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DRAFT

GUIDELINE ON THE INVESTIGATION OF BIOEQUIVALENCE

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This guideline will replace the "Note for guidance on the investigation of bioavailability and bioequivalence" CPMP/EWP/QWP/1401/98 and the related questions in the Q&A document (EMEA/CHMP/EWP/40326/2006). This guideline includes recommendations on BCS-based biowaivers.

Comments should be provided to EWPSecretariat@emea.europa.eu using this template

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GUIDELINE ON THE INVESTIGATION OF BIOEQUIVALENCE

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EXECUTIVE SUMMARY

This guideline defines when bioequivalence studies are necessary and formulates requirements for their design, conduct, and evaluation. The guideline focuses primarily on bioequivalence for immediate release dosage forms with systemic action.

1. INTRODUCTION (background)

Two medicinal products containing the same active substance are considered bioequivalent if their bioavailabilities (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.

In bioequivalence studies, the plasma concentration time curve is used to assess the rate and extent of absorption. Meaningful pharmacokinetic parameters and preset acceptance limits allow the final decision on bioequivalence of the tested products. AUC, the area under the concentration time curve, reflects the extent of exposure. C_{max}, the maximum plasma concentration or peak exposure, and the time to maximum plasma concentration, t_{max}, are parameters that are influenced by absorption rate.

It is the objective of this guideline to define when bioequivalence studies are necessary and to formulate requirements for their design, conduct, and evaluation. The possibility of using in vitro instead of in vivo studies is also addressed.

The concept of bioequivalence forms the basis for approval of generic application, but it may also be applicable to hybrid application, extensions and variations applications, and to different formulations used during the development of a new medicinal product containing a new chemical entity.

For generic applications, the purpose of establishing bioequivalence is to demonstrate equivalence in biopharmaceutic quality between the generic product and a reference medicinal product in order to allow bridging of clinical data associated with the reference medicinal product. The current definition for generic products is found in Directive 2001/83/EC, Article 10(2)(b). In general, a generic product is a product which has the same qualitative and quantitative composition in active substances as the reference medicinal product, the same pharmaceutical form as the reference medicinal product, and whose bioequivalence with the reference medicinal product has been demonstrated by appropriate bioavailability studies. By definition it is considered that 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, various immediate-release oral pharmaceutical forms are considered to be one and the same pharmaceutical form. It is also stated in the Directive that bioavailability studies need not be required if it can be demonstrated that the generic medicinal product meets the relevant criteria for a biowaiver.

Hybrid applications rely on the results of preclinical tests and clinical trials of an approved reference medicinal product and include new data. These new data may include bioequivalence or comparative bioavailability data.

Also applications for extensions such as additional dosage forms, new strengths, new routes of administration often need support of bioequivalence in order to bridge data from the authorised reference medicinal product.

Variations for a change in composition or for significant manufacturing changes which may affect drug bioavailability may also require support of bioequivalence studies.

During development of a new chemical entity, the principles of bioequivalence may be applied in order to bridge data between different formulations e.g. between a formulation used in the pivotal clinical studies and the to-be-marketed formulation. In such situations however, wider acceptance limits may be acceptable if these are justified based on data provided with a complete application,

adequately addressing the clinical relevance of the widening from both a safety and efficacy perspective.

2. SCOPE

This guideline focuses on recommendations for bioequivalence studies for immediate release formulations with systemic action.

Specific recommendations regarding bioequivalence studies for modified release products, transdermal products and orally inhaled products are given in other guidelines (see section 3).

Recommendation for the comparison of biologicals to reference medicinal products can be found in guidelines on biosimilar products. Recommendations for pharmacokinetics of therapeutic proteins are also described in a specific guideline (CPMP/EWP/89249/04).

In case bioequivalence cannot be demonstrated using drug plasma concentrations, in exceptional circumstances pharmacodynamic or clinical endpoints may be needed. This situation is outside the scope of this guideline and the reader is referred to therapeutic area specific guidelines.

Furthermore, this guideline does not cover aspects related to generic substitution as this is subject to national legislation.

3. LEGAL BASIS

This guideline applies to Marketing Authorisation Applications for human medicinal products submitted in accordance with the Directive 2001/83/EC as amended, under Art. 8(3) (full applications), Art 10b (fixed combination), Art. 10 (1) (generic applications), Art 10(3) (hybrid applications), and also for line extension and variation applications in accordance with Commission Regulations (EC) No 1084/2003 and 1085/2003.

This guideline should be read in conjunction with the Annex I of Directive 2001/83/EC as amended, as well as European and ICH guidelines for conducting clinical trials, including those on:

- General Considerations for Clinical Trials (ICH topic E8, CPMP/ICH/291/95)
- Guideline for Good Clinical Practice (ICH E6 (R1), CPMP/ICH/135/95)
- Structure and Content of Clinical Study Reports (ICH E3, CPMP/ICH/137/95)
- CHMP guidance for users of the centralised procedure for generics/hybrid applications (EMEA/CHMP/225411/2006)
- Modified Release Oral and Transdermal Dosage Forms: Section II (CPMP/EWP/280/96)
- Requirements for clinical documentation for orally inhaled products (OIP) including the requirements for demonstration of therapeutic equivalence between two inhaled products for use in the treatment of Asthma and Chronic Obstructive Pulmonary Disease (COPD) (CPMP/EWP/4151/00 rev 1).
- Fixed Combination Medicinal Products (CPMP/EWP/240/95)
- Clinical Requirements for Locally Applied, Locally Acting Products containing Known Constituents (CPMP/EWP/239/95)
- Good manufacturing practice (Eudralex volume 4).

The guideline should also be read in conjunction with relevant guidelines on pharmaceutical quality. The test products used in the bioequivalence study must be prepared in accordance with GMP-regulations.

Bioequivalence trials should be conducted in accordance to Directive 2001/20/EC of the European parliament and of the Council.

Companies may also apply for CHMP Scientific Advice, via the EMEA, for specific queries not covered by existing guidelines.

4. MAIN GUIDELINE TEXT

4.1 Design, conduct and evaluation of bioequivalence studies

In the following sections, requirements for the design, conduct and evaluation of bioequivalence studies investigating immediate release formulations with systemic action are described.

The formulation and the characteristics of the active substance can affect the requirements for bioequivalence studies. When the test product contains a different salt, ester, ether, isomer, mixture of isomers, complex or derivative of an active substance than the reference product, bioequivalence should be demonstrated in appropriate bioavailability studies. However, when the active substance in test and reference products are identical or contain comparable salts, in vivo bioequivalence studies may, in some situations, not be required as described in APPENDIX II (bioequivalence study requirements) and III (biowaiver).

The pharmacokinetic and physico-chemical properties of the substance affect the number of studies needed and the design of the studies. The choice of number of studies and study design should be thoroughly justified based on the physico-chemical characteristics of the substance and its pharmacokinetic properties, discussing especially linearity in pharmacokinetics, activity of metabolites, contribution of metabolites to the effect, the need for enantioselective analysis, and solubility of the active substance. In the context of this guideline, high solubility and low solubility is defined according to the Biopharmaceutics Classification System (BCS) definition of high and low solubility, as defined in APPENDIX III.

The clinical overview of an application for marketing authorisation should list all studies carried out with the product applied for. All bioequivalence studies comparing the product applied for with the reference product of interest must be submitted.

4.1.1 Study design

The study should be designed in such a way that the formulation effect can be distinguished from other effects.

Standard design

If two formulations are going to be compared, a two-period, two-sequence single dose crossover design is the design of choice. The treatment periods should be separated by an adequate wash out period.

Alternative designs

In general, single dose studies will suffice. However, in case of dose or time-dependent pharmacokinetics, resulting in markedly higher concentrations at steady state than expected from single dose data, a potential difference in AUC between formulations may be larger at steady state than after single dose. Hence, a multiple dose study may be required in addition to the single dose study to ensure that the products are bioequivalent regarding AUC also at steady state. However, if the single dose study indicates very similar PK profile for test and reference (the 90% confidence interval for AUC is within 90-111), the requirement for steady-state data may be waived.

In certain cases when a single dose study cannot be conducted in healthy volunteers due to tolerability reasons, and a single dose study is not feasible in patients, conduct of a multiple dose study in patients may be acceptable (see also section 4.1.6 Strength and Dose).

