Notes on the Design of Bioequivalence Study: Lamivudine

Notes on the design of bioequivalence studies with products invited to be submitted to the WHO Prequalification Team – Medicines Assessment Team (PQT/MED) are issued to aid manufacturers with the development of their product dossier. Deviations from the approach suggested below can be considered acceptable if justified by sound scientific evidence.

The current notes should be read and followed in line with the general guidelines of submission of documentation for WHO prequalification. For guidance on issues related to bioequivalence (BE) studies for immediate-release, solid oral dosage forms, see the ICH Harmonised Guideline M13A Bioequivalence for Immediate-Release Solid Oral Dosage Forms (2024). For BE issues outside the scope of the ICH M13A guideline, e.g., for additional strength biowaivers, please consult the "Multisource (generic) pharmaceutical products: guidelines on registration requirements to establish interchangeability" in: *Fifty-seventh report of the WHO Expert Committee on Specifications for Pharmaceutical Preparations,* Geneva, World Health Organization, 2024. WHO Technical Report Series, No. 1052, Annex 8.

Below, additional specific guidance is provided on the invited immediate release products containing lamivudine.

Pharmacokinetics of lamivudine

Maximum lamivudine concentrations are observed in serum within 0.5 to 3.0 hours of dosing in the fasted state (median Tmax of 1 - 1.5 hour). The half-life of lamivudine is 5-7 hours. Co-administration of lamivudine with food results in a delay of Tmax and a lower Cmax (decreased by 47%). However, the extent (based on the AUC) of lamivudine absorption is not influenced. Therefore, lamivudine can be taken with or without food.

Guidance for the design of bioequivalence studies

Taking into account the pharmacokinetic properties of lamivudine the following guidance with regard to the study design should be taken into account:

Design: A crossover design is recommended.

Dose: As the EoI includes lamivudine 150 mg tablets, the bioequivalence study should be conducted on that strength. The bioequivalence study could be waived based on a Biopharmaceutics Classification System (BCS) biowaiver if the corresponding requirements for BCS class III drugs are fulfilled, since lamivudine is considered to be a BCS class III drug (*PQT/MED-specific Annotations for the ICH M9 Guideline for Biopharmaceutics Classification System (BCS)-based Biowaiver Applications*).

For the oral solution or powder for oral solution (50 mg/5ml), the dose of 150 mg should be administered preferably. The bioequivalence study of the oral solution can be waived if the qualitative and quantitative composition of the excipients is similar to that of the comparator, i.e., sucrose 20 % w/v (3 g/15 ml) and propylene glycol. Preservatives (i.e., methyl parahydroxybenzoate and propyl parahydroxybenzoate), buffer agents (i.e. citric acid anhydrous and sodium citrate), and flavours (i.e., artificial strawberry flavour and artificial banana flavour) may differ.

Fasting/fed: As lamivudine can be taken with or without food, a fasted state study is recommended.

Subjects: Healthy adult subjects should be utilized. It is not necessary to include patients in the bioequivalence study.

Parent or metabolite data for assessment of bioequivalence: The parent drug is considered to best reflect the biopharmaceutical quality of the product. Therefore, bioequivalence should be based on the determination of lamivudine.

Sample size: Lamivudine C_{max} seems to be moderately variable (15 - 20% approx.). These data may facilitate the calculation of a sufficient sample size for a cross-over bioequivalence study.

Washout: Taking into account the elimination half-life of lamivudine in healthy volunteers of 5- 7 hours, a washout period of seven days is considered sufficient to prevent carry over.

Blood sampling: The blood sampling should be intensive for the first four hours after administration to properly characterize the C_{max} of lamivudine. It is not necessary to take blood samples beyond 24 hours for the characterization of lamivudine pharmacokinetics. For example, samples can be taken pre-dose and at 0.25, 0.50, 0.67, 0.83, 1.00, 1.25, 1.50, 1.75, 2.00, 2.50, 3.00, 3.50, 4.00, 6.00, 8.00, 12.00, 14.00, 16.00 and 24.00 hours.

Analytical considerations: Information currently available indicates that it is possible to measure lamivudine in human plasma using LC-MS/MS analytical methodology. The bioanalytical method should be sufficiently sensitive to detect concentrations that are 5% of the C_{max} in most profiles of each formulation (test or comparator). See Guideline on bioanalytical method validation and study sample analysis. In: WHO Technical Report Series, No. 1060, Annex 6, or the ICH Harmonised Guideline M10 for more information on bioanalytical recommendations.

Statistical considerations: The data for lamivudine should meet the following bioequivalence standards in a single-dose, crossover design study:

- The 90% confidence interval of the relative mean AUC_{0-t} of the test to reference product should be within 80–125%
- The 90% confidence interval of the relative mean C_{max} of the test to reference product should be within 80–125%.

Biowaiver: A BCS biowaiver for lamivudine is considered a possible alternative to a bioequivalence study, provided the requirements for granting a BCS-based biowaiver are met as outlined in the WHO Guideline "*Biopharmaceutics Classification System-Based Biowaivers*" (TRS1052, Annex 7, 2024) and the PQT/MED guidance "*PQT/MED-specific Annotations for the WHO Guideline on Biopharmaceutics Classification System (BCS)-based Biowaivers*" (2024).