five determinations per concentration. A minimum of three concentrations in the range of expected concentrations is recommended. The precision determined at each concentration level **should not exceed 15% of the coefficient of variation (CV) except for the LLOQ, where it should not exceed 20% of the CV**. Precision is further subdivided into within-run, intra-batch precision or repeatability, which assesses precision during a single analytical run, and between-run, interbatch precision or repeatability, which measures precision with time, and may involve different analysts, equipment, reagents, and laboratories.

g. Calculated concentrations of calibrate samples of the active ingredient in human plasma

h. Matrix effect

i. Anticoagulant (present in the sampling tubes) effect

j. Short term Stability of active ingredient and internal standard in **stock solutions** at different temperatures (such as room temperature, -4 °C, -20 °C).

k. Short term Stability of active ingredient and internal standard in **human plasma** at different temperatures (such as room temperature, -4 °C, -20 °C).

l. Long term stability of active ingredient and internal standard in **stock solutions** at different temperatures (such as room temperature, -4 °C, -20 °C). **This section should cover the period between period I and the end of analytical measurements.**

m. Long term stability of active ingredient and internal standard in **human plasma** at different temperatures (such as room temperature, -4 °C, -20 °C). **This section should cover the period between period I and the end of analytical measurements.**

n. Freeze/thaw stability of active ingredient.

Sponsor should provide stability for n cycles of freeze/thaw at specific temperature.

o. Specificity in the presence of different compounds added: testing for interference of common drugs is required..

p. Robustness.

q. Sensitivity: the lowest limit of detection (LLOD), the lowest limit of quantification (LLOQ)

Note: LLOQ is the lowest concentration of the standard curve that can be measured with acceptable accuracy and precision. The LLOQ should be established using at least five samples independent of standards and determining the coefficient of variation and/or appropriate confidence interval. The LLOQ should serve as the lowest concentration on the standard curve and **should not be confused with the limit of detection** and/or the low QC sample. The highest standard will define the upper limit of quantification (ULOQ) of an analytical method.

r. Concentration of the quality control samples used with their CV: minimum 3 concentrations:

r.1. Low QC

r.2. Medium QC (one or two)

r.3. High QC

8. A minimum of 20% of all subjects analytical spectrums should be provided in the BE report.

9. The materials, solvents and equipment used should be detailed. Method of preparation of the stock solutions, calibration standards and sample handling should be outlined in details.

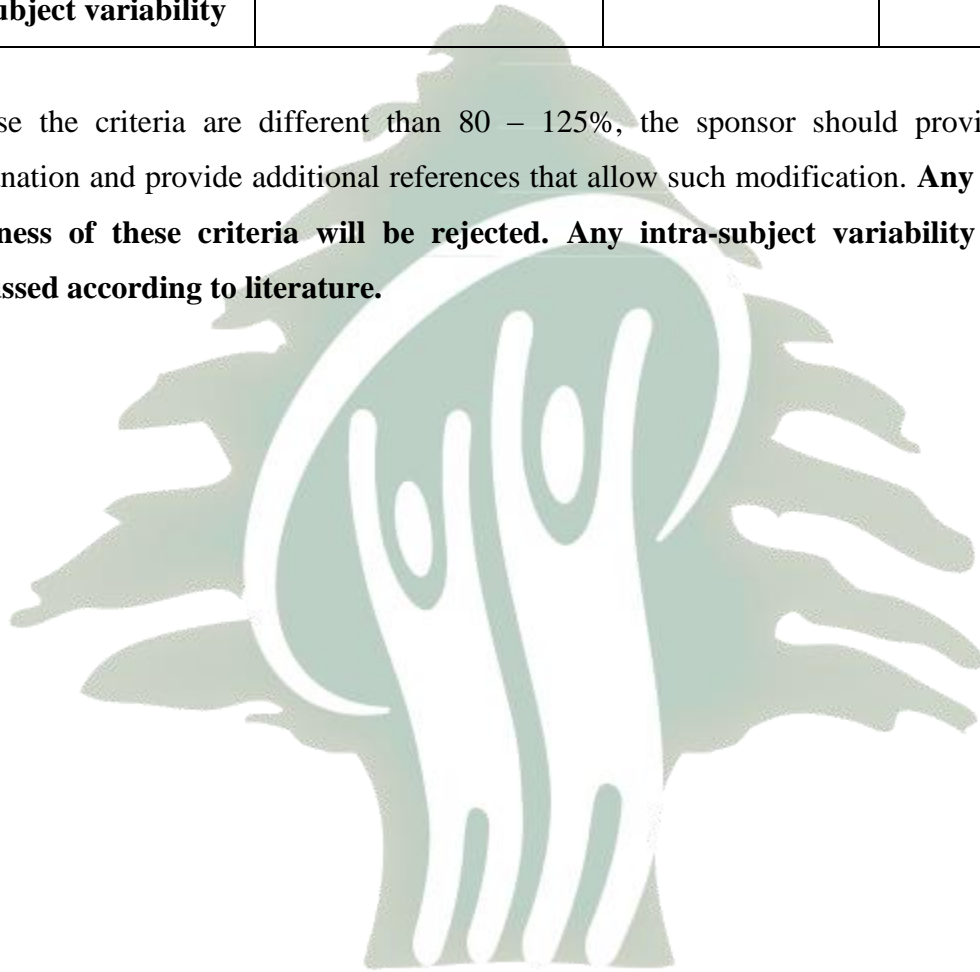
Section V – Pharmacokinetic Analysis Section