A multiple dose study as an alternative to a single dose study may also be acceptable if problems of sensitivity of the analytical method preclude sufficiently precise plasma concentration measurements after single dose administration. As C_{max} at steady state may be less sensitive to differences in the absorption rate than C_{max} after single dose, bioequivalence should, if possible, be determined for C_{max} after the single dose administration (i.e. after the first dose of the multiple dose study) as a measure of

peak exposure while extent of exposure can be based on demonstration of bioequivalence of AUC at steady state.

In steady-state studies the administration scheme should preferably follow the highest usual dosage recommendation (see also section 4.1.6 Strength and dose).

Under certain circumstances, provided the study design and the statistical analyses are scientifically sound, alternative well-established designs could be considered such as parallel design for substances with very long half-life and replicate designs e.g. for substances with highly variable pharmacokinetic characteristics (see section 4.1.10).

4.1.2 Reference and test product

For Article 10(1) and 10(3) applications the chosen reference medicinal product must be a medicinal product authorised in the Community, on the basis of a complete dossier in accordance with the provisions of Article 8 of Directive 2001/83/EC, as amended. The product used as reference product in the bioequivalence study should be part of the global marketing authorisation of the reference medicinal product (as defined in Article 6(1) second subparagraph of Directive 2001/83/EC). The choice of the reference medicinal product should be justified by the applicant in Module 1.2, and Module 1, section 1.5.2.

Test products in an application for a generic product are normally compared with the corresponding dosage form of a reference medicinal product.

In an application for extension of a concerned medicinal product and when there are several dosage forms of this medicinal on the market, the dosage form used for the initial approval of the concerned medicinal product (and which was used in clinical efficacy and safety studies) should be used as comparative product, unless otherwise justified.

For variations of a concerned medicinal product, the comparative medicinal product for use in bioequivalence and dissolution studies is usually that authorised under the currently registered formulation, manufacturing process, packaging etc.

When variations to a generic product are made, the comparative medicinal product for the bioequivalence study should be the reference medicinal product.

The reference and test products should be packed in an individual way for each subject and period. Packaging, which is a manufacturing operation, should be performed and documented in accordance with good manufacturing practice, including Annex 13 to the EU guide to GMP. It should be possible to identify unequivocally the identity of the product administered to each subject at each trial period. Packaging and administration of the products to the subjects should therefore be documented in detail. This documentation should include all precautions taken to avoid and identify potential dosing mistakes.

Batch control results of the test and reference products should be reported. 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. In order to demonstrate that a representative batch of the reference product with regards to dissolution and assay content has been selected, the applicant should present dissolution profiles and content analysis of at least 3 batches of the reference product, unless otherwise justified.

The test product used in the study should be representative of the product to be marketed and this should be justified by the applicant. In the case of oral solid forms for systemic action 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. 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. If the product is subjected to further scale-up, this should be properly validated.

Samples of the product from full production batches should be compared with those of the test batch, and should show similar in vitro dissolution profiles when employing suitable dissolution test conditions (see Appendix I).

The study sponsor will have to retain a sufficient number of all investigational product samples in the study for one year in excess of the accepted shelf life or two years after completion of the trial or until approval whichever is longer to allow re-testing, if it is requested by the authorities.

4.1.3 Subjects

Number of subjects

The number of subjects to be included in the study should be based on an appropriate sample size calculation. The minimum number of subjects in a cross-over study should be 12.

Selection of subjects

The subject population for bioequivalence studies should be selected with the aim to permit detection of differences between pharmaceutical products. In order to reduce variability not related to differences between products, the studies should normally be performed in healthy volunteers unless the drug carries safety concerns that make this unethical. This model, *in vivo* healthy volunteers, is regarded adequate in most instances to detect formulation differences and the results will allow extrapolation to populations in which the reference product is approved (the elderly, children, patients with renal or liver impairment, etc.).

The inclusion/exclusion criteria should be clearly stated in the protocol. In general, subjects should preferably be between 18 - 55 years old and of weight within the normal range according to accepted normal values for the Body Mass Index. The subjects should be screened for suitability by means of clinical laboratory tests, an extensive review of medical history, and a comprehensive medical examination. Depending on the drug's therapeutic class and safety profile, special medical investigations and precautions may have to be carried out before, during and after the completion of the study. Subjects could belong to either sex; however, the risk to women of childbearing potential should be considered on an individual basis. Subjects should preferably be non-smokers and without a history of alcohol or drug abuse. If moderate smokers are included (less than 10 cigarettes per day) they should be identified as such and the consequences for the results should be discussed. Phenotyping and/or genotyping of subjects may be considered for safety or pharmacokinetic reasons.

In parallel design studies, the treatment groups should be comparable in all known prognostic variables that affect the pharmacokinetics of the active substance (e.g. ethnic origin, smoking status, extensive/poor metabolic status). This is an essential pre-requisite to give validity to the study results.

If the investigated active substance is known to have adverse effects and the pharmacological effects or risks are considered unacceptable for healthy volunteers, it may be necessary to use patients, under suitable precautions and supervision, instead. In such case the applicant should justify the alternative.

4.1.4 Study conduct

Standardisation

The test conditions should be standardised in order to minimise the variability of all factors involved except that of the products being tested. Therefore, it is recommended to standardise diet, fluid intake and exercise.

The time of day for ingestion should be specified. As fluid intake may influence gastric passage for oral administration forms, the test and reference products should be administered with a standardised

volume of fluid (at least 150 ml). All meals and fluids taken after the treatment should also be standardised in regard to composition and time of administration during the sampling period. As the bioavailability of an active moiety from a dosage form could be dependent upon gastrointestinal transit times and regional blood flows, posture and physical activity may need to be standardised.

The subjects should abstain from food and drinks, which may interact with circulatory, gastrointestinal, hepatic or renal function (e.g. alcoholic or xanthine-containing beverages or grapefruit juice) during a suitable period before and during the study.

Subjects should not take any other concomitant medication (including herbal remedies) for an appropriate interval before as well as during the study. In case concomitant medication is unavoidable and a subject is administered other drugs, for instance to treat adverse events like headache, the use must be reported (dose and time of administration) and possible effects on the study outcome must be addressed.

In case the study is to be performed under fasting conditions, subjects should fast during the night prior to administration of the products, unless otherwise justified.

Sampling times

A sufficient number of samples to adequately describe the complete plasma concentration-time profile should be collected. The sampling schedule should include frequent sampling around C_{max} to provide a reliable estimate of peak exposure. The sampling schedule should be planned to avoid C_{max} being the first point of a concentration time curve. When partial AUC is to be determined, frequent early sampling is recommended with preferably at least two quantifiable samples before expected t_{max}. The sampling schedule should also cover the plasma concentration time curve long enough to provide a reliable estimate of the extent of exposure which is achieved if AUC_t is at least 80% of AUC_∞. At least three to four samples are needed during the terminal log-linear phase in order to reliably estimate the terminal rate constant (which is needed for a reliable estimate of AUC_∞).

A sampling period longer than 72 h is not considered necessary for any immediate release formulation. Hence, for drugs with a long half-life, comparison of extent of exposure using truncated AUCs at 72 h is acceptable.

Fasting or fed conditions

The study should be conducted during fasting conditions unless the SPC recommends intake of the originator product only in the fed state. If the recommendation of food intake in the SPC is based on pharmacokinetic properties such as higher bioavailability, the bioequivalence study should be conducted in the fed state. Also if the recommendation of food intake is intended to decrease adverse events or to improve tolerability, it is recommended to conduct the bioequivalence study in fed state, although a bioequivalence study under fasting conditions could be acceptable if this has been adequately justified.

For products with enhanced release characteristics differing from conventional immediate release formulations (e.g. microemulsions or solid dispersions), bioequivalence studies performed under both fasted and fed conditions are required.

In cases where information is required in both the fed and fasted states, it is preferable to conduct a four-period single dose crossover design study (both products fed and fasted) rather than conducting two separate bioequivalence studies in fed and fasted state, respectively. In a four-period crossover design study, the food effect on test and reference product can be evaluated which is not the case when conducting two separate two-period, two-sequence single dose crossover design studies under fasting and fed conditions, respectively. In addition to the bioequivalence evaluation of test/reference in fasting and in fed state, the food effect can be presented for test and reference, i.e. the ratio food/fasting and 90% confidence interval for test and reference, respectively.

