Notes on the Design of Bioequivalence Study: Lamivudine / Dolutegravir

Notes on the design of bioequivalence studies with products invited for submission to the WHO Prequalification Team – Medicines (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 dolutegravir and lamivudine.

Pharmacokinetics of dolutegravir and lamivudine

Maximum dolutegravir concentrations are observed in plasma with a median within 2.0 to 3.0 hours of dosing in the fasted state. The half-life of dolutegravir is 14 hours approximately. Food increased the extent and slowed the rate of absorption of dolutegravir. Bioavailability of dolutegravir depends on meal content: low, moderate, and high fat meals increased dolutegravir AUC0-inf by 33%, 41%, and 66%, increased Cmax by 46%, 52%, and 67%, prolonged Tmax to 3, 4, and 5 hours from 2 hours under fasted conditions, respectively. These increases may be clinically relevant in the presence of certain integrase class resistance. Therefore, dolutegravir is recommended to be taken with food by patients infected with HIV with integrase class resistance.

Maximum lamivudine concentrations are observed in plasma 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 dolutegravir, and lamivudine, the following guidance with regard to the study design should be taken into account.

Study design: A single-dose crossover design is recommended.

Dose: The EoI includes lamivudine/dolutegravir 300 mg/50 mg tablets. The bioequivalence study should be conducted with this strength.

Fasted/fed: The bioequivalence study should be conducted in the fasting state as dolutegravir and lamivudine can be taken irrespective of meals.

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

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

Sample size: Dolutegravir, and lamivudine C_{max} seem to be moderately variable (15 - 25% approx.). These data may facilitate the calculation of a sufficient sample size for a cross-over bioequivalence study.

Washout: Taking into account the largest elimination half-life of these drugs, which corresponds to dolutegravir (approximately 14 hours in healthy volunteers), 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 dolutegravir, and lamivudine. It is not necessary to take blood samples beyond 72 h for the characterization of dolutegravir pharmacokinetics, and 24 hours for lamivudine. For example, samples can be taken pre-dose and at 0.25, 0.50, 0.67, 0.83, 1.00, 1.16, 1.33, 1.50, 1.75, 2.00, 2.25, 2.50, 2.75, 3.00, 3.50, 4.00, 5.00, 6.00, 8.00, 12.00, 16.00, 24.00, 36.00, 48.00, and 72.00 h hours.

Analytical considerations: Information currently available to PQT/MED indicates that it is possible to measure dolutegravir, and 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 Cmax in most profiles of each formulation (test or comparator). The bioanalytical method for each analyte should be validated in the presence of the other analyte if the two components are studies in a single BE study (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).

Statistical considerations: The data for dolutegravir and lamivudine should meet the following bioequivalence standards in a single-dose cross-over design study:

- The 90% confidence interval of the relative mean AUC_{0-t} of the test to comparator product should be within 80.00 – 125.00%
- The 90% confidence interval of the relative mean C_{max} of the test to comparator product should be within 80.00 – 125.00%.