1. Sponsor should provide on excel sheet (on attached CD) all raw data related to plasma concentration for all subjects and at all time points. No scanned sheet is allowed.
2. Sponsor should provide the mean plasma concentration vs. time plot in both linear and semi-logarithmic scale (with SEM/SD error bars on each point).
3. Sponsor should provide individual plasma concentration vs. time plot in both linear and semi-logarithmic scale for all subjects.
4. Calculation of pharmacokinetic parameters: $AUC_{0 \rightarrow t}$, $AUC_{0 \rightarrow \infty}$, C_{max} , T_{max} , $t_{1/2}$, K_e , etc ...
 Non-compartmental methods should be used for determination of pharmacokinetic parameters in bioequivalence studies. The use of compartmental methods for the estimation of parameters is not acceptable.
 - a. Actual time of sampling should be used in the estimation of the pharmacokinetic parameters.
 - b. In studies to determine bioequivalence **after a single dose**, $AUC(0 \rightarrow t)$, $AUC(0 \rightarrow \infty)$, residual area, C_{max} and t_{max} should be determined.
 - c. **In studies with a sampling period of 72 h**, and where the concentration at 72 h is quantifiable, $AUC(0 \rightarrow \infty)$ and residual area do not need to be reported; it is sufficient to report AUC truncated at 72h, $AUC(0 \rightarrow 72h)$. In addition, C_{max} and T_{max} should be calculated.
 - d. Additional parameters that may be reported include the terminal rate constant, K_e , and $t_{1/2}$.
 - e. In studies to determine bioequivalence for immediate release formulations at steady state, $AUC(0- |), C_{max,ss}$, and $t_{max,ss}$ should be determined.
 - f. When using **urinary data**, $A_e(0 \rightarrow t)$ and, if applicable, R_{max} should be determined.
5. Sponsor should provide the software used to do the calculation and statistical analysis.

6. Sponsor should complete the following table for the Log-transformed Test/Reference ratios:

Parameters	Cmax	AUC _{0→t}	AUC _{0→∞}
Criteria	80 – 125 %	80 – 125 %	80 – 125 %
Mean			
90% Confidence Interval			
Intra-subject variability			

7. In case the criteria are different than 80 – 125%, the sponsor should provide detailed explanation and provide additional references that allow such modification. **Any unjustified wideness of these criteria will be rejected. Any intra-subject variability should be discussed according to literature.**



Section VI – Statistical Analysis Section

Analysis of variance (ANOVA) should be performed on the pharmacokinetic parameters AUCs, T_{max} and C_{max} . Appropriate statistical models pertaining to the design of the bioequivalence study should be employed. For example, for a conventional two-treatment, two-period, two-sequence (2 x 2) randomized crossover study design, the statistical model often includes factors accounting for the following sources of variation:

1. Sequence (Sometimes called Group or Order).
2. Subjects nested in sequences.
3. Period (or Phase)
4. Treatment (sometimes called Drug or Formulation).

The sequence effect should be tested using the [subject (sequence)] mean square from the ANOVA as error term. All other main effects should be tested against the residual error (error mean square) from the ANOVA. Assumption of the design or analysis should be addressed, and the possibility of differing variation in the formulations should be investigated. This covers investigation of period effects, sequence or carry-over effects, and homogeneity of variance. Outlying observations should be reviewed for their impact on the conclusion. Medical or pharmacokinetic explanations for such observations should be sought.

1. Sponsor should provide the software used to do the ANOVA analysis.
2. Sponsor should complete the table below with corresponding p values.

Source	p values		
	C_{max}	$AUC_{0 \rightarrow t}$	$AUC_{0 \rightarrow \infty}$
Period			
Subject (seq)			
Formulation			
Sequence			

3. Sponsor should provide explanation or additional tests in case any p-value in the table above is < 0.05 (Statistically significant). BE study will be rejected in absence of any explanation/justification.