In studies performed under fed conditions, the composition of the meal should be according to recommendations in the SPC of the reference product. If no recommendation on the composition of

the meal is given in the reference product SPC, the meal should be a "standardized non high-fat meal" (about 650 kcal with about 30% of calories derived from fat). The composition of the meal should be described with regard to protein, carbohydrate and fat content (specified in grams, calories and relative caloric content (%)).

4.1.5 Characteristics to be investigated

Pharmacokinetic parameters

In studies to determine bioequivalence after a single dose, AUC_t, AUC_∞, C_{max} and t_{max} should be determined. Additional parameters that may be reported include the terminal rate constant, λ_z, and $t_{1/2}$.

For products where rapid absorption is of importance, partial AUCs can be used as a measure of early exposure. The partial area can in most cases be truncated at the population median of t_{max} values for the reference formulation. However, an alternative time point for truncating the partial AUC can be used when clinically relevant. The time point for truncating the partial AUC should be pre-specified and justified in the study protocol.

In studies to determine bioequivalence at steady state, AUC_τ, $C_{max,ss,}$ $C_{min,ss,}$ $t_{max,ss}$ and fluctuation should be determined.

Definitions of the pharmacokinetic parameters are given in section 6.

Additional parameters may be presented. The methods of estimating parameters should be specified. The use of compartmental methods for the estimation of parameters is not acceptable.

Parent compound or metabolites

Recommendations for measuring parent compound and metabolite(s) depend on the contribution of parent compound and metabolite(s), respectively, to activity as detailed below and in Appendix IV.

In principle, evaluation of bioequivalence should be based upon measured concentrations of the parent compound. The reason for this is that C_{max} of a parent compound is usually more sensitive to detect differences between formulations in absorption rate than C_{max} of a metabolite.

Also for inactive prodrugs, demonstration of bioequivalence for parent compound is the preferred option when the pharmacokinetics of pro-drug and active metabolite(s) is linear. In this situation, the active metabolite does not need to be measured. However, in case the pro-drug or active metabolites display non-linear pharmacokinetics (or it is difficult to conclude linear pharmacokinetics from available data), it is recommended to demonstrate bioequivalence for the main active metabolite. In such case, the parent compound does not need to be measured provided that it is inactive from efficacy and safety perspectives. Moreover, some pro-drugs may have low plasma concentrations, be quickly eliminated and have high variability, resulting in difficulties in demonstrating bioequivalence for parent compound in a reasonably sized bioequivalence study. In this situation it is acceptable to demonstrate bioequivalence for the main active metabolite without measurement of parent compound. Furthermore, in situations where the pro-drug exposure is low and exposure to active metabolite is very much higher, it is acceptable to demonstrate bioequivalence for the main active metabolite without measurement of parent compound.

The use of a metabolite as a surrogate for an active parent compound can only be considered if the applicant presents convincing arguments demonstrating that it is not possible to reliably measure the parent compound after single dose administration or at steady state. However, as C_{max} of the metabolite is usually less sensitive to differences in the absorption rate than C_{max} of the parent drug, bioequivalence should, if possible, be determined for C_{max} of the parent compound as a measure of peak exposure while extent of exposure can be based on demonstration of bioequivalence of AUC of metabolite. Furthermore, when using metabolite data as a substitute for parent drug concentrations, the applicant should present any available data supporting the view that the parent drug exposure will be

reflected by metabolite exposure and that the metabolite formation is not saturated at therapeutic doses.

In exceptional cases, bioequivalence of active metabolite(s) may need to be demonstrated in addition to parent drug. This is applicable if the metabolite has a major contribution to clinical efficacy of an active substance and metabolite concentrations may reflect differences in formulation which may not be detected in parent compound, such as drugs with linear pharmacokinetics for parent compound and where the active metabolite shows non-linear pharmacokinetics caused by significant saturation of formation and/or elimination.

When evaluating the significance of the contribution of an active metabolite to the clinical efficacy, available information on differences in AUC and pharmacodynamic activity between parent compound and metabolite should be taken into account. Depending on how pharmacodynamic activity has been determined, differences in protein binding between parent compound and metabolite may also need to be taken into account.

Enantiomers

The use of achiral bio-analytical methods is possible when it is demonstrated that both enantiomers show at least one of the following characteristics:

- the same pharmacokinetics,
- the same pharmacodynamics or
- the concentration ratio of enantiomers is not modified by a change in the rate of absorption.

If none of these characteristics is fulfilled or can be asserted with confidence, enantiomeric bio-analytical methods are required. If one enantiomer is pharmacologically active and the other is inactive or has a low contribution to activity, it is sufficient to demonstrate bioequivalence for the active enantiomer. If both enantiomers contribute significantly to activity, bioequivalence should be demonstrated for both enantiomers.

The use of achiral bio-analytical methods is also possible when both products contain the same single enantiomer and there is no inter-conversion in vivo.

The use of urinary data

The use of urinary excretion data as a surrogate for a plasma concentration may be acceptable in determining the extent of exposure in case it is not possible to reliably measure the plasma concentration-time profile of parent compound. However, the use of urinary data has to be carefully justified when used to estimate peak exposure. If a reliable plasma C_{max} can be determined, this should be combined with urinary data on the extent of exposure for assessing bioequivalence.

4.1.6 Strength and dose to be investigated

The strength(s) and dose(s) to evaluate depend on the linearity in pharmacokinetics of the active substance, its solubility, the proportionality in composition between the different strengths and other product related issues described below and in Appendix V.

If a test product constitutes several strengths, it is sufficient to establish bioequivalence with only one strength, provided that all of the below conditions are fulfilled.

a) the pharmaceutical products are manufactured at the same site by the same manufacturer and manufacturing process,
b) linear pharmacokinetics, i.e. proportional increase in AUC and C_{max} with increased dose, over the therapeutic dose range,
c) the qualitative composition of the different strengths is the same,
d) the composition of the strengths are quantitatively proportional, i.e. the ratio between the amount of each excipient to the amount of active substance(s) is the same for all strengths (for

immediate release products, coating components, colour agents and flavours are not required to follow this rule),

e) appropriate in vitro dissolution data should confirm the adequacy of waiving additional in vivo bioequivalence testing (see section 4.2).

If all above conditions are fulfilled, and the drug substance has a high solubility, the dose (and strength) to be tested may be selected based on safety and analytical grounds as the sensitivity to detect a potential difference between products is similar over the dose range.

However, in case of low solubility drug substances, the bioequivalence study should be conducted at the highest dose, using the highest strength, as these conditions are most sensitive to detect a potential difference between products.

For both high and low solubility drug substances, some deviations from condition d) may be accepted as detailed below, provided that the bioequivalence study has been conducted with the highest dose using the highest strength. If the below stated conditions are fulfilled, additional bioequivalence studies at lower strengths can be waived.

- in case the amount of the active substance(s) is less than 5 % of the tablet core weight or the weight of the capsule content and
- the amounts of the different core excipients or capsule content are the same for all strengths and only the amount of active substance is changed or
- the amount of a filler is changed to account for the change in amount of active substance. The amounts of other core excipients or capsule content should be the same for all strengths. However, for BCS class III compounds it should be reassured that the change in filler will not affect the solubility or absorption of the substance (see Appendix III, section IIIb Excipients).

The conditions should be fulfilled for all active substances of fixed dose combinations.

If all conditions except b) above are fulfilled, i.e. pharmacokinetics are non-linear over the therapeutic dose range, bioequivalence between test and reference formulations should be established at the strength(s) and dose(s) most sensitive to identify formulation related differences. Data on linearity in pharmacokinetics is sometimes limited or it may be difficult to conclude linear PK from the available data. If evidence of non-linearity is available or the available data suggest non-linear pharmacokinetics, the strength(s) and dose(s) to be used in the bioequivalence study(s) can be selected as follows:

- the highest dose (using the highest strength) for drugs with a demonstrated greater than proportional increase in AUC or C_{max} with increasing dose.
- the lowest strength (or a dose in the linear range) for drugs with a demonstrated less than proportional increase in AUC or C_{max} with increasing dose, e.g. if this phenomenon is due to saturable absorption. However, if this phenomenon is due to limited solubility of the active substance, bioequivalence should be established also with the highest dose (using the highest strength), i.e. in this situation two bioequivalence studies are needed.

If it cannot be determined which strength(s) and dose(s) are most sensitive to identify formulation related differences based on available data, it is recommended to establish bioequivalence at both the lowest dose using the lowest strength and the highest dose using the highest strength, if possible.

When the pharmacokinetics is non-linear and studies are warranted at the high dose range, they should preferably be performed at the highest commonly recommended dose. If this dose cannot be administered to volunteers, the study may need to be performed in patients. If the study is conducted at the highest acceptable dose in volunteers, the Applicant should justify this and discuss how bioequivalence determined at this dose can be extrapolated to the highest commonly recommended dose. Conduct of the bioequivalence study at a lower dose could be justified if data from this study indicate very similar PK profile for test and reference (the 90% confidence intervals are within 90-111) so that it is unlikely that there will be a risk for non-equivalence at the most sensitive dose.

4.1.7 Chemical analysis

The bioanalytical part of bioequivalence trials should be conducted according to the principles of Good Laboratory Practice (GLP). However, as such studies fall outside the formal scope of GLP, the sites conducting the studies are not required to be certified as part of the GLP compliance certification scheme.

The bioanalytical methods used must be well characterised, fully validated and documented to yield reliable results that can be satisfactorily interpreted. The main objective of method validation is to demonstrate the reliability of a particular method for the quantitative determination of an analyte(s) concentration in a specific biological matrix. The main characteristics of a bioanalytical method essential to ensure the acceptability of the performance and the reliability of analytical results are: (1) stability of the stock solutions and of the analyte(s) in the biological matrix under processing conditions and during the entire period of storage; (2) specificity; (3) accuracy; (4) precision (5) limit of quantification and (6) response function.

The validation of a bioanalytical method should comprise two distinct phases: (1) the pre-study phase in which the compliance of the assay with the characteristics listed above is verified and (2) the study phase itself in which the validated bioanalytical method is applied to the actual analysis of samples from the bioequivalence study in order to confirm the validity of the determinations.

Pre-study phase
As validation involves documenting that the performance of characteristics of the method are suitable and reliable for the intended analytical application, commercial kits need to be re-validated for their use in bioequivalence studies. Similarly, demonstration of stability based on literature data only is not acceptable. The Applicant should discuss the ability of the analytical method to distinguish between the analyte (e.g. parent) and other related substances (e.g. metabolites or co-medication during study phase) that may be formed after the drug administration but are not present in the spiked samples employed in the pre-study phase of the validation. The risk of back-conversion of a metabolite into the analyte during the successive steps of the analysis should also be addressed.

Study phase
A calibration curve should be generated for each analyte in each analytical run and it should be used to calculate the concentration of the analyte in the unknown samples in the run. A sufficient number of separately prepared Quality Control samples should be analysed with processed test samples at intervals based on the total number of samples. In addition, it is necessary to validate the method of processing and handling the biological samples.

The Applicant should discuss the number of samples (and percentage of total number of samples) that have been re-analyzed, the initial value, the reason for reanalysis, the values obtained in the reanalyses, the finally accepted value and a justification for the acceptance. Similarly, the Applicant should discuss the number of chromatograms (and percentage of total number of chromatograms) that have not been automatically integrated, the reason for a different method of integration, the value obtained with the automatic integration and the non-automatic integration and a justification for the acceptance of each individual chromatograms that has not been automatically integrated. Any other deviation of the analytical protocol should also be discussed in the Analytical Report.

All procedures should be performed according to pre-established Standard Operating Procedures (SOPs). All relevant procedures and formulae used to validate the bioanalytical method should be submitted and discussed. Any modification of the bioanalytical method before and during analysis of study specimens may require adequate revalidation; all modifications should be reported and the scope of revalidation justified.

4.1.8 Evaluation

The primary concern of bioequivalence assessment is to compare the bioavailability between a test and a reference product. Two products are considered bioequivalent if their bioavailabilities (rate and extent) after administration in the same molar dose lie within acceptable predefined limits.

The pharmacokinetic parameters should not be adjusted for differences in analysed content of the test and reference batch, i.e. content correction is not accepted, in the evaluation of bioequivalence studies included in applications for generic products.

Statistical analysis

The assessment of bioequivalence is based upon 90% confidence intervals for the ratio of the population geometric means (test/reference) for the parameters under consideration. This method is equivalent to two one-sided tests with the null hypothesis of bioinequivalence at the 5% significance level.

The pharmacokinetic parameters under consideration should be analysed using ANOVA (or equivalent parametric method). The data should be transformed prior to analysis using a logarithmic transformation. A confidence interval for the difference between formulations on the log-transformed scale is obtained from the ANOVA model. This confidence interval is then back-transformed to obtain the desired confidence interval for the ratio on the original scale. A non-parametric analysis is not acceptable.

The precise model to be used for the analysis should be pre-specified in the protocol. The statistical analysis should take into account sources of variation that can be reasonably assumed to have an effect on the response variable. For example, if a two-period, two-sequence crossover design has been used, the terms to be used in the ANOVA model are usually sequence, subject within sequence, period and formulation. The presentation of the findings of a bioequivalence trial should include a 2x2-table that presents for each sequence (in rows) and each period (in columns) means, standard deviations and number of observations for the observations in the respective period of a sequence. In addition, tests for difference and the respective confidence intervals for the treatment effect, the period effect, and the sequence effect should be reported for descriptive assessment. A test for carry-over should not be performed and no decisions regarding the analysis (e.g. analysis of the first period, only) should be made on the basis of such a test. The potential for carry-over can be directly addressed by examination of the pre-treatment plasma concentrations in period 2 (and beyond if applicable). If there are any subjects for whom the pre-dose concentration is greater than 5 percent of the C_{max} value for the subject in that period, the statistical analysis should be repeated with those subjects excluded. Results from both analyses should be presented, but the analysis with the subjects excluded should be considered as primary.

However, if the substance being studied is endogenous, the calculation of pharmacokinetic parameters should be performed using some form of baseline correction so that the calculated pharmacokinetic parameters refer to the additional concentrations provided by the treatment. The method for baseline correction should be pre-specified and justified in the study protocol. In this situation it cannot be directly assessed whether carry-over has occurred, so extra care should be taken to ensure that the washout period is of an adequate duration.

Evaluation of data from several bioequivalence studies

If the application contains some studies which demonstrate bioequivalence and others that do not, the documentation must be considered as a whole. The existence of a positive study does not mean that negative studies can be ignored. In this situation the interpretation of the overall documentation is not straightforward but there are three distinct situations which can be considered:

1. If after the failed trial or trials, some well justified modifications have been made to the product that address the deficiencies that were revealed, then a subsequent bioequivalence study can be assessed without reference to the previous results. A positive study in this situation is not downgraded by the previous negative results.

2. If the failed trial was ambiguous e.g. the confidence intervals were wide and were consistent with both possible bioequivalence and lack of bioequivalence, then a subsequent positive study can be convincing. This is because the new study does not contradict the previous study, but it provides additional information that allows us to be confident that the previous failure was because of lack of

information rather than lack of bioequivalence. It is not acceptable to pool together two ambiguous studies to reach a positive conclusion.

3. If the failed study(s) clearly shows that the test product is bioinequivalent with the reference, a subsequent positive trial will then be a contradictory finding. In this situation, additional study(s) will be needed until the evidence for bioequivalence clearly outweighs the evidence against, indicating that the failed study(s) were simply unlucky chance findings. It is not acceptable to pool together positive and negative studies in a meta-analysis.

Acceptance limits

In studies to determine bioequivalence after a single dose, the parameters to be analysed are AUC_t and C_{max}.

For these parameters the 90% confidence interval for the ratio of the test and reference products should be contained within the acceptance interval of 80-125%.

Confidence intervals should be presented to two decimal places. To be inside the acceptance interval the lower bound should be ≥ 80.00 and the upper bound should be ≤ 125.00.

For products where rapid absorption is of importance, equivalence between test and reference should be supported by demonstration of bioequivalence for partial AUC as a measure of early exposure. The same acceptance interval as for C_{max} applies to partial AUC.

For studies to determine bioequivalence at steady state AUC_τ, $C_{max,ss}$, and $C_{min,ss}$ should be analysed using the same acceptance interval as stated above.

In specific cases of products with a narrow therapeutic range, the acceptance interval may need to be tightened (see section 4.1.9). Moreover, for highly variable drugs the acceptance interval for C_{max} may in certain cases be widened (see section 4.1.10).

Two-stage design

It is acceptable to use a two-stage approach when attempting to demonstrate bioequivalence. An initial group of subjects can be treated and their data analysed. If bioequivalence has not been demonstrated an additional group can be recruited and the results from both groups combined in a final analysis. If this approach is taken appropriate steps must be taken to preserve the overall type I error of the experiment. The analysis of the first stage data should be treated as an interim analysis and both analyses conducted at adjusted significance levels (with the confidence intervals accordingly using an adjusted coverage probability which will be higher than 90%). The plan to use a two-stage approach must be prespecified in the protocol along with the adjusted significance levels to be used for each of the analyses.

Subject accountability

All treated subjects should be included in the statistical analysis, with the exception of subjects in a crossover trial who do not complete at least one period receiving each of the test and reference products (or who fail to complete the single period in a parallel group trial).

The data from all treated subjects should be treated equally. It is not acceptable to have a protocol which specifies that ‘spare’ subjects will be included in the analysis only if needed as replacements for other subjects who have been excluded.

Unbiased assessment of results from randomised studies requires that all subjects are observed and treated according to the same rules, rules that should be independent from treatment or outcome. In consequence, the decision to exclude a subject from the statistical analysis must be made before bioanalysis. Ideally all treated subjects should be included in the analysis provided that the necessary number of treatment periods has been completed. Exclusions can only be made based upon reasons that have been defined in the protocol. Acceptable reasons to exclude a subject are events such as

vomiting and diarrhoea which could render the plasma concentration-time profile unreliable. In exceptional cases, the use of concomitant medication can be a reason for excluding a subject. The search for such explanations must apply to all subjects in all groups. Exclusion of data can never be accepted on the basis of statistical analysis or for pharmacokinetic reasons alone, because it is impossible to distinguish the formulation effects from other effects affecting the pharmacokinetics.

Presentation of data

All individual subject data should be provided. These presentations should include available data from subjects who eventually dropped-out from the study. Drop-out and withdrawal of subjects should be fully documented.

All individual concentration data and pharmacokinetic parameters should be listed by formulation together with summary statistics such as geometric mean, median, arithmetic mean, standard deviation, coefficient of variation, minimum and maximum. Individual plasma concentration/time curves should be presented in linear/linear and log/linear scale.

For the pharmacokinetic parameters that were subject to statistical analysis, the point estimate and 90% confidence interval for the ratio of the test and reference products should be presented.

For single dose studies, the percentage of AUC_{∞} that is covered by AUC_t should be reported for each subject in each period if the observation period is shorter than 72 hours. Subjects should not be excluded from the analysis on the basis of this calculation, but if the percentage is less than 80% in more than 20% of the observations then the validity of the study could be questioned.

The report should be sufficiently detailed to enable the pharmacokinetics and the statistical analysis to be repeated, e.g. data on actual times of blood sampling, drug concentrations, the values of the pharmacokinetic parameters for each subject in each period and the randomisation scheme should be provided.

The analytical report should include a detailed description of the bioanalytical method used, a detailed pre-study validation report and a detailed description of the in study validation results including the results for all standard and quality control samples. A representative number, of chromatograms or other raw data (e.g. for the first 5 subjects) should be included covering the whole concentration range for all standard and quality control samples as well as the specimens analysed. Any manual integration of chromatograms should be justified and listed together with values from the automatic integration.

4.1.9 Narrow therapeutic index drugs

In specific cases of products with a narrow therapeutic index, the acceptance interval may need to be tightened. For the purpose of bioequivalence requirements, narrow therapeutic index drugs (NTIDs) may be considered to be those for which there is a risk of clinically relevant difference in efficacy or safety between two products even when the conventional criteria for bioequivalence (i.e. 90% confidence interval for test / reference ratio for AUC and Cmax within 80-125%) are met. NTIDs often have steep concentration response relationships for efficacy, toxicity, or both. Dosing generally needs to be individualised based on plasma concentration monitoring or titrated according to clinical response and there may be a potential for serious clinical consequences in the event of too low or high concentrations. It is not possible to define a set of criteria to categorise drugs as either NTIDs or not and a judgement must be made in each individual case. Likewise, the need for narrowing the acceptance interval for both AUC and C_{max} or for AUC only should be determined on a case by case basis.

In cases where the acceptance interval needs to be tightened, the acceptance interval for concluding bioequivalence should generally be narrowed to 90-111%. In individual cases alternative or additional requirements might be set.

4.1.10 Highly variable drugs or drug products

In certain cases, C_{max} is of less importance for clinical efficacy and safety compared with AUC. When this is applicable, the acceptance criteria for C_{max} can be widened to 75-133% provided that all of the following are fulfilled:

- the widening has been prospectively defined in the study protocol
- it has been prospectively justified that widening of the acceptance criteria for C_{max} does not affect clinical efficacy or safety
- the bioequivalence study is of a replicate design where it has been demonstrated that the within-subject variability for C_{max} of the reference compound in the study is >30%.

This approach does not apply to AUC.

It is acceptable to apply either a 3-period or a 4-periodcrossover scheme in the replicate design study.

4.2 In-vitro dissolution tests

4.2.1 In-vitro dissolution tests complementary to bioequivalence studies

The results of *in vitro* dissolution tests at least at pH 1.2, 4.5, 6.8 and the media intended for drug product release (QC media), obtained with the batches of test and reference products that were used in the bioequivalence study should be reported. The results should be reported as profiles of percent of labelled amount dissolved versus time.

Unless otherwise justified, the specifications for the in vitro dissolution to be used for quality control of the product should be derived from the dissolution profile of the test product batch that was found to be bioequivalent to the reference product, which would be expected to be similar to those of the reference product (see Appendix I). In this way biorelevance of the chosen in vitro dissolution method may be demonstrated.

4.2.2 In-vitro dissolution tests in support of biowaiver of strengths

Appropriate in vitro dissolution should confirm the adequacy of waiving additional in vivo bioequivalence testing. Accordingly, dissolution should be investigated at different pH values as outlined in the previous section unless otherwise justified. Particular dosage forms may require investigations using different experimental conditions. Similarity of in vitro dissolution should be demonstrated at all conditions

- within the applied product series, i.e. between additional strengths and the strength(s) used for bioequivalence testing, and
- between additional strengths of the applied product and corresponding strengths of the reference product.

At pH values where sink conditions may not be achievable for all strengths in vitro dissolution may differ between different strengths. However, the comparison with the reference medicinal product should then confirm that this finding is drug substance rather than formulation related. In addition, the applicant could show similar profiles at the same dose (e.g. two tablets of 5 mg versus one tablet of 10 mg).

4.3 Variations

If a product has been reformulated from the formulation initially approved or the manufacturing method has been modified by the manufacturer in ways that could be considered to impact on the bioavailability, a bioequivalence study is required, unless otherwise justified. Any justification presented should be based upon general considerations, e.g. as per APPENDIX III, or on whether an acceptable in vivo / in vitro correlation has been established.

In cases where the bioavailability of the product undergoing change has been investigated and an acceptable correlation between in vivo performance and in vitro dissolution has been established, the requirements for in vivo demonstration of bioequivalence can be waived if the dissolution rate in vitro of the new product is similar to that of the already approved medicinal product under the same test conditions as used to establish the correlation (see APPENDIX I). In all other cases bioequivalence studies have to be performed.

As stated in section 4.1.2 *Reference and test product,* the comparative medicinal product for use in bioequivalence and dissolution studies in support of a variation of a concerned medicinal product is usually that authorised under the currently registered formulation, manufacturing process, packaging etc.

When variations to a generic product are made, the comparative medicinal product for the bioequivalence study should be the reference medicinal product.

4.4 Study report

The report of bioequivalence study should be written in accordance with the ICH E3 guideline. The authenticity of the whole of the report should be attested by the signature of the principal investigator in accordance with Annex I of the Directive 2001/83/EC as amended.

Names and affiliations of the responsible investigator(s), the site of the study and the period of its execution should be stated. Audits certificate(s), if available, should be included in the report.

The study report should include evidence that the choice of the reference medicinal product is in accordance with Article 10(1) and Article 10(2) of Directive 2001/83/EC as amended. This should include the reference product name, strength, pharmaceutical form, batch number, manufacturer, evidence of purchase including date and place of purchase and vendor.

Certificates of analysis of batches used in the study, including batch size of the test product, should be submitted and comparative dissolution profiles should be provided. The manufacturing date and, if possible, the expiry date of the test product and the expiry date of the reference product should be stated. In addition, the applicant should submit a signed statement confirming that the test product has the same quantitative composition and is manufactured by the same process as the one submitted for authorisation.

Concentrations and pharmacokinetic data and statistical analyses should be presented in the level of detail described above (section 4.1.8 Evaluation *Presentation of data).*

All individual data (concentrations, pharmacokinetic parameters, randomisation scheme etc.) should be available in electronic format (e.g. as comma separated and space delimited text files or Excel format) to be provided upon request.

DEFINITIONS

C_{max}: maximum plasma concentration;

$C_{max,ss}$: maximum plasma concentration at steady state;

C_{min}: minimum plasma concentration;

$C_{min,ss}$: minimum plasma concentration at steady state;

t_{max}: time until C_{max} is reached;

$t_{max,ss}$: time until $C_{max,ss}$ is reached;

AUC_t: area under the plasma concentration curve from administration to last observed concentration at time t;

AUC_{∞}:	area under the plasma concentration curve extrapolated to infinite time;
AUC_{τ}:	AUC during a dosage interval at steady state;
Partial AUC:	AUC truncated at the population median of t_{max} values for the reference formulation;
$t_{1/2}$:	plasma concentration half-life;
λ_z:	terminal rate constant;
C_{av}:	average steady state concentration ($AUC\tau/\tau$);
Fluctuation:	$(C_{max}-C_{min})/C_{av}$;
SPC:	Summary of Product Characteristics.

APPENDIX I

Dissolution testing

During the development of a medicinal product a dissolution test is used as a tool to identify formulation factors that are influencing and may have a crucial effect on the bioavailability of the drug. As soon as the composition and the manufacturing process are defined a dissolution test is used in the quality control of scale-up and of production batches to ensure both batch-to-batch consistency and that the dissolution profiles remain similar to those of pivotal clinical trial batches. Furthermore, in certain instances a dissolution test can be used to demonstrate bioequivalence. Therefore, dissolution studies can serve several purposes:

i – Testing on product quality

- To get information on the test batches used in bioavailability/bioequivalence studies and pivotal clinical studies to support specifications for quality control.
- To be used as a tool in quality control to demonstrate consistency in manufacture
- To get information on the reference product used in bioavailability/bioequivalence studies and pivotal clinical studies

ii - Bioequivalence surrogate inference

- To support the assumption of similarity between reference products from different Member States provided that the manufacturing process, composition and specifications are similar.
- To demonstrate in certain cases similarity between different formulations of an active substance and the reference medicinal product (biowaivers e.g., variations, formulation changes during development and generic products)
- To investigate batch to batch consistency of the products (test and reference) to be used as basis for the selection of appropriate batches for the in vivo study

The test methodology should be in accordance with pharmacopoeial requirements unless those requirements are shown to be unsatisfactory and/or do not reflect the in-vivo dissolution (i.e. biorelevance). Alternative methods can be considered when justified that these are discriminatory and able to differentiate between batches with acceptable and non-acceptable performance of the product in-vivo.

The recommendations as briefly outlined in the following should be noted as being basic regarding the development of meaningful in vitro dissolution methods. However, current state-of-the-art information must always be considered. If an active substance is considered highly soluble, it is reasonable to expect that it will not cause any bioavailability problems if, in addition, the dosage system is rapidly dissolved in the physiological pH-interval expected after product administration and the excipients are known not to affect the dissolution, stability and absorption processes. A bioequivalence study may in those situations be waived based on similarity of dissolution profiles which are based on discriminatory testing, provided that the other exemption criteria in Appendix III are met. The similarity should be justified by dissolution profiles, covering at least three time points, attained at three different buffers (normally pH 1.2, 4.5 and 6.8).

If an active substance is considered to have a low solubility, the rate limiting step for absorption may be dosage form dissolution. This is also the case when one or more of the excipients are controlling the release and subsequent dissolution step of the active substance. In those cases a variety of test conditions is recommended and adequate sampling should be performed until either 90% of the drug is dissolved or an asymptote is reached. Knowledge of dissolution properties under different conditions e.g. pH, agitation, ionic strength, surfactants, viscosity, osmotic pressure is important since the behaviour of the solid system in-vivo may be critical for the drug dissolution independent of the physico-chemical properties of the active substance. An appropriate experimental statistical design may be used to investigate the critical parameters and for the optimisation of such conditions.

The similarity may be compared by model-independent or model-dependent methods e.g. by statistical multivariate comparison of the parameters of the Weibull function or the percentage dissolved at different time points, or by calculating a similarity factor e.g. the f_2 similarity factor defined below. Alternative methods to prove similarity of dissolution profiles are accepted as long as they are justified:

$$f_2 = 50 \cdot \log\left[\frac{100}{\sqrt{1 + \frac{\sum_{t=1}^{t=n}\left[\overline{R}(t) - \overline{T}(t)\right]^2}{n}}}\right]$$

In this equation f_2 is the similarity factor, n is the number of time points, R (t) is the mean percent drug dissolved of e.g. a reference product, and T(t) is the mean percent drug dissolved of e.g. a test product.

The evaluation of the similarity factor is based on the following conditions:

- A minimum of three time points (zero excluded)
- The time points should be the same for the two formulations
- Twelve individual values for every time point for each formulation
- Not more than one mean value of > 85% dissolved for any of the formulations
- The relative standard deviation or coefficient of variation of any product should be less than 20% for the first point and less than 10% from second to last time point.

An f_2 value between 50 and 100 suggests that the two dissolution profiles are similar. In cases where more than 85% of the drug is dissolved within 15 minutes, dissolution profiles may be accepted as similar without further mathematical evaluation, except in the case of gastro-resistant formulations where the dissolution takes place in the intestine and the 15 minutes for gastric-emptying lacks of physiological meaning.

For immediate release dosage form comparison a sample at 15 min is essential to know if complete dissolution is reached before gastric emptying, i.e. a mathematical calculation is not necessary. In case more than 85% is not dissolved at 15 minutes but within 30 min, at least three time points are required: the first time point before 15 minutes, the second one at 15 minutes and the third time point when the release is close to 85%. For gastro-resistant formulations frequent sampling (e.g. every 5 minutes) is required during the rapid dissolution phase.

In general five to eight sampling times within a 0-60 minutes interval are recommended to achieve meaningful dissolution profiles.

APPENDIX II

Bioequivalence study requirements for different dosage forms

Depending on the type of formulation, there are different requirements regarding support of data from bioequivalence studies as described below.

As stated in section 4.1, when the test product contains a different salt, ester, ether, isomer, mixture of isomers, complex or derivative of an active substance than the reference product, bioequivalence should be demonstrated in appropriate bioavailability studies. However, when active substance in test and reference products are identical or contain comparable salts, in vivo bioequivalence studies may in some situations not be required as described below.

Oral immediate release dosage forms with systemic action

This section pertains to dosage forms such as tablets, capsules and oral suspensions. For these formulations, bioequivalence studies are required unless a biowaiver is applicable (see APPENDIX III). For orodispersable tablets specific recommendations, as detailed below, apply.

Orodispersible tablets

An orodispersable tablet (ODT) is formulated to quickly disperse in the mouth. Placement in the mouth and time of contact may be critical in cases where the active substance also is dissolved in the mouth and can be absorbed directly via the buccal mucosa. Depending on the formulation swallowing of the e.g. coated substance and subsequent absorption from the gastrointestinal tract also will occur.

If the ODT test product is an extension to another oral formulation, the requirements for bioequivalence studies depend on the SPC-claims for the orodispersible tablet. A 3-period study may be required in order to evaluate administration of the orodispersible tablet both with and without concomitant fluid intake.

If the ODT is a generic to an approved ODT reference product, the following recommendations regarding study design applies:

- if the reference product can be taken with and without water, bioequivalence should be demonstrated without water as this condition best resembles the intended use of the formulation. This is especially important if the substance may be dissolved and partly absorbed in the oral cavity. If bioequivalence is demonstrated when taken without water, bioequivalence when taken with water can be assumed.
- if the reference product is taken only in one way (e.g. only with water), BE should be shown in this condition (in a conventional two-way crossover design).
- if the reference product is taken only in one way (e.g. only with water), and the generic applies for additional ways of administration (e.g. without water), the conventional and the new method should be compared with the reference in the conventional way of administration (3 treatment, 3 period, 6 sequence design)

In studies evaluating ODT without water, it is recommended to wet the mouth by swallowing 20 ml of water directly before applying the ODT on the tongue. It is recommended not to allow fluid intake earlier than 2 hours after administration.

Non-oral immediate release dosage forms with systemic action

This section applies to e.g. rectal formulations. In general, bioequivalence studies are required. A biowaiver can be considered in the case of a solution with the same qualitative and similar quantitative composition in active substance and excipients.

Oral solutions

If the test product is an aqueous oral solution at time of administration and contains an active substance in the same concentration as an approved oral solution, bioequivalence studies may be waived, if the excipients contained in it do not affect gastrointestinal transit (e.g. sorbitol, mannitol, etc.), absorption (e.g. surfactants or excipients that may affect transport proteins), solubility (e.g. co-solvents) or in-vivo stability of the active substance. Any differences in the amount of excipients should be justified either by reference to other data or by a bioequivalence study. The same requirements for similarity in excipients apply for oral solutions as for Biowaivers (see Appendix III, Section IVb Excipients).

In those cases where the test product is an oral solution which is intended to be bioequivalent to another immediate release oral formulation, bioequivalence studies are required.

Modified release and transdermal dosage forms

Bioequivalence studies are required in accordance with the guideline on Modified Release Oral and Transdermal Dosage Forms: Section II (Pharmacokinetic and Clinical Evaluation) (CPMP/EWP/280/96).

Fixed combinations dosage forms

Bioequivalence studies are required unless a biowaiver is applicable (see APPENDIX III).

Bioequivalence should be established for all individual active substances. Biowaiver for an additional strength may be applicable when the conditions detailed in section 4.1.6 are fulfilled for all individual active substances.

For generic fixed dose combinations, the reference product in the bioequivalence study should be the originator fixed combination product.

Parenteral solutions

Bioequivalence studies are not required if the test product is to be administered as an aqueous intravenous solution containing the same active substance as the currently approved product. Moreover, the excipients, pH and osmolality have to be the same or, at least, comparable and should not interact with the drug substance (e.g. complex formation).

In the case of other parenteral routes, e.g. intramuscular or subcutaneous, and the test product is of the same type of solution (aqueous or oily), contains the same concentration of the same active substance and the same excipients in similar amounts as the medicinal product currently approved, bioequivalence studies are not required.

Gases

If the product is a gas for inhalation, bioequivalence studies are not required.

Locally acting locally applied products

For products for local use (after oral, nasal, inhalation, ocular, dermal, rectal, vaginal etc. administration) intended to act without systemic absorption, the approach to determine bioequivalence based on systemic measurements is in general not applicable and pharmacodynamic or comparative clinical studies are in principle required (see specific Note for Guidance). In the case of solutions for topical use, e.g. eye drops or cutaneous solutions, and if the test product is of the same type of solution (aqueous or oily), contains the same concentration of the same active substance and the same excipients in the same amounts as the medicinal product currently approved, a biowaiver is acceptable. In certain cases quantitative differences in excipients may be acceptable for these products, if adequately justified.

If the extent of absorption and the bioanalytical method are such that a pharmacokinetic approach is reliable, then a bioequivalence study might provide the best data for the approval of a locally applied/locally acting generic medicinal product.

Whenever systemic exposure resulting from locally applied, locally acting medicinal products entails a risk of systemic adverse reactions, systemic exposure should be measured. It should be demonstrated that the systemic exposure is not higher for the test product than for the reference product, i.e. the upper limit of the 90% confidence interval should not exceed the upper bioequivalence acceptance limit.

APPENDIX III

BCS-based Biowaiver

I. Introduction

The BCS (Biopharmaceutics Classification System)-based biowaiver approach is meant to reduce *in vivo* bioequivalence studies, ***i.e.***, it may represent a surrogate for *in vivo* bioequivalence. *In vivo* bioequivalence studies may be exempted if the equivalence in the *in vivo* performance can be justified by satisfactory *in vitro* data. Provided certain prerequisites are fulfilled as outlined in this document comparative *in vitro* dissolution could be even more discriminative than *in vivo* studies.

Applying for a BCS-based biowaiver is restricted to highly soluble drug substances with known human absorption and considered non-critical in terms of therapeutic range. Hence, those drugs for which tighter acceptance ranges of 90 – 111 % would apply in *in vivo* bioequivalence studies are not eligible for the BCS-based biowaiver approach. Furthermore the concept is applicable to pharmaceutically equivalent immediate release, solid pharmaceutical forms for oral administration and systemic action. However, it is not applicable for sublingual, buccal, orodispersible, and modified release formulations.

BCS-based biowaiver are intended only to address the question of bioequivalence between a test and a reference product. Hence, respective investigations may be useful to prove bioequivalence between early clinical trial products and to-be-marketed products, generics and innovator products, and in the case of variations that require bioequivalence testing.

II. Summary Requirements

BCS-based biowaiver are applicable for an immediate release drug product if

- the drug substance has been proven to exhibit high solubility and complete absorption (BCS-class I; for details see section III) and
- very rapid (> 85 % within 15 min) *in vitro* dissolution characteristics of the test and reference product have been demonstrated considering specific requirements (see section IV.1) and
- excipients are not suspect of having any relevant impact on bioavailability (see section IV.2).

BCS-based biowaiver are also applicable for an immediate release drug product if

- the drug substance has been proven to exhibit high solubility and limited absorption (BCS-class III; for details see section III) and
- very rapid (> 85 % within 15 min) *in vitro* dissolution of the test and reference product has been demonstrated considering specific requirements (see section IV.1) and
- excipients are qualitatively the same and quantitatively very similar (see section IV.2).

Generally the risks of an inappropriate biowaiver decision should be more critically reviewed (e.g. site-specific absorption, risk for transport protein interactions at the absorption site, excipient composition and therapeutic risks) for products containing BCS class III than for BCS class I drug substances.

III. Drug Substance

Generally, sound peer-reviewed literature may be acceptable for known compounds to describe drug substance characteristics particularly required in this biowaiver concept.

Biowaiver may be applicable when the active substances in test and reference products are identical or belong both to the BCS-class I (high solubility and complete absorption; see sections *III.1* and *III.2*) in case of different salts. However, biowaiver may not be applicable when the test product contains a

different ester, ether, isomer, mixture of isomers, complex or derivative of an active substance than the reference product since these differences are likely to lead to different bioavailabilities not deducible by means of experiments used in the BCS-based biowaiver concept.

The drug substance should not belong to the group of 'narrow therapeutic range' drugs (see section 4.1.9 on narrow therapeutic index drugs)

III.1 Solubility

The pH-solubility profile of the drug substance should be determined and discussed. The drug substance is considered highly soluble if the highest single dose administered as immediate release formulation(s) is completely dissolved in 250 ml of buffers within the range of pH 1 – 6.8 at 37±1 °C. This demonstration requires the investigation in at least three buffers within this range (preferably at pH 1.2, 4.5 and 6.8) and in addition at the pKa, if it is within the specified pH range. A minimum of three replicate determinations at each pH condition is recommended (e.g. shake-flask method or other justified method). Solution pH should be verified prior and after addition of the drug substance to a buffer.

III.2 Absorption

Complete absorption (i.e., extent of absorption ≥ 85 %) in humans is preferred for BCS-based biowaiver applications. Complete absorption is generally related to high permeability.

Complete drug absorption should be justified based on reliable investigations in human. Data from

- absolute bioavailability or
- mass-balance

studies could be used to support this claim.

The data should be obtained at the highest therapeutic dose in case of nonlinear PK. However, in case of linear PK data from lower doses are acceptable.

Data from mass balance studies support complete absorption if the sum of urinary recovery of parent compound, Phase 1 oxidative, and Phase 2 conjugative drug metabolites account for ≥ 85 % of the dose. It has also been demonstrated that high Phase 1 (oxidative) and Phase 2 (conjugative) metabolism would support the evaluation of complete absorption if the recovery in urine and faeces account for > 85 % of the dose.

In addition highly soluble drug substances with incomplete absorption, i.e. BCS-class 3 compounds, could be eligible for a biowaiver provided certain prerequisites are fulfilled regarding product composition and *in vitro* dissolution (see also sect. *IV.2* Excipients). The more restrictive requirements will also apply in cases where complete absorption could not convincingly be demonstrated.

Reported bioequivalence between aqueous and solid formulations of a particular compound administered via the oral route may be supportive as it indicates that absorption limitations due to (immediate release) formulation characteristics may be considered negligible. Well performed *in vitro* permeability investigations including a reference standard may also be considered supportive to *in vivo* data.

IV. Drug Product

IV.1 In vitro Dissolution

IV.1.1 General aspects

Investigations related to the drug product should ensure immediate release properties and prove similarity between the investigative products, i.e. test and reference have a similar *in vitro* dissolution considering physiologically relevant experimental pH conditions. However, respective results are not an acceptable way to establish an *in vitro*/*in vivo* correlation. The pH conditions to be employed are at

least pH 1.2, 4.5, and 6.8. Additional investigations may be required at pH values in which the drug substance has minimum solubility. The use of any surfactant is strictly discouraged.

Test and reference products should meet requirements as outlined in the EU guidance on bioavailability and bioequivalence. It is advisable to investigate more than one single batch of the test and reference products in order to ensure that respective results are representative.

Comparative *in vitro* dissolution experiments should follow current compendial standards. Hence, thorough description of experimental settings and analytical methods including validation data should be provided. It is recommended to use 12 units of the product for each experiment to enable statistical evaluation. Usual experimental conditions are e.g.:

- Apparatus: paddle/basket
- Volume of dissolution medium: 500 ml
- Temperature of the dissolution medium: 37±1 °C
- Agitation: paddle apparatus - usually 50 rpm
  basket apparatus - usually 100 rpm
- Sampling schedule: e.g. 10, 15, 20, 30 and 45 min
- Buffer: pH 1.2 (0.1 N HCl or SGF without enzymes), pH 4.5, and pH 6.8 (or SIF without enzymes); (pH should be ensured throughout the experiment; Ph.Eur. buffers recommended)
- Other conditions: no surfactant; in case of gelatin capsules or tablets with gelatin coatings the use of enzymes may be acceptable.

Complete documentation of *in vitro* dissolution experiments is required including a study protocol, batch information on test and reference batches, detailed experimental conditions, validation of experimental methods, individual and mean results and respective summary statistics.

IV.1.2 Evaluation of *in vitro* dissolution results

Drug products are considered 'very rapidly' dissolving when more than 85 % of the labelled amount is dissolved within 15 min. In cases where this is ensured for the test and reference product in all requested media the similarity of dissolution profiles may be accepted as demonstrated without any mathematical calculation. Discussion of dissolution profile differences in terms of their clinical/therapeutical relevance is considered inappropriate since the investigations do not reflect any in vitro/in vivo correlation.

IV.2 Excipients

Although the impact of excipients in immediate release dosage forms on bioavailability of highly soluble and completely absorbable drug substances (i.e., BCS-class I) is considered rather unlikely it can not be completely excluded. Therefore, even in the case of class I drugs it is advisable to use similar amounts of the same excipients in the composition of test like in the reference product.

If a biowaiver is applied for a BCS-class III drug substance excipients have to be qualitatively the same and quantitatively very similar to exclude different effects on membrane transporters.

As a general rule, for both BCS-class I and III drug substances well-established excipients in usual amounts should be employed and possible interactions affecting drug bioavailability and/or solubility characteristics should be considered and discussed. A description on the function of the excipients is required with a justification whether the amount of each excipient is within the normal range. So-called 'active' excipients, like e.g. sorbitol, mannitol, sodium lauryl sulfate or other surfactants, should be identified as well as their possible impact on

- gastrointestinal motility
- susceptibility of interactions with the drug substance (e.g. complexation)
- drug permeability
- interaction with membrane transporters

In cases where critical excipients are relevant the same amount should be used in the test product as in the reference product.

V. Fixed Dose Combinations (FDCs)

BCS-based biowaiver are applicable for immediate release FDC products if all combinational drug substances belong to BCS-class I or III considering specific formulation considerations (see IV.2). Otherwise *in vivo* bioequivalence testing is required.

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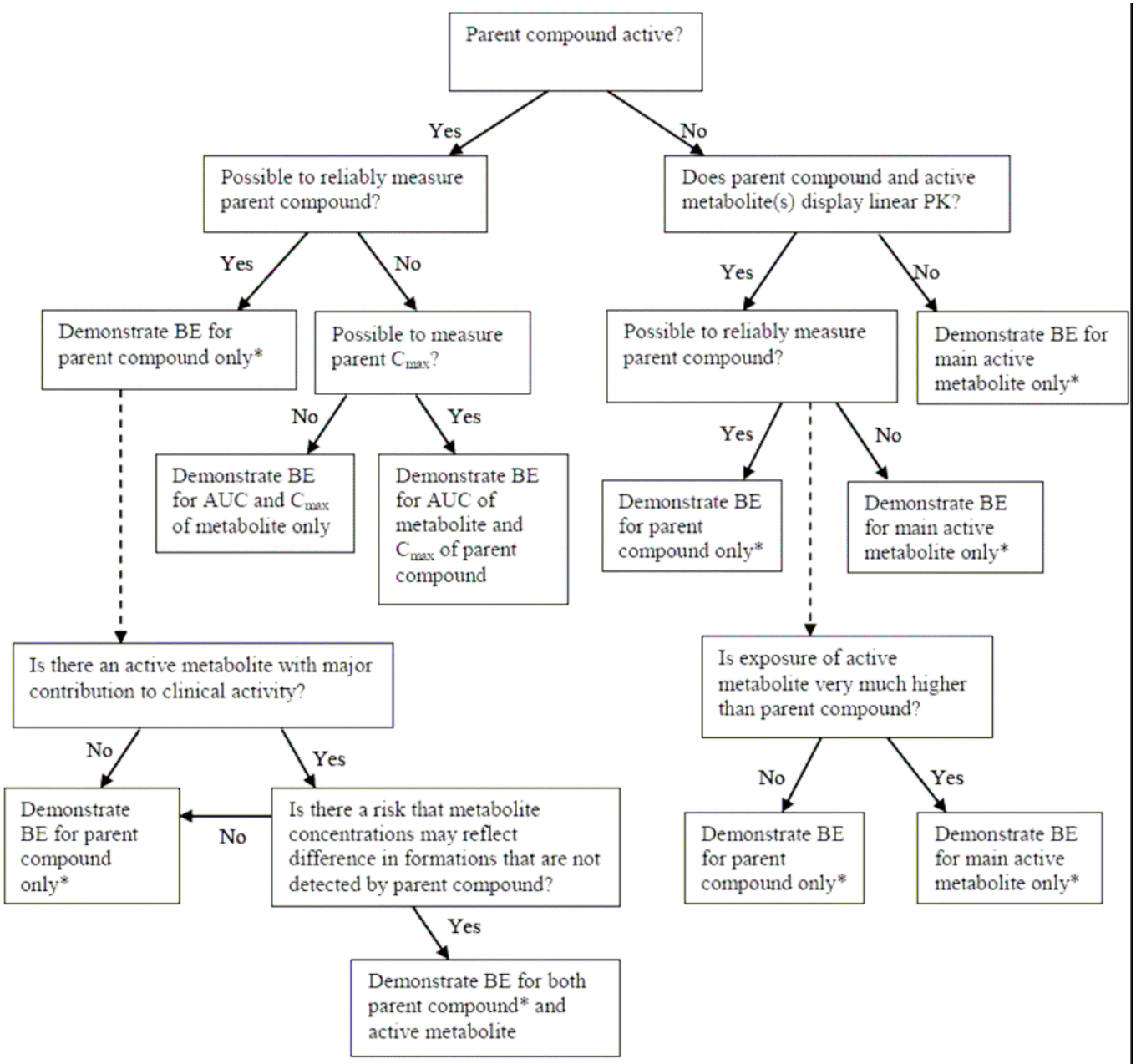
APPENDIX IV

Decision tree on measurement of parent compound or metabolite



*See Appendix V for information on strength and dose to be used in bioequivalence study

APPENDIX V

Decision tree on selection of dose and strength in bioequivalence studies